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TEXTBOOK OF ENDOCRINOLOGY



Textbook
OF
ENDOCRINOLOGY
by
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Nobel Prize Laureate 1947

Buenos Aires, Argentina

Second Edition



ACTA ENDOCRINOLÓGICA INC.
MONTREAL, CANADA

331883/

FIRST EDITION

First printing 1947
Second printing 1947
Third printing 1948
Fourth printing 1948
Fifth printing 1948

SECOND EDITION

First printing 1949
Second printing 1949
Third printing 1949

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PRINTED IN CANADA

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DE
L'UNIVERSITÉ DE MONTRÉAL

PREFACE TO FIRST EDITION

La Endocrinología se ocupa del estudio de la acción de las hormonas, en condiciones normales y patológicas, como reguladoras de funciones importantes del organismo. — Las hormonas son sustancias orgánicas producidas específicamente por las glándulas de secreción interna y que una vez descargadas en los líquidos circulantes (medio interno), alcanzan todas las partes del organismo e influyen marcadamente sobre las funciones de determinadas células o sistemas, obrando en pequeñas cantidades y sin contribuir por ellas mismas importantes cantidades de materia o energía. —

Las hormonas regulan importantes procesos químicos y funcionales del organismo, pudiendo modificar probablemente la marcha de reacciones enzimáticas fundamentales, sin que se haya comprobado que entren a formar parte de los sistemas enzimáticos. —

Estamos lejos de las teorías que prevalecieron desde 1880 hasta cerca de 1910, que atribuían el papel de las secreciones internas a una pretendida capacidad de neutralizar o destruir tóxicos del metabolismo intermedio. — Hoy está demostrado que la función de los órganos endocrinos es elaborar y separar hormonas reguladoras de procesos del metabolismo general o el de órganos determinados. — Basta recordar el papel de la insulina en el metabolismo de los hidratos de carbono o el de las paratiroides sobre el metabolismo de calcio o el de la tiroides en el consumo de oxígeno y la calorigenésis. — En muchos casos, las hormonas regulan procesos químicos que rigen la morfogénesis, como lo demuestran el papel

(English Translation of the Adjacent Text Submitted in Spanish by Professor B. A. Houssay of Buenos Aires, Argentina.)

Endocrinology is concerned with the study of the action of hormones; these are regulators of important functions of the organism under normal and pathologic conditions. — Hormones are organic substances specifically produced by the glands of internal secretion; once discharged into the circulating fluids (internal medium), they reach all parts of the organism and exert an important influence upon the functions of certain cells and systems, working in small quantities and without contributing by themselves important amounts of substance or energy. —

The hormones regulate important chemical and functional processes of the organism, being probably capable of modifying the course of fundamental enzymatic reactions; there is no proof however that they themselves participate in enzyme systems. —

We are far from the theories which prevailed from 1880 to 1910, which attributed to the internal secretions the rôle of neutralizing and destroying the toxic substances arising in intermediate metabolism. — It has now been proven that the function of endocrine organs is to elaborate and secrete regulators of general metabolic processes and of specific organs. — It suffices to mention the rôle of insulin in the metabolism of carbohydrates, or that of the parathyroids upon calcium metabolism, or that of the thyroid upon the consumption of oxygen and upon heat production. — In many cases the hormones regulate

de la hipófisis, la tiroides, el testículo o el ovario sobre el crecimiento y la constitución morfológica. — Las acciones hormonales repercuten también intensamente sobre las funciones nerviosas, el comportamiento y el psiquismo; basta recordar la acción que producen las hormonas masculinas o femeninas y la de la castración, la acción de la tiroides o el atiroidismo, etc. —

El papel regulador de las glándulas endocrinas tiene, en muchos casos, una importancia vital. — Así, la falta de las suprarrenales provoca trastornos químicos que llevan a la muerte si no se administran hormonas, sales adecuadas y una dieta apropiada. — Las glándulas endocrinas tienen también influencia sobre ciertos procesos de inmunidad y resistencia del organismo. —

Cada glándula de secreción interna tiene una regulación que mantiene su secreción a un nivel determinado, de acuerdo con las necesidades del organismo. — Así, la hiperglucemia aumenta la secreción de insulina y la hipoglucemia la disminuye, con lo que la glucemia vuelve al nivel normal y también la secreción de insulina. — Pero aunque muchas glándulas endocrinas intervienen en una función determinada (ej.: metabolismo de los hidratos de carbono) hay un equilibrio entre sus secreciones, lo cual asegura la homeostasis del organismo. — Hay entre las glándulas endocrinas acciones reciprocas que las mantienen en equilibrio funcional (ej.: la relación hipofiso-gonadal). —

Por todos estos mecanismos, las glándulas de secreción interna contribuyen poderosamente a asegurar la unidad del organismo, la correlación de sus partes y el equilibrio de sus funciones. —

En algunos casos patológicos se rompe o desvía esta regulación y este equilibrio, ya sea porque un órgano endocrino no es capaz de segregar bastante cantidad de hormona para man-

chemical processes which direct morphogenesis as shown by the rôle of the hypophysis, the thyroid, the testis or the ovary upon growth and morphologic structure. — The hormones also exert intense actions upon the function of the nervous system, behaviour and psychologic processes; it suffices to recall the action exerted by the male or female hormones or by castration and the actions of the thyroid or of thyroid deficiency, etc. —

In many instances the regulating rôle of the endocrine glands is of vital importance. Thus adrenal deficiency causes chemical changes conducive to death unless appropriate hormones, salts and diets are administered. — The endocrine glands also exert an influence upon certain immunologic processes and upon the resistance of the organism. —

Every endocrine gland possesses a regulating mechanism which maintains its secretion at a certain level in accordance with the requirements of the organism. — Thus hyperglycemia augments the secretion of insulin, while hypoglycemia diminishes it so that both the glycemic levels and the secretion of insulin return to normal. — However, although many endocrine glands intervene in a specific function (e.g., carbohydrate metabolism) there is an equilibrium between their secretions, which assures homeostasis of the organism. — There are between the endocrine glands reciprocal interactions which maintain them in a functional equilibrium. (e.g., the hypophyseogonadal interrelation). —

Through all these mechanisms the glands of internal secretion contribute significantly towards the assurance of the unity of the organism, the correlation of its parts and the equilibrium of its function. —

In some pathologic cases this equilibrium is broken or deranged either because one of the organs is unable to secrete a sufficient quantity of hormone to maintain the homeostasis (e.g., hypo-

tener la homeostasis (ej.: hipotiroidismo, diabetes, insuficiencias sexuales, etc.) o bien, en otros casos, porque los órganos endocrinos pierden la capacidad de moderar los aumentos de su secreción para mantener un nivel hormonal normal y entonces se observan síndromes de hiperfunción (ej.: hiperthyroidismo, hipercorticalismo adrenal, hiperinsulinismo, acromegaly, etc.). — Esto se observa principalmente en casos de adenomas de órganos endocrinos. —

Los órganos endocrinos contribuyen a las reacciones del organismo cuando se producen circunstancias de emergencia. — Tal el caso de la hipersecreción de adrenalina en la hipoglucemia o hipotensión o durante la cólera o el terror o el miedo (Cannon). — El papel de los órganos endocrinos en el síndrome de adaptación general del organismo ante factores vulnerantes físicos o químicos o ante excesivas demandas fisiológicas, ha sido brillantemente demostrado por Selye. — Según sus estudios estas reacciones, que al principio son generalmente favorables, pueden luego por su exceso y repetición, llegar a ser desfavorables y producir estados patológicos, tal como la nefroesclerosis, la hipertensión arterial, lesiones miocárdicas, periarteritis, etc., (Selye). —

En ciertos casos patológicos, se observan secreciones internas que producen enfermedades. — Así, el riñón isquemiado vierte en la sangre renina, la cual en presencia de hypertensinógeno produce hypertensina. — Esta última y quizás algún otro mecanismo humoral provoca una hipertensión arterial.

Ciertos desequilibrios hormonales pueden causar tumores benignos de diverso tipo. — Así, los estrógenos en exceso producen adenoma hipofisario, fibróides subperitoneales, metaplasias endometriales, etc., y a la larga favorecen la producción de cánceres de la mama o del útero, etc. —

thyroidism, diabetes, sexual insufficiencies, etc.) or in other cases, because the endocrine organs lose their ability to moderate rises in their hormone secretion in order to maintain a normal hormone level and then hyperfunctional syndromes arise (e.g., hyperthyroidism, adrenal hypercorticalism, hyperinsulinism, acromegaly, etc.). — This is observed mainly in the case of adenomas of endocrine organs. —

The endocrine organs contribute to the reactions of the organism when an emergency situation arises. — This is the case in the event of adrenaline secretion during hypoglycemia or hypotension, or during rage or terror or fear (Cannon). — The rôle of the endocrine organs in the general-adaptation-syndrome of the organism, when confronted with physical or chemical damaging agents or excessive physiological demands, has been brilliantly demonstrated by Selye. — According to his studies these reactions, which in principle are generally favorable, can become damaging due to excess or frequent repetition; then they result in pathologic conditions such as nephrosclerosis, arterial hypertension, myocardial lesions, periarteritis, etc., (Selye). —

In certain pathologic cases internal secretions can produce diseases. — Thus the ischemic kidney discharges renin into the blood, which in the presence of hypertensinogen produces hypertensin. — The latter, and perhaps also some other humoral mechanism, produces arterial hypertension. —

Certain derangements in the hormonal equilibrium may cause benign tumors of various types. — Thus, excesses of estrogens produce hypophyseal adenomas, subperitoneal fibroids, endometrial metaplasia, etc., and in the long run they favor the production of mammary and uterine cancers, etc. —

Endocrinology is a very young branch of the Biological Sciences. — The first facts to be acquired were in

La Endocrinología es una rama muy joven de las Ciencias Biológicas. — Los primeros conocimientos fueron anatómicos; así por ejemplo se distinguieron los caracteres sexuales y las consecuencias de la castración. — Luego la observación clínica mostró la existencia de enfermedades endocrinas, como ser la de Addison, bocio endémico y exoftálmico, mixedema y cretinismo, acromegalia, etc. — Mas tarde, la experimentación fisiológica permitió analizar las insuficiencias debidas a la ablación glandular y el papel de los extractos. — En este siglo, se descubrieron las hormonas por obra de químicos orgánicos, en especial desde hace 20 años, se prepararon muchas sintéticamente y se encontraron sustancias artificiales de acción semejante a ellas. —

La Endocrinología se ha desarrollado y sigue creciendo debido a las investigaciones científicas fundamentales desinteresadas. — Pero a la vez es una ciencia de aplicaciones cada vez más grandes en la Medicina y la Zootecnia, — Ha dado lugar a una industria química y farmacéutica poderosa, que por un lado ha ayudado económica y técnicamente a su adelanto, pero que ha estimulado el empleo excesivo de la hormonoterapia. — El prestigio de la Endocrinología le ha valido una reputación llena de errores y fantasías. — Por eso son necesarios los libros como éste, que exponen en forma precisa y crítica el estado real de los conocimientos. —

La Endocrinología comprende conocimientos anatómicos, fisiológicos, bioquímicos, genéticos, patológicos, clínicos y zootécnicos. — Como ciencia biológica estudia la función de las glándulas endocrinas y el papel de las hormonas en los organismos vivientes. — Como rama de la Medicina procura asegurar la salud, prevenir las enfermedades, diagnosticarlas precozmente para tratar de curarlas o aliviarlas. — Desde que las hormonas regulan pro-

the realm of anatomy; thus, for example, the sexual characteristics and the consequences of castration became known. — Subsequently, clinical observation showed the existence of endocrine diseases such as Addison's disease, endemic and exophthalmic goiters, myxedema and cretinism, acromegaly, etc. — Later, physiologic experimentation made it possible to analyze the insufficiencies due to ablation of glands and the rôle of extracts. — During the present century hormones have been discovered as the result of the work of organic chemists; especially during the last 20 years many of the hormones have been prepared synthetically and artificial substances have been found whose actions were similar to those of the hormones. —

Endocrinology has developed and continues to grow due to fundamental, detached scientific investigations. — However, at the same time it is a science with ever increasing medical and zootechnical applications. — It has given rise to a powerful chemical and pharmaceutic industry, which in turn helped its economic and technical development, but stimulated the excessive employment of hormone therapy. — The prestige of endocrinology has consequently suffered from the ill-repute of many errors and fantasies. — It is for this reason that books, such as the present one, are necessary to outline in a precise and critical form the real status of our knowledge. —

Endocrinology comprises anatomic, physiologic, biochemical, genetic, pathologic, clinical and zootechnical data. — As a biologic science it studies the functions of the endocrine glands and the rôle of the hormones in the living organism. — As a branch of medicine it endeavours to safeguard health, to prevent diseases and to diagnose them at an early stage in order to cure or alleviate them. — Since the hormones regulate fundamental biochemical processes it is evident that every specialist

cesos bioquímicos fundamentales se comprende que todo especialista en Endocrinología debe serlo del metabolismo y que inversamente todo especialista en enfermedades de la Nutrición debe conocer la Endocrinología. — Por ejemplo, la diabetes es una enfermedad del metabolismo por trastorno endocrino (insuficiencia de insulina, etc.). —

Este libro representa una ordenación crítica y concisa de lo más importante que se conoce en el inmenso causal de hechos de la Endocrinología moderna, que es tan vasta que hay especialistas en numerosos problemas parciales y pocos que conozcan todo su campo de estudio. El autor de este libro no ha procurado exponer sólo los hechos acumulados, sino extraer los principios fundamentales que derivan de ellos, recordando que sólo hay Ciencia de lo General y que de tiempo en tiempo son necesarias las grandes síntesis. —

En un mundo tan vasto de conocimientos es imposible que una sola persona domine con igual competencia todos los aspectos de la endocrinología. — Selye reúne para ello condiciones y aptitudes excepcionales y probablemente únicas. Posee la biblioteca endocrinológica más grande del mundo, admirablemente organizada. — Su dominio excepcional de numerosos idiomas le permite comprender el pensamiento propio de diversos países y culturas y evitar el provincialismo tan frecuente aún en las más grandes naciones. — Además, para que el libro sea una exposición objetiva de la Endocrinología contemporánea, el autor ha hecho revisar cada capítulo por algunos de los más sobresalientes expertos en el tema. — Selye es un brillante expositor y tiene el arte de explicar con claridad y método. — Tiene conocimiento personal de la mayor parte de la endocrinología experimental, en sus aspectos anatómicos y fisiológicos, pues ha contribuido a su adelanto con importantes

in endocrinology must also be one of metabolism and that conversely, every specialist in the diseases of nutrition must know endocrinology. — For example, diabetes is a disease of metabolism due to an endocrine lesion (insulin insufficiency, etc.). —

This book represents a critical and concise, orderly presentation of what is most important in the immense collection of facts of modern endocrinology, a science so vast that there are specialists in numerous branch problems and few who know this entire field of study. The author of this book not only succeeded in describing the accumulated facts but also to crystallize those fundamental principles which are derived from them, thus emphasizing that there is only a science of the general and that from time to time the need for a great synthesis arises. —

In such a vast field of knowledge it is impossible that a single person could dominate all aspects of endocrinology with equal competence. — Selye possesses exceptional and probably unique conditions and abilities for this. — He owns the largest endocrinologic library in the world, a collection which is admirably organized. — His exceptional command of numerous languages allows him to understand the characteristic thoughts and cultures of diverse countries and to avoid the provincialism so common even in the greatest nations. — Furthermore, in order to make the book an objective exposition of contemporary endocrinology, the author has submitted each chapter for revision to some of the most eminent authorities in that particular field. — Selye is a brilliant teacher and knows the art of how to explain things clearly and methodically. — He possesses a personal knowledge of the major part of experimental endocrinology, in its anatomic and physiologic aspects; furthermore, he has contributed important original studies, executed with skilful technique, to the development of the science.

estudios originales realizados con hábiles técnicas. — Su erudición excepcional se aprecia en este libro que es una presentación básica y unitaria del problema. — La escasez del espacio le obliga a veces a una exposición algo dogmática y densa en información, pero ésta podrá ampliarse recurriendo a las citas bibliográficas que aconseja el autor. —

El libro es un atlas, además de un texto, pues se incluyen ilustraciones de todo lo que puede ser fotografiado (histología, cristales de hormonas, experimentos, radiografías y casos clínicos).

Este libro, a pesar de sus cualidades de excepción, no obliga a un acatamiento dogmático, pues no expone conocimientos terminados. Su lectura será un punto de partida indispensable para los principiantes y aún para los especialistas; los primeros completarán luego sus conocimientos, cuando sea necesario, en el estudio de los materiales clínicos o experimentales y en trabajos especiales más detallados. —

Es indudable que este libro tendrá una importancia histórica, pues es la síntesis más completa publicada hasta hoy de los conocimientos endocrinológicos actuales y los difundirá con eficacia no igualada. Además estimulará los estudios y promoverá las investigaciones que harán adelantar la Endocrinología. — Como todos los textos no satisfará en cada punto a todos los especialistas y críticos, pero es indudable que este libro tendrá una influencia decisiva para difundir los conocimientos exactos y para promover las investigaciones que harán adelantar la Endocrinología. —

Bernardo A. Houssay

His exceptional erudition can be appreciated in this book which is a basic and unitarian presentation of the problem. — The limitation of space obliges him sometimes to be somewhat dogmatic in his exposition or to condense the information; however, the latter may be expanded by reference to the bibliography recommended by the author. —

In addition to being a text, the book is also an atlas since it contains illustrations of everything that can be photographed (histology, crystals of hormones, experiments, X-rays of clinical cases).

This book, in spite of its exceptional qualities, does not oblige to a dogmatic adherence since it describes an unfinished field of knowledge. Reading of the book will be an indispensable point of departure for beginners as well as for specialists; the former will subsequently complete their knowledge when necessary by the study of clinical and experimental material and of more detailed treatises. —

It is indubitable that this book will possess an historic importance, since it is the most complete synthesis of endocrinologic facts published up to date and it will disseminate these with unequalled efficiency. It will furthermore stimulate studies and promote investigations which will help the progress of endocrinology. — Like all texts it will not satisfy all specialists and critics in every detail; however, it is indubitable that this book will have a decisive influence upon the dissemination of exact data and the promotion of investigations conducive to the progress of endocrinology. —

Bernardo A. Houssay

PREFACE TO SECOND EDITION

In view of the rapid progress made in the field of endocrinology, it has been considered advisable to prepare a second edition as soon as two years after the first. This provided a welcome opportunity to modify certain sections in accordance with suggestions made by many colleagues throughout the world. At the same time, the most important recent discoveries published during the last two years have been included, new important key references have been added and several of the pictures have been replaced by more adequate illustrations. The section "Commercial Hormone Preparations" had to be completely rewritten. In order to avoid any extensive changes in the complex subject index, this was accomplished without change in pagination.

Truly fundamental changes were not necessary and hence, we cannot recommend the book to those who already own the first edition, unless they are particularly anxious to be kept entirely up to date.

HANS SELYE

Université de Montréal
1949

— TABLE OF CONTENTS —

Introduction	1
• GENERAL ENDOCRINOLOGY •	
Definition and Scope of Endocrinology	9
Definition of Endocrines and Hormones	9
The Endocrine Organs	9
The Hormones	11
Delimitation of Endocrinology	12
The Organization of Contemporary Endocrinology	13
Mechanisms of Hormone Actions	17
Prerequisites of Hormone Actions	17
Directly Acting Hormones	17
Indirectly Acting Hormones	17
Conditionally Acting Hormones	18
Mechanism of Direct Hormone Actions upon Target Organs	20
Hormones are Merely Regulators of Biologic Phenomena	20
Are Hormones utilized while exerting their Effects?	20
Stimuli Regulating the Activity of Endocrine Glands	22
Classic Experimental Procedures in Endocrinology	24
Proof of Endocrine Activity of an Organ	24
Extrication Causes Deficiency	25
Organ Extract Causes Overdosage	25
Efficacy of Substitution Therapy	26
Demonstration of Hormone in Venous Blood of Endocrine Organ	26
Demonstration that Hormone Concentration in the Body and Excretions Depends upon the Condition of the Endocrine Organ	26
Isolation of the Pure Hormone	27
Proof that a Synthetic Substance is Identical with the Natural Hormone	27
Principles of Bioassay	27
Technics of Hormone Administration	31
Route of Administration	31
The Solvent	32
Pellets and Suspensions	32
Activation and Inactivation	33
Sensitization and Desensitization	33
Commercial Hormone Preparations	34
The History of Endocrinology	38
References	41

• SPECIAL ENDOCRINOLOGY •

CHAPTER I

THE STEROIDS

Introduction	47
Occurrence and Role of the Steroids in Nature	47
Chemical Terminology and Classification of the Steroid Hormones	50
The Parent Nuclear Hydrocarbons	50
Numeration and Nomenclature of Substituents	51
Isomerism	53
Common Names	53
Pharmacologic Terminology and Classification of the Steroids	58
Definition and Outline	58
Independent Actions	58
Subordinate Actions	59
Potentially Subordinate Actions	61
The Systematic Table of the Steroids	61

Interrelations between the Various Pharmacologic Properties of the Steroids (Pharmacologic Interrelations)	64
Definition and Outline	64
Synergisms and Antagonisms	64
Manifest and Masked Actions	65
Simple and Complex Actions	66
Inhibitible and Non-Inhibitible Actions	68
Selective Inhibition	69
Interrelations between the Pharmacologic Properties and the Chemical Structure of the Steroids (Pharmacо-Chemical Interrelations)	69
Definition and Outline	69
Pharmacо-Chemical Rules Applicable to all or Several Independent Actions	70
Pharmacо-Chemical Rules Within Each Category of Independent Actions	71
The Folliculoids	71
The Corticoids	71
The Luteoids	72
The Testoids	72
The Spermatogenic Steroids	73
The Anesthetic Steroids	73
General Rules Regulating the Actions of Steroid Hormones	74
Compensatory Atrophy	74
Adaptation	74
Dissociated Adaptation	75
Inverse Response	75
Direct and Mediated Actions	75
Biogenesis and Metabolism of the Steroids	75
Biogenesis	75
Metabolic Factors Influencing the Activity of the Steroids	79
References	85

CHAPTER II

THE ADRENALS

Historic Introduction	86
Adrenaline	86
Sympathin	87
Hyperadrenalinism	87
Addison's Disease	88
Corticoids	88
Hypercorticism	89
Normal Morphology	90
Anatomy	90
Histology	90
Comparative Morphology	93
Embryology	94
Theories concerning the Histophysiology of the Adrenals	95
Chemistry of the Adrenals	96
Chemical composition of the Gland	96
Chemistry of the Adrenal Hormones	96
Chemistry of Adrenaline	96
Chemistry of Adrenaline Derivatives (with their chief Biologic Characteristics)	96
Chemistry of Cortical Hormones	98
General Pharmacology of the Adrenal Hormones	99
Standardization	99
Analytic Methods for the Detection of Adrenaline	99
Analytic Methods for the Detection of Corticoids	100
Bioassay of Adrenaline	100
Bioassay of Corticoids	101
Pharmacology of Adrenaline Derivatives	103
Mode of Administration	104
Adrenaline	104
Corticoids	105
Activation and Inactivation	105
Sensitization and Desensitization	107

Theories concerning the Adrenal Hormones	107
Biogenesis of Adrenaline	107
Biogenesis of the Corticoids	108
Fate of Adrenaline in the Body	108
Fate of Corticoids in the Body	110
Mechanism of Adrenaline Action	110
Mechanism of Corticoid Hormone Action	110
Different Kinds of Adrenaline	113
Different Kinds of Corticoids	115
Experimental Physiology of the Adrenals	115
Explantation of the Adrenals	115
Transplantation of the Adrenals	116
Technic of Adrenalectomy	116
Effects of Adrenalectomy and Treatment with Adrenal Hormones	117
Metabolism of Adrenal Hormones (Adrenal Hormone Content of Body Fluids and Tissues)	127
Adrenaline	127
Corticoids	128
Experimental Pathology of the Adrenal (Stimuli Influencing Adrenal Structure)	130
Extirpation of Endocrine Glands	130
Hormones	131
Diseases	131
Diet	133
Nervous Stimuli	135
Age	135
Sex	135
Estrus, Pregnancy and Lactation	135
Seasonal Changes and Hibernation	135
Other Conditions	135
Rays	136
Diseases of the Adrenals	136
Malformations	136
Vascular Disturbances	137
Degenerations	137
Inflammations	137
Hypocorticism (Addison's Disease)	140
Definition	140
Classification	140
Pathologic Anatomy	141
Incidence	142
Pathogenesis	143
Clinical Course	143
Complications	153
Diagnosis	153
Clinical Manifestations of Corticoid Hormone Deficiency	153
Local Signs of a Lesion in the Adrenal Region	153
Specific Adrenal-Cortical Function Tests	154
Recognition of Certain Systemic Diseases Known Frequently to Affect the Adrenals	156
Differential Diagnostic Considerations	156
Prognosis	157
Therapy	158
Treatment of the Underlying Disease Responsible for the Adrenal Destruction	158
Treatment of the Manifestations of Hypocorticism	158
Treatment and Prevention of the Addisonian Crisis	160
Hypercorticism	161
Definition	161
Classification	161
Pathologic Anatomy	162
Simple Hyperplasia of the Adrenal Cortex	162
Enlargement of Accessory Adrenal-Cortical Tissue	163
Cortical Adenomas	163
Cortical Carcinomas	163
Other Diseases	165
Incidence	166
Pathogenesis	166
Clinical Course	167

Complications	182
Diagnosis	182
Clinical Manifestations of the Adrenogenital Syndrome	182
Local Signs of a Lesion in the Adrenal Region	182
Detection of an Excessive Cortical Hormone Production by Bioassay or Chemical Analysis of Urine or Blood	183
Differential Diagnostic Considerations	183
Prognosis	185
Therapy	185
Spontaneous Hypercorticoidism in Animals	186
Hyperadrenalinism	187
Definition	187
Classification	187
Incidence	187
Pathologic Anatomy	187
Pathogenesis	187
Clinical Course	188
Complications	189
Diagnosis	189
Prognosis	189
Therapy	189
Tumors of the Adrenals	190
Definition	190
Classification	190
Pathologic Anatomy	190
Tumors of the Adrenal Cortex	190
Tumors of the Adrenal Medulla	191
Tumors of the Adrenal Stroma	195
Secondary (Metastatic) Tumors of the Adrenals	195
References	195

CHAPTER III

THE HYPOPHYSIS

Historic Introduction	197
Normal Morphology	199
Anatomy	199
Histology	201
Anterior-Lobe	201
The Pars Tuberlalis	201
Intermediate-Lobe	201
Posterior-Lobe	201
The Stroma	203
Comparative Morphology	203
Embryology	204
Theories Concerning the Histophysiology of the Hypophysis	204
Interdependence of the Various Pituitary Cell types and the Adjacent Brain Centers	204
Interrelations Between the Various Cell Types of the Anterior-Lobe	205
Theories concerning the Pathways of Secretion	205
Which Cell Produces which Hormone ?	206
Chemistry of the Hypophysis	208
Chemical Composition of the Gland	208
Classification and Chemistry of the Hypophyseal Hormones	208
Classification of the Anterior-Lobe Hormones	208
A number of actions of impure anterior-hypophyseal extracts have not been proven to be caused by special hormones	210
Chemistry of Anterior-Lobe Hormones	214
Chemistry and Biologic Characteristics of Intermedin	217
Chemistry and Biologic Characteristics of the Posterior-Lobe Hormones	218
General Pharmacology of the Hypophyseal Hormones	219
Standardization	219
Analytic Methods for the Detection of Hypophyseal Hormones	219
Bioassay of the Follicle-Stimulating Hormone	219
Bioassay of the Luteinizing Hormone	220
Bioassay of Comparatively Impure Gonadotrophin Preparations	221
Bioassay of Luteotrophin	222

Bioassay of Corticotrophin	223
Bioassay of Thyrotrophin	224
Bioassay of Somatotrophin	225
Bioassay of Other Hypophyseal Hormones	226
Bioassay of Vasopressin	227
Bioassay of Oxytocin	227
Mode of Administration	228
Sensitization and Desensitization	229
Anterior-Pituitary Hormones	229
Intermedin	229
Posterior-Lobe Hormones	229
Activation and Inactivation	229
FSH and LH	229
Luteotrophin	230
Intermedin and the Posterior-Lobe Hormones	230
Theories Concerning the Hypophyseal Hormones	230
Biogenesis and Metabolism of the Hypophyseal Hormones	230
Mechanism of Hypophyseal Hormone Actions	230
Different Kinds of Hypophyseal Hormones	231
Experimental Physiology of the Hypophysis	231
Explantation of the Hypophysis	231
Transplantation of the Hypophysis	231
Technic of Hypophysectomy	231
Effects of Hypophysectomy and Hypophyseal Hormone Treatment	234
Metabolism of Hypophyseal Hormones (Hypophyseal Hormone Content of Body Fluids and Tissues)	251
Blood and Urine	251
Hypophysis	253
Placenta	254
Plants	254
Experimental Pathology of the Hypophysis (Stimuli Influencing Hypophyseal Structure)	254
Extirpation of Endocrine Glands	254
Hormones	257
Diseases	258
Diet	259
Nervous Stimuli	259
Age	260
Sex	260
Estrus and Menstruation	260
Pregnancy	260
Lactation	260
Season and Hibernation	260
Drugs	260
Rays	262
Diseases of the Hypophysis	262
Malformations	262
Vascular Disturbances	263
Degenerations	263
Inflammations	263
Tumors	264
Adenomas	264
Anterior-Lobe Hypofunction	266
Definition	266
Classification	266
Pathologic Anatomy	266
Incidence	267
Pathogenesis	268
Clinical Course	271
Complications	282
Diagnosis	282
Manifestations Characteristic of Pituitary Lesions in General	282
Simmond's Disease	282
Pituitary Dwarfism	285
Adiposogenital Dystrophy	287
Prognosis	287

Therapy	288.
Simmond's Disease	288.
Pituitary Dwarfism	288.
Adiposogenital Dystrophy	288.
Posterior-Lobe Hypofunction (Diabetes Insipidus)	288.
Definition	288.
Classification	289.
Pathologic Anatomy	289.
Incidence	289.
Pathogenesis	289.
Clinical Course	290.
Diagnosis	290.
Prognosis	294.
Therapy	294.
Anterior-Lobe Hyperfunction	294.
Definition	294.
Classification	295.
Pathologic Anatomy	299.
Incidence	300.
Pathogenesis	301.
Clinical Course	301.
Complications	314.
Diagnosis	314.
Prognosis	316.
Therapy	317.
Posterior-Lobe Hyperfunction	317.
Intermediate Types of Hyperpituitarism	317.
References	318.

CHAPTER IV

THE OVARY

Historic Introduction	320.
Morphology	320.
Removal and Transplantation of the Ovaries	320.
Folliculoids	320.
Luteoids	321.
Gonadotrophins	323.
Ovarian Diseases	323.
Normal Morphology	325.
Anatomy	325.
Histology	326.
The Germinal Epithelium	326.
The Follicles	326.
The Corpus Luteum	329.
Stroma	330.
Blood Vessels	331.
Nerves	331.
Comparative Morphology	335.
Embryology	335.
Theories Concerning the Histophysiology of the Ovaries	338.
The Mechanism of Ovulation	338.
Migration of the Ovum	338.
The Law of Constant Numbers in Ovulation	338.
Which cell Produces which Hormone ?	339.
Chemistry of the Ovaries	340.
Chemical Composition of the Gland	340.
Chemistry of the Ovarian Hormones	341.
General Pharmacology of the Ovarian Hormones	341.
Standardization	341.
Analytic Methods for the Detection of Folliculoids	341.
Analytic Methods for the Detection of Luteoids	342.
Bioassay of Folliculoids	343.
Bioassay of Luteoids	345.

Pharmacology of Ovarian Hormone Derivatives and of Artificial Ovarian Hormones	347
Folliculoids and Artificial Folliculoids	347
Luteoids	349
Mode of Administration and Chief Indications	350
Folliculoids	350
Luteoids	352
Withdrawal Effects and Permanent Changes Caused by Temporary Treatment	353
Other Pharmacologic Problems	353
Experimental Physiology of the Ovaries	353
Explantation of the Ovaries	353
Transplantation of the Ovaries	354
Technic of Ovariectomy	355
Effects of Ovariectomy and Treatment with Ovarian Hormones	356
Metabolism of the Ovarian Hormones (Ovarian Hormone Content of Body Fluids and Tissues)	372
Occurrence of Folliculoids in Various Body Tissues and Fluids	373
Experimental Pathology of the Ovary (Stimuli Influencing Ovarian Structure)	374
Extrication of Endocrine Glands	374
Hormones	377
Diseases	380
Diet	381
Nervous Stimuli	381
Age	382
Constitution, Race and Heredity	384
Sex	384
Pregnancy and Lactation	384
Season	384
Rays	384
Other Stimuli	385
Ovarian Diseases in General	385
Definition	385
Classification	385
Pathogenesis	386
Clinical Course	386
Complications	392
Diagnosis	392
Therapy	396
Hypogonadism	396
Hyperfolliculoidism	397
Hyperluteoidism	397
Dysmenorrhea	397
Hypermenorrhea (or Menorrhagia)	397
Intermenstrual Bleeding	398
Chronic Cystic Mastitis	398
Vaginitis	398
Sterility and Infertility	398
Special Diseases of the Ovary	399
Malformations and Anomalies	399
Aplasia (or Agenesis)	399
Hypoplasia of the Ovaries	400
Accessory Ovaries	400
Hyperplasia and Hypertrophy	402
Primary Ectopia of the Ovary	402
Ambisexuality (Hermaphroditism and Pseudohermaphroditism)	403
Rare Malformations and Anomalies of the Ovaries	406
Hernia and Prolapse (Secondary Ectopia) of the Ovary	406
Torsion of the Ovary	407
Retrogressive Changes	407
Vascular Disturbances	408
Inflammations	408
Diseases of the Sexual Cycle	409
Definition	409
Diseases of the Pubertal Period	409
Diseases of the Menopause	412
Derangements in the Correlation between Ovulation and Menstruation or Estrus	414
Vicarious Menstruation	415

Ovarian Tumors in General	415
Definition	415
Classification	416
Pathologic Anatomy	416
Incidence	416
Pathogenesis	417
Clinical Course	417
Complications	417
Diagnosis	419
Differential Diagnosis	419
Prognosis	419
Therapy	419
Para-Ovarian Tumors in General	420
Hyperfolliculoidism Associated with Ovarian Growths	
(Follicle Cysts, Small-Cystic Degeneration of the Ovaries, Folliculomas)	420
Definition	420
Pathologic Anatomy	421
Incidence	425
Pathogenesis	426
Clinical Course	427
Diagnosis	430
Prognosis	431
Therapy	431
Follicle Cysts	431
Metropathia Hemorrhagica	431
Ovarian Small-Cystic Degeneration	433
Ovarian Folliculomas	433
Hyperluteoidism and Corpus Luteum Cysts	433
Definition	433
Pathologic Anatomy	434
Incidence	434
Pathogenesis	435
Clinical Course	435
Complications	436
Diagnosis	436
Prognosis	436
Therapy	436
"Hypernephromas," "Luteomas" and Other "Lipid Cell Tumors" of the Ovary	437
Definition	437
Pathologic Anatomy	437
Incidence	439
Pathogenesis	439
Clinical Course	440
Diagnosis	440
Prognosis and Therapy	440
Testoid Hyperthecosis and Leydig Cell Tumors of the Ovary	440
Ovarian Tubular Adenomas (Arrhenoblastomas)	
Classification	441
Pathologic Anatomy	442
Incidence	445
Pathogenesis	445
Clinical Course	446
Diagnosis	447
Prognosis	448
Therapy	448
Ovarian Seminomas (Dysgerminomas) or Embryonic Carcinomas of the Ovary	449
Definition	449
Pathologic Anatomy	449
Incidence	450
Pathogenesis	450
Clinical Course	450
Diagnosis	451
Prognosis	451
Therapy	451
Struma Ovarii	451
Definition	451
Pathologic Anatomy	452
Incidence	452

TABLE OF CONTENTS

XXIII

Pathogenesis	452
Clinical Course	453
Diagnosis	453
Therapy	453
Ovarian Chorionepitheliomas (Chorioncarcinomas?)	453
Definition	453
Classification	453
Pathologic Anatomy	454
Incidence	454
Pathogenesis	454
Clinical Course and Complications	455
Diagnosis	455
Prognosis and Therapy	455
Ovarian Common Cysts	456
Ovarian Common Carcinomas	461
Ovarian Mesonephromas	463
Brenner Tumors or Brenneromas	463
Teratoid Tumors (Dermoids, Solid Teratomas, Embryomas)	463
Non-Epithelial Neoplasms Devoid of Endocrine Function	467
Endometriosis	467
Definition	467
Classification	467
Pathologic Anatomy	469
Incidence	470
Pathogenesis	470
Clinical Course and Complications	472
Diagnosis	472
Therapy	474
References	475

CHAPTER V

THE PANCREAS

Historic Introduction	478
Diabetes Mellitus	478
Insulin	478
Hypophyseal and Adrenal Diabetes	480
Hyperinsulinism	480
Normal Morphology	480
Anatomy	480
Histology	481
Comparative Morphology	482
Embryology	482
Theories Concerning the Histophysiology of the Pancreas	483
Chemistry of the Pancreas	484
Chemical Composition of the Gland	484
Chemistry of Insulin	484
General Pharmacology of Insulin	485
Standardization	485
Chemical Methods	485
Bioassay	485
Pharmacology of Special Insulin Preparations	486
Mode of Administration	486
Sensitization and Desensitization	486
Theories Concerning Insulin	486
Biogenesis	486
Fate of Insulin in the Body	486
Mechanism of Insulin Action	487
Different Kinds of Pancreatic Hormones	489
Experimental Physiology of the Pancreas	489
Explantation of the Pancreas	489
Transplantation of the Pancreas	489
Technic of Pancreatectomy	489
Effect of Pancreatectomy and Insulin Treatment	490
Metabolism of Pancreatic Hormone (Insulin Content of Body Fluids and Tissues)	499

Experimental Pathology of the Pancreas (Stimuli Influencing Pancreatic Structure)	501
Extrication of Endocrine Glands	501
Hormones	502
Diseases	504
Diet	505
Nervous Stimuli	505
Age	505
Pregnancy	505
Drugs	505
Rays	505
Diseases of the Pancreas	507
Malformations	507
Degenerations	507
Inflammations	507
Acute Hemorrhagic Pancreatitis and Pancreatic Necrosis	507
Acute Interstitial Pancreatitis	507
Chronic Pancreatitis	508
Diabetes Mellitus	508
Terminology	508
Definition	508
Classification	508
Pathologic Anatomy	509
Incidence	511
Pathogenesis	512
Clinical Course	513
Complications	524
Diagnosis	525
Physical Findings	525
Laboratory Findings	525
Differential Diagnosis	525
Prognosis	527
Therapy	527
Prevention	527
Diet	527
Insulin	528
Procedure of Standardization	530
Diabetic Coma	531
Insulin Substitutes	532
Therapy of Complications	532
Spontaneous Diabetes Mellitus in Animals	532
Hyperinsulinism	532
Terminology	532
Definition	532
Classification	532
Pathologic Anatomy and Pathogenesis	533
Incidence	534
Clinical Course and Complications	534
Diagnosis	535
Prognosis	535
Therapy	535
Tumors of the Pancreas	536
References	536

CHAPTER VI

THE PARATHYROIDS

Historic Introduction	538
Normal Morphology	539
Anatomy	539
Histology	540
Comparative Morphology	541
Embryology	542
Theories Concerning the Histophysiology of the Parathyroids	542
Chemistry of the Parathyroids	543
Chemical Composition of the Glands	543
Chemistry of the Parathyroid Hormone	543

General Pharmacology of the Parathyroid Hormone	543
Standardization	543
Analytic Methods	543
Bioassay	543
Mode of Administration	544
Sensitization and Desensitization	544
Theories Concerning the Parathyroid Hormone	544
Biogenesis	544
Fate of Parathyroid Hormone in the Body	544
Mechanism of Parathyroid Hormone Action	544
Different Kinds of Parathyroid Hormones	547
Experimental Physiology of the Parathyroids	547
Explantation and Transplantation of the Parathyroids	547
Technic of Parathyroidectomy	547
Effects of Parathyroidectomy and Parathyroid Hormone Treatment	547
Metabolism of Parathyroid Hormone (Parathyroid Hormone content of Body Fluids and Tissues)	557
Experimental Pathology of the Parathyroids (Stimuli Influencing the Parathyroid Structure)	558
Extirpation of Endocrine Glands	558
Hormones	559
Diseases	559
Diet	560
Nervous Stimuli	560
Age	560
Sex	560
Sexual Cycle	560
Pregnancy and Lactation	560
Seasons	560
Drugs	560
Rays	561
Diseases of the Parathyroids	561
Malformations	561
Vascular Disturbances	561
Degenerations	561
Inflammations	562
Hypoparathyroidism	562
Definition	562
Classification	562
Pathologic Anatomy	563
Incidence	563
Pathogenesis	563
Clinical Course	564
Complications	568
Diagnosis	568
Prognosis	569
Therapy	570
Prophylaxis	570
Internal Therapy	572
Parathyroid Transplantation	573
Hypoparathyroidism in Animals	573
Hyperparathyroidism	574
Definition	574
Classification	574
Pathologic Anatomy	575
Incidence	575
Pathogenesis	575
Clinical Course	576
Complications	580
Prognosis	580
Diagnosis	580
Therapy	587
Operative Therapy	587
Pre- and postoperative Therapy	588
Hyperparathyroidism in Animals	588
Tumors of the Parathyroids	589
Definition	589
Classification	589

Pathologic Anatomy	589
(A) Cysts	589
(B) Adenomas	589
(C) Carcinomas	589
(D) Mesenchymal Tumors	590
Clinical Course	590
Diagnosis	590
Therapy	590
References	592

CHAPTER VII

THE PINEAL

Historic Introduction	593
Diseases	593
Normal Morphology	593
Anatomy	593
Histology	593
Comparative Morphology	594
Embryology	594
Theories Concerning the Histophysiology of the Pineal	594
Diseases of the Pineal Body	595
Pathologic Anatomy	595
Incidence	595
Pathogenesis	596
Clinical Course	597
Complications	598
Diagnosis	598
Prognosis	598
Therapy	598
References	599

CHAPTER VIII

THE TESTIS

Historic Introduction	600
Morphology	600
Experimental Physiology	600
Diseases of the Testis	601
Normal Morphology	602
Anatomy	602
Histology	603
The Capsule	603
The Seminiferous Tubules	604
The Stroma	606
The Duct System	606
Comparative Morphology	607
Embryology	607
Theories Concerning the Histophysiology of the Testis	609
Interrelations between the Various Cell Types	609
The Pathways of Testis Hormone Secretion	609
Which Cell Produces the Testis Hormone ?	609
Chemistry of the Testis	610
Chemical Composition of the Gland	610
Chemistry of the Testis Hormones	611
General Pharmacology of the Testis Hormones	611
Standardization	611
Analytic Methods for the Detection of Testoids	611
Bioassay of Testoids	611
Pharmacology of Testis Hormone Derivatives and Artificial Testoids	612
Mode of Administration and Chief Indications	613
Withdrawal Effects and Permanent Changes Caused by Temporary Treatment	614
Other Pharmacologic Problems and Theories Concerning the Testis Hormones	614

Experimental Physiology of the Testis	615
Explantation of the Testis	615
Transplantation of the Testis	615
Technic of Orchidectomy	616
Effects of Orchidectomy and Treatment with Testis Hormones	617
Pregnancy	626
Lactation	626
Hibernation	626
Regeneration and Rejuvenation	626
Tumorigenesis	627
Metabolism of Testis Hormones (Testis Hormone Content of Body Fluids and Tissues)	627
Biogenesis and Fate of the Testis Hormone	627
Occurrence of Testoids and other 17-KS in Body Fluids and Tissues	628
Experimental Pathology of the Testis (Stimuli Influencing Testis Structure)	630
Extirpation of Endocrine Glands	630
Hormones	635
Diseases	638
Diet	639
Nervous Stimuli	640
Age	640
Constitution, Race, Heredity	641
Sexual Intercourse	641
Season	641
Parabiosis	641
Temperature	641
Muscular Work	642
Rays	642
Other Agents	643
Diseases of the Testis	643
Malformations	643
Atrophy, Hypertrophy and Hyperplasia	644
Hemorrhages and other Vascular Lesions	644
Degenerations	645
Inflammations	645
Male Hypogonadism	645
Definition	645
Classification	645
Early Eunuchism	646
Late Eunuchism	646
Hypergonadotrophic Eunuchoidism without a-Leydigism	646
Early Hypogonadotrophic Eunuchoidism	646
Late Hypogonadotrophic Eunuchoidism	648
Simple Delayed Puberty	648
Sterility Without Eunuchoidism	648
The Male Climacteric	649
Pathologic Anatomy	649
Incidence	649
Pathogenesis	649
Clinical Course	650
Diagnosis	663
Prognosis	665
Therapy	665
Hypogonadism in Animals	666
Male Hypergonadism	666
Definition	666
Classification	666
Pathologic Anatomy	668
Incidence	668
Pathogenesis	668
Clinical Course	669
Diagnosis	671
Prognosis and Therapy	671
Hypergonadism in Animals	671
Testis Tumors in General	672
Embryomas	672
Embryoid or Mixed Tumors	672
"False Seminomas" (Embryonal Carcinomas with Lymphoid Stroma, Dysgerminomas, Goniomas)	672

True Seminomas	674
Chorionepitheliomas	675
Adrenal-Cortical Tumors	675
Adenocarcinomas	675
Adenomas	676
Other Testicular Tumors	676
References	676

CHAPTER IX

THE THYMUS

Historic Introduction	678
Diseases	678
Experimental Physiology	679
Normal Morphology	679
Anatomy	679
Histology	680
Comparative Morphology	681
Embryology	681
Theories Concerning the Histophysiology of the Thymus	681
Chemistry of the Thymus	682
Experimental Physiology of the Thymus	682
Explantation and Transplantation of the Thymus	682
Technic of Thymectomy	682
Effects of Thymectomy and Thymus Extract Injections	683
Experimental Pathology of the Thymus (Stimuli Influencing Thymus Structure)	683
Extirpation of Endocrine Glands	683
Hormones	684
Diseases	684
Diet	684
Age	684
Sex	684
Season, Hibernation	684
Estrus, Puberty, Pregnancy and Lactation	684
Other Conditions	685
Diseases of the Thymus	685
Clinical Manifestations	687
Treatment	688
References	688

CHAPTER X

THE THYROID

Historic Introduction	690
Simple Goiter	690
Hypothyroidism	690
Hyperthyroidism	690
Thyroid Hormone	691
Anti-Thyroid Drugs	692
Normal Morphology	692
Anatomy	692
Histology	692
Comparative Morphology	695
Embryology	695
Theories Concerning the Histophysiology of the Thyroid	695
Chemistry of the Thyroid	696
Chemical Composition of the Gland	696
Chemistry of the Thyroid Hormone	696
General Pharmacology of the Thyroid Hormone	697
Standardization	697
Analytic Methods	697
Bioassay	697
Pharmacology of Thyroid Hormone Derivatives	697
Mode of Administration	698
Sensitization and Desensitization	698

Theories Concerning the Thyroid Hormone	698
Biogenesis	698
Fate of Thyroid Hormone in the Body	699
Mechanism of Thyroid Hormone Action	699
Different Kinds of Thyroid Hormones	699
Experimental Physiology of the Thyroid	700
Explantation of the Thyroid	700
Transplantation of the Thyroid	700
Technic of Thyroidectomy	700
Effects of Thyroidectomy and Thyroid Hormone Treatment	700
Metabolism of Thyroid Hormone (Thyroid Hormone Content of Body Fluids and Tissues)	705
Experimental Pathology of the Thyroid (Stimuli Influencing Thyroid Structure)	707
Extirpation of Endocrine Glands	707
Hormones	708
Diseases	708
Diet	708
Nervous Stimuli	709
Age	709
Sexual Cycle	710
Pregnancy	710
Metamorphosis	710
Moultting	710
Hibernation	710
Temperature Changes	710
Drugs	710
Rays	711
Diseases of the Thyroid	711
Malformations	711
Degenerations	712
Amyloid Goiter	712
Inflammations	712
Acute Thyroiditis	712
Chronic Thyroiditis	712
Goiter in General	715
Simple Goiter	715
Definition	715
Classification	715
Pathologic Anatomy	716
Pathogenesis	717
Incidence	717
Clinical Course	717
Complications	718
Diagnosis	718
Prognosis	718
Therapy	718
Hypothyroidism	720
Definition	720
Classification	720
Pathologic Anatomy	720
Incidence	722
Pathogenesis	723
Clinical Course	724
Complications	739
Diagnosis	739
Prognosis	741
Therapy	741
Spontaneous Hypothyroidism in Animals	742
Hyperthyroidism	742
Definition	742
Classification	742
Pathologic Anatomy	744
Incidence	746
Pathogenesis	747
Clinical Course	747
Complications	760
Diagnosis	761
Prognosis	761

Therapy	762
Exclusively Medical Treatment	762
Surgical Treatment	762
X-Ray Treatment	769
Spontaneous Hyperthyroidism in Animals	769
Tumors of the Thyroid	769
Definition	769
Classification	769
Pathologic Anatomy	769
(A) Adenomas	769
(B) Carcinomas	771
(C) Sarcomas	772
(D) Teratomas	773
(E) Secondary (or Metastatic) Tumors	773
Incidence	773
Pathogenesis	773
Clinical Course	773
Diagnosis	774
Prognosis	776
Therapy	776
References	776

CHAPTER XI

HORMONES AND HORMONE-LIKE SUBSTANCES PRODUCED BY SIMPLE ENDO-EXOCRINE AND NON-GLANDULAR ORGANS

Renal Hormones	778
Historic Introduction	778
Methods	778
Chemical Considerations Concerning the Origin of Renal Hypertension	779
Theories Concerning the source of Renal Pressor Hormone	779
Stimuli Influencing the Course of Renal Hypertension	780
Chemical Implications of the Renal Pressor Mechanism	781
References	782
Hormones of the Gastrointestinal Tract	783
Introduction	783
Secretin	784
Enterogastrone	784
Urogastrone	785
Anthelone	785
Gastrin	785
Duodenin	786
Incretin	786
Other Blood-Sugar-Depressing Duodenal Extracts	787
Villikinin	787
Enterocrinin	787
Cholecystokinin	787
Hormones of the Salivary Glands	788
Other Gastrointestinal Hormones	788
References	788
Cardiac Hormones	788
Reference	788
Depressor Substances of Tissues	789
Histamine	789
Acetylcholine	789
Kallikrein	791
Angioxyl	792
Vagotonin	792
Adenosine and Derivatives	792
References	792
Hepatic Hormones	793
The Antianemic Principle	793
Pernicious anemia	793
The Antianemic Factor of Liver	793
Folic Acid	794
The Antianemic Principle of the Stomach	794

Heparin	795
Yakriton	795
Hormones in Invertebrates	796
Plant Hormones	797
Humoral Factors in Inflammation and Wound Healing	797
CHAPTER XII	
CORRELATIONS	
Hibernation and Aestivation	798
Hibernation	798
Aestivation	799
References	799
Puberty, Menarche, Adolescence	799
Definition	799
Course	799
State	799
Growth and Bone Changes	800
Secondary Sexual Characteristics	800
Factors Influencing the development of Puberty	801
Mechanism and Significance of Puberty	801
Diseases of the Puberty Period	801
References	801
The Sexual Cycle	802
Definition	802
Course	802
Duration	802
Metabolism	803
Serologic Changes	803
Blood	803
Nervous System	803
Accessory-Sex-Organs	803
Hormone Metabolism During the Sexual Cycle	811
Stimuli Influencing the Sexual Cycle	812
Mechanism and Significance of the Sexual Cycle	812
Diseases of the Sexual Cycle	813
References	813
Pseudopregnancy	813
Definition	813
Course	813
Hormone Metabolism during Pseudopregnancy	814
Stimuli Influencing Pseudopregnancy	814
Mechanism and Significance of Pseudopregnancy	815
References	815
Pregnancy	815
Definition	815
Course	815
Duration of Pregnancy	815
Fertilization	816
Implantation	816
Metabolic and Organ Changes	820
Hormone Metabolism during Pregnancy	820
Stimuli Influencing Pregnancy	820
Mechanism and Significance of the Pregnancy Changes	823
Diseases of Pregnancy	825
Ectopic Pregnancy	825
Habitual Abortion	826
Hydatidiform Mole and Chorioneopithelioma	826
Hyperemesis gravidarum	826
Pre-Eclampsia and Eclampsia	826
Other Diseases	827
References	827
Lactation	827
Definition	827
Course	828
Hormone Metabolism during Lactation	828
Stimuli Influencing Lactation	828

Neuro-humoral Correlations during Lactation	829
Diseases of Lactation	832
References	832
Antihormones and Other Types of Acquired Resistance to Hormones	833
Historic Introduction	833
The Antihormones	833
Other Types of Acquired Resistance to Hormones	835
Prohormones	836
References	836
The General-Adaptation-Syndrome and the Diseases of Adaptation	837
Definitions and Terminology	837
History	838
Course of the General-Adaptation-Syndrome	840
Metabolism	840
Carbohydrate Metabolism	840
Lipid Metabolism	843
Nitrogen Metabolism	843
Mineral Metabolism	843
Other Metabolites	844
Functional and Morphologic Changes during the General-Adaptation-Syndrome	844
State	844
Growth and Bones	845
Blood	845
Cardiovascular System	846
Lymphatic System	848
Respiratory System	850
Digestive System	850
Urinary System	851
Endocrine Organs	852
Theoretic Interpretation of the General-Adaptation-Syndrome	855
Theories Concerning the Shock-Phase	855
Theories Concerning the Entire General-Adaptation-Syndrome	856
Clinical Implications of the General-Adaptation-Syndrome	858
Hypertension and "Hypertensive diseases"	858
Periarteritis Nodosa	860
Nephrosclerosis	860
Nephritis	863
Rheumatic Diseases	863
Waterhouse-Friedrichsen Syndrome	863
Eclampsia	863
"Accidental Thymus Involution"	863
Appendicitis	863
Tonsillitis	863
Gout	864
Diabetes mellitus	864
Cushing's Syndrome	864
Secondary Shock	864
Gastrointestinal Ulcers	864
Addison's Disease and Simmonds' Disease	864
Primary Renal Disease	864
Conclusions	865
Diseases of Adaptation due to Endocrine Disturbances	866
References	866
Pluriglandular Insufficiency and Allied Conditions	868
General Survey of Hormonal Correlations	869



INTRODUCTION

The purpose of this volume is to act as a standard textbook of vertebrate endocrinology.

The history of medicine shows that whenever a great deal of progress is made in one particular field, individuals (specialists) and books (textbooks and encyclopedic surveys) emerge, with the aim of correlating all the data pertaining to this new subject. These individuals and books depend upon each other. Only a highly specialized physician can write a book concerning an entire branch of science, but, conversely, such books are essential in acquiring the specialist's knowledge. Therefore the work of several generations is usually necessary before a body of data is gradually molded into a recognized separate branch of science.

During the period of its development it is of the greatest importance to delimit the new science from other fields of knowledge, so that it comprises all those facts which lend themselves particularly well for conjoint study by the same individual. The inability of the human mind to master more than a limited field of knowledge is the only justification for the development of specialties and specialists.

A survey of the medical sciences clearly indicates how time-taking it is to draw precise and logical borderlines for a specialty. Anatomy and, to a somewhat lesser extent, its younger sister science histology, are perhaps the best examples of old and well-established specialties, whose contents are clearly outlined by our traditions, based on generations of experience. As a result of this, the anatomists and anatomy books of all countries agree surprisingly well on the data which are pertinent to this science, as well as on the classification and order of presentation of relevant material for didactic purposes. The pertinent facts are either arranged according to organ systems or according to their topographic relations. We do not even think of any other possible classification, although *a priori*, presentation of organs and tissues according to similarities in their chemical composition (water, fat, protein, carbohydrate content, etc.); embryologic development (ectodermal, mesodermal, entodermal organs) and many other points of view might appear to be equally satisfactory. The outlines of histology or pathologic anatomy, representing somewhat younger sciences, are slightly less clearly defined by tradition than those of anatomy. In all these relatively old morphologic sciences, however, the delimitation from other branches of medicine is so generally accepted that most contemporary schools closely agree on the type of routine training they should expect their morphologists to possess and to transmit to their students.

Such standardization of outlines and contents becomes increasingly more difficult with the sciences which are still in the process of active growth. Internal medicine, pediatrics, gynecology and obstetrics for instance, are fairly well standardized specialties, while dietetics, biochemistry, public health and social medicine still expand so rapidly that the specialists and specialized books con-

cerned with these subjects vary greatly in their interpretation of what is relevant to these disciplines.

Endocrinology is one of the youngest and most rapidly expanding fields of medicine; hence, there are comparatively few books which attempt to survey the entire subject and those which exist, exhibit marked differences in their conception of this field. This is partly the cause and partly the result of the fact that in our time there still are only very few professional endocrinologists. There are physicians specializing more or less exclusively in the treatment of diabetes, thyroid diseases, gynecologic endocrinology or even in all of clinical endocrinology, but their interest in the anatomy, embryology or pathologic anatomy of the endocrines is usually limited. There are excellent chemists who have done a great deal to further our understanding of the chemical structure of hormones and to supply us with the many synthetic hormone preparations now available. However, most of them consider the morphologic and clinical aspects of endocrinology entirely outside their field. Zoologists, physiologists, pharmacologists or biochemists may take up one or the other endocrine research problem; and perhaps even teach an endocrinology course at a university, but in the absence of comprehensive standard books and post-graduate courses, they find it almost impossible to acquire the training necessary to master the whole field.

During the sixteen years in which I have taught endocrinology, first at McGill University, and more recently at the Université de Montréal, I have learned that there is a great demand for "professional" endocrinologists to supplant the "amateurs" of this science in universities, hospitals, the pharmaceutical industry, etc. Specialists with a rounded general training in all problems pertaining to the hormones would be singularly well-prepared for the teaching and clinical practice of endocrinology, as well as for investigative work in any of its branches. Our teaching institutions would not think of acknowledging the competence of a surgeon, a radiologist or a dermatologist who has not had the benefit of special post-graduate training in his field. For general medicine, chemistry, physics, etc., we even supply standardized undergraduate courses. However, anyone may call himself an endocrinologist, because no school has organized a systematic post-graduate training schedule for those who wish to become specialists in this subject.

The present textbook is one of three tasks undertaken by our Institute in an attempt to delimit and systematize the field of endocrinology and to help those who wish to specialize in this science.

(1) We have compiled a library containing an almost complete set of all original articles and books dealing with the hormones. This collection which now comprises about 250,000 entries (reprints, books, microfilms, photostats, abstract cards, etc.) serves as a basis for the publication of the "*ENCYCLOPEDIA OF ENDOCRINOLOGY*," which, it is hoped, will ultimately act as a critical survey and a complete guide to the entire endocrinologic literature. Up to now, six volumes of this treatise have appeared in print, and the classification of the literature as well as a large part of the remaining manuscript have been completed for future publication. This treatise is designed mainly as a research tool for the specialist and original investigator.

(2) The present "*TEXTBOOK OF ENDOCRINOLOGY*" represents a miniature of the Encyclopedia, in the form of a concise, and we hope, balanced summary of the most important and best-established facts concerning all branches of our science. It is designed primarily for the medical student and physician,

but also for specialists in endocrinology or in a more general subject (zoölogy, biochemistry, physiology, pathology, etc.) in which endocrinology represents a cognate science.

(3) We have planned a SYSTEMATIC POST-GRADUATE COURSE in endocrinology, which is now given at this Institute. The object of this course — which leads to the Ph.D. degree — is to give both theoretical and practical training in the entire field of endocrinology. In addition to supervised research and post-graduate courses the candidates are required to take a "rotating internship" in the five sections of the Institute (Experimental Surgery, Pharmacology, Biochemistry and Nutrition, General Physiology and Experimental Morphology). Thus they are given an opportunity to familiarize themselves, at least to some extent, with the interest and technics of the investigators in charge of these divisions. After surveying comparable courses in other Universities, it appeared to us that although many centers give excellent post-graduate tuition in certain branches of endocrinology, few, if any, have attempted to offer a systematic post-graduate course covering the entire subject. Hence, we hope that experimentation with such a course in our school may help other centers, if not to emulate our methods, at least to avoid our errors.

But let us return to this textbook. Some readers will perhaps deplore the paucity of references. It is true that only a few outstanding monographs and reviews are quoted at the end of each chapter, but these act as a guide to a large number of pertinent original articles. It is our experience that readers of textbooks rarely consult original articles cited in support of individual statements in the text. If references are given to prove specific points, it is indispensable to quote the entire pertinent literature as we attempted to do in the Encyclopedia of Endocrinology. Indeed, if only selected references are quoted — as is generally the case in textbooks — they tend to mislead the reader into believing there are no other, perhaps contradictory, data in the relevant literature. Such references may help to shift the responsibility from the writer of the textbook to the author of the original article, but they merely mislead those who do not have the time to make a detailed study of each question.

In perusing textbooks dealing with various branches of medicine, it is striking that the number of references quoted is so often inversely proportional to the degree of certainty with which the author feels competent to discuss the matter. Almost no references are given in textbooks of anatomy or histology, and even elementary treatises of internal medicine rarely cite much of the original literature on which the discussions are based. It is unnecessary to quote the authorities who described the branches of the internal carotid artery, the chromaffinity of adrenal medulla cells, the branching of heart muscle fibers, or the classic symptoms and signs of lobar pneumonia. Everybody agrees on these points and the author of the textbook is quite prepared to accept the responsibility for them. On the other hand, it is convenient to quote others when we discuss the biogenesis of the steroid hormones, or the mechanism of parathyroid-hormone action since pertinent theories are likely to be incorrect.

I am quite prepared to state at the onset that the views expressed in this book are mine. I arrived at them, partly by personal observation and partly through the evaluation of literature compiled for the Encyclopedia of Endocrinology. This book will undoubtedly prove to contain errors of omission and commission. Nothing would be gained, however, by shifting the responsibility for any inaccuracies upon a limited number of references, which would have to be selected on an arbitrary, and hence subjective, basis in any case. By

the same token, I have carefully avoided quoting any of my own original publications. Had I done so, the reader could have gained the impression that it is only for the statements made in these that I am prepared to vouch. My own investigations added but little to the subject matter reviewed in this textbook, hence as an original investigator, I cannot be credited or blamed for more than a negligible portion of it. However, in addition to his original observations an author is also responsible for the evaluation of his data and their correlation with other reports in the literature. The interpretation of data and theories is naturally more subject to error than the mere report of observations. Yet the correlation of facts is an integral part in the structure of any science, and in this latter sense, the author of a textbook should also be prepared to be quoted for statements which reflect his judgment. I have undoubtedly made many errors, but I think that the readers of a textbook are more interested in the author's personal views expressed after weighing the relevant data to the best of his ability in the light of his experience, than in his legalistic skill to avoid possible blame by non-committal phrases and quotations.

As a result of these principles, the book suffers from many shortcomings. Firstly it is perhaps too dogmatic. Yet, whenever the evidence failed to convince me, I tried not to give the impression of unwarranted certainty, formulating the definiteness of my conclusions in proportion to the evidence at hand.

Secondly the book can hardly be called amusing. My only excuse for the heaviness of my style, is that my first concern was the concise expression of all the best-established facts. This left no space for colorful descriptions or stimulating speculations, if the book was not to become too bulky. However, I have tried to mitigate the austerity of the text by many illustrations.

If the structure of the book will prove to be a logical outline of the natural borders of Endocrinology and if the readers will be able to find concise and correct statements concerning the most important pertinent facts known to us at present, it will have fulfilled its purpose as a standard text. I shall have to rely upon the teachers of this subject to add, through colorful and stimulating lectures, the enthusiasm which this interesting field of medicine deserves. I shall be entirely satisfied if in turn, the teachers find that by having described the factual matter in the book, they can devote more time during their lectures to digressions from the drab routine.

Finally I should like to ask my readers to *make suggestions concerning possible improvements in this book*. I found it very difficult and timetaking to write the entire volume myself. I undertook to do it, because textbooks in which various chapters are written by different authors, usually lack unity and balance. To some extent, however, a good textbook should be a coöperative enterprise in which readers, and especially experienced teachers and investigators, collaborate with the author in his effort to adjust the volume to current requirements in subsequent editions. In this respect I must count on the help of my colleagues, but conversely, I would like them to feel assured that no effort will be spared to make and keep the book a true spokesman of contemporary endocrinology.

Hans SELYE

Université de Montréal

1947

ACKNOWLEDGMENTS

During the rather arduous task of compiling material for this book, it was one of my most pleasant experiences to note how strong the spirit of co-operation has remained among the scientists throughout this war-torn world. I have called upon many of my colleagues for advice and illustrative material not readily available to me, and it was most gratifying to note that scientists of so many lands,

several of whom I have never met, invariably responded in the most coöperative manner and spared no effort to furnish me with the information or material required. This exhibition of international fraternity among scientists was so heart-warming that I should like to enumerate the names of those who gave me aid, arranging them according to their countries of origin:

ACKNOWLEDGMENTS

	Nielsen, Kai	Institut de Médecine et de Chirurgie expérimentales, Université de Montréal.	Montréal
	Scott, D. A.	Banting and Best Department of Medical Research, University of Toronto, Institut de Médecine et de Chirurgie expérimentales, Université de Montréal.	Toronto
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	Teilum, Gunnar	The University Institute of Pathological Anatomy,	Copenhagen
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	Weber, F. Parkes	Guy's Hospital,	London
	Young, F. G.	National Institute for Medical Research, Hampstead,	London
FRANCE	Courrier, Robert	Collège de France,	Paris
GERMANY	Loeser, A.	Pharmakologisches Institut der Westf., Wihelms — Universität,	Münster
ISRAEL	Joel, C. A.	The Palestine Institute for Sterility Research,	Tel-Aviv.
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	Gardner, W. I.		
	Goldberg, M. B.		
	Gomori, G.		
	Grauer, Robert C.		
	Hall, C. Eric		
	von Haam, Emmerich		
	Henriksen, Erie		
	Karsner, H. T.		
	Kepler, Edwin J.		
	Kochakian, Charles D.		

ACKNOWLEDGMENTS

7

Kraus, Erik Johannes

Levine, Rachmiel
Li, C. H.

Lisser, H.
Mason, Karl E.

McCullagh, E. Perry

McKibbin, John M.

Means, J. Howard

Novak, Emil

Rawson, Rulon W.
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The Shelton Clinic,
Michael Reese Hospital,
Department of Legal Medicine,
Harvard Medical School,
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New York, N.Y.
Los Angeles, Cal.
Chicago, Ill.

Boston, Mass.

Many of these colleagues helped me by criticizing and proof-reading sections of the manuscript while others supplied, illustrative material. Space would not permit me to describe the nature of the participation of each individual in more detail here, but the names of those who furnished me photographs or material for illustrations is specifically listed under the figure which they helped to provide.

I could not write this acknowledgement, however, without specifically emphasizing my most profound indebtedness to Professor *B. A. Houssay* of Buenos Aires for writing the foreword, to Doctor *E. Perry McCullagh* of the Cleveland Clinic for having contributed the largest number of photographs concerning clinical endocrinology.

to the late Doctor *E. Kepler* of the Mayo Clinic for criticizing and improving the chapter concerning the adrenals, to Mr. *K. Nielsen* for having prepared all photographs illustrating material provided by our Institute, to Mr. *Alex. Thérien* for the excellent printing of this book (a task which under the present post-war conditions was not an easy one) and last but not least, to my colleague Miss *Helen Stone* of this Institute, who proof-read and edited the entire manuscript after each chapter had been submitted to the scrutiny of a specialized investigator.

Montreal, May 1949.

H.S.



GENERAL ENDOCRINOLOGY

DEFINITION AND SCOPE OF ENDOCRINOLOGY

DEFINITION OF ENDOCRINES AND HORMONES

It is rather difficult to define the natural confines of endocrinology. According to classic MORPHOLOGIC concepts, endocrinology is a science concerned merely with the glands of internal secretion; on a FUNCTIONAL basis, however, it deals with all hormonal substances even those produced by organs other than the exclusively endocrine glands. These definitions of the field are obviously dependent in turn upon a clear understanding of what we mean by endocrine organs and hormones respectively.

The Endocrine Organs. — On morphologic grounds, the hormone (for definition, see below) producing organs may be classified into three groups, namely:

(1) PURELY ENDOCRINE GLANDS, whose only function is to produce hormones. Among these, we distinguish the "*storage type*" (as exemplified by the thyroid), in which the endocrine cells are arranged in the form of small follicles. The lining cells can secrete directly into the blood stream or store their secretion in the follicular cavity. At times of high hormone requirement, the stored material is reabsorbed by the lining cells and transferred into the blood.

The "*solid type*" of endocrine gland (as exemplified by the parathyroids) contains no storage spaces. The parenchyme consists of massive cords or nests of epithelial cells, whose secretory products are discharged directly into the blood stream. Some degree of storage is possible even in these glands, but

here the hormone accumulation is exclusively intracellular.

(2) ENDO-EXOCRINE GLANDS, which secrete hormones into the blood stream, but, at the same time, also produce an exocrine secretion eliminated through a duct system. This group may be further subdivided into two sub-groups.

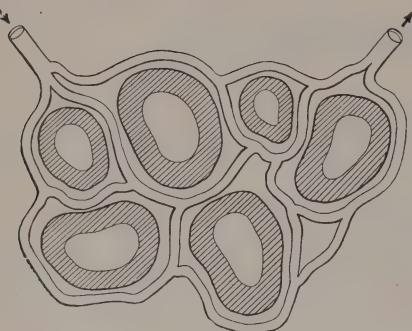
In the "*mixed endo-exocrine glands*" (as exemplified by the pancreas) parts of the organ are purely exocrine (the acinar tissue), while other parts are exclusively endocrine (the Langerhans islet tissue); the latter component, in this instance is a "*solid type*" of an endocrine gland. Anatomically, the two portions are intermixed.

On the other hand, the "*simple endo-exocrine glands*" (as exemplified by the liver) contain only one type of parenchymatous cell, which simultaneously produces both an exocrine secretion eliminated through ducts (bile) and a hormone-like substance secreted directly into the blood stream (the anti-pernicious-anemia factor).

(3) NON-GLANDULAR ENDOCRINE ORGANS such as the adrenergic and cholinergic nerves, produce hormone-like substances (acetylcholine and sympathin) without subserving any exocrine function. Some hormone-like substances (e.g., histamine) can probably be produced in a similar manner by several non-glandular organs.

According to classic concepts, only the purely endocrine cells are regarded as parts of the true endocrine system. The endocrine portions of mixed endo-exocrine glands (e.g., Langerhans islets) are of course always included since they subserve no function other than hormone production.

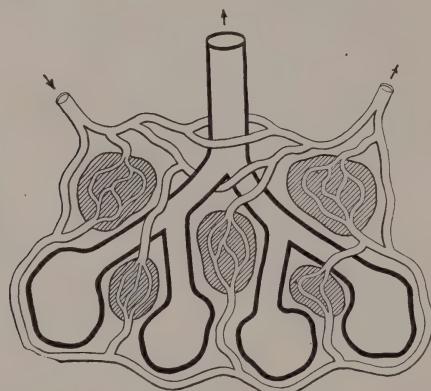
Morphologic classification of the endocrine glands



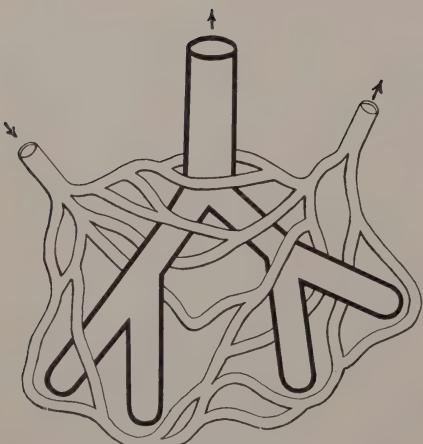
1. STORAGE TYPE OF ENDOCRINE GLAND
as exemplified by the thyroid.



2. SOLID TYPE OF ENDOCRINE GLAND as
exemplified by the parathyroids



3. MIXED ENDO-EXOCRINE GLAND as
exemplified by the pancreas.



4. SIMPLE EXO-ENDOCRINE GLAND as
exemplified by the liver.



5. NON-GLANDULAR ENDOCRINE ORGAN
as exemplified by adrenergic nerve
endings.

The Hormones. — On a purely *functional basis*, endocrinology must include all hormone-producing organs, irrespective of their morphologic structure. The only justification for the purely *morphologic* type of classification is that its outlines are more clear-cut, because it is simpler to recognize purely endocrine structures on a histologic basis than it is to delimit hormones from other active metabolites on functional criteria.

For any functional classification, it is essential to define what we understand by a HORMONE. This term (from the Greek "hormao" = I stir up or stimulate) was introduced by Bayliss and Starling (1902), who defined a hormone as "any substance normally produced in the cell of some part of the body and carried by the blood stream to distant parts which it affects for the good of the organism as a whole." Sharpey-Schafer (1924) introduced the term "AUTOCOID" meaning "a specific organic substance formed by the cells of one organ and passed from them into circulating fluids to produce effects upon other organs, similar to those produced by drugs." He wanted to retain the term "hormone" as a generic designation to include pharmacologically active metabolites produced by any organ. In this sense, CO₂, histamine, acetylcholine, sympathin, etc., would also be hormones, but not autocoids. However, this distinction is somewhat artificial, and the term autocoid has fallen into disuse.

More recently a number of highly active regulators of cellular differentiation and activity (embryonic organizer, wound hormones, etc.) have been regarded by some investigators as "INTRACELLULAR HORMONES." These differ from generally recognized hormones mainly in that they act locally in or around the cells in which they are formed.

The number and cumbersome formulation of the above definitions show how

difficult it is to delimit these concepts. In some sciences (e.g., mathematics) definitions are unchangeable laws, which make a concept what it is. In biology, however, definitions are given merely as concise descriptions of phenomena as they are known at the time. They are formulated with the view of modifying them as soon as further observations necessitate it. With this in view, it may suffice, at this time, to define hormones and endocrines as follows :

HORMONES ARE PHYSIOLOGIC, ORGANIC COMPOUNDS PRODUCED BY CERTAIN CELLS FOR THE SOLE PURPOSE OF DIRECTING THE ACTIVITIES OF DISTANT PARTS OF THE SAME ORGANISM.

ENDOCRINE ORGANS ARE ALL THOSE WHICH PRODUCE HORMONES.

These definitions are formulated to include all the most generally accepted characteristics of hormones and endocrine organs. The following criteria are regarded as essential :

Hormones are *physiologic* compounds, since it is not customary to include among them active substances (e.g., antibodies, toxic metabolites released by injured cells) which are not necessarily produced under physiologic circumstances.

Only *organic* compounds are classed among the hormones, since minerals released by cells are not included in this concept, even though they may be discharged for the sole purpose of directing the activities of distant parts.

Hormones are produced for the *sole purpose of directing, regulating and co-ordinating the activities of the organism*. Substances which direct only by supplying nutritive material and those which are merely catabolites of cells with incidental regulatory functions are not hormones. In the sense of Bayliss and Starling's definition, a nutrient such as glucose produced by hepatic cells would be a hormone, since it is "carried

by the blood stream" and "affects distant parts for the good of the organism as a whole." Sharpey-Schafer's concept of an autocoid hinges upon the, very difficult, definition of what we understand by "drugs." His inclusion of CO₂ among the hormones is likewise contrary to common usage.

We consider it essential that hormones are produced to direct the activities of *distant parts*. It is inadvisable to regard "intracellular hormones" as endocrine substances, since any cell produces a large number of chemical agents which affect the same or adjacent cells. Their inclusion among hormones would deprive this concept of all significance. Such compounds are manifestly not "endocrine" (endon = within; crino = to separate, to secrete), since they are not secreted into internal body fluids, but act locally at the site of their formation.

Finally, it is essential to emphasize that hormones affect distant parts of the *same organism* in order to distinguish them from such regulators of cellular activity as animal poisons, or the odorous substances produced to repel or attract other animals, etc.

If we accept this definition of hormones, the only logical formulation of the concept of endocrine organs is the simple one given above. It is in agreement with common usage, since we speak of the endocrine activity of the liver, kidney, etc., although these glands are not exclusively hormone-producing.

If, in this book, we place special emphasis upon the exclusively endocrine glands and their secretions (more correctly, but less customarily designated as "incretions"), we do so only because these are better known than the hormones of other tissues. It is agreed, however, that in future editions, the scope of this textbook may have to be expanded.

DELIMITATION OF ENDOCRINOLOGY

If we accept the view expressed in the introduction that it is logical and natural, "to delimit a new science from other fields of knowledge, so that it comprises all those facts which lend themselves particularly well for conjoint study by the same individual," then endocrinology must embrace all matters pertaining to the hormones. That means the normal and experimental morphology of the endocrine glands, the pharmacology and chemistry of the hormones, as well as the many clinical problems raised by the diseases of the endocrines. In view of the many correlations between the morphology, chemistry, physiology and pathology of the endocrines, a conjoint study of this vast subject matter by the same individual is sufficiently rewarding to justify this broad formulation of the field. It is impossible, however, to give equal attention to hormones and hormone-like substances produced by organs other than the purely endocrine glands, especially at the present time, when our knowledge concerning these outer areas of our subject is still so limited. As stated in the introduction, it is merely for this reason that our book places its chief emphasis upon the purely endocrine glands.

Those, however, who are about to decide whether they should select endocrinology as their main subject, should think of the broader scope of this science and the trend which its development and applications are taking. The following considerations appear to be of significance in this connection.

For the species, the most important rôle of the hormones is reproduction, but for the individual, it is differentiation and adaptation. It becomes increasingly more obvious, furthermore, that the principal medical application of endocrinology is not the treatment of the primary, but of the secondary diseases of the endocrines. — Tumors and hyperplasias of endocrine glands,

with consequent hormone overdosage, or destruction of incretory organs with the resulting hormone deficiency syndromes, are instructive, simple experiments of nature, which have taught us much about the endocrines. But these are rare diseases in comparison with the hormonal derangements resulting from maladaptation to stress.

The main, fatal syndromes of internal medicine (various cardiovascular, renal, "rheumatic" and old age diseases) may belong to this latter group; they are probably by-products of faulty hormonal adaptive reactions to a variety of non-hormonal pathogenic agents. The apparent cause of illness is often an infection, an intoxication, nervous exhaustion or merely old age, but, actually, a break-down of the hormonal adaptation-mechanism appears to be the most common ultimate cause of death in man.

THE ORGANIZATION OF CONTEMPORARY ENDOCRINOLOGY

In order to obtain a clear view of the scope of contemporary endocrinology, it is also useful to survey the place it takes in our society.

Research and advanced teaching in endocrinology necessitate a rather complex organization of men, laboratories, hospitals, libraries, etc., hence, throughout the world, the study of this subject is still largely restricted to UNIVERSITIES. We need not dwell upon the organization of post-graduate training and university research in endocrinology since this has been discussed in the Introduction.

The participation of PRIVATE INSTITUTIONS (private hospitals not attached to Universities, industrial laboratories) in the development of endocrinology is comparatively limited.

A number of SCIENTIFIC SOCIETIES have been formed, in order to promote research and teaching in the field of endocrinology. Among these are the following :

American Association for the Study of Goiter (U.S.A.).

American Diabetes Association (U.S.A.).
Association for The Study of Internal Secretions (U.S.A.).

British Diabetes Association (Great Britain).
Dansk Endokrinologisk Selskab (Denmark).
Dutch Society for Endocrinology (Holland).
Endokrinologföreningen [Svenska Läkarsällskapets Sektion för Endokrinologi] (Sweden).

Endocrinological Society (England).
Finnish Society for Endocrinology (Finland).

Laurentian Hormone Conference (Canada).
Nippon Naibunpi Gakkai (Japan).

Norwegian Society for Endocrinology (Norway).

Pan-American Endocrinological Congress (South America).

Russian Endocrinological Society (USSR).

Sociedad Argentina de Endocrinología y Nutrición (Argentina).

Sociedad Mexicana de Nutrición y Endocrinología (Mexico).

Société Canadienne d'Endocrinologie (Canada).

Société Française d'Endocrinologie (France).

Many of these have frequent meetings, at which endocrinologic papers are presented, but in most instances, the conventions are held annually or at even longer intervals. Thus the *Association for the Study of Internal Secretions* meets annually, if possible in the city in which the American Medical Association convenes at the same time. Usually, several hundred endocrinologists (mostly from North America) attend; the Meeting being open to all members of the Society, as well as to invited guests. As a rule papers are limited to 10 minutes, followed by a brief discussion. The convention lasts two days.

The *Laurentian Hormone Conference* is sponsored by the American Association for the Advancement of Science (A.A.A.S.) and takes place annually in the United States or one of the Laurentian resorts of Canada. Attendance is by invitation, usually limited to about 100 members. The meetings are purposely organized in a rather leisurely manner, in order to permit ample time for each paper, as well as the subsequent discussion. The

convention lasts one week during which time there is a good deal of opportunity for personal contact and exchange of ideas, even outside of formal meetings.

The Pan-American Endocrinological Congress convenes every three years, in one of the South American University Centers. It lasts several days and in addition to short papers (10 to 15 minute papers), some more extensive surveys are presented by invitation. Although most participants (usually several hundred) are South American, many North American endocrinologists attend.

In addition to the meetings of endocrinological societies, MANY PERTINENT PAPERS ARE PRESENTED AT THE CONVENTIONS OF SCIENTIFIC ORGANIZATIONS WITH A MORE GENERAL SUBJECT MATTER. Most important among these is the *International Physiological Congress*, which represents the largest group concerned with experimental medicine and physiology. It meets once every three years, usually in Europe or America.

The *Federation of American Societies for Experimental Biology* ("Federated Societies"), represents an association of six learned societies of the United States of America (The American Physiological Society, the American Society of Biological Chemists, the American Society for Pharmacology and Experimental Therapeutics, the American Society for Experimental Pathology, the American Institute of Nutrition and the American Association of Immunologists). It features many papers of endocrinologic interest at the annual convention. This group probably has the greatest attendance and presents the largest number of papers (several hundred) among all national societies interested in experimental medicine and physiology. Most of the papers are brief (10 minutes), but a few longer symposia are likewise presented. The Federation convenes once annually in various cities of the U.S.A.

The *Josiah Macy Jr. Foundation* organizes smaller meetings (attendance of 20 to 30), usually in New York City, at which papers are presented by invitation. Many of the subjects discussed are of endocrinologic interest.

The *American Society for Clinical Investigation* meets once a year (usually in Atlantic City). Attendance is limited to the members and some invited guests. A considerable number of the papers are of clinical endocrinologic interest.

It is well for the young endocrinologist to know about societies and conventions of this type, since they offer opportunities to hear scientific discussions in which specialists from various centers participate. At these meetings the student may discuss problems of technic, graduate study, possibilities of inter-laboratory collaboration on endocrine problems, opportunities to obtain laboratory space in institutions at which his work could best be performed, etc. It is important to realize that the meetings (and the published proceedings) of the scientific societies play a very important rôle in the development of any science, and this is particularly true of a rapidly growing young subject such as endocrinology.

The ENDOCRINOLOGIC LITERATURE is so voluminous and grows so rapidly that its publication and classification present many difficult problems of organization.

The following is a list of publications more or less exclusively devoted to endocrinology. Although some of them have temporarily, or even permanently ceased publication, it is important to know about them, since they contain many important papers on hormones :

- Acta Endocrinologica (Canada).
- Acta Endocrinologica (Denmark)
- Acta Endocrinologica (Roumania)
- Acta Endocrinologica et Gynaecologica (Portugal)
- Annales d'endocrinologie (France).
- Archivos de la Clinica e del Instituto de Endocrinologica (Uruguay).

Bulletins et mémoires; section d'endocrinologie de la société roumaine de Neurologie, Psychiatrie, Psychologie et Endocrinologie (Roumania).

Endocrinologie, gynécologie, si obstetrica (Roumania).

Endocrine Round Table (U.S.A.).

Endocrine Survey (Continuing the International Digest of Organotherapy) (U.S.A.).

Endocrinologia e pathologia costituzionale (Italy).

Endocrinologia (Italy).

Endocrinology (U.S.A.).

Endokrinologie (Germany).

Folia endocrinologica (Italy).

Folia endocrinologica japonica (Japan).

Hormones (England).

Japanese Journal of Endocrinology (Japan).

Journal of Clinical Endocrinology (U.S.A.).

Journal of Endocrinology (England).

Ormoni (Italy).

Ormonologia; biologia, patologia clinica (Italy).

Reviews in Endocrinology (U.S.A.).

Revista sud-americana de endocrinologia, immunologia y quimioterapia (Argentina).

Revue française d'endocrinologie (France).

Transactions of the American Association for the Study of Goiter (U.S.A.).

Vitamine und Hormone (Germany).

Vitamines and Hormones (U.S.A.).

Since endocrinology touches upon almost every other field of medicine and physiology, numerous pertinent publications appear in journals not primarily devoted to the endocrines. Among these, we mention the following as important sources of current endocrinologic literature :

Acta Anatomica (Switzerland).

American Journal of Obstetrics and Gynecology (U.S.A.).

American Journal of Physiology (U.S.A.).

Canadian Medical Association Journal (Canada).

Comptes rendus de la Société de biologie de Paris (France).

Journal of the American Medical Association (U.S.A.).

Journal of Physiology (England).

Lancet (England).

Presse Médicale (France).

Proceedings of the Society for Experimental Biology and Medicine (U.S.A.).

Revista Argentina de Biología (Argentina).

Revista Brasileira de Biología (Brazil).

Revue canadienne de Biologie (Canada).

Surgery, Gynecology and Obstetrics (U.S.A.).

In compiling literature concerning an endocrine problem, the following *indices and abstract journals* prove of considerable value :

Annual Review of Biochemistry (U.S.A.).

Annual Review of Physiology (U.S.A.).

Biological Abstracts (U.S.A.).

Chemical Abstracts (U.S.A.).

Excerpta Medica (Holland).

Index Catalogue of the Library of the Surgeon General's Office (U.S.A.).

Physiological Abstracts (England).

Quarterly Cumulative Index Medicus (U.S.A.).

Among the *encyclopedic treatises and handbooks* devoted to endocrinology, the following are noteworthy :

Die Bedeutung der inneren Sekretion für die Frauenheilkunde, von W. Berblinger, C. Clauberg und E. J. Kraus, in Handbuch der Gynäkologie, Dr. W. Stoeckel, Verlag von J. F. Bergmann, München (1936).

Encyclopedia of Endocrinology — by Hans Selye, Richardson, Bond and Wright, Publ., Montreal (1943-1946).

Endocrinology and Metabolism — by Garrison Barker, Appleton Publ. (1922).

Glandes Endocrines, in : Encyclopédie Médico-Chirurgicale — Fondateurs, A. Laffont and F. Durieu, 18, rue Séguier, Paris (6).

Endocrine Medicine — William Engelbach, Springfield, Ill., Baltimore, Md., Charles C. Thomas, Publ. (1932).

Handbuch der inneren Sekretion — Eine umfassende Darstellung der Anatomie, Physiologie und Pathologie der endokrinen Drüsen, ed. by Max Hirsch, Curt Kabitza, Leipzig (1932-1933).

Metabolism, Endocrine Glands, in Nelson New Loose-Leaf Medicine, Thomas Nelson & Sons, Publ. (1941).

Sex and Internal Secretions, a Survey of Recent Research, ed. by Edgar Allen, Charles H. Danforth and Edward A. Doisy, William & Wilkins Company, Publ., Baltimore 1st Ed. (1932); Repr. (1934); 2nd Ed. (1939); Repr. (1944).

The total volume of the endocrinologic literature is difficult to estimate. The endocrinologic library of this Institute, which, though still incomplete, is the most extensive collection of this kind, contains over a quarter of a million references. Additional publications appear at the rate of approximately 5,000 per annum, hence it is evident that the compilation of publications concerned with a certain problem of hormone research represents one of the most difficult tasks met by the contemporary endocrinologist. Once the investigator has succeeded in selecting the references which will be of particular use to him, the Library of the Surgeon General's Office, U.S. Army (Washington, D.C.), is willing to supply microfilms to those who have no library facilities at their disposal. The references can be compiled with some degree of efficiency, using the indices, abstract journals and encyclopedias mentioned above. It is difficult to discuss this problem, however, without calling attention to the fact that the compilation of scientific bibliographies needs a great deal of training and a large body of specialized personnel, if it is to be conducted in a systematic manner. Since the necessary books and library staffs are not at the disposal of most endocrinologists, a centralized, international organization of this work would be most desirable. It would certainly be no more expensive than the duplication of amateurish effort which is now in progress throughout the world in so many research centers, dealing with hormone research. Essentially, the

same could be said about the classification and supply of medical literature in any other field.

The progress of endocrinology has been greatly advanced by SCIENTIFIC FOUNDATIONS, who subsidize pertinent research work by grants-in-aid. Although these foundations do not limit their efforts to endocrinology, the following may be mentioned as contributing considerable sums for research on hormones :

1. Commonwealth Fund (U.S.A.).
2. John and Mary Markle Foundation (U.S.A.).
3. Josiah Macy Jr. Foundation (U.S.A.).
4. National Public Health Service (U.S.A.).
5. Rockefeller Foundation (U.S.A.).
6. Sugar Research Foundation Inc., (U.S.A.) — Mainly in connection with carbohydrate metabolism.

In addition to grants given to institutions for specific research programs, individual *bursaries, fellowships and scholarships* are available for young investigators who wish to specialize in research subjects. The following are enumerated as likely sources of support for promising young endocrinologists who cannot complete their training at their own expense :

Rockefeller Foundation Travelling Fellowships.

American National Research Council Fellowships (usually tenable in the U.S.A.).

Canadian National Research Council Fellowships (usually tenable in Canada).

Banting Fellowships (usually tenable in Canada).

Beit Fellowships (usually tenable in England).

1851 Fellowships (usually tenable in England).

Royal Society Fellowships (usually tenable in England).

Rhodes Scholarships (usually tenable at Oxford, England).

Life Insurance Medical Research Fund Fellowships (tenable in the U.S.A. or Canada).

The PHARMACEUTIC INDUSTRY is taking an ever increasing interest in the manufacture of hormones; many important discoveries concerning, for instance, the isolation, synthesis and bioassay of hormones are due to work performed or subsidized by the major pharmaceutical companies. Among these we may mention the following companies who play a particularly great rôle in supplying the world market with new hormone products :

Abbott Laboratories, Middlesex, England; Montreal, Canada; Chicago, U.S.A.

Armour Company, Chicago, Ill., U.S.A., London, England.

Ayerst, McKenna & Harrison Ltd., New York, N.Y.

British Drug Houses, London England.

Burroughs Wellcome and Co. (U.S.A.) Inc., London, England; New York, N.Y., U.S.A.

Byla Company, Paris, France.

Ciba Pharmaceutical Products, Inc., Summit, N.J., U.S.A.

Eli Lilly and Company, Indianapolis, Indiana, U.S.A.

Harrower Laboratory, Inc., Glendale, Cal., U.S.A.

Parke, Davis and Company, Detroit, Mich., U.S.A.

Reed and Carnick, Jersey City, N.J., U.S.A.

Gedeon Richter Ltd., London, England.

Rhone Poulenc, Paris, France.

Roche Organon Inc., Roche Park, Nutley, N.J., U.S.A.

Schering Corporation, Bloomfield, New Jersey, U.S.A.

Serotherapeutic Institute of Milan, Italy.

E. R. Squibb, New York, N.Y., U.S.A.

Upjohn Company, Kalamazoo, Mich., U.S.A.

Winthrop Chemical Co., New York, N.Y., U.S.A.

This list is of course very incomplete, and perhaps somewhat arbitrarily selected, since hundreds of pharmaceutical companies produce hormone preparations. It is only intended to familiarize the reader with the names of a few major manufacturers of endocrine products.

MECHANISMS OF HORMONE ACTIONS

PREREQUISITES OF HORMONE ACTIONS

The so-called "target organs" or "end organs" do not necessarily react to hormones under all conditions. This is understandable if we consider the mechanisms through which endocrine products exert their actions. From this point of view, we may distinguish three types of hormones :

(1) DIRECTLY ACTING HORMONES. — These are hormones which act upon their targets directly and not through the intermediary of other organs. In general, such hormones affect the receptive cells, even in vitro, since the immediate response they elicit is independ-

ent of the rest of the body. The action of adrenaline upon the blood vessels is an example of such a direct hormone action. It occurs even if adrenaline is directly applied to isolated blood vessels.

(2) INDIRECTLY ACTING HORMONES.

— In themselves, these do not affect the target organ at all. They act upon another organ (usually another endocrine gland) and induce it to produce a hormone which influences the target organ directly. For example, the pituitary produces luteinizing hormone (LH) which stimulates the seminal vesicles, but has no direct effect upon them. It merely causes another endo-

crine structure, the Leydig cells of the testis, to produce testoid hormones, which, in turn, exert a direct action on the target organ, the seminal vesicles. The gonadotrophic, thyrotrophic and corticotrophic hormones may all be cited as examples of such indirectly acting hormones, whose mediated effects depend entirely upon the integrity of the gonads, thyroid and adrenal cortex respectively.

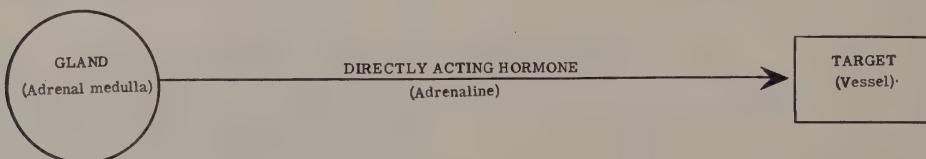
It is for this reason that testosterone, a directly acting hormone, causes seminal vesicle growth in the castrate as well as in the intact male, while luteinizing hormone elicits this same response in intact animals, but is entirely ineffective following castration.

(3) CONDITIONALLY ACTING HORMONES. — These are directly acting hormones, whose effect upon the target organs is not dependent upon the integrity of any intermediate station. Yet, depending upon the circumstances, the receptive cells may or may not respond. Thus, for example, desoxycorticosterone acetate causes nephrosclerosis, but

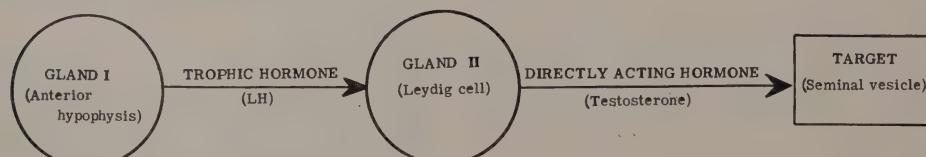
only if the diet of the experimental animals contains sufficient amounts of sodium and chloride. Similarly the direct actions of adrenaline can be modified by denervation which increases, or adrenolytic drugs which decrease the responsiveness of certain target organs to this hormone. To some extent, of course, all hormone actions depend upon the condition of the responsive cells. In some instances, however, the target is almost always optimally sensitive to the hormone, while, in others, the response is greatly influenced by activating or inactivating agents. These modifying agents may themselves be hormones. Thus progesterone, when given by itself, possesses only a very slight progestational effect upon the endometrium, but following pretreatment with minute doses of estradiol (or other folliculoid hormones), small doses of progesterone suffice to cause marked progestational reactions. On the other hand, pretreatment with very large doses of folliculoids completely inhibit the progestational action of the corpus luteum hor-

Mechanisms of hormone actions

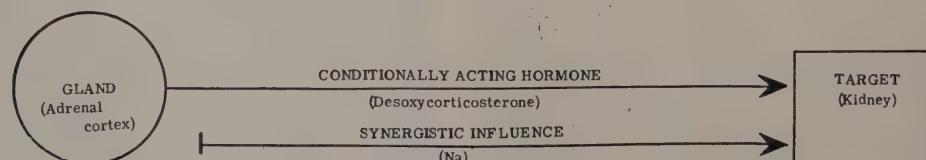
I- DIRECTLY ACTING HORMONE:



II- INDIRECTLY ACTING HORMONE:



III- CONDITIONALLY ACTING HORMONE:



mone. Here, depending upon dosage, one hormone can both increase and decrease the response to another hormone.

In connection with the conditionally acting hormones, the following consideration is important, since disregard of it leads to many errors in the interpretation of observations. If a stimulus which normally causes a response in a target organ fails to do so after extirpation of an endocrine gland, this does not necessarily mean that its effect is mediated by that gland.

It must be kept in mind that if a stimulus cannot influence its normal target organ in the absence of an endocrine gland, there are two possibilities :

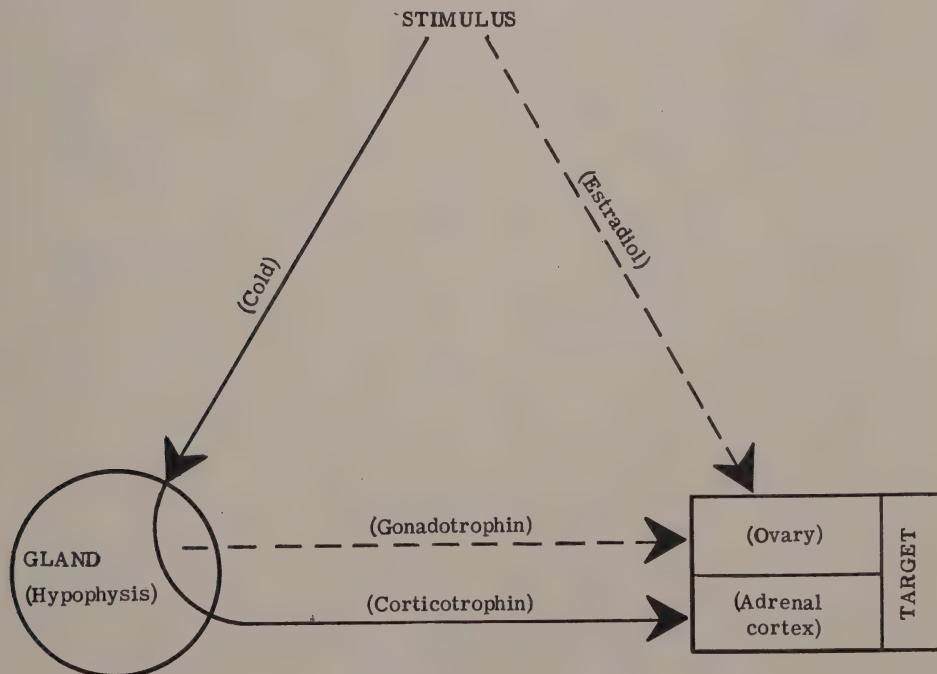
(1) The stimulus is an indirectly acting agent, which affects the target organ merely by modifying the hormone production of the endocrine gland in question.

(2) The stimulus is a conditionally acting agent, whose effect on the target

organ depends upon simultaneous sensitization of the latter by a hormone produced by the endocrine gland in question.

Thus, exposure to cold causes adrenal-cortical enlargement in the rat, but only in the presence of the pituitary. Similarly, large doses of folliculoids elicit the production of large corpora lutea, but again only in the presence of the pituitary. An analysis of these two phenomena has shown that cold actually acts through the intermediary of the pituitary, causing it to produce an increased amount of corticotrophin. In hypophysectomized animals, exposure to cold leads to no cortical hypertrophy, even if their adrenals are maintained in a normal condition by daily corticotrophin injections. On the other hand, large doses of folliculoids, though ineffective in influencing the ovary of untreated hypophysectomized rats, cause enlargement of corpora lutea even after

Possible rôles played by an endocrine gland which is indispensable for the response of a stimulus



ablation of the hypophysis, if the ovary is maintained in an approximately normal condition by treatment with exogenous gonadotrophins. In this case, the folliculoids are conditionally acting agents, whose effect on the target organ (the corpus luteum) depends upon simultaneous sensitization of the latter by gonadotropic pituitary hormones (mainly luteotrophin).

MECHANISM OF DIRECT HORMONE ACTIONS UPON TARGET ORGANS

Hormones are Merely Regulators of Biologic Phenomena. — Almost nothing is known as yet about the intimate mechanisms through which internal secretions influence their target organs. It becomes increasingly more evident, however, that hormones do not bring about any essentially new metabolic activity, but merely regulate the course of phenomena which can progress to some extent, even in their absence. Thus, for example, thyroid hormone increases the basal metabolic rate, but in its absence, basal metabolism proceeds, although at a lower level; adrenotrophin stimulates the activity of the adrenal cortex, but some corticoid hormone production continues even after complete hypophysectomy; estradiol accelerates the proliferation of the vaginal epithelium, although in its absence, the lining cells of the vagina continue to grow at a greatly reduced rate. Even the cornification of vaginal cells supposedly so specific of folliculoids (estrogens) is possible in their absence (e.g., in A-avitaminosis). The effect of hormones has therefore often been compared with that of catalysts or enzymes, whose function is likewise limited to the regulation of reactions which are essentially not dependent upon them. It has even been suggested that the hormones may actually be enzymes, but this has never been definitely demonstrated. The few pertinent data known at the present time would rather suggest that the hormones merely condition the activi-

ty of the enzyme systems which regulate biologic phenomena. Renin, however, is an enzymatic internal secretion of the kidney, which behaves like a conditionally acting hormone. It is inactive in itself but transforms certain blood globulins into the highly potent vasopressor substance hypertensin (or angiotonin).

Are hormones utilized while exerting their effects? — After they enter the blood stream, hormones are partly destroyed in the body, due to chemical degradation or conjugation with other substances. This yields inactive or less active end products; hence the excretions (urine, feces, sweat, etc.) do not contain the total amount of the hormones produced by the endocrines. There is no definite proof, however, that hormones are actually "utilized" by the end-organs, that is, that they are consumed by the tissues as a result of their hormonal activity. Indeed, there is reason to believe that the destruction of hormones proceeds practically unchanged, even if they are prevented from exerting physiologic actions. Thus, the inactivation of exogenously introduced gonadotrophins is not significantly influenced by gonadectomy, although, in the absence of the gonads, these trophic principles can exert no physiologic action. Perhaps the best proof for the relative independence of hormone destruction from hormone activity is the following:

If animals are completely deprived of endogenous gonadotrophins by hypophysectomy, their ovaries involute. In such animals, injection of gonadotrophic hormones causes an ovarian enlargement which, at a certain range, is proportional to the amount injected. Under such conditions however, the increase in the weight of one ovary is the same, whether the other ovary is present or not. If the gonadotrophic hormone were destroyed by the ovary itself (while it performs its stimulating action), obviously a greater effect should be obtained by a given dose

ARE HORMONES "UTILIZED" BY THEIR TARGET ORGANS WHILE THEY EXERT THEIR ACTIONS UPON THEM?

HYPOPHYSIS REMOVED	"PAIRED GLAND" UNDER HYPOPHYSEAL CONTROL (e.g., OVARIES, THYROID, ADRENALS)	UNITS OF TROPHIC HORMONE INJECTED	CONDITION OF PAIRED GLANDS
			ONE PRESENT
			BOTH PRESENT
			3 UNITS
			2 UNITS
			1 UNIT
			3 UNITS

Note that the effect of trophic hormones is independent of the amount of target organ tissue present in the organism.

when only one ovary is present. Under similar experimental conditions, the adrenal enlargement caused by corticotrophin and the thyroid enlargement due to thyrotrophin proved to be independent respectively of the amount of adrenal and thyroid tissue present in the body. In animals whose hypophysis is intact, the removal of one gonad, adrenal or thyroid, causes compensatory hypertrophy of the contralateral gland. This, however, is not because the remaining glandular tissue benefits from an excess of trophic hormone due to non-utilization by the contralateral gland. It is due merely to the above-mentioned compensatory hypertrophy mechanism, that is to say, if one of these paired glands is removed, the pituitary attempts to compensate by an increased production of the corresponding trophic principles. (See below.)

Hormones acting on many distinct target organs do not lend themselves well for similar studies. It has been found, however, that removal of the uterus and most of the vagina does not significantly influence the responsiveness of the vaginal remnant to a threshold dose of a folliculoid hormone, nor does ablation of most of the male accessory-sex-organs noticeably alter the sensitivity of the remaining sex-organs to a given amount of testoid material.

STIMULI REGULATING THE ACTIVITY OF ENDOCRINE GLANDS

Humoral and nervous stimuli help to adjust the activity of the endocrine glands to changing conditions in the organism or its surroundings.

Among the HUMORAL STIMULI, hormones play a particularly important rôle. Thus, the anterior-lobe of the pituitary is almost exclusively responsible for the normal function of the adrenal cortex, the thyroid, the gonads, and to some extent, even the Langerhans islets of the pancreas. All these endocrine glands are under the controlling in-

fluence of so-called trophic hormones of the anterior-lobe. Their name is derived from the Greek "trophe" = nourishment, although they are not nutrients in the ordinary (caloric) sense of the word. The anterior-pituitary has therefore been compared with the central nervous system, which plays a similar rôle in the integration of nervous activities. In order to adjust the function of the various "peripheral" endocrines to the needs of the organism, such a central control by one "master gland" is advantageous.

In order to stabilize the activity of those glands of internal secretion which are under hypophyseal control, the production of trophic hormones by the pituitary is in turn regulated by the peripheral endocrines. Thus, for instance, an increase in the gonadotrophin secretion of the pituitary augments the folliculoid hormone production of the ovary, but these folliculoids act back upon the pituitary to decrease its gonadotrophin secretion. In this manner the ovarian stimulation is maintained at a fairly steady level by a self-regulating mechanism. Such interrelations between the hypophysis and various peripheral endocrines are responsible for many of the phenomena of so-called *compensatory atrophy* and *compensatory hypertrophy*. Thus, thyroid hormone administration depresses the endogenous production of thyrotrophin by the anterior-lobe and causes a compensatory atrophy of the thyroid; partial thyroideectomy and the consequent decrease in circulating thyroid hormone acts as a stimulus for the compensatory increase in thyrotrophin production and thus, helps to re-establish a normal thyroid hormone concentration in the blood through the stimulation of the thyroid remnant. Many other examples of compensatory atrophy and hypertrophy will be mentioned in the sections devoted to the individual endocrine glands. Indeed, the menstrual cycle may merely represent a spe-

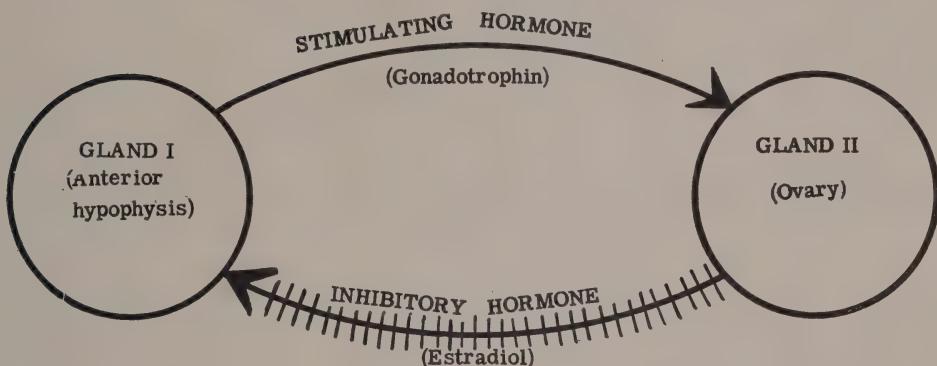
cial instance of this mechanism. Some investigators believe that an increase in ovarian-hormone secretion automatically depresses the gonadotrophin secretion of the pituitary; since this is followed by a decreased ovarian hormone

production, the gonadotrophin secretion is augmented during the next phase and so forth. (See : Sexual Cycle.)

The following drawings illustrate these mechanisms of compensatory hypertrophy and atrophy :

Compensatory hypertrophy and atrophy

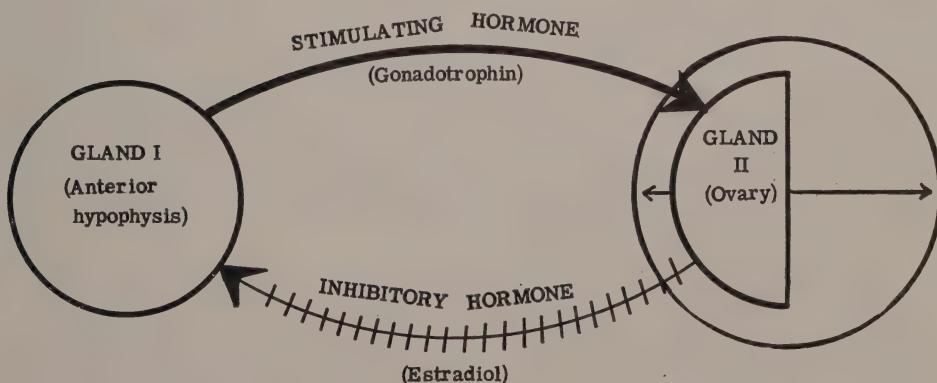
I — Normal balance between glands I and II



The normal balance between gland I (e.g., anterior-hypophysis) and gland II (e.g., ovary). Gonadotrophin stimulates the ovary to produce estradiol which inhibits the anterior-hypophysis.

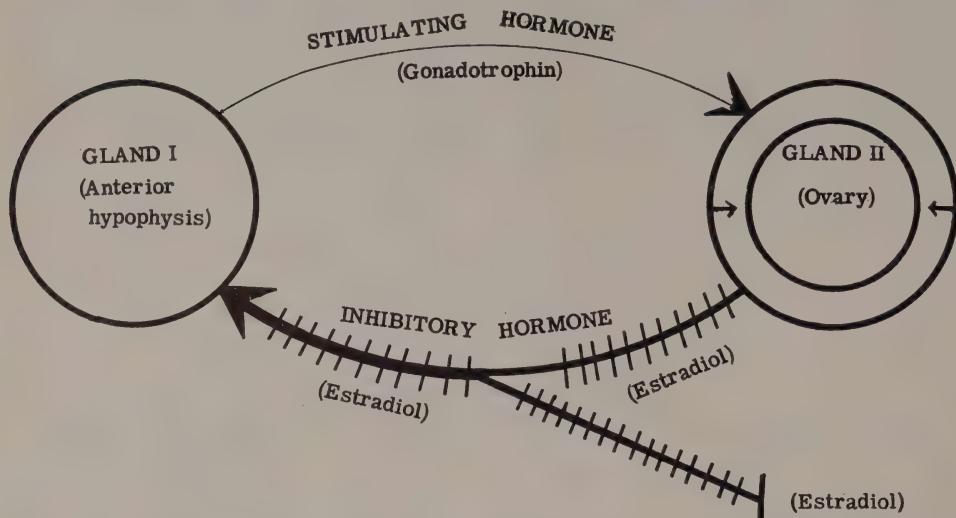
(Inhibition is indicated by cross-hatched arrows.)

II — Effect of partial ablation of gland II



Following partial ablation of gland II the inhibitory-hormone production is diminished and hence an increased amount of stimulating-hormone is secreted by gland I; this leads to compensatory hypertrophy of gland II.

III — Effect of exogenously introduced hormone of gland II



Exogenous administration of the inhibitory-hormone (e.g., estradiol) causes inhibition of gland I and thus diminishes the stimulating-hormone production; this leads to compensatory atrophy of gland II.

Many OTHER HUMORAL AGENTS exert a regulating influence upon the secretion of the endocrines. Thus glucose increases the production of insulin, while hypoglycemia causes a discharge of adrenaline. Since insulin depresses while adrenaline raises the blood sugar level, these responses are eminently suited for the self-regulation of the blood sugar level.

NERVOUS STIMULI likewise play an important rôle in the regulation of hormone production. For example, stimulation of the splanchnic nerve causes adrenaline liberation apparently because this nerve contains secretory fibers regulating the hormone production of the adrenal medulla. Similarly, the nervous stimulus of nursing has been proven to regulate the secretion of pro-

lactin by the hypophysis, thus adjusting the amount of milk produced to the requirements of the nursling. The hormone production of many endocrine glands, however, is remarkably independent of nervous stimuli; for instance complete denervation of the parathyroids, thyroids, adrenal cortex and gonads causes no significant change in their ability to secrete hormones or even to adjust the quantity of their hormone production to varying requirements.

In summary it may be stated that the elaboration of hormones is mainly dependent upon : 1) the amount of circulating hormones in the blood; 2) the blood concentration of certain metabolites whose utilization depends upon hormones; 3) nervous mechanisms.

CLASSIC EXPERIMENTAL PROCEDURES IN ENDOCRINOLOGY

PROOF OF ENDOCRINE ACTIVITY
OF AN ORGAN

A series of experiments has to be performed to prove the endocrine activity of an organ. Only in a few cases is it technically possible to furnish all the

proofs enumerated below, but generally, the three first-mentioned procedures are accepted as giving adequate information. It will be borne in mind that essentially the same type of experimental evidence is necessary to establish the

hormonal causation of a change (e.g., testicular atrophy) or the hormonal nature of a biologic substance (e.g., pituitary extract).

Extirpation Causes Deficiency. — One of the most important proofs for the endocrine activity of an organ is that its surgical removal causes specific deficiency symptoms. This is due to the fact that most hormones are produced in one organ only, so that after complete destruction of the latter, hormone production ceases. Thus, after extirpation of the testes, there is involution of the seminal vesicles, prostate and other male accessory-sex-organs, because only the testis produces significant amounts of testoid (androgenic) hormones. Similarly, removal of the hypophysis induces gonadal, adrenal-cortical and thyroid atrophy, since only the trophic hormones of the pituitary have the ability to stimulate these glands directly. It becomes increasingly more evident, however, that the organ specificity of hormone production is by no means an absolute rule. Thus, certain amounts of hypophysoid (gonadotrophic, luteotropic) and folliculoid hormones can be produced by the placenta, testoids by the adrenal cortex, etc. Some organs elaborate hormones which can also be secreted by other tissues. Therefore, if extirpation of a gland does not cause any specific deficiency symptoms, this should not be interpreted as absolute proof against its endocrine nature. For instance, the impossibility of producing specific signs of deficiency by thymectomy or pinealectomy can not be regarded as absolute proof against their hormone-producing ability.

If in such instances complete extirpation of all endocrine cells is impossible, the production of similar hormones by several organs must be proven by the procedures mentioned below (e.g., specific overdosage symptoms by glandular extracts, demonstration of hormone in the venous blood of the gland, proof that the hormone concentration in the

body and its excretions depends upon the degree of development of the supposedly endocrine tissues, etc.).

Organ Extract Causes Overdosage.

— After suitable purification and concentration, the active principles of endocrine organs can be shown to possess physiologic activities, similar to those normally exerted by the organ itself. Indeed, if excessive doses of such preparations are injected into experimental animals, usually hormone-overdosage results. This procedure lends itself even for the demonstration of endocrine activity in tissues which, for technical reasons, cannot be completely eliminated and hence do not lend themselves to proof of their internal secretion by the production of an experimental hormone-deficiency.

In evaluating the results of such experiments, it must be kept in mind, that although large amounts of hormones are secreted by certain tissues, their active principles are not necessarily stored to any extent in the cells which elaborate them. Under such conditions, it may be very difficult to obtain the necessary concentration of hormone activity in gland extracts. For instance, the ovaries contain only negligible quantities of folliculoid hormones, so that crude ovarian extracts reveal themselves as inactive in tests of this type. On the other hand, if a definite biologic activity is demonstrated in an organ extract, this does not prove that the tissue normally secretes such principles into the blood. Various active metabolites accumulate in cells and a variety of poisons prepared from the organs of animals or plants can act as drugs. Their activity does not necessarily bear any direct relationship to the normal physiologic function of the tissues from which they were obtained. It is for this reason that the effect of organ extracts, injected into intact test animals, must simulate the normal physiologic function of the tissue from which they are

derived before the experiment can be interpreted as indicative of endocrine activity.

Efficacy of Substitution Therapy. — It is a particularly convincing proof of the endocrine activity of tissues if extracts prepared from them can correct the deficiency symptoms produced by extirpation of the organ in question. Such "substitution therapy" shows most clearly that the extracts truly *imitate the physiologic function of organs*.

The classic procedure is to remove an endocrine gland from an experimental animal, note the resulting deficiency syndrome, and then restore conditions to normal by the administration of purified organ-extract concentrates.

For instance, the atrophy of the accessory-sex-organs subsequent to castration, is restored by the administration of active testicular extracts. This type of proof is again not applicable in the case of endocrine organs which cannot be removed, completely enough, to produce a definite deficiency syndrome.

Demonstration of Hormone in Venous Blood of Endocrine Organ.

— In certain instances, it is actually possible to demonstrate a high concentration of hormones in venous blood coming from an endocrine organ. This is significant only if the concentration in the veins of the endocrine organ is much higher than in the general systemic circulation.

Thus, unusually high concentrations of adrenaline or corticoids have been demonstrated in the adrenal veins by direct bioassay. This experimental procedure is particularly suitable for the study of changes in hormone secretion induced by various stimuli (see below). It has been shown, for instance, that the adrenaline concentration in the adrenal veins is increased after splanchnic stimulation, insulin administration, emotional stimuli, etc. From this it could be concluded that probably a discharge of this hormone also occurs phy-

siologically under the direct influence of such stimuli.

Demonstration that Hormone Concentration in the Body and Excretions Depends upon the Condition of the Endocrine Organ. — Dependence upon an organ of the hormone concentration in the blood, the tissues and excretions, may help to identify it as a hormone producer. Thus, partial or complete *extirpation of a gland or its destruction by disease* decreases, while *stimulation of its activity* (by trophic hormones, nervous stimuli and other agents) demonstrably increases hormone production, approximately in proportion with the amount of glandular tissue present.

Other technics may help to demonstrate that a biologic change depends upon the presence of certain endocrine cells in the organism; among these are, *transplantation* of endocrine organs (in the form of free grafts or by anastomosis of the transplant's vessels with those of the host), or the establishment of a *cross-circulation* or *parabiosis* between the body of an animal in which an endocrine gland has been removed, with a second animal in which this gland is present.

Explantation of endocrine cells (tissue cultures) may reveal the formation of hormones by the cells *in vitro*; similarly in organ cultures (perfusion of isolated endocrine glands) hormonal principles can be elaborated and secreted into the perfusing fluid, where they become demonstrable by bioassay or chemical tests.

After gonadectomy for instance, the concentration of gonadal hormones in the blood and urine can be shown to diminish; transplantation of the gonads into gonadectomized hosts restores conditions to normal and cross-circulation or parabiosis between a gonadectomized and a normal animal reveals the transition of gonadal hormones from the latter to the former. Similarly, thyroid cells produce thyroid hormone *in vitro* and if a whole thyroid is perfused, the

gland can be shown to secrete biologically demonstrable quantities of thyroid hormone into the perfusion fluid.

Isolation of the Pure Hormone — The best proof for the endocrine activity of an organ is the isolation of its pure hormone or hormones. This can be accomplished by chemical or physical means. The pure product is usually a crystalline, physically homogenous substance which accurately imitates the normal endocrine activity of the organ. In intact animals, it simulates the endocrine activity of the gland by producing overdosage effects, while in animals rendered deficient in the secretions of the gland (e.g., by extirpation) it gives complete substitution therapy. Non-protein hormones usually form typical crystals, whose melting point, crystal form, optic rotation, etc., help to identify them (e.g., steroid hormones, adrenaline, thyroxin). The crystallization of protein hormones (e.g., insulin), is difficult, however, and in itself not a conclusive criterion of purity. Usually, homogeneity can only be demonstrated by several physical constants, for instance osmotic pressure, solubility, electrophoresis, etc., (e.g., growth hormone, prolactin).

PROOF THAT A SYNTHETIC SUBSTANCE IS IDENTICAL WITH THE NATURAL HORMONE

It has been possible to synthesize several hormones. In order to demonstrate that such synthetic compounds are actually identical with the naturally occurring hormones, it is essential to show that they imitate all biologic and chemical actions of the latter. In the case of crystalline substances (e.g., steroids), the melting point of the artificial compound must be identical with that of the hormone prepared by extraction and purification of glandular tissue; even after mixing the artificial and natural substances, the "mixed melting point" must remain the same.

Synthetic protein-hormones have not yet been prepared.

PRINCIPLES OF BIOASSAY

It is not within the scope of this book to give a detailed account of bioassay technics or of the mathematical principles upon which we base their interpretation, but the following fundamental considerations are indispensable.

Purely technical reasons often preclude the use of accurate, sensitive and specific chemical tests for the estimation of hormone concentrations in biologic materials. In such instance the hormones are better identified and estimated on the basis of their biologic activities.

In principle, we may distinguish between "internal" and "external" bioassays. INTERNAL BIOASSAYS are based upon the observation of target organs in the individual whose hormone production we wish to estimate. Thus, vaginal smears or uterine biopsies taken from a woman are internal indicators of her own ovarian-hormone production. In certain instances, the responsiveness of such internal indicators can be increased by special sensitizing methods. Thus, the responsiveness of the iris, or the heart, to endogenously produced adrenaline is augmented by denervation.

All these internal bioassay methods have the advantage that no loss of hormone is incurred, since they require no extraction from the tissues of the donor or transfer to a recipient test animal. They are simple technics in which the hormone production of the same individual can be repeatedly tested under identical conditions and they give an over-all picture of the effective hormone concentration in the body. They lend themselves well to serial determinations of the endogenous hormone production of an individual under varying conditions.

Their great disadvantage is that they are less suitable than external bioassays

for exact quantitative determinations on a statistically significant basis. Furthermore, they give no indication concerning the distribution of the hormone in the various body fluids and tissues.

Conversely, the EXTERNAL BIOASSAYS are based upon the removal from a donor, of certain specimens of body fluids or tissues, which are tested by their effects on other animals. They can be administered as such, or after suitable purification and concentration of the active material.

It is advisable, furthermore, to USE MODERATE DOSES for bioassay purposes, that is, quantities just sufficient to produce a definite biologic reaction. If large quantities are given, the relationship between the dose injected and the result obtained is less definite, while threshold doses give erratic results.

In all bioassay technics, it is important to ADMINISTER THE HORMONE IN A MANNER ASSURING GOOD ABSORPTION, with as little destruction as possible (see : Technics of Hormone Administration, below).

In selecting a suitable test object it is noteworthy that ANIMALS DEFICIENT IN A CERTAIN HORMONE ARE USUALLY MOST SUITABLE for the bioassay of that hormone. If adequate amounts of a hormone are present in the body, the addition of small doses fails to cause a significant change. For this reason, the folliculoid hormones are assayed on spayed or immature animals which have no significant endogenous source of such hormones. For similar reasons, hypophyseal hormones show their activity much better in hypophysectomized than in intact animals. The use of animals deprived of the specific endocrine gland in question is also advisable since compensatory changes due to endogenous hormone production cannot take place during the bioassay and hence, the effect of the injected hormones is not blurred by internal adaptive mechanisms.

CONDITIONALLY ACTING HORMONES SHOULD BE TESTED UNDER CIRCUMSTANCES MOST FAVORABLE FOR THE EXERTION OF THEIR EFFECTS. Thus insulin, whose hypoglycemic action is largely dependent upon dietary factors, should be assayed in fasting animals; progesterone in animals sensitized by folliculoids, etc.

Hormones to which ADAPTATION (e.g., progesterone), TACHYPHYLAXIS (e.g., vasopressor posterior-lobe extracts) or ANTI-HORMONE FORMATION (e.g., hypophysoid gonadotrophins) occurs, must be assayed on animals not desensitized by previous treatment. Very PROLONGED HORMONE DEFICIENCY MAY RENDER ANIMALS COMPARATIVELY INSENSITIVE to certain internal secretions. This must be taken into account for instance in the assay of folliculoids on rats spayed long before the test. In such cases a single injection of folliculoids a few days before the test, usually suffices to restore normal responsiveness.

It is advisable to COMPARE THE ACTIVITY OF THE UNKNOWN HORMONE CONCENTRATION WITH THAT OF A MEASURED STANDARD PREPARATION, under similar laboratory conditions. International standards of pure hormone preparations have been established by the League of Nations and samples of these are placed at the disposal of interested investigators.

In general, it is best to base bioassays on the determination and use of a DOSAGE-RESPONSE LINE. For this purpose, at least three dosage levels must be tested, since it requires at least three points to define a line, with which the points obtained by individual determinations can subsequently be compared. With most bioassay procedures, sigmoid curves are obtained when the arithmetic dose is plotted against the arithmetic response. The dose level at which the tests are performed should fall within the steepest portion of the curve, because the response to varying

dose levels is not very different within the initial and final flattened portions of such curves. It is advisable to transform the original sigmoid curve into a straight line, since the usual statistical computations assume that over the dosage range used, the dose-response relationship follows a straight line.

If the test is based on a "yes or no" response (e.g., number of deaths in toxicity tests, number of animals showing cornified smears in folliculoid assays), this can be accomplished by plotting the log of the dose against the probit of the response. Convenient tables have been prepared for this purpose.

Other types of bioassays give *graded responses*, that is, even among the positive reactors, the degree of response

depends upon the amount of hormone administered (e.g., adrenal weight in the assay of adrenotrophic hormone, duration or depth of anesthesia in the assay of anesthetic steroids). In these instances, the dosage-response curve can be transformed into a straight line by plotting the dose against the arithmetic response. Sometimes, however, the log or some other function of the response, must be used to fit the desired relationship over the dosage-range in question.

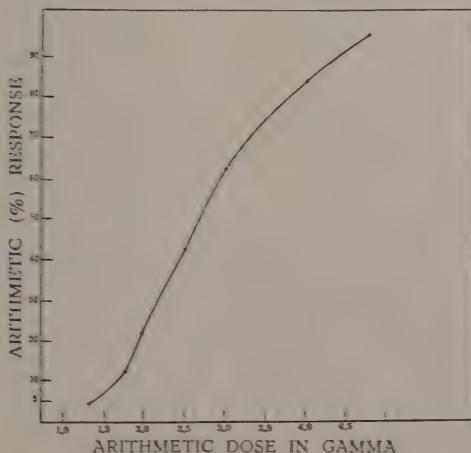
The following two examples, based on actual bioassays (courtesy of Dr. L. Pugsley), illustrate these points. The first gives the data and curves of an estrone assay (vaginal cornification), the second of an androsterone assay (capon-comb growth);

Dosage-response data for Estrone using castrated female rats

No. of rats	Dose (in γ)	Log dose	Response in %	Response Probits
25	1.4	.1461	4	3.2493
25	1.8	.2553	12	3.8250
23	2.0	.3010	22	4.2278
26	2.5	.3979	42	4.7981
26	3.0	.4771	62	5.3055
25	4.0	.6021	84	5.9945
22	5.0	.6990	95	6.6449

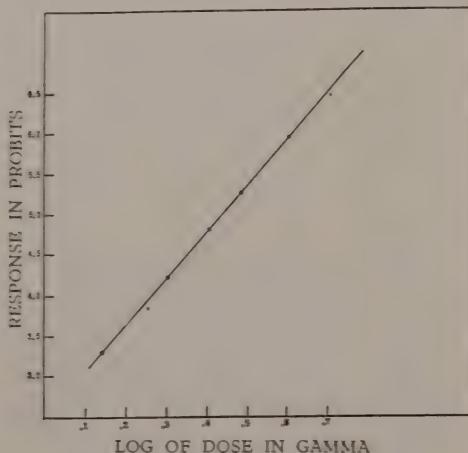
Dosage-response curve of Estrone
(quantal response)

Arithmetic dose against arithmetic response



Dosage-response line of Estrone
(quantal response)

Log arithmetic dose against probit response

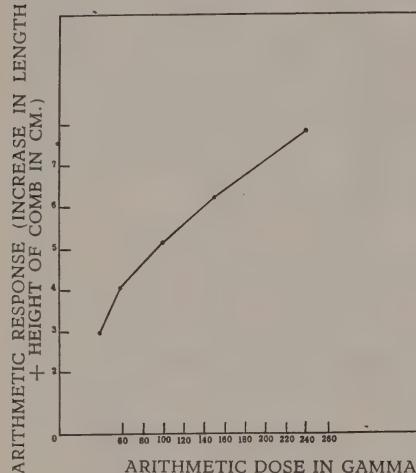


Dosage-response data of Androsterone, using Capons

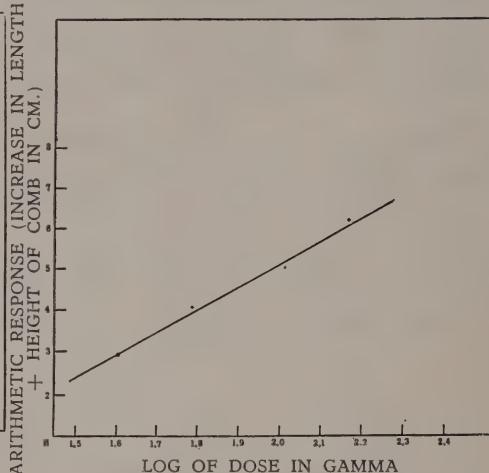
No. of capons	Dose (in γ)	Log dose	Response Length and Height of comb in cm.
10	40	1.6021	2.9
10	60	1.7782	4.0
10	100	2.000	5.1
10	150	2.1761	6.2
10	250	2.3979	7.8

Dosage-response line of Androsterone
(graded response)

Arithmetic dose against arithmetic response

Dosage-response line of Androsterone
(graded response)

Log of dose against arithmetic response



In the case of hormone preparations for which no international standards are available, it is necessary to express the results of bioassays in terms of arbitrary UNITS. Whenever possible these units are expressed as equivalents of a certain weight of the crystalline pure hormone. Thus, one international unit of luteoid activity corresponds to 1 mg. of pure crystalline progesterone; the international unit of testoid activity is 100γ of androsterone; the international unit of folliculoid activity is 0.1γ of estrone, etc.

Finally, it is indispensable that the results of the bioassays be subjected to ANALYSIS BY SUITABLE STATISTICAL METHODS. For this, it is indispensable that the variations in the individual assays be expressed as the "standard deviation" or one of its modifications.

It does not suffice to indicate the range and mean of the individual results, since this precludes the possibility of a QUANTITATIVE MEASUREMENT OF THE VARIATION.

In biologic assays we encounter two types of variation. The *individual variations* are due to the fact that various animals of the same species do not respond in exactly the same manner when exposed to the same treatment. *Aperiodic variations* are those which affect entire groups of animals of the same species, at irregular time intervals. The former are frequently due to hereditary factors, diseases, etc., which affect individual animals, the latter to irregular variations in temperature, diet, climate, season, etc., which affect the entire group of test animals. The former increase the standard deviation of individ-

ual results in single tests, while the latter raise or depress the average response of the entire group receiving the same hormone treatment.

In connection with the statistical evaluation of any biologic finding it is important to emphasize that these can only tell us whether the difference between two groups of results is significant or merely apparent. Thus, if in studying the growth-inhibiting effect of folliculoids most, but not all, the treated animals are smaller than their controls, statistical evaluation of the results can tell us whether the growth inhibition was real or not. It cannot tell us, however, that folliculoids inhibit growth. In the above example it could be possible that the animals treated with the folliculoids were unwittingly also injected with some toxic or infectious material, which was accidentally introduced into the solution, or that their growth was inhibited due to a latent infectious disease, prevalent in their cage but not in that of the controls, etc.

Many serious abuses of statistical evaluation, not only of bioassays but also of other scientific observations, are due to the tendency of experimenters to disregard these self-evident facts and to imply that the evaluation of their data, and not the data themselves, is supported by statistical analysis.

Conversely, it must be kept in mind that lack of statistical significance merely means that we can not prove that our observation *is not* due to chance; this does not mean that we proved it *is* due to chance. Furthermore, not every type of observation must be "statistically significant" in order to be important. Thus single case-reports of rare diseases can be very instructive, although the changes depicted do not lend themselves to statistical analysis. To illustrate this point by a simple, although fantastic, example — if only a single animal would indefinitely survive complete hepatectomy, all our current theories of intermediate metabolism

would have to be revised, although the observation would not be statistically significant.²⁰

TECHNICS OF HORMONE ADMINISTRATION

Route of Administration. — The route of administration exerts a considerable influence upon the activity of hormone preparations. Certain hormones (e.g., insulin, vasopressin, oxytocin, parathyroid hormone, etc.) are ineffective when given BY MOUTH, while others (e.g., thyroid hormone, certain folliculoids and corticoids) are highly effective by this route. Ineffectiveness following oral administration is probably due in some cases to destruction by the digestive enzymes of the gastrointestinal tract, in others, to inactivation by the liver to which orally given hormones are carried directly through the portal vein.

As regards parenteral administration of hormones, it may be said that INTRAVENOUS injections are most rapidly effective, but generally exert more transitory effects, than INTRAMUSCULAR and SUBCUTANEOUS injections.

Certain hormones which are comparatively ineffective when given orally may be administered by direct application to MUCOUS MEMBRANES. In a sense, this is a procedure intermediate between the oral and the intravenous route of administration. From the oral, nasal or tracheal mucous membranes, certain hormones (e.g., adrenaline, steroids, posterior-lobe hormones) are comparatively readily absorbed without much local destruction by the surface epithelium.

Several hormones are readily absorbed through the SKIN (e.g., estradiol, testosterone), but this route of administration is rarely indicated since it is wasteful and does not permit accurate dosage.

Comparatively large amounts of certain hormone preparations can be rapidly absorbed from the PERITONEUM.

Therefore, in the case of water-insoluble hormones, intraperitoneal administration may be the most effective means of ascertaining a sudden and very pronounced increase in the hormone concentration of the blood. This was the basis for the intraperitoneal administration of the water-insoluble steroids for anesthetic purposes. The sudden and pronounced rise in blood steroid concentration necessary for the production of anesthesia is difficult to obtain by other means.

The DIRECT APPLICATION OF HORMONES TO THEIR TARGET ORGANS is especially advantageous, when an exclusively local action is desired. Thus, adrenaline is applied directly to wounds in order to decrease bleeding through its local vasoconstrictor effect; similarly, testosterone applied directly to the capon's comb causes an increase in the weight of this accessory-sex-organ, without influencing the other secondary sex-characteristics of the bird.

The local administration of hormone preparations has the additional advantage that comparatively small doses suffice to produce definite effects. This has often been used to advantage in bioassay technics. Thus, extremely small doses of luteoid hormones can be detected, by the local progestational proliferation in the rabbit endometrium, when they are directly applied to it.

The Solvent. — The solvent in which a hormone is parenterally administered plays an important rôle in determining its efficacy. Certain hormones are much more rapidly absorbed than the solvents in which they are given and since it is time-taking for the organism to extract the hormones selectively from their solvents, activity is delayed by slowly absorbable solvents. It has been shown that the folliculoids (as well as other steroids) can be absorbed selectively from oil, since the hormone concentration of the parenterally administered oil gradually decreases before the solvent disappears

completely. It is because of this that estrone sulphate given subcutaneously, in aqueous solution, has a much more rapid and evanescent effect than when it is given in oily solution. For intravenous administration, only aqueous solutions are to be recommended. Certain water-insoluble steroids have been administered intravenously in other solvents (e.g., glycols), but this is inadvisable, since after dilution with serum, such solutions become unstable and the hormones tend to precipitate. This introduces the danger of hormone-embolisms.

Pellets and Suspensions. — If delayed and very continuous hormone absorption is desired, compressed PELLETS of hormone crystals can be implanted under the skin. The compounds are gradually absorbed from the surface of such pellets and this assures a continuous, steady hormone supply. In cases of eunuchoidism, testoids; in Addison's disease corticoids, have advantageously been administered in this manner. This saves the patient the annoyance of continuous injections and at the same time, reduces the cost of treatment, since comparatively small amounts of hormone suffice to produce a definite effect under these conditions. It must be kept in mind that in the case of repeated hormone administration in readily absorbable form, a great deal of the hormone is wasted, because at certain times, the blood concentration of the substance is excessive and much of it is lost through the excretions, or through excessive destruction in the tissues. Between injections, on the other hand, the hormone concentration tends to fall below the effective level.

The administration of SUSPENSIONS of crystalline or amorphous hormone preparations in fluids in which they are not soluble, represents a modification of the pellet-implantation technic. Its advantage is that by introducing a large number of comparatively small hormone particles, the absorption surface

is increased and more hormone can be assimilated than from pellets, yet the absorption rate remains continuous. If aqueous suspensions are administered, the solvent is rapidly absorbed and the minute hormone deposits come into direct contact with the tissues. In the case of oily suspensions, only a few particles are in actual contact with the absorbing tissues and the mechanism of hormone uptake is somewhat different. In this case, the suspended particles merely assure that the hormone concentration in the oil always remains near the saturation point, in spite of selective absorption of that portion of the hormone which is in solution.

Activation and Inactivation. — Certain hormone preparations become more effective if administered in the form of their ESTERS. Thus, testosterone propionate is more active than testosterone, mainly because hydrolysis of the propionate progresses slowly and hence, esterification delays and prolongs the activity of a single injection. In other instances, esterification activates the hormone because it increases its resistance to destruction when administered orally. Thus, estrone sulfate is more active than estrone when both are given by mouth.

Other hormones are activated by AD-MIXTURE OF CERTAIN SUBSTANCES, without there being any actual compound-formation between the hormone and the activator. Thus, when given simultaneously, certain heavy metals augment the activity of gonadotrophic preparations, zinc increases the effect of insulin, etc. Some hormones are activated by other hormones. In the previously cited instance of progesterone, for instance, pretreatment or simultaneous treatment with folliculoids results in a great increase in activity. Somewhat confusingly, this type of activation has often been described as "sensitization" (see below).

Sensitization and Desensitization.

— The term "SENSITIZATION" is employed to describe an increase in sensitivity, following pretreatment with the same hormone principle. It has been found for instance that some time after spaying, rats loose a great deal of their sensitivity to folliculoid hormones, but a single folliculoid hormone injection suffices to re-sensitize them. This sensitization should be distinguished from the cumulative effect of certain hormone preparations. The latter is merely due to the fact that threshold doses of a slowly acting hormone may not elicit any detectable action in themselves, but if several such doses are administered consecutively, a sufficient hormone concentration is gradually built up in the body to elicit a manifest change. Thyroid hormone, which exerts its actions very slowly, is subject to such cumulative effects following chronic administration. This must be taken into account in determining the adequate dosage for the hormonal treatment of hypothyroidism, since quantities ineffective during the first days of treatment may actually result in overdosage phenomena after sufficient time has elapsed for the cumulation of their actions.

Conversely, DESENSITIZATION to hormone preparations may be accomplished by previous treatment with the same substances. In certain instances, a single injection of a hormone is rapidly followed by brief periods of comparative insensitivity, which has been described as "*tachyphylaxis*." The mechanism of this phenomenon has not yet been fully explained. It may be due to a temporary refractoriness of the target organ, or to a depletion of the body in necessary "co-hormones" or activators, which are essential for the activity of conditionally acting hormones. Vasopressin tachyphylaxis is probably due to the first-mentioned mechanism, while the ineffectiveness of renin, following repeated injections, is presumably due to depletion of the reninactivator (blood-globulin).

ANTI-HORMONE formation to certain endocrine products may also lead to desensitization, as we shall see in the chapter devoted to the anti-hormones.

In some cases the mechanism of desensitization is not known and then the phenomenon is usually designated by the non-committal term of "ADAPTATION." Thus, the somatic-growth-inhibiting effect of moderate doses of folliculoids in the rat, or the sex-skin stimulation by the same hormones in certain monkeys, vanishes after a period of continuous treatment. We do not know, however, through what mechanism this adaptation is effected.

Commercial Hormone Preparations

(Compiled with the kind assistance of Miss Helen Stone, Montreal, Schering Corporation, Bloomfield, New Jersey, Roche-Organon, Nutley, New Jersey and Frank W. Horner Limited, Montreal).

The following is a list of commercially available endocrine products and their distributors. However, almost continually, some products are taken off the market while others are added, hence the list is necessarily incomplete. It is given here mainly to help physicians to identify hormone preparations whose trade-name is not self-explanatory.

I - ADRENALINE

Epinephrine	(U.S.P. XIII.)
Adrenalin	(Parke, Davis)
Adrin	(Sharp & Dohme)
Biosurrenal	(Instituto Opoterapico Nazionale)
Endosurrenal Inhalant	(Harrower)
Epinephrine	(Wilson)
Epinephrine HCl	(Abbott, Biorganic, Boyle, Breon, Bristol, Buffalo, Burroughs Wellcome, Chicago Pharm. Denver Mud, Endo, Estro, Gold Leaf, Gotham, Grant, Harrower, Hart, Ingram, Kremers-Urban, Lakeside, Lederle, Massengill, McNeil, Merrell, Metropolitan, Miller, National Drug, Phys. Drug, Picker, Premo, C. D. Smith, Smith-Dorsey, Solex, Supreme, U. S. Standard, Upjohn, Warren-Teed, Winthrop)
Supraneprin	(Rorer)
Suprarenalin	(Armour)
Suprarenin	(Winthrop)
Tonhormon	(Byk-Gulden)

II - ADRENAL CORTICAL HORMONES

A. DESOXYCORTICOSTERONE ACETATE	
Desoxycorticosterone	(U.S.P. XIII.)
Acetate	
Cortate	(Schering)
Cortenil	(Farben)
Cortiron	(Schering)
Doca	(Roche-Organon)
Neocortine	(Godeau)
Percorten	(Ciba)
Syncortyl	(Herman-Labor)
B. ADRENAL CORTEX EXTRACTS	
Adrenal Cortex	(Armour, Bellevue, Breon, Christina, Endo, Estro, Harrower, Lakeside, Marvell, Pitman-Moore, Prof. Prod., Smith-Dorsey, Upjohn, Wilson)
Adrenal Cortical Ext.	(Connaught)
Adreno-Cortin	(Endocrine, Harrower)
Adrenofer	(Istituto Opoterapico Nazionale)
Cortalex Tabs	(Upjohn)
Cortedrin	(Harvey)
Cortical "C"	(Istituto Opoterapico Nazionale)
Cortical Extract	(Gold Leaf, Kremers-Urban, United Labs)
Cortidyn	(Kretschmer)
Cortin	(Degewop, Roche-Organon)
Cortine Naturelle	(Laroche-Navarron)
Cortinoral	(Harrower)
Cortisorbate	(Reed & Carnick, Schieffelin)
Cortithydrin	(Harrower)
Eschatin	(Parke, Davis)
Glycortal Pills	(Schieffelin)
Interrennin	(Marvell)
Lipo-Adrenal Cortex	(Upjohn)
Novocortex	(Iscovesco)
Suprarenal Cortex	(Armour, Burroughs Wellcome, Carnrick, Lafayette, Massengill, Miller, Pro-Medico, Upjohn, U.S. Standard, Wilson)

III - ANTERIOR PITUITARY HORMONES

A. THYROTROPHIC HORMONE (THYROTROPHIN)	
Pretiron	(Schering)
Thyractin	(Winthrop)
Thyrogan	(British Drug Houses)
Thyrotropic Factor	(Armour, Ayerst McKenna & Harrison)
B. ADRENOTROPHIC HORMONE (CORTICOTROPHIN)	
Adrenotrophic Factor	(Armour, Ayerst McKenna & Harrison)

C. SOMATOTROPHIN AND PREPARATIONS CONTAINING SEVERAL ANTERIOR-LOBE PRINCIPLES IN COMBINATION WITH SOMATOTROPHIN

Accretin	(Harrower)
Antuitrin-G	(Parke, Davis)
Growth Complex	(Armour)
Growth Factor	(Ayerst, McKenna & Harrison)
Phyne	(Wilson)
Pituitary Extract "S" Fraction	(Miller)

D. LACTOGENIC HORMONE (PROLACTIN, LUTEOTROPHIN)

Praelacton	(Promonta)
Prolactin	(Armour, Ayerst McKenna & Harrison, Schering, Squibb)
Suppletan	(Boehringer)

E. HYPOPHYSEAL GONADOTROPHIC EXTRACTS

Accretin	(Harrower)
Ambinon	(Roche-Organon)
Antelobine	(Byla)
Anterior Pituitary Ext.	(Alpinol, A.P.C., Armour, Bellevue, Bishop, Breon, Buffalo, Cole, Direct Sales, Empire, Endo, Ennes, Estro, Fitch, Flint Eaton, Gotham, Haist, Harrower, Harvey Pittenger, Ingram, Kirk, Lakeside, Lilly, Marvell, Massengill, Maury, McNeil, Metropolitan, Miller, National Drug, Parke Davis, Park Drug, Phys. Drug, Pitman-Moore, Premo, Prof. Prod., Pynosol, Sharp & Dohme, Sherman, C. D. Smith, Smith-Dorsey, Squibb, United Labs, United Standard, Upjohn, Verax, Vitarine)
Antex Leo	(Lovens Kemiske Fabrik)
Anteparsine	(Gremy)
Antrone	(Endo)
Entridin	(Endo)
Entromone	(Endo)
Equiphysin	(Harvey)
Gonadophysin	(Searle)
Gonadothyn	(Flint Eaton)
Gonado-Trone	(Miller)
Gonadotrope	(Forbes, Straub)
Gonadotrophic Factor	(Armour, Ayerst McKenna & Harrison, Kirk, Premo)
Gonatin B	(Spanner)
Gynantrin	(Searle)
Hormogen-S	(Mallard)
Lutrogen	(Difco)
Megalin	(Difco)
Phyketrone	(Squibb)
Physex Leo	(Lovens Kemiske Fabrik)
Pituitary Gonadotrophin	(Squibb)
Pituitras-C	(Verax)
Polyansyn	(Armour, Ayerst McKenna & Harrison)
Prephysin	(Chappel, Stearns)
Synapoidin	(Parke Davis)

F. CHORIONIC GONADOTROPHIN (ANT. PITUITARY-LIKE EXTRACTS, HYPOPHYSOID HORMONES OF PREGNANCY URINE OR PLACENTA)

Antegone	(Abbott)
Antuitrin-S	(Parke, Davis)
Apestrin	(Harrower)
Apitor	(Estro)
A.P.L.	(Ayerst, McKenna & Harrison)
Anterior Pituitary-Like Hormone	(Bellevue, Bishop, Pynosol, U.S. Standard)
Asprodin	(Smith-Dorsey)
Choranid	(Smith)
Chorocomon	(Mack)
Chorgon	(Massengill)
Chorigonin	(Lakeside)
Chorione	(Hospital Liquids)
Chorionex	(Horton & Converse)
Chorionic Gonadotrophin	(Armour, Bellevue, Breon, Cole, Empire Drug, Endo, Estro, Hygiene Inc., Kirk, Lakeside, Maury, McNeil, Pitman-Moore, Prof. Prod., Upjohn, United Labs, U.S. Standard)

Choriotropin	(Metropolitan Lab.)
Follutein	(Squibb)
Gestasol	(National Drug)
Glanthropin	(Ingram)
Gonadotrophin	(Flint Eaton)
Gonan	(British Drug Houses)
Gonatrin B	(Spanner)
Korotrin	(Winthrop)
Lyovac Vacules	(Sharp & Dohme)
Marvantrine G	(Marvell)
Neo Apoidin	(Parke Davis)
Placestrin	(Italian Drug)
Praehormon	(Promonta)
Pranturon	(Schering)
Pregenyl	(Degewop)
Pregnyl	(Roche-Organon)
Prolan	(Farben)
G. EQUINE GONADOTROPHIN (PREGNANT MARE SERUM)	
Anteron	(Schering)
Antex	(Ayerst, McKenna & Harrison)
Apoidina	(Parke Davis)
Gestyl	(Roche-Organon)
Gonadin	(Cutter)
Gonadogen	(Upjohn)
Serogan	(British Drug Houses)

IV - POSTERIOR-PITUITARY HORMONES

A. PRESSOR-PLUS OXYTOCIC FACTORS	
Posterior Pituitary	(U.S.P. XIII)
Inj.	
Infundin	(Burroughs Wellcome)
Pitone	(N. V. Organon-Oss)
Pituitrin	(Parke Davis)
Post-Hypophyse	(Choay)
Posterior Pituitary desic.	(Armour, Lilly, Parke Davis, Wilson)
Posterior Pituitary Extract	(Abbott, A.B.C., Alpinol, Armour, Bellevue, Bishop, Blue Line, Breon, Bristol, Buffalo, Cheplin, Chicago Pharm., Christina, Cole, Endo, Estro, Flint Eaton, Frosst, Gold Leaf, Haist, Harrower, Harvey, Horton & Converse, Ingram, Intra Prod., Kremers-Urban, Lakeside, Lilly, Mallard, Marvell, Massengill, McNeil, Merrell, Metropolitan, Miller, National Drug, Parke Davis, Phys. Drug, Pitman-Moore, Premo, Prof. Prod., Rorer, Sharp & Dohme, Sherman, C.D. Smith, Smith-Dorsey, Solex, Squibb, Torigian, Tosse, United Labs, Upjohn, U.S. Standard, Verax, Vitarine, Warner, Warren-Teed, Wilson, Wyeth)
Posterior Pituitary Ob & Surg.	(Miller)
B. PRESSOR FACTOR (β -HYPOPHAMINE, VASOPRESSIN)	
Pitressin	(Parke Davis)
Pitressin Tannate	(Parke Davis)
Tonephin	(Farben)
Vaso-Pituitgan	(Henning)
C. OXYTOCIC FACTOR (α -HYPOPHAMINE, OXYTOCIN)	
Hypophysin	(Bayer)
Neo-Pituitairine	(Optima)
Physormon	(Promonta)
Pitocin	(Parke Davis)
Pituglandol	(Roche)
Pituitgan	(Henning)
Pituitgan forte	(Henning)
Orastin	(Farben)

V - FOLLICULOIDS

1) FOLLICULOID HORMONES AND THEIR ESTERS, ESTROGENIC HORMONES

a. Estradiol	(U.S.P. XIII)
Estradiol	(National Drug)
Aquadiol	(Barrington)
Bar-Estro	(Barrington)
Bar-Estro-A	(Barrington)
Dimenformon	(Roche-Organon)
Estradiol " suspension	(Brothers Pharm.)
Estrinol	(Maurty, Verax)
Estrogenic Hormone	(Cabot)
Estrogenic suspension	(Southern Med.)
Estrovin	(Bio-Intrasol)
Gynoestrol	(Rousel, Herman-Labor)
Macro-Pellets	(Cabot)
Ovasterol	(Frosst)
Ovestrin	(Reed)
Ovoglylin	(Ciba)
Progynon-DH	(Schering)

Progynon-Micropellets	(Schering)
Estradiol + Estrogenic Substance	(Metropolitan)
Estradiol suspension	(McNeil)
Estrogenic substance	(Endo)
Femacrine	(Phys. Drug)
Hormegen	(Kurt)
Kurtestrin	(Durst)
Mikrogen	(Vitamix)
Viestra	(Lakeside)
Estradiol + Estrone	b. Estradiol Benzoate
Compestrin	(U.S.P. XIII)
Endocreme	(Byla)
Estrogenic Hormone	(Hirestra Labs)
Estradiol-trimethyl acetic acid ester	(Cole)
Under	
Estradiol Benzoate	
Benzhormovarine	(Godeau)
Difollisterol	(Roche-Organon)
Dimenformon Benzoate	(Italian Drug)
Ectofoliculina	(Horton & Converse)
Estradiol Benzoate	(Ciba)
Fenacyclin	(Degewop)
Follitulin-Menformon	(British Drug Houses)
Oestroform	(Hoffman LaRoche)
Oestroglandol	(Frosst)
Ovasterol B	(Loven Kemiske Fabrik)
Ovex	(Boehringer)
Ovoglylin Benzoate	(Ciba)
Perlatan	(Schering)
Progynon-B	(Farben)
c. Estradiol Dipropionate	
Dimenformon Dipropionate	(Roche-Organon)
Diprovex	(Byla)
Estradiol Dipropionate	(Verax)
Ovoglylin-Dipropionate	(Ciba)
Progynon-DP	(Schering)
d. Ethynod Estradiol	
Ethynd Estradiol	(Under consideration for U.S.P. XIV)
Estinyl	(Schering)
Eticylol	(Ciba)
Lynoral	(Roche-Organon)
Ronol	(Horner)
e. Estrone	
Estrone	(U.S.P. XIII)
Estrone	(Abbott, Brothers Pharm., Christina, Lilly, Medi-Synth, Miller, National Drug, Warren-Teed)
Estrugenone	(National)
Hiestrene	(High)
Theelin	(Parke Davis)
Thixol	(Horton & Converse)
Estrone, Aqueous	
Aquacrine, suspension	(Endocrine)
Estrone, suspension	(Abbott, Breon, Christina, Lilly)
Theelin, suspension	(Parke Davis)
f. Estrone Sulfate	
Conestron	(Wyeth)
Linesterol	(Lincoln Prop.)
Oestrilin	(Desbergers)
Premarin	(Ayerst McKenna & Harrison)
Questerin Compound	(Frosst)
g. Estriol	
Estriol	(Abbott, Lilly)
Theelol	(Parke Davis)
h. Estriol glucuronidate	
Emmenin	(Ayerst McKenna & Harrison)
Emmenoplex	(Ayerst McKenna & Harrison)
i. Estrogenic Substances (mixed)	
Estrogenic Substances	(Under consideration for U.S.P. XIV.)
Amniotin	(Squibb)
Carrion's	(Fougera)
Cavestragen	(Cavendish)
Cyrestrone	(Winthrop-Stearns)
Di-Folliculine	(Union Chimique Belge)
"Meurice"	
Estras	(Verax)
Estrinol	(Bellevue)
Estrofol	(Premo)
Estrogen-Miller	(Miller)
Estrogenic Cartridges	(Southern Med.)
Estrogenic Hormone	(Reed & Carnick)

Estrogenic Substance (A.P.C.) Ayerst McKenna & Harrison, Barry, Biorganic Labs, Boericke Runyon, Boyle, Breon, Bristol, Buffington, California Pharm., Cheplin, Chicago Pharm., Cole, Crevules, Direct Sales, Drug Prod., Duffield, Elder, Empire Drug, Endocrine, Estro, Eton, Farnsworth, Forbes, Gotham, Haist, Harrower, Harvey, Horton & Converse, Kirk, Lakeside, Lederle, Lincoln, Maitine, Maury, Medi-Synth, Merrell, McNeil, Miller, Morris, National Drug, Premo, Prof. Prod., Pynosal, Reed & Carnrick, Rorer, Sharp & Dohme, Sherman, Smith-Dorsey, Southern Med., Straub, United Labs, U.S. Standard, Warren-Teed, Wilson, W'eth)

Estrogenol (Chemico)
Estrogyn (Estro)
Estro-Hart (Hart Drug)
Estrolin (Lakeside)
Estromone (Endo)
Estronat (National Drug)
Estrophorol (Cabot)
Estropalex (Sonoral)
Estrotoron (Pitman-Moore)
Estrovarin (Warren-Teed)
Estrovin (Bio-Intrasol)
Estrovite (Vitarine)
Estrageneone (Kremers-Urban)
Estrusol (C.D. Smith)
Femestrone (Kirk)
Folestrin (Armour)
Follacor (Schieffelin)
Follestrtol (Blue Line)
Follicomone (Premo)
Follidrin (Astra)
Folliplex (Roland)
Follugen (Gotham)
Folumone (Endocrine)
Genestrolin (Christina)
Gynestrol (Penick)
Harvestrin (Harvey)
Hormestrin (Miller)
Hormobex (Lakeside)
Hormotone-T (Carnrick)
Hormovarine (Byla)
Kal-Estrin (Medi-Synth)
Kestrone (Buffington)
Lentabs (Lederle)
Managen (Parke Davis)
Menestrin (Harris Prod.)
Menformon (Roche-Organon)
Met-Estrin (Metropolitan)
Natestrin (Upjohn)
Neo-Amniotin (Squibb)
Novestrine (Gremy)
Ova-Estrin (Hospital Liquids)
Ovarian Substances (A.P.C., Cole, Flint Eaton, Hildebrand, Hynson, Lafayette, Marvell, Purity, Rorer, Sharp & Dohme, Sherman, Smith-Dorsey, United Labs, U.S. Standard, Zemmer)
Ovestrin (Amer. Parentrasol)
Ovestrinolin (Reed Pharm.)
Ovhormone (Alpinol)
Ovifollin (Nyegaard)
Ovogen (Solex)
Plestrin (Harrower)
Prolagen (Drug Prod.)
Prolicalin (Merrell)
Semestrin (Massengill)
Thelestrin (Carnrick)
Tri-Estrin (Endocrine)
Urestrin (Upjohn)
Varium (Burroughs Wellcome)

2) ARTIFICIAL FOLLICULOIDS DIETHYLSILBESTROL

(Δ -3,4-di-(p-hydroxyphenyl)-hexane)

a. Diethylstilbestrol (U.S.P. XIII)
Des (Grant Chem.)
Ayren B (Farben)
Diestrene (Frost)
Dietabs (Bever)
Diethylol (Merrell)
Diethylstilbestrol (Abbott, A.B.C., Alpinol, Anglo-Canadian, Armour, Barlow-Maney, Bates, Bio-Intrasol, Blue Line, Boyle, Breon, Bristol, Buffalo, Buffington, Burroughs Wellcome, Carnrick, Chicago Pharm., Ciba, Cole, Columbus Pharm, Cowley, Daniels, Drug Prod., Endo, Ennes, Estro, Gold Leaf, Harco, Harrower, Harvey, Hildebrand, Horner, Horton & Converse, Ingram, Intra Prod., Kremers-Urban, Lakeside, Lederle, Lilly, Massengill, Maury,

McNeil, Merck, Merrell, Miller, Penick, Pitman-Moore, Premo, Purity, Rorer, Schieffelin, Seaver, Sharp & Dohme, Sherman, C.D. Smith, Smith-Dorsey, Solex, Squibb, Stearns, Strasenburgh, Torigian, Upjohn, U.S. Standard, Van Pelt & Brown, Veray, Warner, Warren-Teed, Wendt, Bristol, Winthrop, Wyeth, Zemmer.)

Dioestrol (Frost)
Distilbene (Borne)
Endestrol (Endocrine)
Estrobone (Ayerst McKenna & Harrison)
Estrol Cream (Estrol)
Estrosyn (Buffington)
Glo-Bestrol (Globe)
Meteostrol (Metro)
Milestrol (Miller)
Neo-Oestranol (Crookes)
Sibol (Beebe, Bristol, Chicago Pharm., Daniels)
Stibulum (Desbergers)
Stilbestralp (Alpinol)
Stilbestrol (A.P.C., Dagra, Phys. Drug, British Drug Houses).
Synthogen (National Drug)
b. *Benzestrol* (2,4-di(p-hydroxyphenyl)-3-ethyl hexane)
Benzestrol (Lederle, Schieffelin)
Cycloestrol (Bruneau)
c. *Dienestrol* (3,4-di(p-hydroxyphenyl hexadiene-2,4); dehydrodiethylstilbestrol)
Cycladiene (Bruneau)
Dienesterol (Biophady, British Drug Houses, Frosst, Ortho, Rare, White)
Novoestrol (Mees)
d. *Hexestrol* (3,4-di(p-hydroxyphenyl)-n-hexane)
Dioestrol-DH (Frost)
Hexesterol (British Drug Houses, Cowley, Direct Sales, Horton & Converse, Lanteen, Massengill, Merrell, Ortho, Penick, Prof. Prod., Rorer, Vitamins Inc.)
e. *Dimethyl Hexestrol Dipropionate*
Meprane (Reed & Carnrick)
f. *Hexesterol + Phenobarbital*
Hexesterol Phenobarbital (Buffalo, Columbus Pharm., Lanteen, Massengill, Phys. Drug)
Hexestrofen (Merrell)
Hexital (Ortho Prods.)
Thelesterol (Carnrick)
g. *Diethylstilbestrol Monomethyl Ether*
Monomestrol (Wallace & Tierman)
h. *Diethylstilbestrol Dipalmitate*
Stilpalmitate (Abbott)
i. *Diethylstilbestrol Dipropionate*
Diethylstilbestrol Dipropionate (Blue Line, Breon, Burroughs Wellcome, Lakeside, Penick, Winthrop)
Estrobene-DP (Ayerst McKenna & Harrison)
Stilbestronate (Blue Line)
Stilbestrol Dipropionate (Burroughs Wellcome)

VI ~ LUTEOIDS

1) PROGESTERONE AND CORPUS LUTEUM EXTRACTS

<i>Progesterone</i>	(U.S.P. XIII)
<i>Corlutin</i>	(Pro. Medicus)
<i>Corlutone</i>	(Gold Leaf)
<i>Corpolutin</i>	(Astra)
<i>Corpomone</i>	(Premo)
<i>Corpus Luteum Ext.</i>	(Abbott, A.P.C., Armour, Bristol, Cole, Drug Prod., Endo, Haist, Harrower, Hosp. Liquids, Lafayette, Lakeside, Lilly, Mallard, Marvell, Merrell, National Drug, Pitman-Moore, Purity, Rorer, Sharp & Dohme, Sherman, Shores, Smith-Dorsey, Stoddard, Wilson, Wyeth, Zemmer)
<i>Engestin</i>	(Harrower)
<i>Gestormone</i>	(Godeau)
<i>Glanestin</i>	(Ingram)
<i>Homoflavine</i>	(Byla)
<i>Lipo-Lutin</i>	(Parke Davis)
<i>Luteal</i>	(Istituto Opoterapico Nazionale)
<i>Lutein Ext.</i>	(Hynson, Westcott & Dunning)
<i>Luteocrin</i>	(Harrower)
<i>Luteoglandol</i>	(Hoffman LaRoche, Kretschmar)

THE HISTORY OF ENDOCRINOLOGY

37

<i>Luteolin</i>	(Nyegaard)	<i>Parathyroid Gland</i> (Burroughs Wellcome)
<i>Luteolipex</i>	(Kretschmar)	<i>Parathyroid Hormone</i> (A.P.C., Armour, Lilly,
<i>Luteosan</i>	(Kretschmar)	Parke Davis, Squibb, United Labs, Wilson)
<i>Luteotransann</i>	(Kretschmar)	<i>Paroидин</i> (Parke Davis)
<i>Lvthorn</i>	(Upjohn)	
<i>Lutocycline</i>	(Ciba)	IX — TESTOIDS (MALE SEX HORMONES, ANDROGENS)
<i>Lutocyclin</i>	(Ciba)	
<i>Lutogestrin</i>	(Solex)	A. CRYSTALLINE TESTOSTERONE AND ITS DERIVATIVES
<i>Lutogyl</i>	(Rousel, Herman-Labor)	<i>a. Testosterone</i>
<i>Lutolin</i>	(Spanner)	<i>Aceto-Sterandryl</i> (Rousel, Herman-Labor)
<i>Lutren</i>	(Farben)	<i>Hormogen C</i> (Mallard)
<i>Lutromone</i>	(Endo)	<i>Male Sex Hormone</i> (Hoosier Pharm.)
<i>Lutrone</i>	(Buffington)	<i>Oreton F</i> (Schering)
<i>Nalutron</i>	(Winthrop)	<i>Perandrene</i> (Ciba)
<i>Progesterol</i>	(Cabot, C.D. Smith)	<i>Testin</i> (Nyegaard)
<i>Progesterone</i> (Abbott, A.B.C., Alpinol, Armour, Ayerst McKenna & Harrison, Bellevue, Biorganic, Bishop, Boyle, Breon, Cabot, Carnrick, Chemico, Chicago Pharm., Cowley, Dalton, Denver Mud, Direct Sales, Drug Prod., Empire, Endocrine, Ennes Pharm., Estro, Fitch, Flint Eaton, Forbes, Frost, Glidden, Gotham, Harrower, Hart Drug, Hema, Horton & Converse, Intra Prod., Kings County, Kremers-Urban, Lakeside, Leader, Lederle, Lilly, Loyal, Massengill, Maury, McNeil, Medi-Synth, Metropolitan, Miller, Morris, Parentasol, Phys. Drug, Prof. Prod, Reed & Carnrick, Schieffelin, Sherman, Squibb, Torigian, United Labs, Upjohn, Warren-Teed)	<i>Testodrin</i> (Astra)	
<i>Progrestin</i>	(Desbergers)	<i>Testosterone</i> (Ayerst McKenna & "Lingusors" Harrison)
<i>Progrestin</i> (Abbott, Barrington, Bio-Intrasol, Buffington, Drug Prod., Flint Eaton, Gotham, Harrower, Hospital Liquids, Kirk, Lakeside, Lilly, Medi-Synth, National, Purity, Pynosol, Roche Organon, Smith-Dorsey, U.S. Standard, Yale)	<i>Testosterone</i> (Biorganic, Cabot, Chicago Pharm., Duffield, Estro, Gotham, Loyal, Morris, Phys. Drug, Vitamix)	
<i>Progrestine</i>	(N.V. Organon-Oss)	b. Methyl-testosterone
<i>Progrestone</i>	(Carnrick)	<i>Methyl-testosterone</i> (U.S.P. XIII)
<i>Progesteron</i>	(Pitman-Moore)	<i>Glosso-Sterandryl</i> (Rousel)
<i>Progistas</i>	(Verax)	<i>Metandren</i> (Ciba)
<i>Progonasyl</i>	(Progonasyl)	<i>Metosterone</i> (Ayerst McKenna & Harrison)
<i>Prolatol</i>	(Schering)	<i>Methyl-Testosterone</i> (Armour, Parke Davis, Phys. Drug, Rare, Upjohn, Southern Med., Vitamix)
2) PREGNENINOLONE		<i>Neo-Hombreol-M</i> (Roche-Organon)
Anhydroxydroxy- progesterone	(U.S.P. XIII)	<i>Orchisterone-M</i> (Frost)
<i>Lutocyclol</i>	(Ciba)	<i>Oreton-M</i> (Schering)
<i>Pranone</i>	(Schering)	
<i>Progesterol</i>	(Roche-Organon)	c. Testosterone propionate
<i>Progesterin</i>	(Frost)	<i>Testosterone</i> (U.S.P. XIII) propionate
3) ETHYNYL-ANDROSTENEDIOL		<i>Androtestone</i> (Godeau)
<i>Sabacol</i>	(Horner)	<i>Ludin</i> (Harrower)
4) PROGESTERONE + ESTRADIOL BENZOATE		<i>Orchisterone-P</i> (Frost)
<i>Di-Pro</i>	(Roche-Organon)	<i>Neo-Hombreol-P</i> (Roche-Organon)
<i>Progestriol</i>	(Ciba)	<i>Oreton</i> (Schering)
<i>Prometron</i>	(Schering)	<i>Perandrene</i> (Ciba)
5) PROGESTERONE + ESTROGENIC SUBSTANCE		<i>Testhormone</i> (Alpinol)
<i>Cyclogesterin</i>	(Upjohn)	<i>Testosterone propionate</i> (Armour, Chicago Pharm., Denver Mud, Direct Sales, Estro, Gotham, Parke Davis, Phys. Drug, Rare, Vitamix)
<i>Estrogen & Progesterone</i>	(Breon, Empire, Smith-Dorsey)	<i>Testodrin</i> (Astra)
<i>Estrone-Progesterone</i>	(Miller)	<i>Testostinate</i> (Cabot)
<i>Estroplex</i>	(Sonoral)	<i>Virosterone</i> (Endo)
<i>Estrotate</i>	(Lakeside)	
<i>Femestro-Lutin</i>	(Kirk)	d. Testosterone, Aqueous
<i>Procyco</i>	(Metropolitan)	<i>Aqueous Testrone</i> (Miller)
<i>Proculin</i>	(Cabot)	<i>Malogen</i> (Testagar)
<i>Pro-Estrin</i>	(Vitamix)	<i>Testacrine</i> (Columbus)
<i>Progenol</i>	(Metropolitan)	<i>Testosterone</i> , (Endo) suspension
<i>Promegen</i>	(Phys. Drug)	
VII — a. INSULIN, REGULAR, CRYSTALLINE OR NON-CRYSTALLINE		B. TESTICULAR EXTRACTS
<i>Insulin Injection</i>	(U.S.P. XIII)	<i>Androgenic Hormone</i> (Chicago Pharm., Pynosol)
<i>Iletin</i>	(Lilly)	<i>Androgenol</i> (Chemico Lab.)
<i>Insulin</i> (Bayer, Connaught, Degewop, Mack, Mulford, Searle, Sharp & Dohme, Squibb, Ulm)		<i>Andropex</i> (Christina)
b. INSULIN, MODIFIED		<i>Androstin</i> (Ciba)
<i>Protamin Zinc</i>	(U.S.P. XIII)	<i>Andrusol</i> (Smith)
<i>Insulin Inj.</i>		<i>Cavotestrone</i> (Cavendish)
<i>Globin Insulin with Zinc</i>	(Burroughs Wellcome, Squibb)	<i>Glandular Comp. Male</i> (Carnrick)
<i>Globin Insulin</i>	(Lilly)	<i>Gleydig</i> (Schiefflin)
<i>Protamin Zinc Iletin</i>	(Lilly)	<i>Homovir</i> (Anglo-French)
<i>Protamin Zinc Insulin</i>	(Sharp & Dohme, Squibb)	<i>Lypocin</i> (Stoddard)
<i>Zinc Iletin Crys.</i>	(Lilly, Sharp & Dohme)	<i>Male Sex Hormone</i> (Bellevue)
<i>Zinc Insulin Crys.</i>	(Squibb)	<i>Malestrome</i> (Kirk)
VIII — PARATHYROID HORMONE		<i>Natural Orchic</i> (Miller)
<i>Parathyroid Injection</i>	(U.S.P. XIII)	<i>Orchiber</i> (Marvell)
<i>Hytakerol</i>	(Winthrop)	<i>Orchic Concentrate</i> (Drug Prod., U.S. Standard)
<i>Parathyroid Ext.</i>	(Armour, Lilly, Wilson)	<i>Orchic Extract</i> (Lafayette, Lakeside, National Drug, Wilson)

X — THYROID HORMONE

A. THYROID EXTRACT

Thyroid (.17-.23% (U.S.P. XIII)
iodine)

Desi-Thyroid

(Frosst)

Elyractin

(Boyer)

Endothyrin

(Harrower)

Glythoid Pills

(Schieffelin)

Ityphen

(Straus)

Proloid

(Maltine)

Thygentabs

(Carnrick)

Thyracloids

(Reed & Carnrick)

Thyracitin

(Winthrop)

Thyranon

(Roche-Organon)

Thyrobrom

(Van Patten)

Thyroid (Abbott, A.B.C., A.P.C., Armour, Artaco, Bates, Bellevue, Biorganic, Bishop, Blue Line, Boyle, Breon, Bristol, Buffalo, Buffington, Burroughs Wellcome, Carnrick, Cheplin, Chicago Pharm. Clinic Line, Daniels, Drug Prod., Endo, Endocrine, Harco, Harrower, Harvey, Hildebrand, Horton & Converse, Hynson Westcott & Dunning, Jamieson, Kremers-Urban, Kretschmar, Lakeside,

Lederle, Lilly, Massengill, Maury, McNeil, Merrill, Metro, Miller, National Drug, Park Drugs, Parke Davis, Petroline, Pitman-Moore, Premo, Prof. Prod., Purity, Rorer, Schering, Schieffelin, Seal, Seaver, Sharp & Dohme, Sherman, Smith-Dorsey, Solex, Squibb, Stayner, Stoddard, Success, Supreme, Testagar, United Labs, Upjohn, Van Pelt & Browne, Viobin, Warren-Teed, Wendt Bristol, Wilson, Wyeth)

Thyroidal (C.D. Smith)
Thyronal (Desbergers)
Thyroprotein (Parke Davis)

B. THYROXIN (64% Iodine)
Thyroxin (U.S.P. XIII)
Thyroxine (British Drug Houses, Roche-Organon, Roche-Schering-Henning, Squibb)
Thyroxin Fraction (Squibb)

C. DI-IODO-TYROSINE
Di-Iodo-Tyrosine (Roche-Organon)
Elityran (Farben)
Thyreoidin (Merck)
Thyreoglandol (Hennig-LaRoche)
Thyrowop (Degewop)

THE HISTORY OF ENDOCRINOLOGY

A good deal can be learned from the manner in which endocrinologic discoveries were made in the past, since problems similar to those which have already been solved present themselves to us now and will probably arise again in the future. In choosing our problems and in preparing our plans for medical research, it is useful to profit by the experience of our predecessors. History can teach us to recognize promising and important subjects at an early date, by revealing the form under which such problems appeared, much before their implications were fully understood and proven.

This brings us to the most fundamental question an investigator must consider: what is an important research problem? I believe that experimental medicine is not an abstract, but an applied science. Admittedly, in the early stages, it is often difficult to foretell whether a problem will have important applications to practical medicine, but at least we should select our problems keeping in mind that practical applicability is their most noble goal. Medical research undertaken merely as a sophisticated type of mental gymnastics — a viewpoint often defended by the slogan "science for science's own sake" — appears unworthy of the traditions of the medical profession, whose primary aim has always been, and should remain, to help the sick.

In choosing a topic for medical research I have found it useful to be guided by the adage: "*The most important problem is that which means most to most people.*" Applying this dictum to endocrinology, the best problems for investigation would be those most likely, directly or indirectly, to help in the therapy of the most common and serious diseases. It must not be disregarded, however, that investigations which lend themselves par-

ticularly well to generalizations and the formulation of laws applicable to a large number of endocrine processes, are also of great import in accordance with our maxim. Even tentative generalizations help to formulate hypotheses and theories which preliminarily connect a number of apparently unrelated and inexplicable facts. The history of endocrinology clearly shows that the *horror of interpretative thought* — which developed as a reaction against the purely speculative, dialectic approach to medicine prevalent in past centuries — is unjustified. It is always advantageous to connect cognate facts through a preliminary hypothesis. Interrelations — so important in endocrinology — can only be elucidated after we have sketched a temporary plan which coordinates the known facts as well as it is possible on the basis of the knowledge available at the time. The history of endocrinology has clearly demonstrated, furthermore, that even incorrect theories are often of great help in unveiling the secrets of nature, as long as we regard them merely as concrete formulations of possibilities, which, by virtue of their concreteness, lend themselves to be proven or disproven by subsequent observation. The hypothetical nature of such theories, or of any link in a chain of theoretic interrelations, must always be clearly emphasized, however, and the investigator must be prepared to change his theories without any feeling of remorse or resentment, as soon as new facts require a modification. The hazy outlines of indistinct concepts become sharp when they crystallize into a definite theory, and it is only then that we can subject their correctness to experimental proof.

It is an unfortunate fad among many contemporary scientists to limit publications to the mere registration of observations, spurn-

ing their interpretation as vain verbiage. It must be pointed out that the listing of facts is not science. By definition, science is "accumulated knowledge systematized and formulated with reference to the discovery of general truths or the operation of general laws" (Webster's Dictionary). The mere enumeration of facts is undoubtedly the "safest" procedure but an author who chooses this technic for the publication of his results must realize that he contributes nothing to science until a bolder colleague attempts their evaluation at the risk of being blamed for a possible error.

If, for a moment, I may abuse of my privileged position as an author who has no editor to put him in his place, I would also like to mention that most of the pleasure which the investigator derives from his work lies in the "artistic" interpretation of his observations. In music, for instance, single tones have no artistic value; melodies result only if tones are connected in a certain order; the more sophisticated musician will even want to harmonize the melody by supporting notes and several independent melodies must be coördinated to create counterpoint. Very much the same can be said about the various degrees of refinement in the artistic enjoyment and evaluation of scientific discoveries. If we substitute facts for notes and coördination of several facts in the theoretic connection of a "chain of events" for melodies, then the elucidation of independent factors influencing any one link in the chain of events would correspond to chords. To create counterpoint we have to integrate several independent chains of events which, at certain points, coöperate for the good of the organism. (Note to students: please forgive this, I promise you will not be "held responsible" for it at examination time!)

Endocrinology as we know it to-day is a young science hardly more than fifty years of age. It would be difficult, however, to determine exactly when and where the thought has originated that there is an integration of organs through humoral means.

In an historic survey it is almost inevitable that great discoveries are connected with the names of certain individuals. We must realize, however, that the historian of wars, as that of peaceful endeavors, selects most of his "heroes" more or less arbitrarily. When we review the story of an accomplishment it is almost impossible to determine in retrospect exactly what the hero's exploits were, and to what extent the work of those whose names are pointed out to posterity were dependent upon the accomplishments of the many "unknown soldiers" who went before them.

Some harmonious and lawful coöperation, between the different organs of the body, has been suspected since times immemorial and was designated as the "consensus partium."

It has long been anticipated furthermore that certain organs contain substances (or "spirits" or "potentialities") which exert beneficial actions when introduced into the body. It is on the basis of such indistinct concepts that in ancient medicine, the organs of animals, or even of enemies killed in battle, have been ingested in order to give strength and courage or to cure diseases. One of the oldest medical texts, the Egyptian *Papyrus of Eber*, enumerates many organ extracts among the 700 drugs which it discusses. Such ORGANO-THERAPY has also been advocated by the Greek philosopher Aristotle (384-322 B.C.), and the Roman Pliny the Elder (23-79 A.D.) devoted all of books XXVIII-XXXII of his Natural History to "Materia Medica," an enumeration of medicines derived from the bodies of men and animals.

Paracelsus (1493-1541) (his true name was Theophrastus Bombastus von Hohenheim) a Swiss physician, often described as the father of pharmaceutic chemistry, was apparently the first, however, to justify such practices by a scientific hypothesis, characterized by his slogan "Similia similibus curantur," according to which a diseased organ is best cured by administration of a similar organ. Thus, we arrive at a fairly clear formulation of SUBSTITUTION THERAPY.

The therapeutic administration of animal organs, and organ extracts, constantly gained in popularity. According to Winkler, the pharmacies of Innsbruck (Austria) still carried 122 official preparations of this type, as late as 1765. Even human organ extracts, such as "Cranium humanum preparatum" or "Oleum cranii humani" were prepared from the bodies of executed men and distributed through the pharmacies.

At this time, we are still very far from the true understanding of ENDOCRINE ACTIVITY AS A PHYSIOLOGIC PROCESS WHICH HELPS TO CORRELATE the parts of the body. The integration of the activities of distant organs has chiefly been considered as a function of the nervous system ever since the French philosopher *Descartes*, in the 17th century, and later the Viennese physiologist *Prochaska* (1784) developed the concept of nervous reflexes as the "reflexion of sensory into motor impulses."

New possibilities of integration were raised through the discovery by the English physician *Harvey* (1628) of the blood circulation.

In his treatise "Analyse Médicinale du Sang" (1775), *Théophile de Bordeu* of Montpellier, expressed the view that every organ produces specific substances which enter into the blood and that these are useful to the organism. He also suspected that deficiency symptoms after castration might be due to a failure of humoral substances produced by the sex glands. He expressed his premonition, furthermore, that ANOMALIES IN HUMORAL SECRETIONS PLAY AN IMPORTANT RÔLE IN PATHOLOGY by saying that "C'est au médecin à suivre et à classer les divers reflux qui surviennent par la faute de chaque organe en particulier." (It is up to physicians to follow and classify the divers ebbs which supervene due to failure of each particular organ).

FROM A PURELY MORPHOLOGIC VIEWPOINT, most glands of internal secretions have been known for a very long time. Only a few have been discovered comparatively recently, e.g., the adrenals, by *Eustachius* (1563) in his treatise "de Glandulis Quae Renibus Incumbunt," and the parathyroids by *Sandström* (1879). It was not clearly understood, however, that these organs are specialized for the production of internal secretions.

In his textbook of physiology (1844) the German physiologist, *Johannes Müller* described as "ganglia sanguineo-vasculosa" the thyroid, the adrenals, the spleen and the placenta. He states that these ductless glands: "exert a plastic influence upon the humors which circulate through them and return from them to the general circulation; they have no relations with the exterior, as have the other glands."

However,* all the evidence cited above was based either upon mere speculation, or only upon the interpretation of the morphologic structure of endocrine glands. *John Hunter* in England, probably around 1771, and *A. A. Berthold*, (Germany) in 1849, were apparently the first to demonstrate EXPERIMENTALLY that the virilizing effect of the testis is actually due to an endocrine activity. They found that castrated cocks retained their normal male appearance, libido, fighting instincts, comb and wattles — that is, they remained masculine — if the testes were re-implanted into a different part of the body. From this, Berthold concluded that the "consensus" in question is due to the influence of the testes upon the blood, which, then, in its turn, influences the organism in general. It is noteworthy that the first proof for an "internal secretion" was furnished by the elimination of all other possibilities. All other connections of the transplanted testes (nerves, ducts, etc.) having been severed, the glands could act only by influencing the blood brought to them by the invading vessels.

These early observations were forgotten because a few subsequent investigators failed to confirm them. The theory of an internal secretion did not receive general attention until the great French physiologist, *Claude Bernard* (1813-78) first clearly expressed the thought that in addition to the "sécrétions externes" of the ordinary glands, all organs produce a "sécrétion interne" through which they influence the "milieu intérieur" whose composition they help to maintain invariable (1857). Hence, we

must credit Bernard, not only for clearly formulating the concept of endocrine activity, and for creating the now current term "internal secretion," but also for having first expressed one of the most fundamental physiologic laws, that of the importance of maintaining the normal composition of the "milieu intérieur," the body humors.

He failed to differentiate, however, between actual hormone production, as we now understand it, and the discharge into the blood of metabolites, nutrients or even the blood cells themselves. Among the purely endocrine glands, he listed the adrenal, the thyroid, the spleen, thymus and lymph nodes; among the partially-endocrine organs, the liver, which secretes glucose into the blood, and the lungs, which oxygenate it.

The actual birthday of endocrinology is traditionally listed as June 1st, 1889, when the distinguished French physician Brown-Séquard, then 72 years old, made his now historic communication to the Société de Biologie de Paris. He reported a truly astonishing degree of rejuvenation after having treated himself with subcutaneous injections of a Pasteur-filtered, aqueous, dog-testis suspension. However, on the basis of what we now know about the chemical properties of testis hormone, his extracts could not possibly have contained a sufficient amount to produce any detectable effect. Testosterone, the hormone in question, is comparatively insoluble in water and its concentration in the testis is so low that an adequate dosage could never be injected in the form of a crude suspension. Nevertheless, the venerable old gentleman described the process of his alleged rejuvenation, in its most intimate details, with so much satisfaction and contagious enthusiasm that the medical world began to take an active interest in the possibilities of endocrinology. It must be admitted, furthermore, that he so clearly outlined the scope of the endocrines as an independent integration-mechanism separate from the nervous system, that he gave that final impetus to medical thinking

along these lines which was necessary to establish endocrinology as a science. He said : "Nous admettons que chaque tissu et plus généralement, chaque cellule de l'organisme, secrète pour son propre compte, des produits ou des ferment spéciaux, qui sont versés dans le sang et qui viennent influencer, par l'intermédiaire de ce liquide, toutes les autres cellules rendues ainsi solidaires les unes des autres par un mécanisme autre que le système nerveux."

It is especially noteworthy that at the time of Brown-Séquard's famous lecture, the meticulous observations and the well-founded deductions of Claude Bernard were already known, as were the deductions of anatomists and histologists who postulated an internal secretion on the basis of morphologic evidence. We might add that in England Thomas Addison (1855) had already described the disease which now bears his name; Hilton-Fagge (1871) spoke of sporadic cretinism due to absence of the thyroid and W.-W. Gull (1873) pictured spontaneous myxedema in adults; the brothers Reverdin (1879) in Switzerland noted "post-operative myxedema" following thyroideectomy for goiter. — Any one of these earlier discoveries could have been the cradle of endocrinology, but it is an historic fact that they were not. Apparently the strictly endocrinologic implications of these discoveries were not described with sufficient poignancy to interest the medical profession in the basic concepts of endocrinology. It is somewhat humiliating to note that our science began with an error due to subjective interpretation of observations and no one will condone Brown-Séquard's lack of objectivity. Yet, there is a lesson to be learned from his paper by those so fearful of "over-dramatizing" their observations that they write drab and monotonous papers, "catalogues of facts" without an effort of synthetic interpretation and evaluation.

These are the salient historic events in the evolution of the concept of internal secretion. Additional data concerning individual glands will be found in the "Historic Introduction" to the corresponding sections.

REFERENCES

ALBEAUX-FERNET, M.: *Les Hormones en Thérapeutique*. 2e Ed. Legrand et Cie., Publ., Paris (1948).

A textbook (353 pages, numerous figures, charts and tables, no references) mainly written for the practising endocrinologist. (In French.)

ALBEAUX-FERNET, M., J. BROUET-SAINTON, F. COSTE, J. DECOURT, J. C. DREYFUS: *Traité de Médecine*. Tome XIII. Maladies des Glandes

Endocrines. Masson et Cie., Publ., Paris (1948).

An extensive treatise (1119 pages, numerous figures, few references) compiled by several clinical endocrinologists, based mainly on clinical experience. (In French.)

ASCHNER, B.: *Dic Blutdrüsenerkrankungen des Weibes*. J. B. Bergmann, Publ., Wiesbaden (1918).

Merely of historic interest.

BARKER, L.-F. (Ed.) : *Endocrinology and Metabolism. Presented in their scientific and practical clinical aspects by ninety-eight contributors.* 5 volumes. D. Appleton & Company, Publ., New York (1922).

The first encyclopedic treatise (4770 pages, numerous illustrations and references) on endocrinology in the English language. The book is of historic interest.

BAUER, J. : *Innere Sekretion, ihre Physiologie, Pathologie und Klinik.* J. Springer, Publ., Berlin & Wien (1927).

Merely of historic interest.

BEACH, F. A.: *Hormones and Behavior.* A survey of Interrelationships between Endocrine Secretions and Patterns of Overt Response. P. B. Hoeber, Publ., New York (1948).

An excellent monograph (368 pages, 852 references) summarizing the literature and the author's extensive personal experience concerning influence of hormones upon the more complex patterns of animal behaviour. Highly recommended.

BERBLINGER, W., C. CLAUBERG AND E. J. KRAUS : *Die Bedeutung der inneren Sekretion für die Frauenheilkunde.* In Stoeckel, W. Handbuch der Gynakologie. 9, (1936). J. F. Bergmann, Publ., München (1936).

A treatise (1107 pages, 305 illustrations, numerous references) concerning gynecologic endocrinology. (In German.)

BEST, C. H. AND N. B. TAYLOR : *The physiological basis of medical practice.* 4th Ed. The Williams & Wilkins Company, Publ., Baltimore (1945).

A textbook of physiology (1169 pages, 497 figures, 83 tables, 2 plates, numerous references) containing a brief, but very adequate section on endocrinology.

BIEDL, A. : *Innere Sekretion. Ihre physiologischen Grundlagen und ihre Bedeutung für die Pathologie.* 4th Ed. Urban & Schwarzenberg, Publ., Berlin (1922).

One of the first comprehensive textbooks of endocrinology (818 pages, 44 figures and numerous references). It lists almost the entire endocrine literature up to 1922 and is a treatise of great historic interest. (In German.)

BOMSKOV, C. : *Methodik der Hormonforschung.* Volume I, 1937; volume II, 1939. Georg Thieme, Publ., Leipzig (1937 & 1939).

A treatise (1732 pages, 525 illustrations and numerous references) in two large volumes, intended to review all methods useful in hormone research. Now largely outdated.

BROWN, SIR W. L. : *The endocrines in general medicine.* London (1927).

Merely of historic interest.

BROWN, SIR W. L. : *The integration of the endocrine system.* Cambridge (1935).

Mainly of historic interest.

CAMERON, A. T. : *Recent advances in endocrinology.* 5th Ed. The Blakiston Company, Publ., Philadelphia (1945).

An outline (415 pages, 73 figures, numerous references) of endocrinology for medical students and physicians. The first edition (1933) was continuously brought up-to-date in subsequent years, but since the book was never completely rewritten, it became difficult to maintain its uniform structure.

DEL CASTILLO, E. B., J. R. MEMBRIVES, F. A. DE LA BALZE AND C. G. MAININI : *Endocrinología clínica.* Librería y editorial "El Ateneo." Publ., Buenos Aires (1944).

An excellent textbook (560 pages, 94 figures, numerous references) mainly concerned with the clinical problems of endocrinology. (In Spanish.)

CAWADIAS, A. P. : *Clinical Endocrinology and Constitutional Medicine.* F. Muller, Publ., London (1947).

A textbook (362 pages, several drawings and references) which in the description of factual matters is frequently at variance with currently accepted views and lays much emphasis upon philosophical problems.

COBB, I. G. : *The organs of internal secretion.* 4th Ed. Baillière, Tindall & Cox, Publ., London (1933).

Mainly of historic interest.

COLD SPRING HARBOR SYMPOSIA ON QUANTITATIVE BIOLOGY (BY SEVERAL AUTHORS). Volume X, *The relation of hormones to development.* The Biological Laboratory, Publ., Cold Spring Harbor, L. I., N.Y. (1942).

Proceedings of a symposium on the rôle of endocrines in growth and differentiation (167 pages, numerous illustrations and references); it contains 18 papers by leading authorities in this field.

COUNCIL ON PHARMACY AND CHEMISTRY OF THE AMERICAN MEDICAL ASSOCIATION (various authors). *Glandular physiology and therapy.* American Medical Association, Publ., Chicago (1942). — Spanish translation by F. F. de Eandi : *Glandulas Endocrinas su fisiología y Terapéutica.* Editorial Futuro, Publ., Buenos Aires (1944).

An outline (571 pages, numerous references, no illustrations) consisting of 31 papers by various authors, in which the most important problems of endocrinology are concisely outlined. It acts mainly as a guide to the pertinent literature.

COOPER, E. R. A.: *The histology of the more important human endocrine organs at various ages.* London (1925).

Merely of historic interest.

COUNCIL ON PHARMACY AND CHEMISTRY OF THE AMERICAN MEDICAL ASSOCIATION: *New and Nonofficial Remedies, 1946.* American Medical Association, Publ., Chicago (1946).

A book (770 pages) listing those drugs (including hormones) which the Council on Pharmacy and Chemistry has found acceptable. In general, standard drugs described in the United States Pharmacopeia are not included, so that this volume (known as N. N. R.) complements the latter. The book also contains a description of various bioassay methods, units, and details for the preparation of recommended hormone products.

CURSCHMANN, S.: *Endocrine disorders.* London (1929).

Merely of historic interest.

DODDS, E. C. AND F. DICKENS: *The chemical and physiological properties of the internal secretions.* Oxford University Press, Publ., Oxford (1925).

A textbook (214 pages, numerous figures and references) of historic interest.

DUNCAN, G. G. (ED.): *Diseases of metabolism. Detailed methods of diagnosis and treatment.* W. B. Saunders Company, Publ., Philadelphia (1942).

A treatise (985 pages, 158 figures, numerous tables and references) containing very authoritative sections concerning the rôle of the hormones upon metabolism and its diseases. The chapters on diabetes mellitus, diabetes insipidus, hyperinsulinism, obesity, as well as those on carbohydrate, protein, lipid, mineral and water metabolism are particularly instructive for the endocrinologist.

ENGELBACH, W.: *Endocrine medicine.* C. C. Thomas, Publ., Springfield (1932).

An extensive treatise (1912 pages, 933 illustrations, numerous references) in three volumes and an index volume. The book contains an enormous amount of information such as anthropologic measurements as influenced by age or endocrinotrophies, numerous case reports, etc.; although the theoretic interpretations are often very subjective, it is still of more than historic interest.

FALTA, W.: *Die Erkrankungen der Blutdrüsen.* In Bergmann, G. and R. Staehelin's *Handbuch der inneren Medizin 4, 1035* (1927). 2nd Ed. Julius Springer, Publ., Berlin (1927).

A monograph (361 pages, 43 illustrations, numerous references) containing excellent descriptions of the endocrine diseases, by one of the great masters in this field. In spite of its date the volume is still of more than historic interest.

GEY, E.: *Les grands problèmes de l'endocrinologie.* Librairie J.-B. Baillière et fils, Publ., Paris (1926).

An outline (178 pages, 13 figures, few references) by one of the great masters of endocrinology; now merely of historic interest. (In French.)

GEY, E.: *The internal secretions, their physiology and application to pathology.* (Translated from the French.) P. B. Hoeber, Publ., New York (1918).

Merely of historic interest.

GOLDZIEHER, MAX A.: *The endocrine glands.* Rudolf Shick Publishing Company, Publ., New York (1945).

A booklet (49 pages) which accompanies a set of endocrine wall charts designed for teaching purposes.

GOODMAN, L. AND A. GILMAN: *The pharmacological basis of therapeutics.* The Macmillan Company, Publ., New York (1941).

A textbook of pharmacology (1383 pages, 126 figures, 67 tables, numerous references) which is a valuable source of information concerning the pharmacodynamics aspects of hormones.

GOULD, G. M. AND W. L. PYLE: *Anomalies and curiosities of medicine.* Sydenham, Publ., New York (1937).

An instructive and amusingly written treatise (968 pages, 295 illustrations, 747 references) describing rare and curious diseases, many of which are of endocrinologic interest.

GREENBLATT, R. B.: *Office Endocrinology.* 3rd Ed. Charles C. Thomas, Publ., Springfield (1947).

An outline (313 pages, few figures and references) intended for the practitioner.

GREGORY, J.: *A B C of the Endocrines.* Williams & Wilkins Company, Publ., Baltimore (1935).

An outline (126 pages, numerous illustrations, 65 references) of endocrinology, mainly based on highly simplified, but amusing charts and cartoons.

GROLLMAN, A.: *Essentials of Endocrinology.* 2nd. Ed. J. B. Lippincott Co., Publ., Philadelphia (1947).

An excellent textbook (644 pages, 132 illustrations and a selection of key references) highly recommendable to medical students and physicians.

HAMBLEN, E. C.: *Endocrinology of woman.* Charles C. Thomas, Publ., Springfield (1945).

An authoritative textbook (571 pages, 157 figures, numerous references) primarily devoted to endocrine problems in gynecology; it also contains a clear and concise summary of other aspects of endocrinology.

HARROW, B. AND C. P. SHERWIN : *The chemistry of the hormones*. Baillière, Tindall & Cox, Publ., London (1934).

Mainly of historic interest.

HARROWER, H. R. : *Endocrine Diagnostic Charts*. The Harrower Laboratory, Inc., Publ., Glendale, California (1929).

A brief outline (144 pages, no illustrations or references) of endocrine diagnosis, mainly in the form of synoptic charts.

HEREDIA, P. : *Tratado de Endocrinología*. 2 Volumes. Editorial Vasquez, Publ., Buenos Aires (1947).

An extensive treatise (1742 pages, 319 figures and charts, numerous references) principally concerned with clinical problems. (In Spanish.)

HIRSCH, M. (Ed.) : *Handbuch der inneren Sekretion. Eine umfassende Darstellung der Anatomie, Physiologie und Pathologie der Endokrinen Drusen*. C. Kabiszsch, Publ., Leipzig (1932).

An encyclopedia (5714 pages, 1105 illustrations, numerous references) in five large volumes which treats both the theoretic and clinical aspect of endocrinology. The various chapters are written by different authors and are of very variable quality. Unfortunately, many of the references are incomplete, so that the book does not serve as a good guide to the literature.

HOSKINS, R. G. : *Endocrinology, the glands and their functions*. W. W. Norton & Company, Inc. Publ., New York (1941).

An amusingly written outline (388 pages, few illustrations and references) of endocrinology which may serve as an introduction to this subject.

HOUSSAY, B. A., J. T. LEWIS, O. ORIAS, E. HUG, E. B. MENÉNDEZ AND V. G. FOGLIA : *Fisiología humana*. Libreria y editorial "El Ateneo". Publ., Buenos Aires (1946).

A textbook of physiology (1343 pages, 497 figures, 82 tables, numerous references) containing a particularly instructive summary of endocrinology. (In Spanish.)

LAQUEUR, E., G. E. DE JONGH, M. TAUSK, J. H. GAARENSTROOM, M. B. C. MANUS : *Hormonologie*. Physiologie en Pharmacologie van de Hormonen. N. V. Noord-Hollandsche Uitgevers Maatschappij, Publ., Amsterdam (1948).

An excellent textbook (485 pages, few illustrations and references) compiled by leading Dutch endocrinologists. (In Dutch.)

LEREBOUILLÉT, P., P. HARVIER, A. C. GUILLAUME, M. CARRION, J. CHABRUN AND BARIÉTY : *IX — Sympathique et Glandes Endocrines. Traité de Pathologie Médicale et de Thérapeutique appliquée*. Publié sous la direc-

tion de Emile Sergeant, L. Ribadeau-Dumas et L. Babonneix.

A treatise (570 pages, 25 figures, no references) mainly designed to outline the relationships between the endocrine glands and the sympathetic nervous system. (In French.)

LIPSCHUTZ, A. : *The internal Secretions of the Sex Glands*. W. Heffer & Sons Ltd., Publ., Cambridge (1924).

A textbook of experimental endocrinology (513 pages, 142 figures, numerous references) which is now merely of historic interest.

LOESER, A. : *Hormontherapie*. S. Hirzel, Publ., Leipzig (1947).

A brief outline (152 pages, 7 illustrations, key references) concerning the pharmacology of the hormones. German trade preparations are considered almost exclusively. (In German.)

LOEWENBERG, S. A. : *Clinical Endocrinology*. 2nd Ed. F. A. Davis Company, Publ., Philadelphia (1941).

A voluminous textbook (883 pages, 194 figures, 37 charts and tables, few references) describing the diseases of the endocrines mainly on the basis of the author's own experience.

LUTZ, W., H. W. SIEMENS AND J. STRANDBERG : *Vererbung innere Sekretion Stoffwechsel*. J. Jadassohn's Handbuch der Haut- und Geschlechtskrankheiten. 3, (1929). Verl. von Julius Springer, Berlin (1929).

A monograph (389 pages, 57 figures, numerous references) which deals mainly with the rôle of hormones in dermatology. (In German.)

MARTIN, L. and M. HYNES : *Clinical Endocrinology*. J. & A. Churchill, Publ., London (1948).

A brief booklet (222 pages, 8 plates, 22 figures, key references) principally concerned with problems of clinical diagnosis and therapy.

MOULTON, F. R. (Ed.) : *The chemistry and physiology of hormones*. American Association for the Advancement of Science, Publ. (1944).

The proceedings (243 pages, numerous illustrations and 1200 references) of a research conference. It is a most instructive up-to-date summary, concerning the chemistry of the hormones, compiled by 17 leading authorities in this field.

PARTHON, C. I. AND M. GOLDSTEIN : *Traité d'Endocrinologie*. Jassy "Viata Romineasca". S. A. Publ., (1923).

A textbook of endocrinology (467 pages, 17 figures, no references) which is now merely of historic interest. (In French.)

PENDE, N.: *Endocrinologia. Patologia e clinica degli organi a secrezione interna.* Parte I. e II. 3rd Ed. Dottor Francesco Vallardi, Publ., Milano (1923, 1924).

A textbook of endocrinology (1172 pages, 141 figures, 24 tables, no references) in two volumes; the first deals with theoretic, the second with clinical aspects. The book was one of the first large treatises on this subject. (In Italian.)

PINCUS, G. and K. V. THIMANN (ED.): *The Hormones. Physiology, Chemistry and Applications.* Academic Press Inc. Publ., New York (1948).

The first volume (886 pages, numerous tables and references, few figures) is a series of excellent papers by highly competent specialists, but without any attempt to correlate the chapters into an integrated synopsis. The chapters on hormones in plants and invertebrates (195 pages) probably represent the best synopsis of this field now available. Clinical problems are not discussed although they will receive a limited amount of space in the second volume, which is now in preparation.

PUGSLEY, L. I.: *The application of the principles of statistical analysis to the biological assay of hormones.* Endocrinology, 39, 161 (1946).

A most useful article (15 pages, 7 illustrations, 14 references) concerning statistical considerations in the evaluation of hormone assays. It also contains a valuable list of monographs dealing with biologic statistics.

RAAB, W.: *Innersekretorische Störungen und Organotherapie.* Berlin (1932).

Merely of historic interest. (In German.)

REISS, M.: *Die Hormonforschung und ihre Methoden.* Urban & Schwarzenberg, Publ., Berlin and Wien, (1934).

A textbook (415 pages, 26 figures, few references) concerning technics and methods of endocrinology. Mainly based on the author's own experience. (In German.)

RIDDLE, O.: *Endocrines and Constitution,* in Doves and Pigeons. Carnegie Institution of Washington Publication 572, Washington (1947).

Summary (306 pages, 7 illustrations, many charts and tables, numerous references) of the author's extensive personal investigations concerning the influence of race, sex and genetic constitution upon the development of endocrines in certain birds. Recommended only for specialists.

ROLLESTON, H. D.: *The endocrine organs in health and disease with an historical review.* Oxford University Press, Publ., Oxford, (1936).

A textbook (521 pages, numerous references) mainly devoted to clinical problems

in endocrinology. 35 of the 45 figures depict prominent endocrinologists, most of whom exhibit no overt endocrinopathy. The book is now somewhat outdated, but especially the historic sections still provide interesting and instructive reading.

ROWE, A. W.: *The differential diagnosis of endocrine disorders.* London (1933).

Merely of historic interest.

SAINTON, P., H. SIMONNET AND L. BROUHA: *Endocrinologie. Clinique, thérapeutique et expérimentale.* 2nd Ed. Masson et Cie., Publ., Paris (1942).

A textbook (912 pages, 216 figures, no references) which contains an extraordinarily large number of data, but places almost equal emphasis upon doubtful and fully established observations; hence it is most useful to those having sufficient experience to evaluate the data for themselves. (In French.)

SAMUELS, J.: *Endogenous Endocrinotherapy,* including the Causal Cure of Cancer. Holdert & Co., Publ., Amsterdam (1947).

An astonishing opus (539 pages, 30 figures, 324 references) in which the author comes to the conclusion that "by restoring the balance of the pituitary gland, tumour growth is arrested and the patient saved".

SELYE, H.: *Encyclopedia of Endocrinology.* Section I, The Steroids. Volumes 1-4. A. W. T. Franks, Publ., Montreal (1943). Section IV. Volumes 7 and 7 references. Ovarian Tumors, Richardson, Bond & Wright, Publ., Montreal (1946).

An encyclopedic treatise planned eventually to cover the whole field of endocrinology. Section I (in 4 volumes) The Steroids (728 pages, formulae and synoptic charts) deals with the chemistry and pharmacology of the steroids. Of section IV (The Ovary) only the Ovarian Tumors (in 2 volumes) have been completed (876 pages, numerous illustrations and more than 15000 references). The individual parts are self-contained monographs independent of the other sections which have not yet been published. The encyclopedia is edited in looseleaf form and intended to act as a critical guide to the entire endocrine literature. Additional pages will be printed in order to keep the work up-to-date.

SEVRINGHAUS, E. L.: Endocrinology in The 1945 year book of neurology, psychiatry and endocrinology. The Year Book Publishers, Chicago (1946).

An outline (332 pages, 31 figures, few references) summarizing the most important recent advances in clinical endocrinology; partly based upon case reports.

SEVRINGHAUS, E. L.: Endocrine Therapy in General Practice. The Year Book Publishers, Inc., Publ., Chicago (1942).

A small outline of clinical endocrinology (243 pages, 47 illustrations, no references) mainly intended for the practitioner.

SÉZARY, A. AND M. LABBÉ (ED.) : *Glandes endocrines. Maladie de la Nutrition.* (By various authors). In A. Laffont and F. Durieux's *Encyclopédie médico-chirurgicale*. Masson et Cie., Publ., Paris (1935).

A voluminous loose-leaf tome concerned primarily with the clinical problems of endocrinology. It will constantly be brought up-to-date by the addition of new loose-leaf sheets. (In French.)

SÉZARY, A. ET J. LENÈGRE: *Précis de Pathologie Médicale.* Tome VIII. *Maladies Endocrinianes.* Masson et Cie., Publ., Paris (1948).

A textbook (714 pages, 155 figures, no references) dealing mainly with clinical problems of endocrinology. (In French.)

SHARPEY-SCHAFFER, E. A.: *The endocrine organs.* 2 volumes, 2nd Ed., Longmans, Green & Co., London (1924).

Merely of historic interest.

STOCKARD, C. R.: *The genetic and endocrinian basis for differences in form and behavior.* Wistar Institute of Anatomy and Biology, Publ., Philadelphia (1941).

A classic treatise (775 pages, 128 figures, 113 plates, numerous tables and references) concerning the rôle of heredity in endocrinology.

TRENDELENBURG, P.: *Die Hormone, ihre Physiologie und Pathologie.* 2 volumes, Berlin (1929, 1934).

Merely of historic interest.

TURNER, C. D.: *General Endocrinology.* W. B. Saunders Co., Publ., Philadelphia (1948).

An excellent textbook (604 pages, 164 charts and figures, 1367 references) written mainly from the zoologist's point of view. The section on endocrines in invertebrates (73 pages), and several other often neglected fields of theoretical endocrinology, are especially well written, but clinical problems are given little prominence (e.g., myxedema, 1 page; diabetes mellitus, 2 pages).

TWOMBLY, G. H. AND G. T. PACK (ED.): *Endocrinology of Neoplastic Diseases.* A Symposium by Eighteen Authors. Oxford University Press, Publ., New York (1947).

A symposium (392 pages, numerous charts and figures, many references) on the relationship between hormones and neoplastic disease. It represents a good summary of this field, but is slightly outdated.

UNITED STATES PHARMACOPOEIAL CONVENTION: *The Pharmacopoeia of the United States of America.* Twelfth Revision. Board of

Trustees, Publ., Mack Printing Company, Easton (1942).

This book (880 pages, numerous tables) as well as the British Pharmacopoeia, represent authoritative manuals describing the characteristics of those drugs (including hormones) whose utility is most definitely established. It also contains sections on: reference standards, international standards, patented and trade-mark products and regulations governing the manufacture and sale of drugs.

VALLERY-RADOT, P. J., J. HAMBURGER ET F. L'HERMITTE: *Pathologie Médicale.* Livre quatrième: *Le Diabète.* Livre cinquième: *Glandes Endocrines.* Flammarion et Cie., Publ., Paris (1948).

An extensive treatise of medicine (few figures and references) by prominent French clinicians, in which two sections are devoted to diabetes (60 pages) and other endocrine problems (128 pages) respectively. (In French.)

VIÉGAS, A. PINTO: *Endocrinologia Clínica.* Livraria Editora Paulo Bluhm, Belo Horizonte, 1941.

A textbook (292 pages, 53 illustrations, few references) mainly intended as a guide for the clinical endocrinologist. (In Portuguese.)

VINCENT, S.: *Internal secretions and the hormonal glands.* 3rd Ed., Edward Arnold & Co., Publ., London (1924).

Merely of historic interest.

WERNER, A. A.: *Endocrinology. Clinical application and treatment.* 2nd. Ed., Lea & Febiger, Publ., Philadelphia (1942).

A textbook (923 pages, 327 illustrations, numerous references) of clinical endocrinology primarily for the practicing physician.

WOLF, W.: *Endocrinology in modern practice.* 2nd Ed. (1939).

A voluminous textbook primarily intended for practitioners.

YATER, W. M.: *Fundamentals of internal medicine.* 2nd. Ed., D. Appleton-Century Company, Inc., Publ., New York (1944).

A textbook of internal medicine (1204 pages, 275 figures, numerous tables and references) in which the diseases of the endocrines and of metabolism are particularly clearly described.

ZONDEK, H.: *The diseases of the endocrine glands.* 4th Ed. Edward Arnold & Co. Publ., London (1944).

A textbook (496 pages, 180 figures, numerous references) in which rather prominent emphasis is placed upon inadequately investigated subjects and upon the author's personal observations.

SPECIAL ENDOCRINOLOGY

I THE STEROIDS

INTRODUCTION

A special section of this book is devoted to a general discussion of the steroids because of the close chemical, physiologic, pharmacologic and metabolic relationships which exist between the individual members of this group.

The chemical and pharmacologic terminology of the steroids; the classification of the compounds themselves and of their actions; the interrelations between the chemical structure and the pharmacologic activity of steroids are all subjects which best lend themselves to synoptic discussion.

It will also be kept in mind that the same steroid hormone may be elaborated by several endocrines (e.g., gonads, adrenal cortex and placenta) hence, many steroids, as well as their metabolites, cannot be regarded as the specific products of any one endocrine gland. Furthermore, the overlap between the pharmacologic actions of chemically different steroids is so considerable that it appears best to devote a separate section to the interrelations which exist between the members of this group.

In the sections dealing with the individual, steroid-producing endocrine glands, namely the Ovary, Testis, Adrenals and Placenta (see : Pregnancy), the reader will find additional data concerning those steroid hormones which are particularly characteristic of the activity of any one of these organs.

In the present section we wish to deal mainly with the following subjects :

(1) Occurrence and rôle of the steroids in nature.

- (2) Chemical terminology and classification of the steroids.
- (3) Pharmacologic terminology and classification of the steroids.
- (4) Interrelations between the various pharmacologic properties of the steroids (pharmaco-pharmacologic interrelations).
- (5) Interrelations between the pharmacologic properties and the chemical structure of the steroids (pharmaco-chemical interrelations).
- (6) Biogenesis and metabolism of the steroids.

OCCURRENCE AND RÔLE OF THE STEROIDS IN NATURE

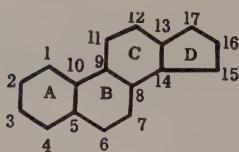
The steroids are derivatives of the hydrocarbon cyclopentanoperhydrophenanthrene, sometimes also referred to as norestrane (see below). Derivatives of this basic four ring compound are very widely distributed in nature and many of them are characterized by outstanding physiologic and pharmacologic effects.

THE NATURALLY OCCURRING GROUPS OF THE STEROIDS are :

- (1) Animal sterols (Zoösterols).
- (2) Plant sterols (phytosterols).
- (3) Bile acids.
- (4) Steroid hormones.
- (5) Odoriferous steroids.
- (6) Cardiac aglucones.
- (7) Sapogenins (e.g., Digitalis sapogenins).
- (8) Genins of the toad venoms.

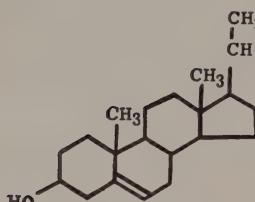
The "EVOCATOR" substances, emitted by certain "organizer" tissues for structural organization in early embryonic life are also claimed to behave like steroids.

Numbering of carbon atoms and lettering
of rings in parent hydrocarbon of the
steroids

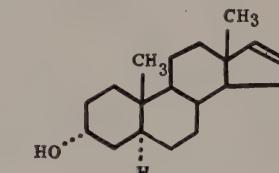


Cyclopentanoperhydrophenanthrene
(Norestrane)

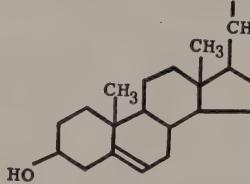
Representatives of the naturally occurring groups of steroids



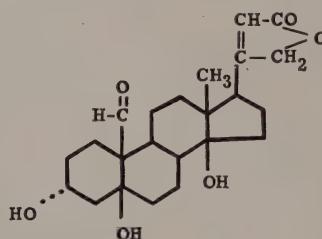
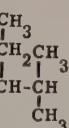
1. — Cholesterol
(Zoosterols)



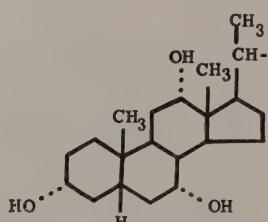
5. — Δ^{16} -Androstene-3 (α)-ol
(Odoriferous substances of testis)



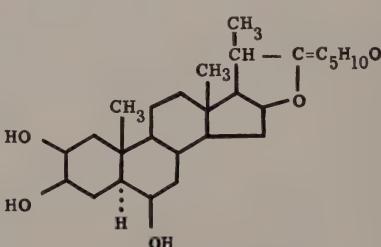
2. — Stigmasterol
(Phytosterols)



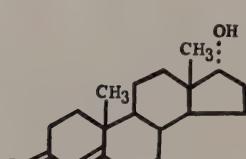
6. — Strophanthidin
(Cardiac Aglucones)



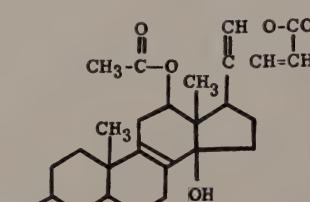
3. — Cholic Acid
(Bile Acids)



7. — Digitogenin
(Digitalis Sapogenins)



4. — Testosterone
(Steroid Hormones)



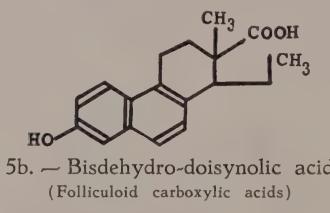
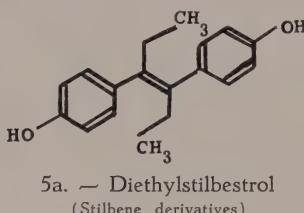
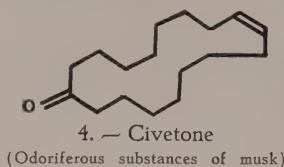
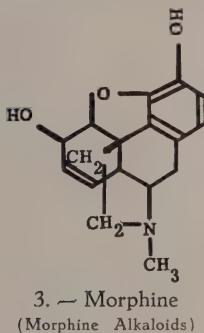
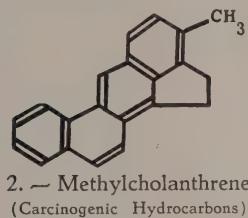
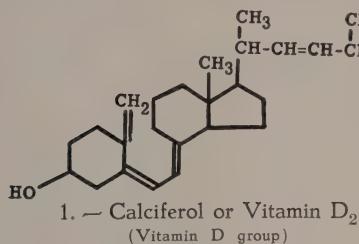
8. — Cinobufagin
(Genins of Toad Venoms)

A number of other biologically important compounds are RELATED TO THE STEROIDS, although the original four ring system of norestrane is not maintained, for instance:

- (1) The vitamin D group.
- (2) Carcinogenic hydrocarbons.

- (3) Morphine alkaloids (incl. codeine and its derivatives).
- (4) Odoriferous substances of musk.
- (5) Artificial hormone-like substances (a. stilbene derivatives, b. folliculoid carboxylic acids).

Representatives of biologically important groups of compounds related to the steroids



larity between these and some of the constituents of musk which are responsible for its odor (Civetone).

The metabolism of the steroid hormones proceeds along somewhat different lines even in closely related vertebrates. Thus the metabolic end-products of estradiol are different even in so closely related species as the rat and rabbit. Yet steroids show no definite species specificity as regards their pharmacologic actions. For instance, estradiol stimulates the growth of the female accessory sex organs — while testosterone promotes the development of the male genitalia — in fish, amphibia, reptiles, birds, as well as in mammals.

Even if we limit our considerations to the comparatively small group of the

Representatives of the steroid hormones, themselves, occur in all the vertebrates and probably even in some invertebrates, plants and microbes. While zoösterols and phytosterols manifestly fulfil important functions in the lives of animals and plants, the biologic significance of steroid hormones has not been adequately studied, as yet, except in vertebrates.

Some steroids isolated from the testis proved to be odoriferous (musk-like odor) and there is a structural simi-

steroid hormones and their immediate derivatives, the variety of their functions in nature is truly astonishing. Such diverse phenomena are dependent upon the activities of steroid hormones as the pre- and postnatal growth and function of the accessory reproductive organs, the development of the ovaries and testes during the earliest stages of embryonic life, the striking nuptial coloring which certain fishes assume during the breeding season, the picturesque, sex-linked plumage pattern of many birds, the beard growth, baldness and muscular development of virile men, the crowing and the fighting instinct of the rooster, the woman's breast and the sexual drive. But as we shall see in the following sections, not only sex, but the development and metabolism of the entire body, as well as its resistance and adaptability to exposure and disease, are influenced by the steroid hormones of the gonads, the adrenal cortex and the placenta.

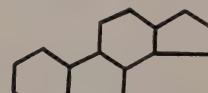
CHEMICAL TERMINOLOGY AND CLASSIFICATION OF THE STEROID HORMONES

There are several systems according to which the steroids can be named and classified with respect to their chemical structure; none of these is completely satisfactory. The procedure followed in this book was selected to conform primarily with the requirements of the endocrinologist. It lists all compounds as substitution products of the parent nuclear hydrocarbons. This helps to compile each in a group, the biologically important derivatives of androstane and estratriene, segregating them from other groups of lesser pharmacologic significance (etiocholane derivatives, D.homo-androstane, pyroandrostane, urane).

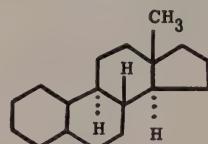
The Parent Nuclear Hydrocarbons.

— The parent compounds of all the steroids are saturated nuclear hydrocarbons, that is they consist exclusively of carbon and hydrogen and contain

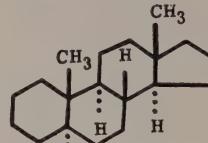
no double bonds. Depending upon the presence or absence of so-called angular methyl groups at C₁₀ and C₁₃, and depending upon the relative steric (spatial) position of the four rings, a number of such nuclear hydrocarbons is possible. From the endocrinologist's point of view, the following are of the greatest importance :



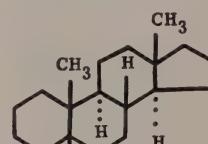
1. Norestrane



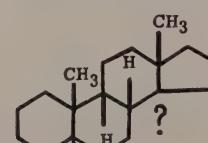
2. Estrane



3. Androstane



4. Etiocholane



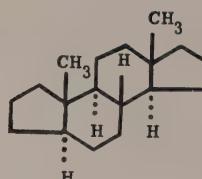
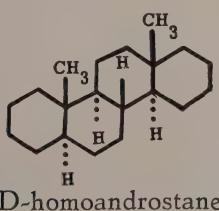
5. Urane
(9-epietiocholane)

The relative spatial position of two adjacent rings is difficult to show on a plain paper sheet, hence, it is customary to indicate it by the position of the hydrogen atom attached to the carbon atom which is common to two adjacent

rings. If this is assumed to be behind the plane of the paper it is called " α " and is shown attached to the nucleus by a dotted line; in the reverse case it is designated by the letter " β "—and shown attached to the nucleus by a solid line. The other hydrogens on the nucleus — which do not deserve special attention — are customarily omitted from the formula.

It will be kept in mind that in the molecule of norestrane all 17 carbon atoms are saturated with hydrogen. This means that 2 hydrogen atoms are attached to each carbon, except those (5, 10, 8, 9, 13, 14) which are common to adjacent rings. These carry only a single hydrogen whose spatial position is dependent upon that of the two rings. (See : Nuclear isomerism, below.)

It is evident that in addition to the five nuclear parent hydrocarbons mentioned above, several other steric variations are possible; these are not mentioned here since they are of no obvious biologic significance. It will also be kept in mind that if an additional carbon is introduced into the five membered ring D (thus transforming it into a six membered ring) or if one carbon is removed from the six membered ring A, thus transforming it into a five membered (pentane) ring, additional parent nuclear hydrocarbons can be prepared by partial synthesis but these will likewise not be discussed in this book since they are not proven to be naturally occurring. Suffice it merely to mention D-homoandrostan e and pyroandrostan e, the parent compounds of the series with a six-membered ring D and a five-membered ring A, respectively.

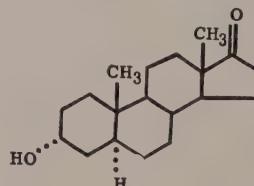


Numeration and Nomenclature of Substituents.—

1. THE NUCLEUS. As illustrated above, the letters A, B, C. and D designate the four rings of the cyclopentanoperhydrophenanthrene skeleton and the numbers 1 to 17, the nuclear carbon atoms. The carbons of the angular methyl groups at C₁₀ and C₁₃ are labelled respectively 19 and 18.

Alcoholic ($-OH$) and ketonic ($=O$) substituents in the nucleus are designated by the suffixes " $-ol$ " and " $-one$ ", respectively, preceded by the number of the carbon atom bearing the function :

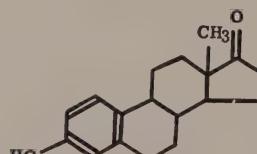
e.g., *androstane-3(α)-ol-17-one* :



Numeral indices, following the sign Δ , indicate the position of double bonds in the nucleus; where the linkage extends between carbon atoms not consecutively numbered, both numbers are recorded :

e.g., *estrone* =

$\Delta^{1,8,5:10}$ -*estratriene-3-ol-17-one* :



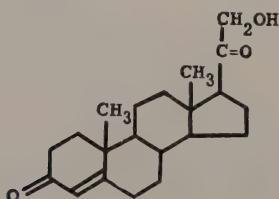
It will be noted that functional groups attached to a benzenoid ring are neces-

sarily in the same plane as the latter, hence in the above example for instance the — OH group is neither α , nor β .

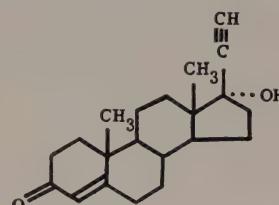
Saturated, nuclear hydrocarbons and their derivatives are designated by terms ending in “-ane”, while unsaturated derivatives are designated with terms ending in “-ene” (compounds possessing two double bonds are “-dienes”, those with three double bonds, “-triene”, etc.). The name of the parent hydrocarbon is printed in bold or italic letters to render it more conspicuous.

Etiocholane derivatives with a double bond at C₄ or C₅ are classified as androstenes. They could be designated as etiocholenes with equal justification since the relative position of rings A and B is equalized by such a double bond and the characteristic C₅ hydrogen eliminated.

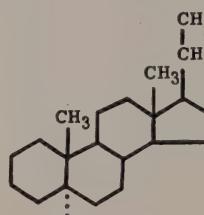
2. THE SIDE-CHAIN. Aliphatic side-chains, both saturated and unsaturated, are designated by the name of the appropriate alkyl radical, the carbon atoms of which are numbered in sequence from the point of attachment. Functions substituted in the side-chain are named in accordance with the International Union Rules for the Naming of Organic Compounds. The prefixes hydroxy-, keto-, aldo- and carboxy- (respectively for alcohols, ketones, aldehydes and acids) are placed in square brackets with the name of the alkyl radical which precedes that of the nuclear hydrocarbons; in the absence of functions in the side-chain, the brackets are omitted. e.g., desoxycorticosterone = 17(β)-[1-keto-2-hydroxyethyl]- Δ^4 -androstene-3-one :



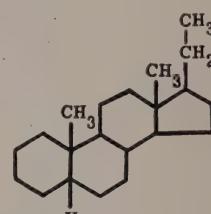
Ethynyl-testosterone = 17(β)-ethynyl- Δ^4 -androstene-3-one-17(α)-ol :



Among the naturally occurring steroid hormones and their derivatives, alkyl radicals (other than the angular methyl groups) — if present — are attached only on carbon atom 17. The most important, naturally occurring alkyl substituted nuclear hydrocarbon derivatives are those of 17(β)-ethyl-androstan e and 17(β)-ethyl-etiocholane, compounds also known under the shorter common names of allo-pregnane and pregnane, respectively :



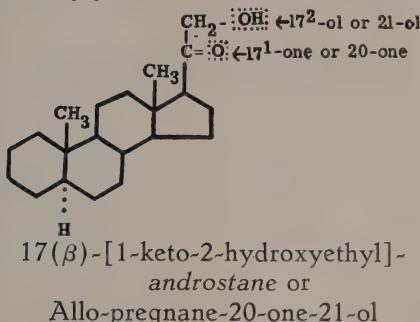
17(β)-ethyl-androstan e
(allo-pregnane)



17(β)-ethyl-etiocholane
(pregnane)

Derivatives of functions in the side chain are indicated by numerals with indices; the number gives the position of the alkyl radical, and the index that of the individual carbon atom in the side-chain to which the function is attached. In the pregnane, allo-pregnane nomenclature, the numeration of the carbon atoms of the nuclear parent hydrocarbon (which, with its two angular methyl groups goes up to 19) is merely continued so that carbon atom 17¹ becomes carbon atom 20 and 17², carbon atom 21. The following formula illustrates the two methods of numeration. That with indices for side-chain carbon atoms is more convenient, since it is generally

applicable to all types of organic compounds, while the other is simpler and generally preferred in clinical literature.



Isomerism. — 1. NUCLEAR ISOMERISM. Consistent with the burden of evidence, it is assumed that in the androstane series, the substituents of each pair of adjacent asymmetric carbon atoms (i.e., 5:10, 9:10, 8:9, 8:14, 13:14) bear in space the opposite or "trans" relation to each other. Accordingly the two angular methyl groups fixed at C₁₀ and C₁₃ lie on the same side of the flat plane of the molecule (arbitrarily the near side) and serve as points of reference. As stated above, dotted lines are used to denote valence bonds which project behind the plane of the paper (the "trans" configuration with respect to the angular methyl groups), and solid lines to indicate those which stand forward (the "cis" configuration).

ETIOCHOLANE differs from ANDROSTANE only in the orientation about C₅. Accordingly it is formulated with a solid line issuing therefrom. In androstane (as in allo-pregnane or 17(β)-ethyl-androstane) the relative positions of ring A:B, B:C, and C:D, is "trans" throughout. Such compounds are customarily designated as "trans-trans-trans".

In all naturally occurring *estrane* ring A is unsaturated, hence there is no hydrogen at C₅. Here the relative orientation of rings A and B cannot vary as ring A is flat and attached in the plane of the molecule. The relative positions of rings B:C and C:D is again "trans".

2. ISOMERISM OF THE SUBSTITUENTS AND OF THE SIDE-CHAIN. For the designation of the steric arrangement of substituents at all centers of asymmetry, brackets follow the number of the sub-

stituted carbon atom. Contained therein is the index *a* or *β*; to denote the stereo-chemical relation to the rest of the molecule of a functional group (usually -OH) or a side-chain. *β* designates substituents which lie on the same side of the flat plane of the molecule as the angular methyl groups; *a* denotes the opposite direction. As regards such substituents, the terms *β* or "cis", on the one hand and *a* or "trans", on the other, are now used synonymously. All of the chemically stable, naturally occurring and biologically highly active derivatives of allo-pregnane are 17(β)-ethyl-androstane derivatives, while the 17(*a*)-ethyl-androstane compounds include the chemically labile, biologically less active, artificially prepared steroids.

Common Names. — The correct chemical designation of steroids has the great advantage of giving complete details concerning their structure. However, as we saw above, these chemical terms are rather clumsy; hence, abbreviated common names are generally in use for the steroid compounds to which frequent reference is made. We have already encountered examples of these for instance, desoxycorticosterone and testosterone. Minor chemical changes in the molecular structure of such common compounds lead to derivatives which can still rather conveniently be referred to by modifications of the common name of the parent compound. Thus methylation of testosterone yields "methyl-testosterone"; introduction of an ethynyl group into estradiol gives us "ethynodiol-estradiol"; a compound which is isomeric with testosterone differing from the latter merely in the steric position of the hydroxyl, is called "isotestosterone," etc.

For the convenience of the reader, the structure formulæ, systematic chemical names and common names of the most frequently mentioned steroid compounds are listed (in alphabetic order) in the following table.

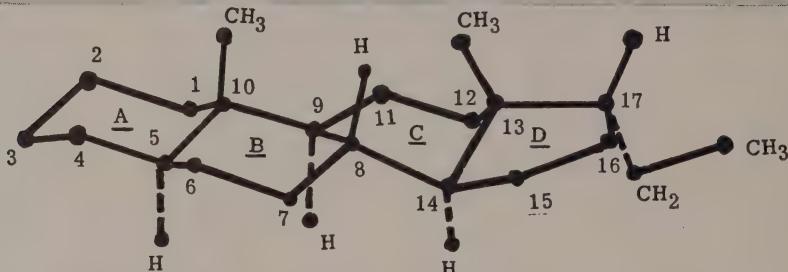
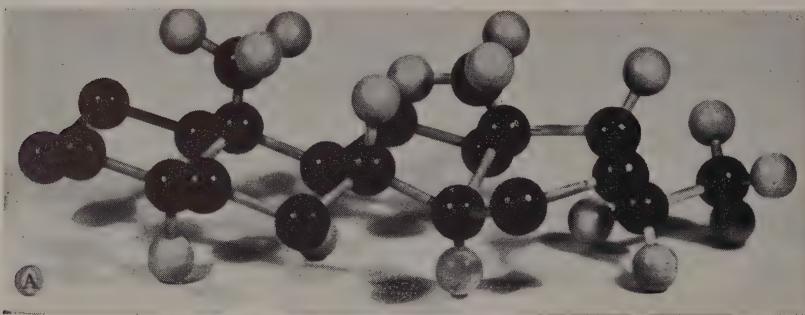
LIST OF MOST COMMONLY MENTIONED STEROID HORMONES

Structure	Common Name	Systematic Chemical Designation	Natural occurrence	Predominant activity
	ADRENOSTERONE	$\Delta^4\text{-androstene-3,11,17-trione}$	adrenal	testoid
	ANDROSTERONE	<i>androstane-3(α)-ol-17-one</i>	urine	testoid
	CORTICOSTERONE	$17(\beta)\text{-[1-keto-2-hydroxyethyl]-}\Delta^4\text{-androstene-3-one-}11(\beta)\text{-ol}$	adrenal	gluco-corticoid
	DEHYDROCORTI-COSTERONE	$17(\beta)\text{-[1-keto-2-hydroxyethyl]-}\Delta^4\text{-androstene-}3,11\text{-dione}$	adrenal	gluco-corticoid
	DEHYDRO- <i>iso</i> -ANDROSTERONE	$\Delta^5\text{-androstene-3}(\beta)\text{-ol-17-one}$	urine	testoid
	DESOXYCORTI-COSTERONE OR 11-DESOXYCORTI-COSTERONE	$17(\beta)\text{-[1-keto-2-hydroxyethyl]-}\Delta^4\text{-androstene-3-one}$	adrenal	mineralo-corticoid
	ESTRADIOL	$\Delta^{1,8,5,10}\text{-estratriene-3,17(a)-diol}$	ovary placenta testis urine	folliculoid
	ESTRIOL	$\Delta^{1,8,5,10}\text{-estratriene-3,16}(\beta), 17(a)\text{-triol}$	placenta urine pussy-willows	folliculoid
	ESTRONE	$\Delta^{1,8,5,10}\text{-estratriene-3-ol-17-one}$	urine testis placenta palm kernels	folliculoid
	ETIOCHOLANOLONE	<i>etiocholane-3(a)-ol-17-one</i>	urine	anesthetic (hormonally inactive)
	HYDROXY-PREGNENOLONE	$17(\beta)\text{-[1-keto-2-hydroxyethyl]-}\Delta^5\text{-androstene-3}(\beta)\text{-ol}$	not found in tissues	mineralo-corticoid

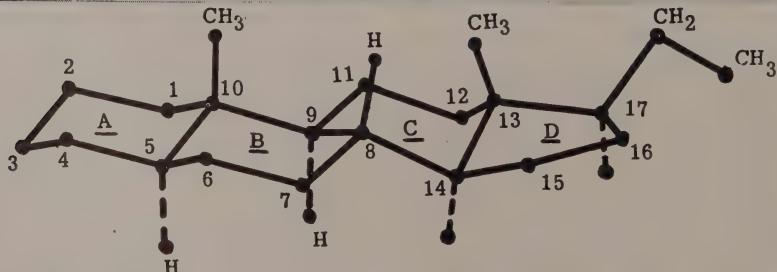
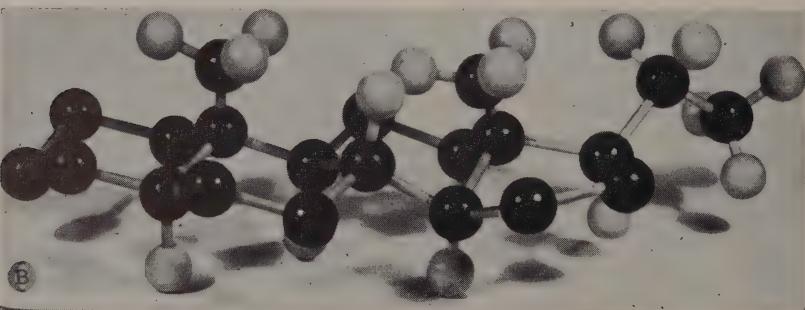
Structure	Common Name	Systematic Chemical Designation	Natural occurrence	Predominant activity
	iso-ANDROSTERONE	<i>androstane-3(β)-ol-17-one</i>	urine	testoid
	KENDALL'S CPD. "E" 17-HYDROXY-11-DEHYDRO CORTICOSTERONE	$17(\beta?)\text{-[1-keto-2-hydroxyethyl]-}\Delta^4\text{-androstene-3,11-dione-17(a?)-ol}$	adrenal	gluco-corticoid
	METHYL-TESTOSTERONE	$17(\beta)\text{-methyl-}\Delta^4\text{-androstene-3-one-17(a)-ol}$	not found in tissues	testoid
	PREGNANEDIOL	$17(\beta)\text{-[1(a)-hydroxyethyl]-etiocholane-3(a)-ol}$	urine	anesthetic (hormonally inactive)
	PREGNANEDIONE	$17(\beta)\text{-[1-ketoethyl]-etiocholane-3-one}$	urine	anesthetic (hormonally inactive)
	PREGNANOLONE	$17(\beta)\text{-[1-ketoethyl]-etiocholane-3(a)-ol}$	urine	anesthetic (hormonally inactive)
	PREGNENINOLONE OR ETHYNODIOL-2-ONE OR ANHYDRO-HYDROXY-PROGESTERONE	$17(\beta)\text{-ethynyl-}\Delta^4\text{-androstene-3-one-17(a)-ol}$	not found in tissues	luteoid
	PREGNENOLONE	$17(\beta)\text{-[1-ketoethyl]-}\Delta^5\text{-androstene-3(β)-ol}$	testis	spermatogenic
	PROGESTERONE	$17(\beta)\text{-[1-ketoethyl]-}\Delta^4\text{-androstene-3-one}$	ovary	luteoid
	TESTOSTERONE	$\Delta^4\text{-androstene-3-one-17(a)-ol}$	testis	testoid

SPACE MODELS OF THE PRINCIPAL STEROID TYPES

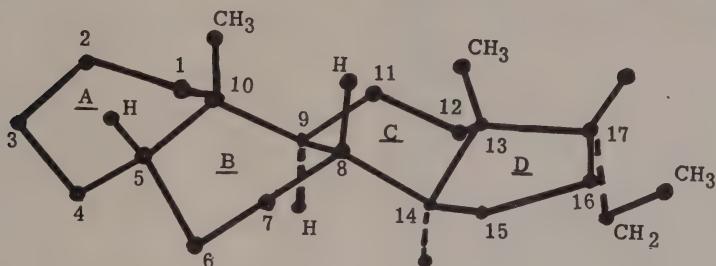
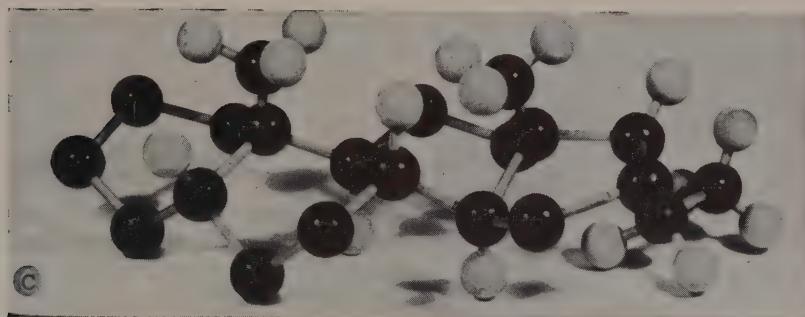
The following are actual photographs of space-model reconstructions of the 4 principal hydrocarbons from which the steroid hormones are derived. Under each photograph is a schematic drawing, showing the connections (valence bonds) between the various atoms, viewed precisely from the same angle as the corresponding photographs. In order to avoid too much overlap between individual atoms, all four molecule reproductions have been photographed and drawn from a position almost parallel with the "plane" of the molecule.



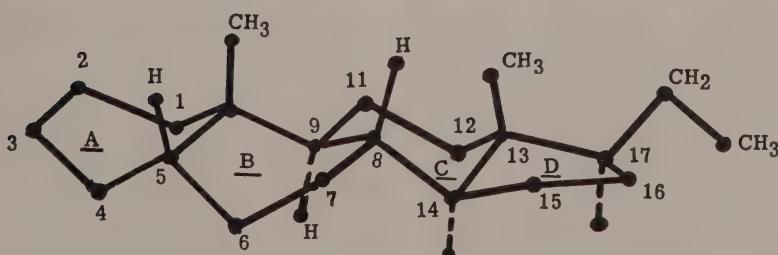
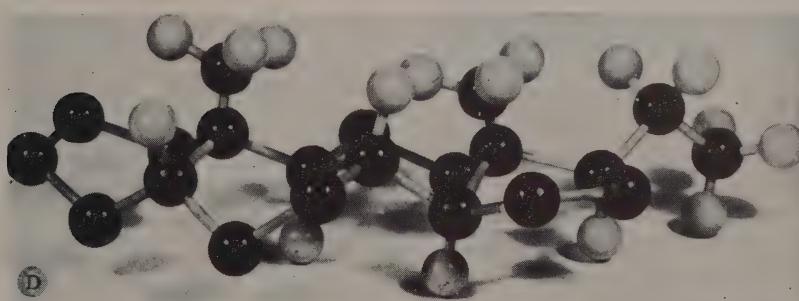
A.-17(α)-ethyl-ANDROSTANE. Note alpha position of the hydrogens at C₅, C₉ and C₁₄. This is the so-called "trans-trans-trans" arrangement ("cholestane type"). The ethyl side-chain is also attached in the alpha position.



B.-17(β)-ethyl-ANDROSTANE. The steroid skeleton is the same as in the previous compound, but the ethyl side-chain is attached in the beta position.



C.-17(α)-ethyl-ETIOCHOLANE. Note that steric arrangement of the 4 rings is the same as in previous compounds, except for the relative position of ring A to ring B. Correspondingly, the hydrogen at C₅ is in beta position. ("Coprostane type".) The ethyl side-chain is attached in alpha position.



D.-17(β)-ethyl-ETIOCHOLANE. Steric arrangement of the steroid ring system is the same as in previous compound, but the side-chain is attached in beta position.

**PHARMACOLOGIC TERMINOLOGY
AND CLASSIFICATION OF
THE STEROIDS**

Definition and Outline. — We have seen that the complicated chemical structure of the steroid molecule made it necessary to devise a rather intricate system of terminology and classification. When we come to consider its complex pharmacologic characteristics we shall find that these also require careful systematization before they can be clearly understood or even rationally enumerated.

The manifold pharmacologic activities of the steroids and the fact that almost any of them possess an apparently unpredictable combination of such activities, tend to give the impression of a complete lack of orderliness. In other words, it appears as though there were no correlation between the chemical structure of a compound and its pharmacologic activities (pharmacо-chemical correlations), nor between the several pharmacologic effects themselves (pharmacо-pharmacologic correlations) which a single compound may exhibit. Yet, certain general lawful correlations of this kind have already been elucidated and found to hold true for all hormone-like steroids examined up to the present time. This type of study is perhaps the most fascinating and, from a practical point of view, the most important aspect of contemporary steroid hormone research. An almost unlimited number of steroids could be made available by partial synthesis from the known compounds, but since the only value of these compounds is their therapeutic efficacy we must learn more about the structural prerequisites of their pharmacologic activities in order to direct the work of the synthetic chemists into profitable channels.

The following paragraphs are merely a sketch of the fundamental prerequisites for the study of this important field. They attempt to outline the main principles according to which the ac-

tions of steroid hormones are named and classified.

Independent Actions. — The basic principle according to which the pharmacologic activities of steroids are classified is that certain actions are independent of each other while others are merely the subordinate manifestations of such independent actions and hence dependent upon them. It must be clearly understood that independent actions are characterized by the fact that every one of them can be exhibited independently of any of the others; that is to say, there is no direct parallelism between the degree to which a compound exhibits the various independent actions. In this sense we recognize the independent nature of the following actions :

- (1) **FOLLICULOID** (estrogenic, gynæcogenic, estromimetic or follicular-hormone-like).
- (2) **TESTOID** (androgenic, andromimetic or male hormone-like).
- (3) **LUTEOID** (progestational, corpus luteum-hormone-like).
- (4) **CORTICOID** (adrenal-cortical hormone-like).
- (5) **SPERMATOGENIC** (having the ability to stimulate the spermatogenic epithelium and mainly to protect it against atrophy caused by deficiency in gonadotrophic hypophysoid hormones).
- (6) **RENOTROPHIC** (nephrotrophic, having the ability to increase kidney size due mainly to hypertrophy of the convoluted tubules).
- (7) **ANESTHETIC**.
- (8) **ANTI-FIBROMATOGENDIC** (Inhibiting fibroma formation by folliculoids — see: p. 370).

It will be noted that for those independent actions which imitate the function of an organ of internal secretion, terms are used which suggest a specific connection with that particular endocrine gland. The Greek ending “-oid”, which is added to the name of a gland, means “similar to” without implying that the hormone is necessarily derived

from that particular gland. It merely suggests that the compound simulates the gland's activity, and this is true by definition. It would be misleading, for instance, to designate hormones of the testoid type as "testicular hormones" since such compounds are also elaborated by the adrenal cortex and probably even by ovarian and placental tissues. It would be equally misleading to term them "androgenic" since the most potent natural "androgen" testosterone, causes testis atrophy in experimental animals. Thus, far from being masculinizing it is actually "demasculinizing." Similarly, in many animal species, the so-called "estrogens" do not in themselves cause estrus or heat without simultaneous progesterone treatment, hence the latter hormone could be called "estrogenic" with almost equal justification. Furthermore, folliculoids interrupt the estrous cycle in the intact rodent so that they are actually "anti-estrogenic" under ordinary circumstances of bioassay. The term "gynaecogenic" is no more adequate for them since they cause ovarian atrophy.

In the light of the above considerations, it appears unsatisfactory to designate these hormones either according to their source of origin or on the basis of one particular action on a certain target organ. Hence, we give preference to the terminology which is based upon the natural classification of the independent steroid hormone actions according to the gland whose function they simulate.

Subordinate Actions. — As indicated above, each independent action has numerous distinct subordinate manifestations inasmuch as it affects a variety of target organs. Thus a folliculoid hormone (e.g., estradiol) which produces vaginal cornification in the rodent, invariably — and to a degree which runs parallel with the intensity of its effect on the vagina — causes enlarge-



Steroid hormone anesthesia. Photograph of a female rat (body weight 70 gm.) under profound anesthesia after intraperitoneal administration of 10 mg. of progesterone in 0.5 cc. of oil. Note marked muscular relaxation.

ment of the uterus, the oviducts, the adrenals, the nipples, hyperemia of the monkey's sex skin, atrophy of the testes, etc. All these manifestations are termed subordinate actions in order to emphasize that they are dependent upon, and the direct consequence of, a single independent action (in this case, the folliculoid action). In the same sense, the ability to stimulate the seminal vesicles, the prostate, the capon's comb, etc., are subordinate to the testoid action. For the sake of simplicity, organ growth stimulating and inhibiting subordinate actions are designated by the

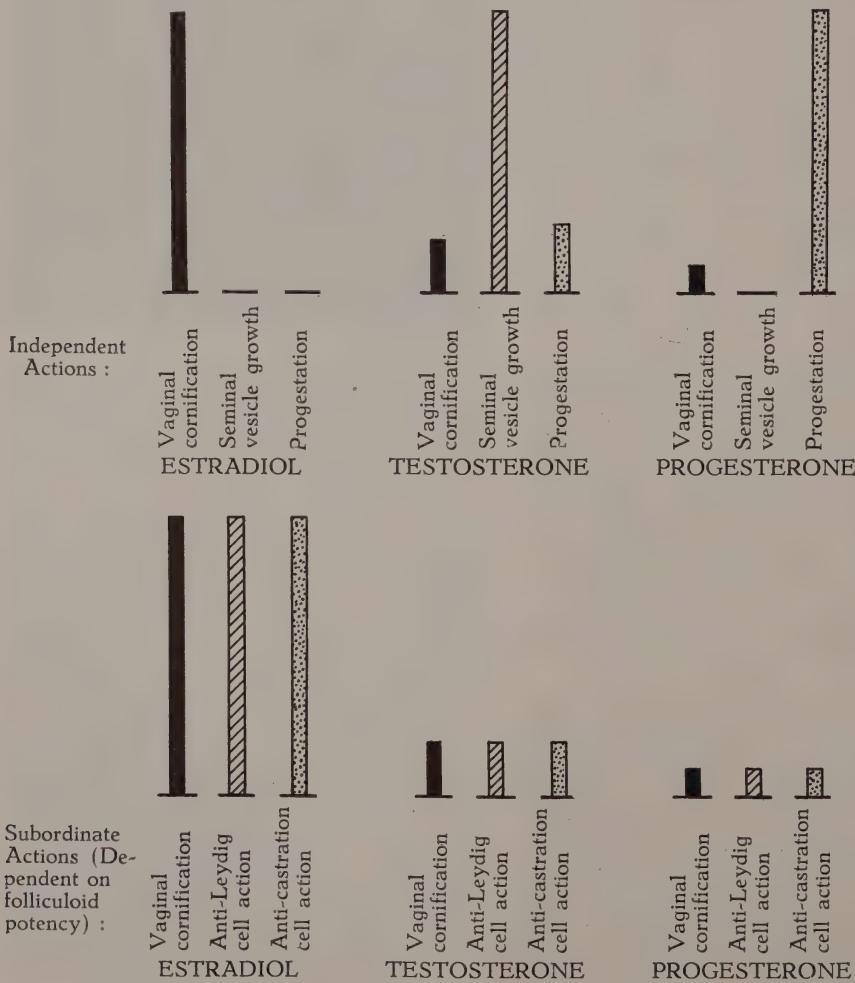
prefix, "pro-" and "anti-", respectively, followed by the name of the affected organ; (e.g., the Leydig cell atrophy-producing property is termed anti-Leydig-cell, the adrenal enlarging effect, "pro-adrenal" action). This merely

represents an extension to morphogenetic effects of the principle underlying the designation of biochemical changes by the prefixes "hyper-" and "hypo-", respectively, as in hyperglycemia, hypochloremia, etc.

Graph representing interrelations between independent and subordinate (dependent) actions of steroid hormones.

The principal independent actions of estradiol, testosterone and progesterone — used as examples here — are folliculoid, testoid and luteoid respectively. These can be estimated by one of the typical subordinate effects of these three independent actions, for instance, vaginal cornification (folliculoid), seminal vesicle growth (testoid) and uterine progestation (luteoid). The complete lack of interdependence between these activities is clearly demonstrated by the graphs of the top row.

The graphs of the bottom row represent bioassays for three subordinate actions of the folliculoid potency : vaginal cornification, anti-Leydig-cell action, anti-castration-cell action. Estradiol is much more effective in all these respects than is testosterone, while progesterone is least active. Yet all three compounds exhibit the three subordinate folliculoid actions in the same relative proportions. This illustrates their interdependence.



Potentially Independent Actions.

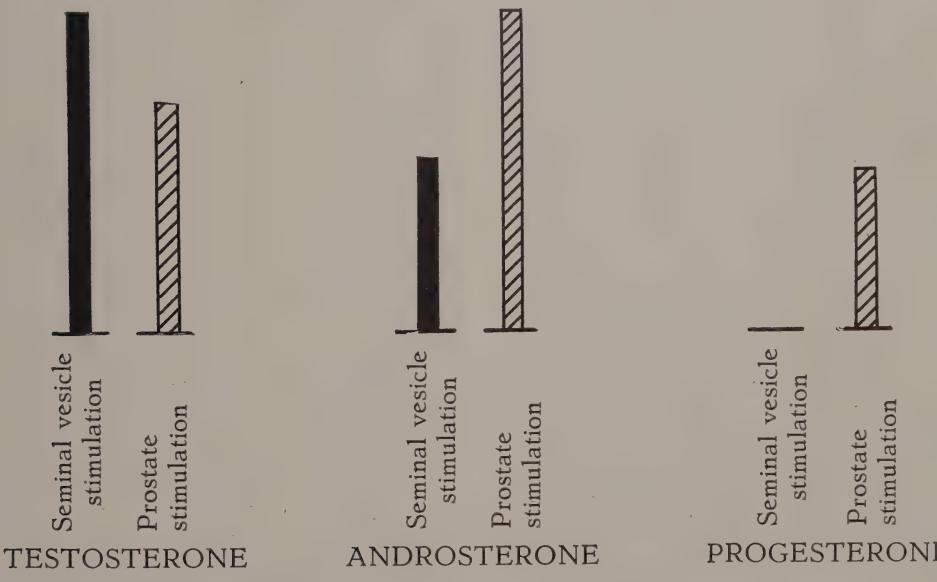
— There is a third type of effect which appears to be intermediate between the independent and the subordinate actions. This has been termed a "potentially independent" effect. Let us take an independent action I, one of its typical subordinate actions S and one of its potentially subordinate actions P. S is subordinate to I, since it never occurs without other manifestations of the latter, while P is independent of S, because it may occur with or without evidence of S, yet P is only potentially independent of I, since all compounds of the group I necessarily exhibit the action P, but compounds with action P do not necessarily exhibit other actions characteristic of group I. For instance,

no testoid (I) compound is known to stimulate the seminal vesicles of the rat (S), without simultaneously increasing the weight of the prostate and preputial glands (P). There are compounds, however, which stimulate the latter two organs without affecting the seminal vesicles (e.g., progesterone).

Similarly, among the corticoids, some compounds have a particularly pronounced effect on sugar and others on mineral metabolism, but all the corticoids exert a beneficial effect on life maintenance after adrenal deprivation. Hence, both the sugar and the mineral metabolism influencing (gluco-corticoid and mineralo-corticoid) activities are potentially independent of the life-maintaining potency.

Graph representing interrelations between two potentially subordinate actions.

Seminal vesicle stimulation is a characteristic subordinate effect of the testoid potency. It is exhibited by all testoid compounds. Prostate stimulation is potentially independent of testoid action. It is exhibited by all compounds, having typical testoid effects (e.g., seminal vesicle stimulation), for instance, by testosterone and androsterone. However, this potentially independent action may also be exhibited by compounds which are devoid of such typical testoid effects (e.g., progesterone).



TESTOSTERONE

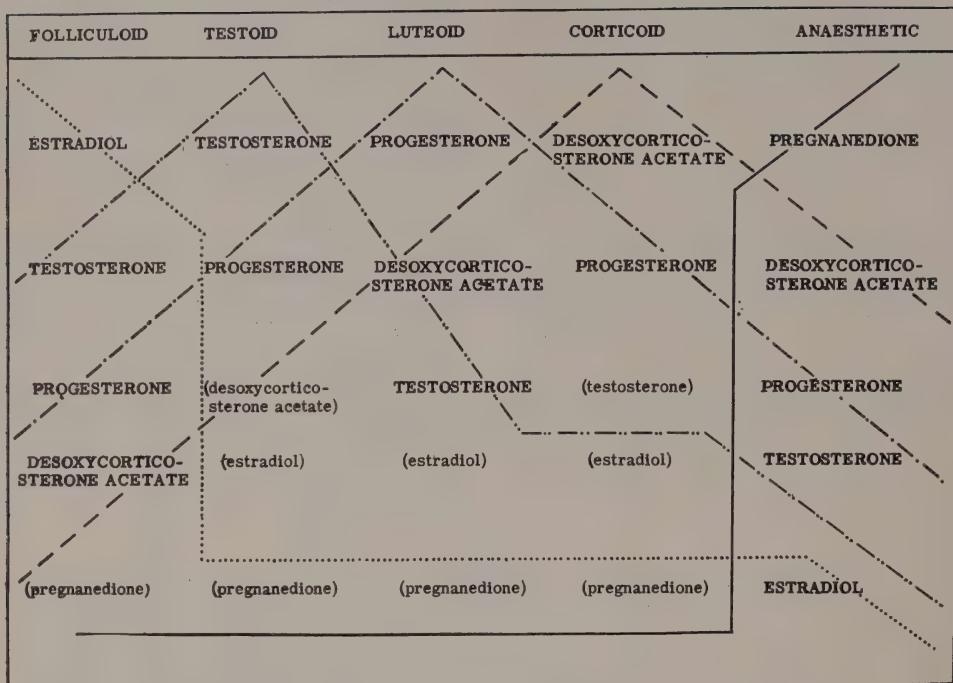
ANDROSTERONE

PROGESTERONE

The Systematic Table of the Steroids. — The fact that steroids can exhibit only the anesthetic and the folliculoid activities in a pure form (that is in the absence of all other independ-

ent actions), as well as the degree and type of overlap between the other independent actions is visualized by the systematic table.

Systematic table of the steroids



In constructing this table, the most active representatives of five independent actions have been selected and arranged from left to right according to decreasing order of folliculoid potency. (Since the most potent representatives of the renotrophic and spermatogenic series have not been definitely identified as yet, these actions are not considered here. However, according to the folliculoid effect of the known prominent representatives of these groups, they should both be inserted next to testosterone.) It will be noted that the folliculoid activity of all these compounds, except estradiol, is "masked" and detectable only under certain experimental conditions. Furthermore, for the purpose of this discussion prostatic enlargement is considered sufficient for the demonstration of testoid activity. Desoxycorticosterone acetate has been chosen as the most active representative of the corticoid series as it is the only highly active member of this group which has been adequately studied for

all five independent actions. In each column representing a certain type of activity the inert compounds are inserted in brackets below the active compounds of the group.

When the steroids are so arranged according to decreasing folliculoid activity in the first line of the table, the most active representatives of the folliculoid, testoid, luteoid, corticoid and anesthetic compounds appear in the order stated. The second most active representatives of each of these five activities were placed in the second line, third place being given to the next most active compounds, and so forth.

Perusal of the table indicates that, if the steroids are arranged in the first column according to decreasing order of folliculoid activity, they are automatically in increasing order of anesthetic potency. The position of each steroid was traced by a line through the five columns. This revealed that estradiol and pregnandione are at the bottom of the graph, except in the column in

which they are placed on top as the most active folliculoid and anesthetic compound respectively. The three remaining compounds slowly lose height in both directions with approximately the same slope. Thus the curve described by the position of any steroid in the table exhibits a single maximum.

The table expresses the empirical fact that only folliculoid or anesthetic potencies may be exhibited to the exclusion of all other independent actions, since these two are on the outer limits of the system and hence do not have to overlap with other effects. Compounds not included in this table also obey the same rules although, of course, depending upon the degree of their potency, they range above or below the curves given here. Thus ethynodiol-estradiol, which is somewhat more folliculoid than estradiol, describes a line parallel with that of estradiol although somewhat higher than the latter, while pregnenolone, a compound having an activity which is qualitatively similar but quantitatively inferior to that of progesterone, parallels the curve of the latter at a somewhat lower level. Acetoxypregnendiol, a comparatively inactive corticoid, parallels the desoxycorticosterone acetate curve in a similar manner, and so forth. Among all steroids studied no exception could be found to the rule that all compounds describe curves with a single maximum when inserted in this system.

Disregarding all possible *a priori* arguments from chemical relationships, statistical analysis showed that the regularities in the whole scheme would occur by chance only in 1 out of 2,304 trials, allowing for the selection of the top entry in each column and giving no "credit" for possible orderliness in the compounds found inactive in each type of test. Hence, it may be said that this particular arrangement is not due to chance but is apparently dependent upon certain inherent natural relationships between the steroids. The main

weakness of the classification so far detected is that the activity curve of a certain compound may skip one or more points on the curve, perhaps because this particular action is so "masked" that it is not detectable with our bioassay methods.

The biologic significance of these correlations among the steroids is not easy to interpret. It may be that the molecular structure necessary for any one activity necessarily carries within itself other pharmacologic properties and that the intensity of these decreases in direct proportion to their distance from the primary activity in the table. It is also possible that the compound placed at the peak of any one curve is partly transformed in the body into compounds with neighbouring actions which in turn yield smaller amounts of steroids in the columns next to them. Thus the degree of activity would gradually diminish in proportion to the distance in the table from the position of the original compound injected. In this sense the pure folliculoids and anesthetics might be considered as metabolic end-products incapable of re-transformation into compounds occupying more central positions in the table. This is graphically expressed by their marginal position and the fact that the most active folliculoids and anesthetics exhibit no other activities. That a compound may skip a point on the curve could be explained by assuming that certain steroids may go through a pharmacologically inactive stage during their metabolism.

Considering the limited data available at this time, it would scarcely be justified to base any far-reaching speculations on the regularities observed. The only purpose of the table is to direct attention to the fact that if the steroids are arranged according to the degree of their folliculoid activity, they fall into a natural system which permits — within limits — a prediction of their other activities.

INTERRELATIONS BETWEEN THE VARIOUS PHARMACOLOGIC PROPERTIES OF THE STEROIDS (PHAR-MACO-PHARMACOLOGIC INTERRELATIONS)

Definition and Outline. — By pharmaco-pharmacologic interactions among the steroids we understand the property of two independent actions to influence each other. The subordinate effects of any one independent action cannot detectably influence each other since they are only different manifestations of the same pharmacologic property. On the other hand, if an animal is simultaneously treated with two steroid compounds, having different independent actions, one compound may inhibit or augment the effects of the other. An inhibition may result from a simple diminution or abolition of the effect (quantitative inhibition), but it may also be due to a modification of the action (qualitative alteration). In the case of an augmentation or synergism, there may be pure summation (mere addition of two similar actions) or true potentiation (synergism greater than could be expected by merely adding the two effects).

Even two independent actions exerted by a single chemical compound may mutually influence each other. In this event, treatment with one compound will cause a complex response, namely the resultant of this interaction.

Knowledge of the pharmaco-pharmacologic interrelations is of great practical value, since it is often possible to increase the selectivity of hormone actions by using drug combinations which accentuate the desired property but minimize undesirable side effects.

Synergisms and Antagonisms. — Synergisms, in the sense of mere summation of effects, are comparatively common among the steroid compounds. In fact most steroids which have certain actions in common, are additive as regards these pharmacologic properties. Thus, the effect of estradiol and estrone

upon the vagina, or that of testosterone and androsterone upon the seminal vesicles, is additive if the compounds are given simultaneously in submaximal doses.

On the other hand, there are only few instances of clear-cut POTENTIATION of steroid hormone actions. As a pertinent instance, we may mention the potentiation of the luteoid effect of progesterone by folliculoids. The pregestational transformation of the endometrium, the mucification of the vaginal epithelium, the proliferation of the mammary glands, the relaxation of the symphysis pubis and the production of deciduomas, following endometrial trauma, are all characteristic effects of luteoid compounds. However, enormous doses of progesterone are required in order to elicit these changes unless the test animals are pretreated, or simultaneously treated with minute doses of folliculoids. Therefore, in all these instances, we may well speak of a true potentiation of the luteoid compounds by the folliculoids.

There are many more instances of ANTAGONISMS between steroid hormones. Thus, the ability of corticoids to maintain adrenalectomized animals, is counteracted by simultaneous treatment with folliculoids. This is merely due to the fact that the latter are very toxic for the adrenal-deficient animal and raise its corticoid hormone requirement. The adrenal-cortical enlargement elicited by folliculoids is also inhibited by corticoids. Similarly, the testis atrophy produced by folliculoids in normal males is diminished, or even completely prevented, by concurrent treatment with spermatogenic steroids, such as pregnenolone.

It is important to realize that the manner in which two steroid hormones influence each other's actions depends upon three factors :

- (1) The ratio of the two compounds in a mixture.

- (2) The dose level at which the mixture is administered.
- (3) The target organ whose response to the mixture is studied.

To illustrate the importance of the RATIO of two interacting steroids in a mixture, suffice it to mention that small doses of estradiol enhance certain actions of progesterone (e.g., pregestational proliferation of the endometrium, vaginal mucification, deciduoma formation, maintenance of pregnancy in ovariectomized females) while large doses of the same folliculoid completely prevent these luteoid effects.

The DOSAGE, in which a given mixture is administered, is likewise important. Thus, in spayed rats, a solution containing small amounts of estradiol and large quantities of progesterone causes continuous vaginal cornification at low, but mucification at high dose levels. In other words, at the low daily dose level the folliculoid, while at the high dose level the luteoid effect of the mixture predominates.

Similarly, if a solution containing small doses of estradiol and large doses of testosterone is given to intact male rats, it causes especially pronounced atrophy of the seminiferous tubules at low daily dose levels, but results in no tubular damage at high dose levels. Apparently, at the low dose level, the anti-spermatogenic action of estradiol and of testosterone is additive, while at high dose levels, the anti-spermatogenic effect of estradiol is counteracted by the spermatogenic property of testosterone. In this antagonism the dose of estradiol is of no great importance. This folliculoid appears to act upon the testis by inhibiting the gonadotrophin production of the hypophysis and even doses many times greater than the minimal amount required for this "functional hypophysectomy" cannot accomplish more than a surgical hypophysectomy can. Since moderate doses of spermatogenic steroids (e.g., testosterone)

maintain the tubules even after complete ablation of the hypophysis it is understandable that they are equally effective in rats in which the testis atrophy is produced by any dose of estradiol.

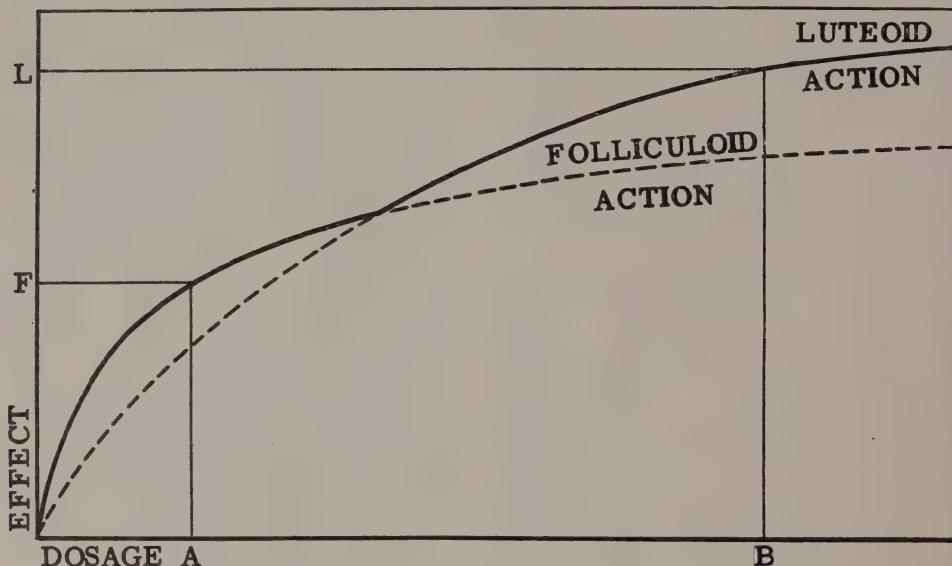
Other things being equal, the effect of a steroid hormone mixture is largely dependent upon the TARGET ORGAN whose response we study. Thus, desoxycorticosterone acetate or ethynodiol-testosterone, given in high doses, completely inhibit the vaginal cornification normally produced by estradiol. Hence, as regards this target organ, they are antagonists of folliculoids. On the other hand, these same compounds do not inhibit the anti-spermatogenic effect of estradiol.

Such observations are of fundamental importance. They clearly demonstrate that steroids which antagonize folliculoid actions, do not completely annul them (as an acid would neutralize an alkali, or glucose would antagonize all the manifestations of insulin hypoglycemia); they merely interfere with certain effects upon specific target organs. Indeed, we know of no instance in steroid pharmacology where all effects of a certain compound would be completely nullified by simultaneous treatment with another steroid.

Manifest and Masked Actions. — We have seen that chemically pure steroid compounds may exhibit several independent pharmacologic actions. We also learned that in mixtures containing two steroids — each exhibiting a different independent action — the effects of the two components may be synergistic or antagonistic, depending upon the ratio, the dose and the target organ. In view of these facts, it is not surprising that two independent actions of a single chemical substance may likewise influence each other. Obviously, here, the ratio of the two activities is fixed since both actions are due to the same molec-

Graph illustrating interrelations between manifest and masked activities of steroids.

If a mixture containing certain proportions of a folliculoid compound (e.g., estradiol) and a luteoid substance (e.g., progesterone), or a single compound possessing both these activities (e.g., testosterone), is administered at various dose levels, the folliculoid actions (F) will predominate at low doses (A), while at high dose levels (B), the luteoid (L) actions become manifest. As the graph implies, this may be due to differences in the slope of the dose-effect relationship of luteoid and folliculoid activity respectively, so that at low dose levels, the former effect would be "masked" by the latter.



ular structure; only dose level and target organ are subject to variation.

For the analysis of such pharmacologic interactions between two independent properties — whether exhibited by a single steroid or a mixture — it is convenient to distinguish between manifest and masked activities. An activity is considered to be MANIFEST, or overt, under all experimental conditions in which it is detectable. Thus, for instance, the testoid activity of testosterone is manifest if the compound is administered to immature, or castrate rats.

Certain steroid hormone actions are not demonstrable under ordinary circumstances, yet they are real since they become evident under special experimental conditions. In such cases, we speak of MASKED activities and assume that the pharmacologic property is inherent in the compound though not in a manifest form. For instance, in tes-

tosterone, the vagina-cornifying effect is not detectable under ordinary conditions of bioassay because it is masked by the vagina-mucifying effect. During the first days of treatment, however, the compound causes a transitory vaginal cornification in the spayed or immature female rat; in hypophysectomized females, the cornification usually becomes permanent. In this manner, it is possible to "unmask" an otherwise hidden property. It was to be expected that any vagina-cornifying action of testosterone would be masked, under the standard experimental conditions prevailing in the spayed female, since even the most potent folliculoids (e.g., estradiol or estrone) are inactivated in this respect by simultaneous administration of a vagina-mucifying compound such as testosterone (see above).

Simple and Complex Actions. — The differentiation of manifest and masked properties leads us to another

fundamental consideration, namely the distinction between simple and complex activities. Certain steroids appear to possess only their manifest actions which are detectable under all circumstances. Others are endowed with a multiplicity of pharmacologic properties, some of which are manifest while the rest are masked or modified. The compounds belonging to the former category are pharmacologically "SIMPLE" since they possess only overt properties. The compounds of the second category are pharmacologically "COMPLEX" because, in addition to their overt manifestations, they possess masked properties which may modify the others. As previously mentioned, the best representatives of "simple" actions are found among the predominantly folliculoid and anesthetic compounds; hence, the problem can best be clarified by making a pharmacologic comparison between these and the main representatives of all other independent groups of our classification.

It appears that the predominantly folliculoid compounds differ from the principal representatives of all other independent groups in that there are no qualitative differences in the pharmacologic properties of hormonally active estrane compounds of high folliculoid potency or their synthetic derivatives. Indeed even non-steroid compounds (e.g., the stilbene derivatives) are qualitatively equal, although quantitatively the degree of their activity varies. In sharp contrast with this uniformity the principal representatives of all other independent groups possess highly variable properties. Thus the two most active natural testoids, testosterone and androsterone, differ markedly in that the former is comparatively more active in stimulating the capon's comb and the rat's seminal vesicles, while the latter possesses a relatively greater pro-prostatic effect. Similarly, among the corticoids, corticosterone has a greater gluco-

corticoid and desoxycorticosterone a greater mineralo-corticoid activity.

The comparative pharmacologic simplicity of the primarily folliculoid compounds is especially striking if we consider that none of the active estranes show any pharmacologic overlap with the hormonal properties characteristic of other independent groups. Neither estradiol nor estrone, estriol or even the stilbenes possess the slightest luteoid, corticoid, testoid, spermatogenic or renotrophic activity. This is very remarkable if we remember that not a single compound is known which would selectively exhibit any one of these latter five activities. It should be remarked parenthetically that the seminal-vesicle-increasing effect of hormonally active estranes is by no means a "testoid" action, as it is merely due to disproportionate fibro-muscular growth.

There is only one other independent action which may be considered to be "simple" in the sense in which the word is used here. This is the anesthetic effect which is selectively exhibited by some steroids (e.g., pregnanediol) to the exclusion of all known hormonal properties.

All the steroids with other independent actions [corticoid, luteoid, testoid, spermatogenic or renotrophic] are pharmacologically complex. No compound is known which would exhibit any one of these latter activities in a pure form, without possessing some other action as well. It is impossible therefore to decide whether it is merely coincidental that all known steroids with predominantly corticoid, luteoid, testoid, spermatogenic or renotrophic activity, exhibit some subsidiary actions or whether these pharmacologic effects themselves are inherently dependent upon the simultaneous manifestation of several activities. In other words, the problem is, are these actions single tones or chords? Should compounds become known which possess only one of these independent activities, then this action will

be proven to be simple; until then, however, we must consider the possibility that the actions themselves are inherently complex.

Let us illustrate this point. As we have stated before, the main (luteoid) activity of progesterone, the principle representative of the luteoids, is greatly enhanced by minute quantities of folliculoids. It has also been demonstrated that progesterone itself possesses some folliculoid effect although it is masked under ordinary circumstances. It is quite possible, therefore, that what we consider the "luteoid" or "progestational" effect is inherently the sum of a slight folliculoid and some other (yet unknown) pharmacologic potency. Until quite recently, when very large doses of progesterone were assayed, this compound was believed to be inert unless given in conjunction, or following pretreatment with folliculoids. It is conceivable that the manifestation of luteoid activity, in the case of treatment with progesterone alone, necessitates unduly large doses merely because great amounts have to be administered to supply the folliculoid potency required for the manifestation of the luteoid effect.

In the light of these considerations, it appears possible that important hormonal activities may be detected in some apparently inert steroids (e.g., pregnanediol) by combined administration with other substances which complement their effect. This type of activation is by no means without precedent in endocrinology. Thus, for instance, the luteinizing hormone of pregnancy urine has no effect upon the granulosa cells in the ovary of the hypophysectomized rat unless some preliminary development is first induced by treatment with the follicle-stimulating hormone. At first sight it may appear that the so-called "x-substance" (which can be extracted from testis tissue and is probably identical with some of the

highly unsaturated fatty acids) belongs to this group, inasmuch as it is inactive in itself but increases the effect of testoids. Yet this type of activation is somewhat different; it manifests itself only if the two compounds are injected at the same site and hence it is probably due merely to the resultant delay in hormone absorption.

Inhibitable and Non-Inhibitable Actions. — We have already had occasion to mention the existence of antagonisms between different steroid hormones and the fact that even the diverse actions of a single chemical compound may likewise antagonize each other in such a manner as to produce "masked" or modified actions. Indeed, in certain instances, such a mutual antagonism between two properties of the same molecule may be regarded as almost definitely proven. For instance in the case of desoxycorticosterone, which possesses both corticoid and folliculoid actions, it may be taken for granted that the anti-adrenal effect, which is subordinate to the former, and the pro-adrenal effect, characteristic of the latter property, partly antagonize each other. Similarly the vagina-cornifying property of progesterone is usually inhibited by its own mucifying effect, which — as was previously mentioned — is so strong that it even inhibits the cornification otherwise caused by estradiol. However, some actions of estradiol (e.g., the anti-thymus, anti-Leydig-cell and anti-castration-cell effects) are not inhibitable by simultaneous treatment with other steroids.

In view of these considerations it is interesting to note that all the non-inhibitable folliculoid actions are common to all steroid hormones. Conversely (with the exception of the anesthetic effect which is not strictly speaking a hormone-like property), all the adequately studied actions common to all steroid hormones are primarily folliculoid properties, inasmuch as

they are exhibited most readily (at the lowest dosage level) by compounds which are predominantly folliculoid.

Many of the inhibitable folliculoid actions are not manifest in compounds with other independent activities, since the latter can counteract or "mask" the former even when produced by pure folliculoid estrane derivatives. Thus testosterone shares with the folliculoids such non-inhibitible actions as the anti-thymus, anti-castration cell, pro-mammary gland and plumage-feminizing effects. These cannot be inhibited by testosterone even when they are elicited by estradiol itself. On the other hand, testosterone elicits little or no persistent vaginal cornification, pro-adrenal or anti-spermatogenic effects and these are precisely the properties of estradiol which are inhibited by simultaneous testosterone administration. Hence, the evidence available at this time, is in agreement with the assumption that all steroid hormones possess some folliculoid actions (see : Systematic Table of the Steroids) but that among these, only the non-inhibitible properties are always manifest, while the others are demonstrable only under certain circumstances.

Selective Inhibition. — The different subordinate manifestations of one independent action of a certain steroid are not always equally inhibited by another independent action of the same compound. Thus, for instance, ethynodiol-diethylstilbestrol can cause vaginal mucification and progestational transformation of the endometrium, that is, changes dependent upon the predominance of its luteoid over its folliculoid properties. Unlike progesterone, ethynodiol-diethylstilbestrol is anti-spermatogenic at all dose levels; that is, in this respect its folliculoid properties predominate. Other luteoids (e.g., progesterone) inhibit both their own folliculoid actions and those of simultaneously given folliculoids, not only on the

vagina (causing mucification rather than cornification) and uterus (progestational rather than estrus changes) but also on the testis (inhibition of anti-spermatogenic effect). Hence, we may conclude that in the case of ethynodiol-diethylstilbestrol, which possesses both folliculoid and luteoid effects, the former are selectively inhibited by the latter on some target organs (e.g., vagina, uterus) but not on others (e.g., testis).

INTERRELATIONS BETWEEN THE PHARMACOLOGIC PROPERTIES AND THE CHEMICAL STRUCTURE OF THE STEROIDS (PHARMACO-CHEMICAL INTERRELATIONS)

Definition and Outline. — The study of the correlations between the chemical structure and pharmacologic activity of the steroids is also of great practical importance. It is the only means by which the efforts of the chemists, interested in the synthesis of biologically useful steroids, can receive a promising orientation. Very little has been done along these lines as yet, apart from a fairly systematic study concerning pharmaco-chemical correlations among the anesthetic steroids and among the testoid androstane derivatives.

A possible explanation for the deplorable lack of interest displayed in this field is that when pharmacologists attempt to study correlations between chemical structure and biologic action in any series of related compounds, they are often discouraged by striking instances which appear to contradict all their conclusions. An apparent instance of this type has already been encountered in the steroid field, inasmuch as certain stilbene derivatives share all the actions of the natural folliculoids (claims for allegedly corticoid and luteoid stilbene derivatives have been refuted) although they possess an entirely different chemical structure. It must be kept in mind, however, that pharmaco-chemical correla-

tions established for a certain chemical group do not necessarily hold for another series. Let us not forget that in many other fields of pharmacology (morphine derivatives, adrenaline derivatives, etc.) important new drugs became available to the medical profession only as a result of painstaking pharmaco-chemical studies, which gave a lead to the synthetic chemist concerning the type of compound which is likely to possess the desired pharmacologic property. We hope to show that even in the complex steroid molecule certain definite pharmaco-chemical correlations are already clearly demonstrable.

Pharmacological Rules Applicable to All or Several Independent Actions. — ALL ETIOCHOLANES ARE DEVOID OF HORMONAL ACTIONS. This is also true of all C₁₇ substituted etiocholanes such as the pregnanes. Etiocholanes may however, exhibit a pronounced anesthetic effect. Androstanane configuration at C₅ or unsaturation at Δ⁴ or Δ⁵, which removes the C₅ hydrogen characteristic of the etiocholanes, renders the steroid potentially capable of hormonal actions.

A 6 MEMBERED RING D IS COMPATIBLE BOTH WITH ANESTHETIC AND HORMONAL ACTIVITY as shown by the example of the D-homoandrostanes.

NO STEROID POSSESSING A LONG C₁₇ SIDE-CHAIN HAS ANY HORMONAL OR ANESTHETIC ACTIVITY. 5 and 6 carbon-side-chain compounds have not been adequately studied as yet, but "21-ethyl-progesterone"; that is, 17(β)-[1-ketobutyl]-Δ⁴-androstene-3-one with a 4 carbon atom side-chain, proved highly active as an anesthetic and also exhibited some folliculoid activity, while Δ⁴-nor-cholestene-3,25-dione, with a 7 carbon atom side-chain, proved entirely inert.

The STERIC POSITION OF THE SIDE-CHAIN IS VERY IMPORTANT, the natural 17(β) position being usually prefer-

able. Thus natural, that is 17(β)-progesterone and 17(β)-desoxycorticosterone are highly active as luteoids and corticoids respectively, while the corresponding isomeric 17(α)-alkyl compounds are practically inert. The synthetic 17(β)-alkyl derivatives are also very potent as shown by 17(β)-ethynyl-testosterone, 17(β)-ethynyl-estradiol, 17(β)-methyl-testosterone.

17-METHYL OR 17-ETHYNYL-SIDE-CHAINS TEND TO INCREASE THE ORAL ACTIVITY of compounds which are otherwise comparatively inactive by this route. Thus methyl-testosterone, ethynyl-estradiol and ethynyl-testosterone are more active when taken by mouth than estradiol or testosterone. However, such a side-chain may cause a qualitative change in the pharmacologic properties as shown by the fact that unlike testosterone, ethynyl-testosterone exhibits intense luteoid, but only slight testoid properties.

ESTERIFICATION CAUSES NO QUALITATIVE CHANGE IN ACTIVITY but may either increase or decrease the potency of a steroid. It usually acts by changing the rate of absorption, utilization or elimination of the free compound. In the case of most hormonal activities, potency is increased if esterification leads to an appropriate delay in the absorption rate and vice versa. Conversely, the anesthetic effect is most readily obtained if the organism is suddenly flooded with the compound. As regards the testoid effect of testosterone, enol-esters proved less potent, the corresponding hydroxy-esters more potent than the free compound.

OXIDATION AT BOTH EXTREME POLES OF THE MOLECULE APPEARS TO BE ADVANTAGEOUS FOR ALL BIOLOGIC ACTIVITIES (C₃ and C₁₇ among the estranes and androstanes and C₃ and C₁₇¹ or C₁₇² among the C₁₇ alkyl substituted androstanes).

In general it may be said that, judged by the evidence available at this time,

QUALITATIVE CHANGES IN PHARMACOLOGIC ACTIVITY MAY ONLY BE EFFECTED BY ALTERING CARBON TO CARBON LINKAGES OR THE STATE OF OXIDATION of a steroid.

Pharmaco-Chemical Rules Within Each Category of Independent Actions. — In this section we wish to review only the most important pharmaco-chemical correlations which appear to be responsible for each of the independent pharmacologic actions. We shall not repeat the general rules mentioned in the previous chapter, but it must be remembered that they also apply to all the independent actions.

Since only very little work has been done under strictly comparable bioassay conditions, most of the available data of the literature are unsuitable for this type of analysis. We summarize a few of the main facts, merely as examples of how such pharmaco-chemical correlations can be worked out.

The most convincing type of evidence is derived from the comparative bioassay of two compounds differing from each other only with respect to one detail in chemical structure. For instance, the comparison of two isomeric alcohols can show us the importance of the steric position of the hydroxyl in question. Comparison between a hydroxyl and the corresponding ketone tells us which of these two groups are more likely to increase potency. It must be realized, however, that these correlations are necessarily valid only if the rest of the molecule is identical. For instance, a 3-hydroxyl may be more advantageous for the production of a certain effect than the corresponding 3-ketone if the rest of the molecule has a certain structure, while the reverse relationship may obtain after introduction of another group at a different part of the molecule. Extrapolations to other steroids are permissible only if a certain pharmaco-chemical relationship has been noted in a large number of compounds.

The folliculoids. — All naturally occurring pharmacologically pure (simple) folliculoids are ESTRANE derivatives and conversely, all biologically active estrane derivatives are *pure* folliculoids devoid of any other activity.

A SIDE-CHAIN AT C₁₇ does not necessarily decrease folliculoid activity in the estrane series and under certain circumstances, it may even increase it as in ethenyl-estradiol or ethynodiol-estradiol.

All naturally occurring pure folliculoids are comparatively highly UNSATURATED, having three or more double bonds and a PHENOLIC HYDROXYL group. But neither the high degree of unsaturation, nor the phenolic hydroxyl, are essential for some degree of folliculoid activity in a compound having other independent actions as well. This is shown, for instance, by androsterone which is completely saturated, and devoid of a phenolic hydroxyl, yet possesses (usually masked) folliculoid activity. The unimportance of the phenolic hydroxyl has also been proven within the estrane group since Δ^{5,7,9:10}-estratriene-3(a), 17-diol is strongly folliculoid though less so than estradiol.

It has been claimed that the DISTANCE BETWEEN THE TWO POLAR OXYGENS (e.g., 3 and 17 in estrone and estradiol) is the chief prerequisite of folliculoid activity; however, some of the carcinogenic hydrocarbons, triphenylchloroethylene etc., are folliculoid although they possess no oxygen.

Attachment of a METHYL group at C₁₀ is compatible with folliculoid potency as shown by the androstane and allo-pregnane derivatives which possess this activity.

OPENING OF RING D is compatible with the highest degree of folliculoid activity as shown by the doisynolic acid derivatives.

The corticoids. — A 2-CARBON ATOM SIDE-CHAIN AT C₁₇ proved essential for corticoid activity in all compounds tested.

AN α,β UNSATURATED KETONE GROUP AT C₃ is not essential for corticoid activity since both pregnenolone and acetoxy pregnenolone proved to possess this property. Even completely saturated compounds (dihydro-corticosterone, that is 17(β)-[1-keto-2-hydroxyethyl]-androstane-3-one-11(β)-ol) may exhibit slight corticoid activity of the pro-muscular efficiency type. However, the α,β unsaturated ketone group is apparently advantageous since all the most active corticoids possess it.

A HYDROXYL, OR ACETOXY GROUP AT C₁₇² is beneficial as shown by the fact that desoxycorticosterone and its acetate are more potent than progesterone and acetoxy pregnenolone is more potent than pregnenolone.

OXYGEN AT C₁₁ in the form of a hydroxyl or ketone, proved indispensable for the gluco-corticoid and the pro-muscular efficiency activity of all steroids so far examined. Its importance is best shown by comparing corticosterone with desoxycorticosterone, since the latter differs from the former only in that it is deprived of the alcoholic oxygen at C₁₁. This loss leads to the disappearance of gluco-corticoid activity in desoxycorticosterone although the compound is highly active with regard to the life-maintaining and mineralo-corticoid effects.

The luteoids. — Only androstane derivatives possess luteoid activity but the possession of a C₁₇ SIDE-CHAIN, though very beneficial, is not indispensable since testosterone and Δ^4 -androstene-3,17-dione proved effective.

AN α,β UNSATURATED KETONE GROUP AT C₃ is advantageous and present in all of the most active luteoids (progesterone, ethynodiol-17 β -oate). Yet it is not essential since 17(β)-ethyl-androstane-3-one-17(α)-ol and 17(α)-ethyl-androstane-3(β)-17(α)-diol both proved effective.

SHIFT OF THE DOUBLE BOND from Δ^4 to Δ^5 (Δ^5 -iso-progesterone) greatly

diminishes or destroys luteoid potency. The addition to the Δ^4 of a Δ^6 -double bond as in 6-dehydro-progesterone, or a Δ^{11} double bond as in 11-dehydro-progesterone, cause only a slight decrease in potency. On the other hand an additional Δ^{16} double bond as in 16-dehydro-progesterone results in complete inactivation.

ANY DEVIATION FROM THE PROGESTERONE STRUCTURE decreases activity. 3-desoxy-progesterone proved inactive in the doses tested. Reduction of the C₁₇¹-ketone to a hydroxyl group likewise destroys activity. Addition of a C₆(α) or C₁₂ hydroxyl decreases but does not destroy luteoid potency. The same is true of the introduction of a 6-keto group. Addition of a C₁₁ or C₁₇-hydroxyl is claimed to destroy luteoid activity completely.

The testoids.— A C₁₇ SIDE-CHAIN is usually detrimental, but under certain conditions methyl-testosterone proved actually more active than testosterone; even -ethyl-testosterone and ethynodiol-17 β -oate — though less active than testosterone — possess marked testoid potency. The oral activity of androstanes improves by C₁₇-alkyl substitution. Methyl-testosterone is particularly active by mouth. Among C₁₇-ethyl substituted androstanes the prostatic effect is more common than the seminal vesicle stimulating potency. This is quite obvious in progesterone, pregnenolone and Kendall's Compound "E", that is 17(β)-[1-keto-2-hydroxyethyl]- Δ^4 -androstene-3,11-dione-17(α)-ol.

UNSATURATION is not of great importance for testoid activity; one of the most potent naturally occurring testoids (androsterone) is fully saturated. A shift of the double bond from Δ^4 , as in testosterone, to another position diminishes, but does not abolish activity as shown by the fact that the double bond isomerides Δ^1 -iso-testosterone (Δ^1 -androstene-3-one-17(α)-ol) and Δ^5 -iso-

testosterone are both endowed with some testoid activity. It is also true that if an additional double bond is introduced into testosterone, as in $\Delta^{4,6}$ -androstanediene-3-one-17(α)-ol, activity diminishes without disappearing.

A SINGLE OXYGEN suffices to endow androstanane with some testoid activity, as shown by the fact that the 17-amine of androstan-3-ol and the 3-chloro derivative of androstan-17-one are both slightly active, as is Δ^5 -androstene-3(β)-ol. The completely reduced parent hydrocarbon, androstanane, has not been assayed as yet. Introduction of MORE THAN THE USUAL TWO OXYGENS decreases activity, but adrenosterone (Δ^4 -androstene-3,11,17-trione) still possesses about one-fifth the potency of androsterone and androstan-3,17-dione-11-ol as well as androstan-3(β)-11-diol-17-one are highly testoid.

The presence at C₃ and C₁₇ of either ALCOHOLIC OR KETONIC OXYGEN is compatible with a high degree of testoid potency. Indeed, all not-alkyl-substituted androstanes or androstenes containing only 2 (alcoholic or ketonic) oxygens, one at C₃ and one at C₁₇, possess some testoid activity.

THE STERIC POSITION OF THE C₃ AND C₁₇ HYDROXYLS plays a very important rôle in determining the degree of testoid potency. In the great majority of the compounds investigated, the alpha position is preferable at both these locations.

A 17²-ACETOXY GROUP is definitely detrimental inasmuch as both desoxycorticosterone acetate and acetoxy-pregnenolone fail to exhibit the pro-prostatic effect which is so clear in the case of progesterone and pregnenolone. It will be remembered that the former two compounds differ from the latter two, only in that they have an additional 17²-acetoxy group. The free 17²-hydroxy compounds have not been adequately tested as yet, but presumably they act in essentially the same manner as the acetylated alcohols.

The spermatogenic steroids.—A C₁₇ SIDE-CHAIN is not essential, indeed not even of much consequence for this effect, since one of the most active compounds proved to be androstenediol, the other pregnenolone.

Completely SATURATED COMPOUNDS (androsterone), as well as Δ^4 - (testosterone, progesterone) or Δ^5 -unsaturated steroids (androstenediol, pregnenolone) were found to possess a high degree of spermatogenic activity.

The addition of an ACETOXY GROUP AT C₁₇² completely destroys spermatogenic activity in progesterone and pregnenolone as shown by the inactivity of desoxycorticosterone and acetoxy-pregnenolone.

The anesthetic steroids.—EVEN ETIOCHOLANE DERIVATIVES possess this effect.

A C₁₇ SIDE-CHAIN is beneficial, but not indispensable as shown by the fact that 17-ethyl substituted etiocholanes and androstanes are among the most active anesthetics, yet etiocholanes and androstanes, which are not alkyl substituted, also proved highly active.

One DOUBLE BOND, if it is situated in ring A or B, does not appear to interfere seriously with the anesthetic effect but 2 or more double bonds in these 2 rings or 1 double bond in ring D are detrimental.

The highest anesthetic effect is exhibited by STEROIDS OXYGENATED ONLY AT THE TWO EXTREME ENDS OF THE MOLECULE.

The STERIC POSITION OF THE HYDROXYL GROUP IN POSITION C₃ appears to be without importance for the anesthetic effect as shown by the observation that the two isomeric androsterones are of equal activity, as are the two isomeric etiocholane-3-ol-17-ones.

A 5-MEMBERED RING D is not indispensable for the anesthetic effect as shown by the high activity of D-homo-androstan derivatives.

GENERAL RULES REGULATING THE ACTIONS OF STEROID HORMONES

Compensatory Atrophy. — As with most other hormonal compounds, exogenous administration of steroid hormones causes compensatory atrophy of the cells normally concerned with their elaboration. This is a useful compensatory arrangement, since in the presence of excessive exogenous quantities of a certain hormone, there is no need for its production by the organism. In this compensation it is the pharmacologic activity which counts and not the chemical structure of the compound. Thus ovarian atrophy is also produced by stilbene derivatives, which bear no close chemical relationship to the folliculoids and Leydig cell atrophy is induced by synthetic testoids (e.g., methyl-testosterone), which normally neither occur nor give rise to physiologic testoid metabolites in the body.

It is customary to distinguish between SIMPLE AND TRANSFERRED COMPENSATORY ATROPHY. A compensatory atrophy is simple when it affects the cell type which normally elaborates the hormone administered. It is transferred, when it affects a cell type which normally does not produce the hormone given, but one having similar pharmacologic properties. Thus, for instance, the Leydig cell atrophy produced by testosterone is a simple compensatory atrophy, because the Leydig cells normally elaborate this substance. On the other hand, the Leydig cell atrophy produced by stilbestrol represents an example of transferred compensatory atrophy.

It is noteworthy that the compensatory atrophy of an endocrine cell, which normally produces several pharmacologically different hormones, often interferes not only with the secretion of the hormone given but also with that of these other, pharmacologically different, steroids. This is also a type of transferred compensatory atrophy. Thus, involution of the adrenal-cortical cells, due to desoxycorticosterone treatment,

interferes not only with the secretion of mineralo-corticoids, such as desoxy-corticosterone itself, but also with that of gluco-corticoids of the corticosterone type. This may explain the singular phenomenon that animals overdosed with desoxycorticosterone may exhibit signs of cortical deficiency (inasmuch as they have a great tendency to develop hypoglycemia), perhaps because of subnormal gluco-corticoid production.

It will be kept in mind that the terms simple and transferred compensatory atrophy bear no relation to the designations "direct" and "mediated" (or indirect) hormone actions. Compensatory atrophy may — as so many hormone actions — be due to the immediate, direct effect of a hormone upon the target organ or it may be mediated through another gland (e.g., the hypophysis).

Adaptation. — There is no clear-cut evidence, as yet, to prove that treatment with steroid hormones of any kind gives rise to the formation of true anti-hormones. However, following prolonged pretreatment with a steroid hormone, some kind of adaptation to it may occur. Thus, during chronic treatment with estradiol, the adrenal cortex at first undergoes marked hypertrophy and the body weight declines. Later, however, both the adrenal size and the body weight revert towards normal in spite of continued treatment. This is not an immunity to estradiol, as such, since animals pretreated with this compound become comparatively resistant, not only to this hormone, but also to the chemically unrelated stilbestrol.

Similarly, chronic treatment with anesthetic doses of steroid compounds produces resistance not only to the narcotic effect of the steroid with which pretreatment occurred, but also to that of other steroids. These observations clearly indicate that adaptation may occur, not only to chemical substances,

but also to certain pharmacologic actions.

Dissociated Adaptation. — It has been found that following treatment with steroid compounds, the organism may become selectively resistant to some of their actions, without acquiring insensitivity to others. This is termed "dissociated adaptation."

Thus, following repeated intraperitoneal injections of certain steroids, rats can acquire resistance to the anesthetic effect of these compounds without becoming insensitive to their hormonal actions. Similarly, rats chronically treated with desoxycorticosterone may become resistant to some properties of this compound (involution of the thymico-lymphatic apparatus) although the treatment continues to cause compensatory atrophy of the adrenals, hypochloremia, etc. Such examples of selective acquired resistance give additional support to the concept that adaptation to steroid hormones is not due to antihormone formation, since antihormones nullify all the actions of the corresponding hormones.

Inverse Response. — Depending upon circumstances, the same steroid hormone may exhibit two diametrically opposed pharmacologic actions. Observations which illustrate this fact have already been cited above, in other connections. Thus, we said that small doses of folliculoids enhance, while large doses inhibit the effects of luteoids. We have also seen that small doses of testosterone cause atrophy of the seminiferous epithelium, while large doses protect the spermatogenic elements against atrophy. The mechanism of this inverse response is not yet fully understood and it is rather probable that its cause is not the same in every instance.

Direct and Mediated Actions. — Some of the pharmacologic actions of steroids are apparently DIRECT. Thus, the effect of estradiol upon the vaginal epithelium or the endometrium appears

to be due to the direct action of this hormone upon the responsive cells, since it is most readily elicited by local application of the hormone. Similar experiments have proven the direct effect of progesterone upon the endometrium and of testosterone upon the capon's comb.

Other actions are MEDIATED OR INDIRECT. Thus, large doses of folliculoids cause corpus luteum formation and pregestational proliferation of the endometrium in intact females. These actions are apparently mediated through the anterior lobe of the pituitary whose gonadotrophic hormone and prolactin production is increased by them. This results in the formation of large, "pregnancy type" corpora lutea which in turn affect the endometrium through excessive secretion of progesterone. Here we have a clear-cut example of the indirect actions which are dependent, in the case of the ovarian response, upon one (hypophysis) and in the case of the endometrial reaction, upon two (hypophysis and ovary) intermediate stations, whose integrity is essential for the response.

Another type of mediated action is that which depends, not upon the stimulation of the hormone formation in an intermediate organ, but upon transformation in the body of an inactive into an active steroid. Thus certain folliculoid hormones are ineffective when directly applied to the vaginal epithelium in comparatively high doses, yet when injected subcutaneously, they cause vaginal cornification at dosage levels which could not give rise to a similar concentration in the vagina. Here, we must assume that it is within the organism that the injected steroid is transformed into an active folliculoid.

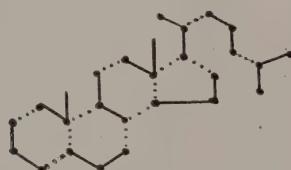
BIOGENESIS AND METABOLISM OF THE STEROIDS

It is obviously of the greatest importance to determine the manner in which the body forms the steroid hormones

(biogenesis) and the mechanism through which these are eventually destroyed or eliminated (metabolism).

Biogenesis. — There are two principal schools of thought concerning the mechanism through which the body makes steroid hormones. Some investigators believe that these are synthetized directly from smaller molecules, while others consider it more probable that they are formed from cholesterol by degradation of the side chain.

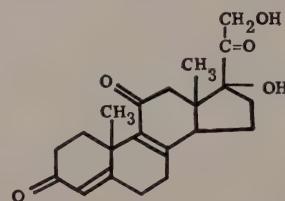
Reichstein expressed the view that the isolation of the numerous highly oxidized adrenal-cortical steroids — many of which still contain actual sugar remnants in their molecule — make the former hypothesis more likely. He considers it possible that the adrenal steroids are FORMED FROM SUGARS and are progressively reduced to cholesterol-like compounds. In this connection he emphasized that not only the hormones of the 21 carbon atom series, but numerous other physiologically important steroids, such as cholesterol and the bile acids, contain a skeleton in which the number of carbon atoms is divisible by 3 and could theoretically be reconstructed from three or six carbon-atom chains, such as are typical of sugars (e.g., dihydroxy-acetone, glyceric aldehyde). This is illustrated by the following formula :



Rittenberg described experiments on mice in which a constant deuterium content of 1.5 atom percent was maintained. After 60 days the cholesterol in the body of these mice showed a relation between deuterium and hydrogen content which was half that found in body fluids. It was concluded that at least 22 H atoms of cholesterol are exchangeable with the deuterium of the

body fluids in some step of its biogenesis. This fact was interpreted to mean that cholesterol is SYNTHETIZED IN THE BODY FROM SMALLER MOLECULES. Furthermore, deuterium has been found in pregnanediol, isolated from the urine of a woman to whom deuterio-cholesterol had been administered (*Bloch*). Apparently, $\frac{1}{2}$ to $\frac{2}{3}$ of the pregnanediol excreted arose by degradation of this cholesterol. The possibility of hydrogen-deuterium exchange reactions has been eliminated, since the amount of deuterium present in the cholesterol and pregnanediol thus formed, is greater than could thus be accounted for. All these findings suggest that cholesterol is first built up from one, two or three carbon compounds and subsequently, degraded to the steroid hormones. In the body, these reactions do not proceed automatically, however, since otherwise, an increased intake of such 1-3 carbon compounds, or of cholesterol, would necessarily lead to an increased steroid hormone production. Probably, the gonado- and adrenotropic hormones play an important rôle in determining not only the total amount, but also the type of the steroid hormones produced from whatever their precursor may be.

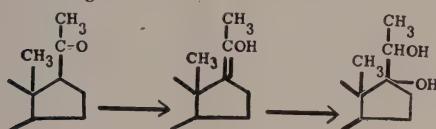
Marker suggested that at least all cortical steroids may be FORMED BY REDUCTION OF A HITHERTO NOT YET ISOLATED HYPOTHETIC PARENT COMPOUND :



This highly oxygenated steroid "may be the as yet unisolated cortical hormone." However, this theory is not supported by any established fact.

Marrian expressed a somewhat different view and suggested that the STEROIDS WITH C_{17} TERTIARY HYDROXYLS

ARE SECONDARILY FORMED BY THE ADDITION OF H₂O TO THE DOUBLE BOND OF AN ENOLIZED C₁₇¹ KETO-STEROID in the following manner:



If Marrian's theory of C₁₇ side-chain oxidation is correct, corticoids could be formed from progesterone and testoid androstanes could arise from 17-ethyl-androstanes, by oxidation between C₁₇ and C₁₇¹ which could lead to C₁₇ oxygenated androstan derivative.

Several investigators speculated upon the POSSIBLE INTERCONVERSIONS OF STEROID HORMONES WITHIN THE BODY. In the chart below we summarized what we consider a possible route for the biogenesis of the different types of hormonal compounds, assuming that they originate from pregnenolone which in turn may arise either from cholesterol by degradation of the side-chain, or by direct synthesis from smaller molecules. It was felt that such a synopsis would help to visualize the possible hormone interconversions which may take place within the organism, as well as the form in which the hormones can be recovered from urine. It must be admitted that many steps indicated in the table rest on insufficient evidence. However, some of these interconversions have definitely been proven by injecting a certain chemical compound and recovering its metabolites from the urine. Only those urinary steroids are listed which were actually isolated from normal human urine and are presumably of major importance. These observations proved beyond doubt the pharmacologically most important fact, that the biologic results elicited by a certain compound are not necessarily caused by this compound, as such, but may be due to other steroids formed within the body from the injected compound.

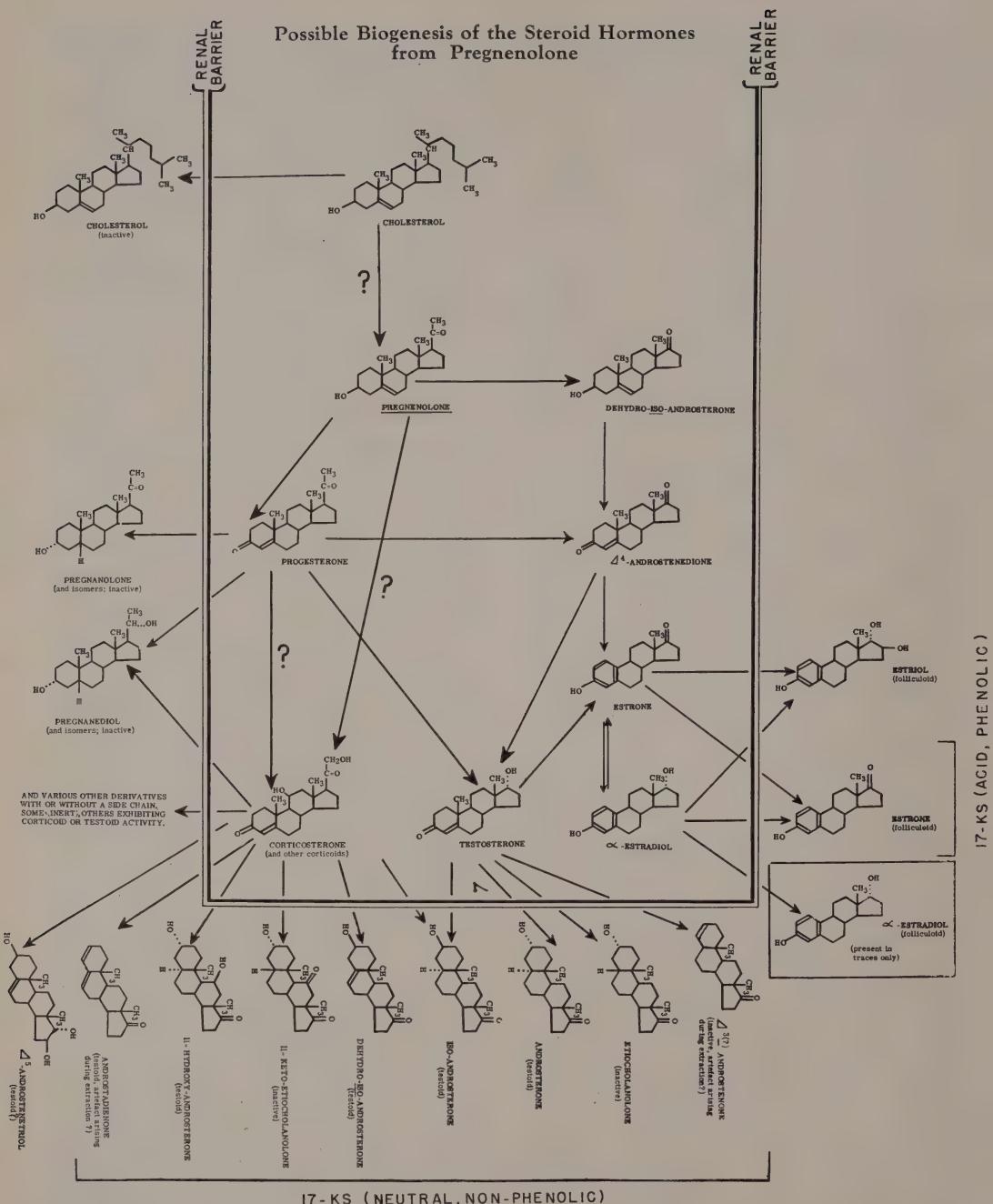
A study of the chart shows the prominent rôle played by pregnenolone. If the hormones are derived from cholesterol this compound would appear to be the logical parent substance of all steroid hormones, as it differs from cholesterol only in the side-chain. Pharmacologically, pregnenolone also occupies a rather unique position as it is the only compound known to possess all the independent steroid hormone actions. (The testoid action is somewhat doubtful as pregnenolone stimulates only the prostate in castrate rats.) It appears that the specialization for a certain pharmacologic action occurs gradually and always at the expense of other activities which are present — even if only to a slight degree — in the parent compound. This differentiation is rather reminiscent of the evolution of highly specialized cells from "multi-potent" but undifferentiated embryonic cells.

It will be noted that the direction of the conversions which occur in the body obviously cannot be merely automatic. If this were the case, pregnenolone — or any other steroid which would prove to be the hypothetical parent substance — would always give rise to the same hormone combination. This would not allow for the hormonal differences between male and female organisms and the qualitative changes in the type of hormone production during the various phases of the estrous cycle, pregnancy, adaptation to changes in environment, etc. It is highly probable that the power to direct such interconversions is within the steroid hormone producing endocrine cells, which in turn are mainly under pituitary control. Conversely, through hormonal mechanisms, such as those responsible for compensatory hypertrophy or atrophy, the steroid-producing cells can act back on the pituitary, thus regulating its "trophic" hormone production.

Many puzzling pharmacologic problems may perhaps be solved by the elucidation of these hormone interconver-

THE STEROIDS

Possible Biogenesis of the Steroid Hormones from Pregnenolone



sions in the body. Thus there is a surprising difference in the folliculoid action of saturated and unsaturated androstanes. Compounds having a double bond in rings A or B tend to exhibit a marked pro-vaginal corni-

fication effect, while little or no such potency could be detected in saturated androstanes. This may mean that the folliculoid activity of androstanes is, at least partly, due to conversion into the highly unsaturated estratrienes.

Metabolic factors influencing the activity of the steroids. — It is not within the scope of this book to discuss, in detail, the extensive literature concerning the metabolism of steroids. The many species differences in the manner in which the body handles these compounds, the isolation from urine, or tissues, of incompletely identified steroids, etc., are of interest only to the specialist. Hence, we shall limit ourselves to a brief survey of the salient metabolic factors which are likely to influence the activity of the steroids.

In general, enterally or parenterally given steroids are well ABSORBED. Unlike the protein hormones, they are not readily destroyed by the gastrointestinal juices, and unlike for cholesterol, bile is not essential for the intestinal absorption of the steroids. That most steroids are comparatively inactive when given by mouth is not due to destruction in the intestine or slow absorption but, on the contrary, to unduly rapid absorption and consequent speedy destruction and excretion (in urine and bile) before their full effect could take place. To some extent the relative inefficiency of orally administered steroids is also due to the circumstance, that they are directly led to the liver through the portal vein.

The LIVER plays an important rôle in the detoxification of all steroid hormones. After partial hepatectomy both the hormonal and the anesthetic effects of steroids are greatly increased. Furthermore, subcutaneous implantation of hormone pellets is more effective than intra-splenic implantation, presumably because in the latter case all the absorbed material is immediately exposed to the detoxifying hepatic cells as it passes through the portal circulation. We do not know as yet what chemical factors are responsible for this action of the liver, but judged by in vitro experiments on the inactivation of estratrienes by liver slices, it is highly prob-

able that enzymes play an important part. The detoxification may be achieved by degradation to inactive compounds or by conjugation, which make the steroids less active or increase the rate of their elimination.

In the case of desoxycorticosterone acetate and progesterone, HYPOPHSECTOMY or ADRENALECTOMY increases the animal's sensitivity towards the anesthetic effect, while no such increase is obtained by NEPHRECTOMY or THYROIDECTOMY. Apparently the hypophysis and the adrenal also play an important part in the detoxification of the steroids and perhaps these glands exert their effect partly through their action upon the hepatic cells.

The general depression of metabolism caused by thyroidectomy and the interference with renal elimination occasioned by nephrectomy on the other hand, cause no important change in the sensitivity of the organism to steroids. It must be kept in mind that probably a large part of the excreted steroid hormone degradation products and conjugated steroids are inactivated even before they are excreted.

Hypophysectomy may even cause a qualitative change in the response to certain steroids as shown by the example of testosterone which normally causes mucification but after hypophysectomy induces persistent cornification of the rat vagina. It remains to be seen whether the folliculoid activity assumed by the compound after hypophysectomy is due to an improvement in its transformation into an active estratriene, or whether removal of the pituitary merely inhibits those actions of the testosterone molecule which normally "mask" its own folliculoid properties.

The above-mentioned examples clearly demonstrate that factors within the organism are of the greatest importance in determining the pharmacologic response to a steroid compound.

I. Steroids isolated from the adrenal *

<i>Systematic name in terminology of this book</i>	<i>Other names</i>
$\Delta^{1,3,5,10}$ -estratriene-3-ol-17-one	Estrone
$\text{androstan-3}(\beta),11\ (\beta)$ -diol-17-one	Reichstein's mono-ketone m.p.: 236°
Δ^4 -androstene-3,11,17-dione	
Δ^4 -androstene-3,11,17-trione	Adrenosterone
$[7(\beta)-[1\text{-ketoethyl}]\text{-androstane-3}(\beta)]\text{-ol}$	Allo pregnanolone
$[7(\beta)\text{-}[1(a)\text{-hydroxyethyl}]\text{-androstane-3}(\beta),17(a)\text{-diol}$	Reichstein's cpd. "O"
$17(\beta)\text{-}[11(\beta)\text{-hydroxyethyl}]\text{-androstane-3}(\beta),17(a)\text{-diol}$	Reichstein's cpd. "J"
$17(\beta)\text{-}[1\text{-ketoethyl}]\text{-androstane-3}(\beta),17(a)\text{-diol}$	Reichstein's cpd. "L"; Wintersteiner's cpd. "G".
$17(\beta)\text{-}[11(\beta),2\text{-dihydroxyethyl}]\text{-androstane-3}(\beta),17(a)\text{-diol}$	Reichstein's cpd. "K".
$17(\beta)\text{-}[1\text{-keto-2-hydroxyethyl}]\text{-androstane-3}(\beta),11(?)\text{-diol}$	Reichstein's cpd. "R".
$17(\beta)\text{-}[1\text{-keto-2-hydroxyethyl}]\text{-androstane-3}(\beta),17(a)\text{-diol}$	Reichstein's cpd. "P".
$17(\beta)\text{-}[1\text{-keto-2-hydroxyethyl}]\text{-androstane-3}(\beta)\text{-ol-11-one}$	Reichstein's cpd. "N"; Wintersteiner's cpd. "H".
$17(\beta)\text{-}[1(?)\text{-2-dihydroxyethyl}]\text{-androstane-3}(\beta),11(\beta),17(a)\text{-triol}$	Reichstein's cpd. "A"; Kendall's cpd. "D"; Wintersteiner's cpd. "A".
$17(\beta)\text{-}[1\text{-keto-2-hydroxyethyl}]\text{-androstane-3}(a),11(?)\text{-17(a)-triol}$	Reichstein's cpd. "C"; Kendall's cpd. "C"; Wintersteiner's cpd. "D".

* Continued on next page.

<i>Systematic name in terminology of this book</i>	<i>Other names</i>
17(β)-[1-keto-2-hydroxyethyl]- <i>androstane-3(β),11(β),17(α)-triol</i>	Reichstein's cpd. "V".
17(β)-[1-keto-2-hydroxyethyl]- <i>androstane-3(β),17(α)-diol-11-one</i>	Reichstein's cpd. "D"; Kendall's cpd. "G"; Wintersteiner's cpd. "B".
17(β)-[1-ketoethyl]- \triangle^4 - <i>androstene-3-one</i>	Progesterone
17(β)-[1-ketoethyl]- \triangle^4 - <i>androstene-3-one-17(α)-ol</i>	17(β)-hydroxy-progesterone
17(β)-[1-keto-2-hydroxyethyl]- \triangle^4 - <i>androstene-3-one</i>	Desoxycorticosterone; Reichstein's cpd. "Q"; Kendall's desoxy cpd. "B".
17(β)-[1(?)2-dihydroxyethyl]- \triangle^4 - <i>androstene-3,11-dione</i>	Reichstein's cpd. "T".
17(β)-[1-keto-2-hydroxyethyl]- \triangle^4 - <i>androstene-3-one-11(β)-ol</i>	Corticosterone; Reichstein's cpd. "H"; Kendall's cpd. "B"
17(β)-[1-keto-2-hydroxyethyl]- \triangle^4 - <i>androstene-3-one-17(α)-ol</i>	Reichstein's cpd. "S".
17(β)-[1-keto-2-hydroxyethyl]- \triangle^4 - <i>androstene-3,11-dione</i>	Dehydro-corticosterone; Kendall's cpd. "A".
, β -Unsaturated ketone ($C_{21}H_{28-30}O_4$) constitution unknown	
17(β)-[1(?)2-dihydroxyethyl]- \triangle^4 - <i>androstene-3-one-11(β),17(α)-diol</i>	Reichstein's cpd. "E".
17(β)-[1-keto-2-hydroxyethyl]- \triangle^4 - <i>androstene-3-one-11(β),17(α)-diol</i>	17-hydroxy-corticosterone; Reichstein's cpd. "M"; Kendall's cpd. "F".
17(β)-[1(?)2-dihydroxyethyl]- \triangle^4 - <i>androstene-3,11-dione-17(α)-ol</i>	Reichstein's cpd. "U".
17(β)-[1-keto-2-hydroxyethyl]- \triangle^4 - <i>androstene-3,11-dione-17(α)-ol</i>	Reichstein's cpd. "Fa"; Kendall's cpd. "E"; Wintersteiner's cpd. "F"; 17-hydroxy-dehydrocorticosterone.

Note that the configuration of the 17-hydroxyls is designated contrary to hitherto accepted convention which described them as " β ".

II. Steroids isolated from urine *

<i>Systematic name in terminology of this book</i>	<i>Other names</i>	<i>Sources</i>
<i>Estrane-3(?)₁₇(a)-diol</i>	Octahydro-estrone; Estranediol B	Man ♀
<i>Estrane-3(?)₁₇(?)₋diol</i>	Estranediol A	Man ♀
$\triangle^{1,3,5,10-} \text{-estratriene-3-17(a)-diol}$	α -estradiol	Pregnant: woman, mare
$\triangle^{1,3,5,10-} \text{-estratriene-3,17(\beta)-diol}$	β -estradiol	Pregnant mare
$\triangle^{1,3,5,10-} \text{-estratriene-3-ol-17-one}$	Estrone	Man ♂ stallion, cattle ♂ ; pregnant: woman, mare
$\triangle^{1,3,5,10-} \text{-estratriene-3-ol-17-one-Na, sulphate}$	Estrone as Na. sulphate	Pregnant mare
$\triangle^{1,3,5,10-} \text{-estratriene-3,16(\beta),17(a)-triol}$	Estriol	Pregnant woman
$\triangle^{1,3,5,10-} \text{-estratriene-3,16(\beta),17(a)-triol-Na glucuronide}$	Estriol as Na. glucuronide	Pregnant woman
$\triangle^{5,10,6,8-} \text{-estratriene-3(\beta)-ol-17-one}$	—	Pregnant mare
$\triangle^{1,3,5,10,7-} \text{-estratetraene-3-ol-17-one}$	Equilin	Pregnant mare
An isomer of above	Hippulin	Pregnant mare
$\triangle^{1,3,5,10,6,8-} \text{-estratetraene-3,17(\beta)-diol}$	17β -dihydro-equilenin	Pregnant mare
$\triangle^{1,3,5,10,6,8-} \text{-estratetraene-3-ol-17-one}$	Equilenin	Pregnant mare
$\triangle^{1,3,5,10,6,8-} \text{-estratetraene-11,17-dione}$	3-desoxy-11-keto-equilenin	Pregnant mare
<i>Androstone-3(α)₁₇(a)-diol</i>	—	Man ♂
<i>Androstone-3(β)-ol-x-one</i>	—	Pregnant mare
<i>Androstone-3(α)-ol-17-one</i>	Androsterone, cis-androsterone	Man ♂ , δ / c, ♀ , ♀ with adrenal tumor; cattle ♂ ; pregnant: woman, cow
<i>Androstone-3(α)-ol-17-one-Na, sulphate</i>	Androsterone as Na. Sulphate	Man ♂ with Leydig cell tumor
<i>Androstone-3(β)-ol-17-one</i>	Iso-androsterone, trans-androsterone	Man ♀ , ♀ with adrenal hyperplasia; ♀ with ovarian cysts, ♂ with cancer
<i>Androstone-3,17-dione</i>	Androstanedione	Man ♂ , ♀ with adrenal carcinoma
<i>Androstone-3(α)11(β)-diol-17-one</i>	11-hydroxy-androsterone	Man ♂ , adrenal carcinoma
$\triangle^{2?} \text{-androstene-17-one}$	$\triangle^{2?}$ -androsterone	Man: ♂ , ♂ with cancer, ♀ / c, (Artifact?)
$\triangle^5 \text{-androstene-3(\beta)-ol-17-one}$	Dehydro-iso-androsterone	Man ♂ , ♂ / c, ♀ , ♀ with adrenal tumor; cattle ♂ ; pregnant: cow

<i>Systematic name in terminology of this book</i>	<i>Other names</i>	<i>Sources</i>
$\Delta^{11}\text{-}androstene\text{-}3(\alpha)\text{-ol}\text{-}17\text{-one}$		Man ♀ with adrenal tumor
$\Delta^5\text{-}androstene\text{-}3(\beta)\text{-}16(\beta)\text{-}17(\beta)\text{-triol}$		Man with adrenal carcinoma
$\Delta^{3,5}\text{-}androstadiene\text{-}17\text{-one}$		Man ♂, ♀ and ♀ with adrenal tumor, (Artefact?)
$17(\beta)\text{-}[1(\alpha)\text{-hydroxyethyl]}\text{-}androstane\text{-}3(\alpha)\text{-ol}$	Epi- <i>allo-pregnane-diol</i> , <i>Allo-pregnane-3(\alpha),20(\alpha)-diol</i>	Man ♀, cattle ♂ ; pregnant: woman, cow, mare
$17(\beta)\text{-}[1(\alpha)\text{-hydroxyethyl]}\text{-}androstane\text{-}3(\beta)\text{-ol}$	<i>Allo-pregnane-3(\beta),20(\beta)-diol</i>	Cattle ♂ ; pregnant: woman, cow, mare
$17(\beta)\text{-}[1(\beta)\text{-hydroxyethyl]}\text{-}androstane\text{-}3(\beta)\text{-ol}$	<i>Allo-pregnane-3(\beta),20(\beta)-diol</i>	Pregnant mare
$17(\beta)\text{-}[1\text{-ketoethyl]}\text{-}androstane\text{-}3(\alpha)\text{-ol}$	Epi- <i>allo-pregnanolone</i>	Pregnant woman
$17(\beta)\text{-}[1\text{-ketoethyl]}\text{-}androstane\text{-}3(\beta)\text{-ol}$	<i>Allo-pregnanolone</i>	Pregnant: woman, sow, mare
$17(\beta)\text{-}[1\text{-ketoethyl]}\text{-}androstane\text{-}3\text{-one}$	<i>Allo-pregnandione</i>	Pregnant mare
$17(\beta)\text{-}[1(\text{?})\text{-hydroxyethyl]}\text{-}androstane\text{-}3(\alpha)\text{-}16(\text{?})\text{-diol}$	Pregnanetriol "B"	Pregnant mare
$17(\beta)\text{-}[1(\text{?}),2\text{-dihydroxyethyl]}\text{-}androstane\text{-}3(\beta)\text{-}11(\text{?})\text{-diol}$	<i>Allo-pregnane-3(\beta),11,20,21-tetrol</i>	Horse ♂
<i>Etiocolane</i> -3(α),17(α)-diol		Man ♂
<i>Etiocolane</i> -3(α)-ol-17-one	Etiocolanolone	Man ♀, ♀/c, ♂/c, ♀ with adrenal tumor, with breast cancer
<i>Etiocolane</i> -3(α)-ol-11,17-dione	11-keto- <i>etiocolanolone</i>	Man ♂, adrenal carcinoma
$17(\beta)\text{-ethyl-}etiocolane\text{-}3(\alpha)\text{-ol}$	Epi- <i>pregnanol-3</i>	Pregnant: woman
$17(\beta\text{-}[1(\alpha)\text{-hydroxyethyl]}\text{-}etiocolane\text{-}3(\alpha)\text{-ol}$	<i>Pregnanediol</i>	Pregnant: woman, chimpanzee, mare, cow
$17(\beta)\text{-}[1(\alpha)\text{-hydroxyethyl]}\text{-}etiocolane\text{-}3(\alpha)\text{-ol-Na. glucuronide}$	Pregnanediol as Na. glucuronide	Man ♀, ♀/c, ♂ ; cattle ♂
$17(\beta\text{-}[1\text{-ketoethyl]}\text{-}etiocolane\text{-}3(\alpha)\text{-ol}$	Epi- <i>pregnanolone</i>	Pregnant: woman, sow
$17(\beta\text{-}[1\text{-ketoethyl]}\text{-}etiocolane\text{-}3\text{-one}$	<i>Pregnandione</i>	Pregnant: mare
$17(\beta)\text{-}[1(\text{?})\text{-hydroxyethyl]}\text{-}etiocolane\text{-}3(\alpha),17(\alpha)\text{-diol}$	Pregnanetriol	Man ♀, ♀ with adrenal tumor
$17(\beta)\text{-ethyl-}9\text{-epietiocolane-}3(\beta),11(\text{?})\text{-diol}$	Urine-3(β).11()-diol	Pregnant mare (structure uncertain)
$17(\beta)\text{-ethyl-}9\text{-epietiocolane-}3\text{-one-}11(\text{?})\text{-ol}$	Urine-3-one-11()-ol	Pregnant mare (structure uncertain)
$17(\beta)\text{-}[1(\alpha)\text{-hydroxyethyl]}\text{-}9\text{-epietiocolane-}3(\alpha),$ $11(\beta)\text{-diol}$	Urine-3(α).11(β).20(α)-triol; Pregnanetriol "A"	Stallion; Pregnant: woman, mare (structure uncertain)
Structure unknown	Active urinary corticoid	Man ♂, ♀, adrenal tumor, alarm reaction

III. Steroids isolated from the testis

<i>Systematic name in terminology of this book</i>	<i>Other names</i>
$\Delta^{1,3,5:10}\text{-estratriene-3,17}(\alpha)\text{-diol}$	α -estradiol
$\Delta^{1,3,5:10}\text{-estratriene-3-ol-17-one}$	Estrone
<i>Androstane-3,17-dione</i>	—
$\Delta^4\text{-androstene-3-one-17}(\alpha)\text{-ol}$	Testosterone
Steroid of unknown structure $C_{21}H_{32}O_3$	Testalolone
$17(\beta)\text{-[1-ketoethyl]-}\Delta^5\text{-androstene-3}(\beta)\text{-ol}$	Pregnenolone
$17(\beta)\text{-[1-ketoethyl]-etiocholane-3}(\beta)\text{-ol}$	Allo-pregnane-3(β)-ol-20-one
$\Delta^{16}\text{-androstene-3}(\alpha)\text{-ol}$	—
$\Delta^{16}\text{-androstene-3}(\beta)\text{-ol}$	—

IV. Steroids isolated from the ovary

<i>Systematic name in terminology of this book</i>	<i>Other names</i>
$\Delta^{1,3,5:10}\text{-estratriene-3,17}(\alpha)\text{-diol}$	α -estradiol
$17(\beta)\text{-[1-ketoethyl]-androstane-3}(\beta)\text{-ol}$	Allopregnanolone
$17(\beta)\text{-[1-ketoethyl]-}\Delta^4\text{-androstene-3-one}$	Progesterone

V. Steroids isolated from the placenta

<i>Systematic name in terminology of this book</i>	<i>Other names</i>
$\Delta^{1,3,5:10}\text{-estratriene-3,17}(\alpha)\text{-diol}$	α -estradiol
$\Delta^{1,3,5:10}\text{-estratriene-3-ol-17-one}$	Estrone
$\Delta^{1,3,5:10}\text{-estratriene-3,16}(\beta),17(\alpha)\text{-triol}$	Estriol (also occurs as glucuronide)

REFERENCES

DODDS, E. C.: *Possibilities in the Realm of Synthetic Estrogens*. Vitamins and Hormones. Advances in research and applications. Ed. by R. S. Harris and K. V. Thimann. 3, 229 (1945). Academic Press Inc. Publ. New York (1945).

A very brief (6 pages), but most authoritative and stimulating summary, of problems in the field of artificial folliculoids.

FIESER, L. F.: *The Chemistry of Natural Products Related to Phenanthrene*. Reinhold Publishing Corporation. New York, U.S.A. (1936).

This volume (358 pages) deals with the chemistry of all phenanthrene derivatives from the chemical point of view; only occasional annotations concerning biologic activity are included. The volume is very authoritative, but outdated.

MASSON, GEORGES: *Les Hormones artificielles*. Rev. Canad. de Biol. 3, 491 (1944).

A detailed (90 pages) review concerning the artificial folliculoids and their derivatives. It lists the principal chemical and pharmacologic properties of these compounds and gives a brief synopsis of pertinent bioassay methods as well as of the pharmaco-chemical correlations within this group. It is useful mainly to research workers who specialize in this field. (In French.)

PINCUS, GREGORY AND WILLIAM H. PEARLMAN: *The Intermediate Metabolism of the Sex Hormones*. Vitamins and Hormones. Advances in Research and Applications. Ed. by R. S. Harris and K. V. Thimann. 1, 294 (1943). Academic Press Inc. Publ., New York (1943).

Very authoritative review (47 pages, 244 references), concerning the biogenesis and metabolism of folliculoid and testoid hormones. Numerous synoptic tables and charts help to illustrate this very readable and instructive article. It does not attempt to survey the pertinent literature completely.

REICHSTEIN, T. AND C. W. SHOPPEE: *The Hormones of the Adrenal Cortex*. Vitamins and Hormones. Advances in Research and Applications. Ed. by R. S. Harris and K. V. Thimann. 1, 346 (1943). Academic Press Inc. Publ. New York (1943).

A rather detailed masterly review (52 pages, 207 references). A brief introductory

section deals with the biologic importance and bioassay of the corticoids, but most of the text is devoted to the isolation, structure and partial synthesis of the corticoids and their derivatives.

SELYE, HANS: *The pharmacology of Steroid Hormones and their Derivatives*. Rev. Canad. de Biol. 1, 577 (1942).

Fairly extensive review concerning the pharmacology of the steroid hormones and their derivatives, with a brief section which deals with their metabolism.

SELYE, HANS: *Encyclopedia of Endocrinology, Section I, Classified Index of Steroid Hormones and Related Compounds*. Vol. 1-4, A.W.T. Franks, Publ., Montreal (1943).

An index listing the main chemical and pharmacologic properties of the steroid hormones and their derivatives. The volumes are presented in loose-leaf form, a separate page being assigned to each of the (more than 700) parent compounds. — The introductory section gives a concise description of the chemical and pharmacologic nomenclature and system of classification. An appendix contains synoptic charts (which list the steroids naturally occurring in various biologic materials), a dictionary of bioassay techniques, etc. These volumes are useful only to research workers, specializing in the steroid hormone field.

SOBOTKA, HARRY: *The Chemistry of the Steroids*. The Williams & Wilkins Company, Baltimore (1938).

An extensive (634 pages) treatise describing the principal chemical properties of the steroids. This interesting volume is most useful to those interested in steroid chemistry, but is somewhat out of date.

STRAIN, WILLIAM H.: *Chapter 19. The Steroids*. Organic Chemistry. An Advanced Treatise. Ed. by Henry Gilman 2, 1341 (1943). 2nd Ed. John Wiley & Sons, Inc. Publ. New York, (1943).

A large (190 pages) and excellently written chapter which deals with the chemistry of the steroids. This is undoubtedly one of the best, comparatively up-to-date, descriptions of this field. It is most useful to those interested in the purely chemical aspects of the subject, and does not attempt to discuss biologic and pharmacologic problems.

THE ADRENALS

HISTORIC INTRODUCTION

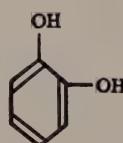
The adrenals (or suprarenals) were apparently first described by *Eustachius* (1563) in his book "De Glandulis Quæ Renibus Incubunt". It took more than 300 years, however, before any indication of their probable action was obtained. In those early days, when the public considered it sinful to dissect the human body, autopsies had to be performed in secret, often several days after death. By that time the medulla, which is highly subject to autolysis, was usually transformed into a viscous dark fluid ("atra bile"), within the more resistant cortex. Hence, the adrenals were thought to be capsules filled with liquid ("capsula suprarenalis").

Adrenaline.—*Vulpian* (1856) discovered that the cells of the adrenal medulla differ, in their staining ability, from those of the cortex. He noted that if a slice across an adrenal is immersed into a ferric chloride solution, the medulla takes a greenish tinge, while the cortex does not. Subsequently, *Henle* (1865) observed that certain granules in the medullary cells give a reddish-brown precipitate with dilute solutions of potassium bichromate, the so-called "chromaffin reaction." The use of this method enabled *Wiesel* (1904) to describe the chromaffin system as we know it today.

Pressor adrenal extracts were first prepared by *Oliver & Schäfer* (1894). The active principle of the adrenal medulla was subsequently isolated, in the form of its very insoluble benzoylate, as an amorphous but highly potent powder, by *Abel* (1899), who gave the

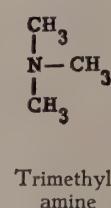
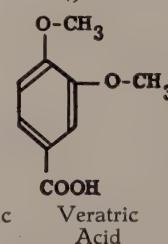
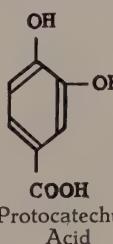
name "epinephrine" to this ester. Still amorphous, but highly purified preparations of the hormone, were obtained by precipitation of the insoluble metallic salts of adrenaline and subsequent splitting of the latter (*v. Fürth*, 1897-1903). Pure crystalline preparations of natural 1-adrenaline were first described by *Aldrich* (1901) and *Takamine* (1901). The hormone itself was named suprarenin (*v. Fürth*) or adrenaline (*Takamine*). The preparation of the hormone led to the determination of its empirical formula as $C_9H_{13}O_3N$.

The characteristic ferric chloride reaction (*Vulpian*, 1856), raised the suspicion that adrenaline is a pyrocatechol derivative.



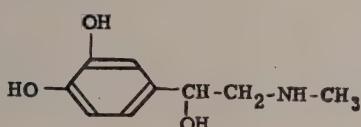
Pyrocatechol

This was confirmed (*Takamine*, 1902) by showing that, on melting adrenaline with potassium hydroxide, protocatechuic acid is obtained, while, upon exhaustive methylation and subsequent oxidation, trimethylamine and veratic acid are formed (*Jowett*, 1904).



Subsequently, *Stolz* (1905) and *Dakin* (1905) simultaneously, succeeded in preparing racemic *dl*-3, 4-dihydroxyphenylethanomethylamine — that is to say, racemic adrenaline — at a time when their views concerning the chemical structure of the compound were not yet precise.

These observations are of considerable historic interest, since they represent for the first time, the isolation of a pure crystalline hormone and its synthesis. They also show that the synthetic preparation of a hormone, can succeed even on the basis of very uncertain concepts concerning its structure. It was actually the synthesis of adrenaline and the subsequent work of *Friedmann* (1906), which definitely established that the hormone possesses the formula :



This was subsequently separated into the two optical isomers (*Flächer*, 1908).

Sympathin. — In 1902, *Langley* observed the similarity between the actions of adrenal extracts and those that follow sympathetic nerve stimulation. *Elliott* (1904) came to the conclusion that "adrenaline might then be the chemical stimulant liberated on each occasion, when the impulse arrives at the periphery," because of the great similarity between the effects of adrenaline administration and sympathetic nerve stimulation. But it was not until 1921, when *Loewi* published the results of his now classical experiments, that the sympathin theory obtained experimental confirmation. *Loewi* showed that Ringer's solution, circulating through the isolated heart of a frog, can acquire sympathicomimetic properties following stimulation of the sympathetic fibers in the vagosympathetic nerve.

(In the frog, vagal and sympathetic fibers course in a common nerve.) Such Ringer solution increases the pulse rate and the altitude of contractions, when applied to a second heart. *Loewi* termed the hypothetic substance, liberated during sympathetic stimulation, "accelerans substance" or, in German, "Acceleransstoff." In the same year *Cannon* and *Uridil* (1921) observed that stimulation of hepatic nerves liberates a substance into the blood stream, which increases the pulse rate of the denervated heart in the cat.

Subsequent investigators furnished ample evidence showing that sympathetic stimulation causes the liberation of a substance at the nerve endings in many organs innervated by "adrenergic," or adrenaline-imitating nerves. This substance has been termed "sympathin" (*Cannon* et al. 1931). (Cf. p. 114.)

Hyperadrenalinism. — *Fränkel* was probably the first, in 1886, to describe a case of an adrenal tumor, which led to typical symptoms of hyperadrenalinism, that is, attacks of cardiac palpitation, anguish, dizziness, headache, hypertension and frequent epistaxis. Death occurred suddenly in collapse. Autopsy revealed tumors in both suprarenals. The neoplasms were thought to be angiosarcomas, but from the description given, it is evident that they were tumors of the chromaffin tissue.

The first cases, in which the causative neoplasm was correctly identified by the pathologists, were those of *Kolisko* (1910), *Herde* (1912) and *Helly* (1913).

In 1927, *C. H. Mayo* saw a woman, who suffered from repeated attacks of paroxysmal hypertension, pulmonary edema, pallor and vomiting. Exploratory operation revealed a tumor in the region of the left adrenal. Although the nature of this growth was not recognized at the time of the operation, it was removed and the patient

became symptom-free. In the first publication, the pathologic diagnosis was "malignant blastoma," but subsequent studies, by the same author, showed it to be a pheochromocytoma.

Addison's Disease. — Descriptions of clinical cases, exhibiting the syndrome of adrenal-cortical insufficiency, have been described in the earliest medical literature. It was not until 1855, however, that the English physician, *Thomas Addison*, called attention to the accompanying adrenal lesions, which he considered of pathogenic significance. *Addison's* admirably concise description of the disease, which now bears his name, deserves to be quoted :

"The leading and characteristic features of the morbid state, to which I would direct attention are, anemia, a general languor and debility, a remarkable feebleness of the heart's action, irritability of the stomach and a peculiar change of colour of the skin occurring in connection with a diseased condition of the suprarenal capsule."

His publication was of the greatest importance for the development of endocrinology as a whole, since it gave the first tangible proof of the important rôle played by the endocrines in internal medicine. Even before *Addison*, some morphologists theorized on the possible functional significance of the endocrine glands in general and the adrenals in particular, but his now classical monograph, gave one of the strongest motives for the work of the pioneers, who directed their attention to this field. *Sir William Osler* (1896) was the first to use (successfully?) an adrenal extract in the treatment of *Addison's* disease, but it was not until about 20 years ago that consistently active cortical preparations became available.

Corticoids. — The first adrenalectomies were performed by *Brown - Séguard* (1856), who concluded that these glands are essential for the main-

tenance of life. However, it appears very probable that the death of his animals was due to inappropriate surgical technic, since even unilateral adrenalectomy was fatal in his hands. As in his famous report on the activity of the testis extracts, the "father of endocrinology" succeeded in drawing a correct conclusion from faulty experiments.

That adrenaline is not the only hormone of the adrenals, and that the substance necessary for the maintenance of life actually originates in the cortex, has been demonstrated on elasmobranch fish, such as the torpedo, in which the cortex forms a separate organ. Here the cortex can be removed without injury to the medulla and this is followed by the appearance of typical deficiency symptoms and death (*Biedl*, 1910). It was later shown, in the United States, that adrenalectomized dogs and cats can be kept alive almost indefinitely, by the continued administration of adrenal-cortical extracts (*Rogoff* and *Stewart*, 1925; *Hartman* et al. 1928; *Swingle* and *Pfiffner*, 1931). Subsequent work led to the isolation from the cortex (mainly of cattle) of several crystalline compounds with high corticoid activity (*Grollman* and *Firor*, 1933; *Kendall* et al. 1935; *Wintersteiner* and *Pfiffner*, 1935). Several of the early crystalline fractions were probably mixtures, but they were sufficiently pure to be identified as steroids and hence as close relatives of the ovarian and testicular hormones.

In 1936 *Mason*, *Kendall* et al. in the U.S.A., isolated their compounds "A" and "B," which are identical with DEHYDROCORTICOSTERONE and CORTICOSTERONE respectively. Almost simultaneously, in Switzerland, *Reichstein* (1936) succeeded in isolating corticosterone from the adrenal cortex and the same author showed that several cortical steroids can be transformed into "ADRENOSTERONE," a testoid compound,

which also occurs naturally in the adrenal cortex. This transformation helped to prove the close relationships existing between the testoids and corticoids and to elucidate the structure of the latter.

Since that time, a large number of steroids have been isolated from the adrenal cortex. While some of these are apparently inactive precursors or metabolites of hormones, others exhibit corticoid, testoid, luteoid, folliculoid and other pharmacologic activities. (See : The Steroids.)

The first *in vitro* production of a corticoid was accomplished by the partial synthesis of DESOXYCORTICOSTERONE (*Steiger and Reichstein, 1937*), from the plant sterol, stigmasterol, after first degrading the latter to 3-hydroxy-*etio*-cholanic acid.

There still is considerable doubt concerning the true hormonal nature of desoxycorticosterone, although steroids with similar pharmacologic actions, are undoubtedly produced by the adrenal cortex.

It is of historic interest that the alleged isolation of this compound from the gland — and thus, the demonstration of its possible natural occurrence as a hormone — was, in any case, reported only after its synthesis (*Reichstein and von Euw, 1938*).

More recently, 11-DEHYDROCORTICOSTERONE (*Lardon and Reichstein, 1943*); CORTICOSTERONE (*v. Euw, Lardon and Reichstein, 1944*); 17-HYDROXY-11-DEHYDROCORTICOSTERONE (*Kendall's "compound E"*) (*Sarett, 1945*), have been prepared from desoxycholic acid. A good deal of work along these lines is now in progress in various laboratories, so that we have reason to believe that many additional naturally occurring cortical steroids will be available by synthesis and that the discouragingly small yields obtained will improve.

Hypercorticoidism. — The first proven case of adrenal-cortical tumor was described by *Tilesius* (1803). In 1811, *W. Cooke* described the case of a 7-year-old girl with bilateral adrenal tumors, adiposity and marked hirsutism, especially around the external genital organs.

During the subsequent century, several other cases were, more or less accurately, described but the subject received serious attention only after 1905, when *Bullock and Sequeira* published their famous report on 11 children, all under 15 years of age.

Bovin was probably the first, in 1909, to "cure" a female pseudohermaphrodite by removing an adrenal tumor.

It was not until quite recently, however, that chemists have shown us the existence of a variety of steroids in the adrenal cortex and pharmacologic assays revealed that many of these possess qualitatively distinct activities. This has raised the possibility that the various manifestations of the adrenogenital syndrome (pseudohermaphroditism, hypertension, diabetes, adiposity, etc.), are not necessarily due to the excess production of one adrenal-cortical hormone. The relative preponderance of one or the other symptom, in individual cases, thus becomes understandable.

The production of nephrosclerosis and hypertension, in the absence of any sexual anomaly, in experimental animals treated with desoxycorticosterone, led us to suppose that a selective increase in the endogenous mineralo-corticoid production of the cortex, may be the cause of corresponding syndromes in man. The concept of the "diseases of adaptation" was largely based upon this observation, since it is known that certain corticoids are produced in excess, during adaptation and defence to any type of damaging agent.

NORMAL MORPHOLOGY

ANATOMY

In man the adrenals are paired organs situated in the retroperitoneal space, close to the upper pole of each kidney and separated from the latter by a layer of fat tissue. The right adrenal has the shape of a pyramid and is situated directly above the upper pole of the kidney, while the left adrenal is crescent-shaped and lies more on the anterior and medial kidney surface. Both kidney and adrenal are surrounded by the same firm renal fascia and a common adipose capsule. The suprarenals are located at the height of the 11th or 12th thoracic to 1st lumbar vertebrae; the right adrenal is adherent to the liver and the inferior vena cava, the left is completely covered with peritoneum on its anterior surface and does not adhere to any organ.

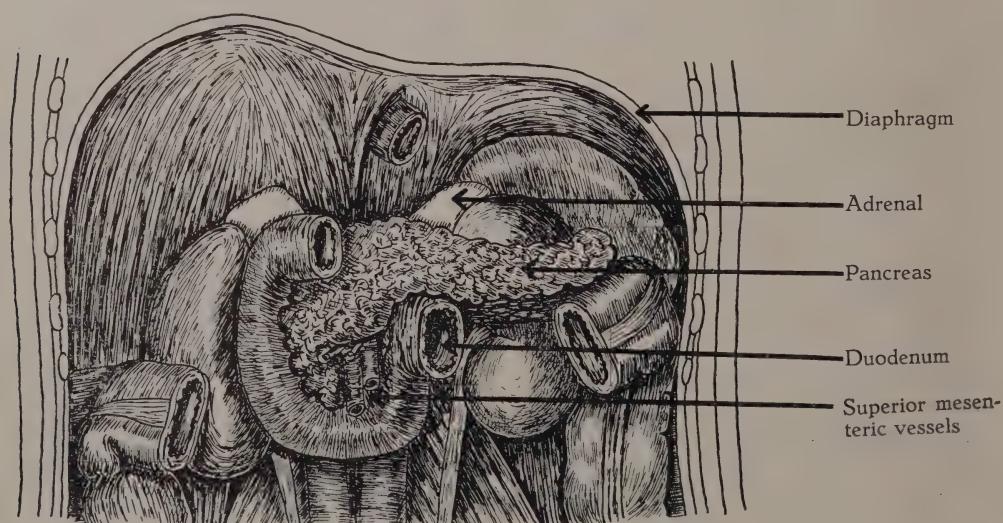
The surface of the adrenal is irregular, with many furrows. On the cut surface, the naked eye readily distinguishes the yellow outer CORTEX from the reddish-brown inner cortex and the grey MEDULLA. The latter is highly subject to post-mortem autolysis. Although

there is no clear-cut evidence of any direct functional interrelation between cortex and medulla, the former surrounds the latter in a capsule-like manner in all mammals. In some of the lower vertebrates, the two types of cells tend to be even more closely intermingled. (See : Comparative Morphology.)

The SIZE of the adrenal is subject to great individual variation, but averages about $45 \times 25 \times 6$ mm. in width, height and thickness, respectively; its total volume is about 5 cc. The weight of the two adrenals averages about 10 gm. in the normal individual. The higher "normal" weights, given in some textbooks, are derived from observations on patients who died from diseases, almost all of which cause adrenal enlargement, owing to the general-adaptation-syndrome elicited by them.

HISTOLOGY

The histologic structure of the ADRENAL MEDULLA is quite different from that of the cortex. Hence differentiation between the two is not difficult, although cell cords of the cortex may reach deep into the medulla and vice versa, thus

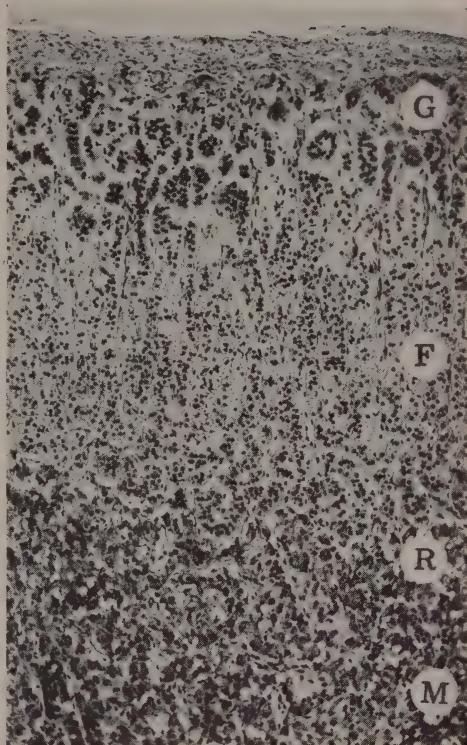


Position of adrenals and pancreas in relation to other abdominal organs.
(Redrawn after H. Gray: "Anatomy of the Human Body", Lea & Febiger, 1942).

making the border line between them rather indistinct. The medullary cell is polygonal; it measures about $18-30\mu$ and its vesicular nucleus about $6-8\mu$ in diameter. One of the most characteristic features of the medullary cells is their chromaffinity, which is a good index of their adrenaline content. The bichromate reaction is based upon the fact that the intracellular adrenaline granules give a typical color reaction, with dilute solutions of potassium bichromate. The reddish-brown tinge of the reaction product is a combination of the brown color of the reduced



Histologic appearance of the normal adrenal (monkey): — Under the connective tissue capsule are the acinus-like, dark cell-groups of the glomerulosa (G); these are followed by the almost straight, light (richest in lipids) cell-columns of the fasciculata (F) and then by the interlacing strands of the reticularis (R). Only a small part of the medulla (M) is visible underneath the cortex.



Histologic appearance of the normal adrenal (human): — Note distinct glomerulosa (G); and fasciculata (F); while reticularis (R) is less prominent. Medulla (M) shows signs of beginning autolysis.

chromate and the red of the adrenaline oxidation products. Since, in the test tube adrenaline gives a similar "chromaffin reaction" with potassium bichromate solutions, it is generally agreed that the hormone itself, or precursors closely related to it, are responsible for this histo-chemical reaction.

The ADRENAL CORTEX consists of three structurally different layers. The external zone, immediately under the connective tissue capsule, is called the *zona glomerulosa*. Its cells are small, poor in cytoplasm and more or less irregularly arranged. The nuclei are small and rich in chromatin. Inside this layer is the *zona fasciculata*, which consists of regular rows of large polygonal cells with vesicular nuclei. The cell

columns are radially arranged, and run parallel with each other, from the outer zone towards the medulla. Between them are radially coursing sinusoids, whose walls are studded with littoral, reticulo-endothelial cells. *The zona reticularis* is the innermost layer of the cortex, immediately adjacent to the medulla. It consists of very irregular strands, which form a network of small cells with dark nuclei. Many melanin and iron-pigment containing phagocytes are found in this zone. The development of the glomerulosa and reticularis are subject to great individual variations, but the fasciculata is always the widest zone.

Because of the numerous lipid granules, the cortical cells, especially in the fasciculata, have a vacuolized cytoplasm and are designated as "spongicytes." Around the central vein, the cortex may be invaginated into the medulla, thus forming the "central cortex" or "inverted cortex."

The REGENERATION and growth of the adrenal cortex has been claimed to occur mainly from the glomerulosa, whose cells gradually migrate through the fasciculata into the reticularis and eventually succumb in this latter zone. However, this view is not unanimously accepted. Regenerative phenomena in the medulla are probably diffuse and do not occur in any one especially predisposed zone.

Islets of LYMPHATIC TISSUE and round cell infiltration are rather common in human adrenals, especially under certain pathologic conditions. (See : Addison's Disease.)

Three adrenal ARTERIES, derived respectively from the aorta, inferior phrenic and renal arteries, supply each of the two adrenals. They form a rich subcapsular plexus, from which the blood flows through the cortical sinusoids into the medulla; here it gathers in a large central VEIN, leaving the organ at the hilum. The right adrenal vein dis-

charges its blood directly into the inferior vena cava, while the left empties into the renal vein. Hence, tumors of the left adrenal tend to invade this vein. The adrenal arteries enter the gland at various points of the surface, the vein emerges at the hilum. The adrenals are extraordinarily well supplied with blood, receiving about 6 to 7 cc. per gm. of tissue, per minute. All of this blood flows through the cortex before reaching the medulla, so that the latter is exclusively supplied by venous blood, maximally saturated with the metabolites of the cortical cells. The physiologic significance of this arrangement is not known. In addition to the large central veins, small venules leave the gland at various points of the capsular surface; some of these form anastomoses with the renal veins. This may be of physiologic importance, since through these veins blood, saturated with adrenal hormones, may directly enter the kidney. There are important muscular sphincters in the wall of the central vein. Their periodic contraction and relaxation helps to collect and release large amounts of hormone saturated blood, in accordance with the requirements of the organism.

The LYMPHATICS emerge from the gland, around the central vein, in the hilum.

The adrenal NERVES are derived from the great splanchnic, after having passed through the suprarenal plexus. They are preganglionic, medullated fibers which are not interrupted by cell stations along their course. Transection of the splanchnics, above the semilunar ganglion, causes degeneration of the adrenal nerves, which proves that these pass through the ganglion without interruption. Thus they appear to differ from other sympathetic pathways. Actually the nerve cells, and the embryologically related chromaffin cells of the medulla, correspond to the ganglion and the postganglionic fibers. Less im-

portant nerve tracts course to the adrenals from the semilunar ganglion and the renal plexus. All adrenal nerves reach the medulla and form intimate connections with its cells after merely traversing the cortex. The cortical cells themselves receive no important nerve endings. (See also p. 115.)

The vagus is not known to participate in the innervation of the adrenals.

A large number of **GANGLION CELLS** are seen on the surface and in the medulla of the human adrenal, especially in the hilum region. Most of these are multipolar, a few bi- or unipolar. They probably represent peripheral nerve centers.

COMPARATIVE MORPHOLOGY

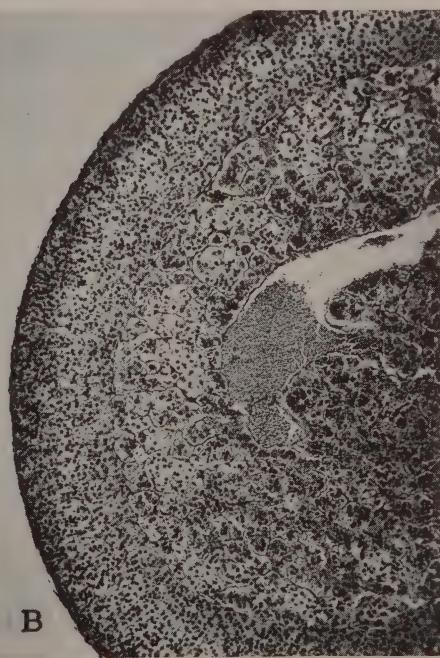
The presence of the adrenaline-containing, chromaffin cells has been demonstrated in **INVERTEBRATES** (most annelides, mollusca, cephalopodes) and in cyclostomata, where they are usually found in close connection with large

vessels and nerve tracts. Cortical cells, on the other hand, do not appear in animals lower than the vertebrates.

In **SELACHIANS**, unlike in most other animals, the adrenal cortex (or "interrenal body," as it is called in these fish) is anatomically distinct from the chromaffin accumulations. This is of special interest because it permits the separate removal of cortical tissue without injury to medullary cells.

In **AMPHIBIA** there is no distinct demarcation between cortical and medullary cells, the two being intimately intermixed. In some amphibia, adrenaline-like substances are produced in the mucous glands of the skin, but it remains to be seen whether there is any relationship between these glands and the adrenal system. In **REPTILES** and **BIRDS** the cortical and medullary cells are also irregularly intermingled, there being no separate cortex and medulla.

Among the **MAMMALS**, the adrenal medulla is always surrounded by a dis-



Adrenal with "X-zone". — A. Dark, cellular X-zone in reticularis region of the adrenal in a castrate male mouse. — **B.** Adrenal of a similar castrate male mouse 25 days after subcutaneous implantation of a 12 mg. pellet of methyl-testosterone. Note complete disappearance of "X-zone".

tinct cortex, but the size and shape of the gland is extremely variable. In the small laboratory rodents, the adrenals are roundish or oval and their surface is smooth, while in most of the larger mammals the cortical surface is corrugated, as it is in man. The *guinea pig* adrenal is noteworthy because of its extraordinarily large size. In certain strains of *mice* the reticularis is especially prominent and often designated as the "X-Zone." There is reason to believe that the cells of this zone are the source of adrenal testoids. It is interesting that the cortex is much larger in the wild, than in the laboratory *rat*. (Exposure to strain? Diet?)

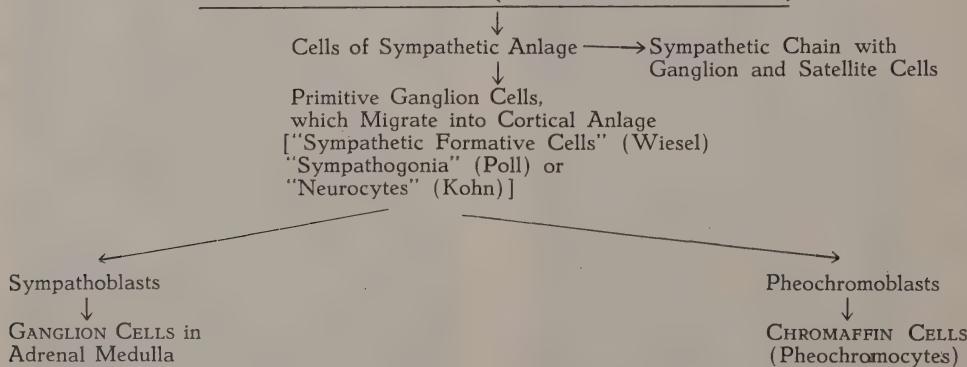
EMBRYOLOGY

The adrenal CORTEX is formed from a thickening of the celomic epithelium, in the region between the two renal (pronephros) primordia. It begins to be clearly distinguishable in embryos of 5.6-8.5 mm. crown-rump length. Lipid

granules are first observed in 2.5-5 cm. embryos. The fetal adrenals (in common with similar tissues, such as the corpus luteum) have a particular affinity for ponceau fuchsin. The postnatal persistence of fuchsinophilic granules in the reticularis is frequently associated with pseudohermaphroditic traits, as we shall see later. (See p. 163.)

The MEDULLA develops from the primordium of the sympathetic nervous system, which in turn arises from the neuroectoderm. The primitive sympathetic anlage is the common precursor of the sympathetic chains, the ganglion cells in the medulla and the chromaffin cells themselves. The terms "sympathogonia" (Poll), "sympathetic formative cells" (Wiesel) and "neurocytes" (Kohn) have all been used to designate intermediate cell types in the course of the development of the adrenal medulla. This development may be schematically illustrated as follows:

NEUROECTODERM (OF NEURAL CREST)



Any of the above cell types may give rise to tumor formation.

In man the invasion of the adrenal cortex, by the primitive ganglionic cells, occurs in embryos of 15-17 mm. crown-rump length. The invading cells continue to proliferate and to differentiate in their new location, even during early postnatal life.

The PARAGANGLIA are slightly chromaffin cell accumulations found, in contact with sympathetic ganglia, outside the adrenal medulla. They develop from the same type of primitive ganglionic cells which form the adrenal medulla. These extra-adrenal chromaffin cells represent an additional source of adrenaline, yet their ability to

compensate for the loss of adrenal chromaffin cells is very limited, as judged by the behaviour of adrenalectomized animals.

The SIZE of the adrenals, in proportion to the other organs of the body, reaches a certain maximum in 4-week-old human embryos. In these the adrenal is as large, or larger, than the kidney. In the new-born the adrenal is one-third, and in the adult $1/28$, as large as the kidney. A particularly pronounced and rapid involution, especially of the cortical portion, occurs during the first few days of postnatal life; perhaps because of the sudden withdrawal of maternal hormonal influences. (See p. 135.)

THEORIES CONCERNING THE HISTOPHYSIOLOGY OF THE ADRENALS

It is generally conceded that the CHROMAFFIN GRANULES in the medullary cells are actually adrenaline itself or its precursor. In any case, the amount of chromaffin material histologically demonstrable in the medullary cells, closely corresponds to the amount of adrenaline, as determined by analytic or bioassay methods. Histologic observations show that the adrenaline granules are discharged directly into the medullary sinusoids.

The LIPID GRANULES in the cortical cells are not the hormones of the cortex, but consist mainly of cholesterol and neutral fats. Since, however, the cortical hormones are highly fat-soluble (much more soluble in fats than in plasma) it is probable, that the visible lipid granules act as solvents in which the cortical hormones are stored.

It remains unexplained why, in all vertebrates (except certain fish), the primordia of the medulla travel a long distance in order to unite with those of the cortex. All the blood reaching the medulla, has to travel through the

cortical sinusoids first; this suggests that products of the cortical cells may have to undergo further transformation in the medulla, but such a possibility is entirely conjectural. Since both cortical and medullary hormones play an important rôle in the general-adaptation-syndrome, the demand for both types is especially great during exposure to non-specific damage. (See : General-Adaptation-Syndrome.) Perhaps the ANATOMIC UNION OF THE TWO GLANDS is advantageous because, during emergencies, relaxation of the sphincters in the adrenal veins suddenly supplies the general circulation with large amounts of both cortical and medullary hormones, stored in the gland's spacious sinuses.

The tendency of the adrenals to acquire a flattened corrugated shape, in the course of evolution, results in the development of a very large contact-surface between cortex and medulla. This may also be cited as suggesting the existence of close functional correlations between the two parts of the glands.

Stimulation of the adrenal NERVES elicits an immediate release of chromaffin granules into the blood stream, without causing any detectable secretion of cortical lipid granules. Conversely, denervation of the adrenals prevents the secretion of adrenaline, without interfering with cortical function. Hence, it may be concluded that the secretion of cortical hormones is not significantly influenced by specific secretory nerves, while that of adrenaline is almost entirely dependent upon nervous stimuli.

The high concentration of ASCORBIC ACID in the adrenals suggests that this vitamin may play an important rôle in the biogenesis of cortical hormones and adrenaline, but this has not been proven.

CHEMISTRY OF THE ADRENALS

CHEMICAL COMPOSITION OF THE GLAND

The chemistry and biogenesis of the adrenal hormones will be discussed in subsequent chapters. Here we shall merely consider the most important data concerning the general chemical composition of the gland.

The adrenals — like most other tissues — contain approximately 80% WATER.

The CARBOHYDRATE content of the adrenals is extremely low, although there are traces of glycogen and rather large amounts of LACTIC ACID (about 0.2% of the wet weight) in it.

The cortex is particularly rich in LIPIDS. Most of these form distinct cytoplasmic granules. Considerable quantities of fatty acids, cholesterol, cholesterol esters, phosphatides and lipochromes (e.g., carotene and xanthophyll), have been isolated from the adrenal cortex. These lipids and lipochromes are responsible for the light, yellowish color of the cortex.

About two-thirds of the dry material of adrenal tissue is PROTEIN, but so far its specific characteristics have not been adequately studied.

Among the INORGANIC CONSTITUENTS, a comparatively high concentration of iodine, bromine, iron and sulphur are noteworthy. The latter two probably are of importance in the formation of the pigment granules in the zona reticularis.

Various ENZYMES have been demonstrated in the adrenals. Lipases appear to play an important rôle in the discharge of lipid granules. A high concentration of proteases has been held responsible for the unusually rapid, postmortem autolysis characteristic of these glands, while oxidases, decarboxylases, etc., appear to play an important part in the biogenesis of adrenaline and its subsequent transformation

into inactive oxidation products. (See : Biogenesis of Adrenaline.)

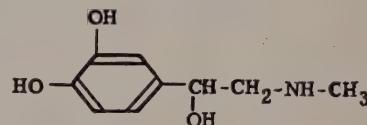
The high VITAMIN C content has already been mentioned in connection with histologically demonstrable cell inclusions.

The fact that the adrenals are rich in "PLASMALOGENS", CHOLINE and GLUTATHIONE has provoked much speculation concerning the possible rôle of these compounds in adrenal physiology, but this question still awaits clarification.

CHEMISTRY OF THE ADRENAL HORMONES

Chemistry of Adrenaline. — The only hormone known to be produced by the adrenal medulla is adrenaline.

It exists in two optically isomeric forms, but only l-adrenaline occurs in nature. Its structure is :



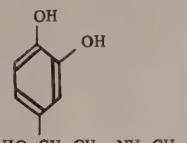
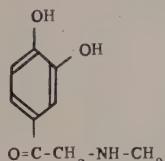
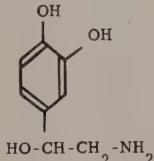
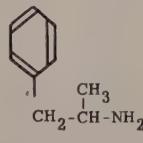
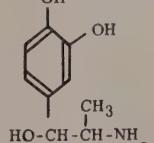
Adrenaline is very sensitive to the action of oxidizing agents. (See : Fate of Adrenaline in Body.) It is usually administered in the form of the highly water-soluble hydrochloride.

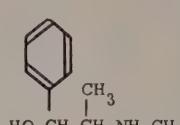
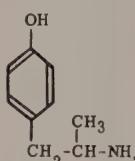
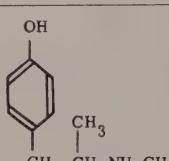
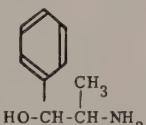
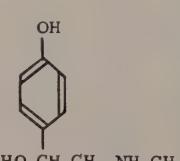
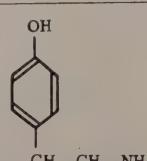
Chemistry of Adrenaline Derivatives (with their chief biologic characteristics). — It is not within the scope of this book to discuss the chemistry of the, very numerous and pharmacologically important, derivatives of adrenaline. It should be emphasized, however, that some derivatives of the hormone are valuable sympathomimetic drugs, that is, they stimulate structures innervated by adrenergic nerves. Many of these compounds possess definite advantages over adrenaline. Some exhibit one or the other desired action in a more specific manner than the hormone itself (that is, with less of the undesired side effects), while others have a compara-

tively greater oral activity, a more prolonged action or a lesser toxicity. Many of the pharmacologically important, adrenaline derivatives occur naturally in plants (ephedrine), as animal poisons (toad venoms) or at sympathetic nerve endings (sympathin), while others are artificial compounds obtainable only by synthesis. — Little is known about the mechanism of their action. Some (e.g.,

ephedrine) are claimed to act merely because they are destroyed by the same amineoxydase which inactivates adrenaline. Thus they protect the hormone from rapid destruction, since they successfully compete with it for this enzyme.

The most important adrenaline derivatives, with some of their outstanding characteristics, are listed below:

Name of Compound	Formula	Characteristics
ADRENALINE 1-hydroxy-1-(3',4'-dihydroxyphenyl)-2-methylaminoethane <i>Syn</i> : Adrenalin, Epinephrin, Suprarenine	 HO-CH-CH ₂ -NH-CH ₃	Naturally occurring hormone of the adrenal medulla. Its characteristics are described in the text, the compound being mentioned here merely for purposes of comparison with its derivatives. Presumably identical with "sympathin I", which has also been termed "Sympathin A". (Cf. p. 114.)
ADRENALONE 1-keto-1-(3',4'-dihydroxyphenyl)-2-methylaminoethane <i>Syn</i> : Kephrine, Stryphon	 O=C-CH ₂ -NH-CH ₃	The ketone corresponding to adrenaline. It has marked sympathomimetic effects, although its pressor action is comparatively mild. Used mainly for its local action as a hemostatic, because of its prolonged action and low systemic toxicity.
ADRENOXINE		An oxidation product of adrenaline obtained by treatment with tyrosinase. It exerts acetylcholine-like, negative chrono- and inotropic effects and decreases the blood pressure. It has been advocated as an anti-hypertensive drug. (Cf. p. 109.)
ARTERENOL 1-hydroxy-1-(3',4'-dihydroxyphenyl)-2-aminoethane <i>Syn</i> : Nor-adrenaline	 HO-CH-CH ₂ -NH ₂	A demethylated adrenaline. It was claimed to be identical with "Sympathin B", because it elicits mainly excitatory effects, which cannot be inhibited by ergotamine. Its pressor action is even greater than that of the natural hormone. It has also been designated as "Sympathin N". (Cf. p. 114.)
BENZEDRINE 1-phenyl-2-aminopropane <i>Syn</i> : Mecodrin, Simpanin, Amphetamine	 CH ₃ CH ₂ -CH-NH ₂	Pharmacologically similar to ephedrine, differing from the latter mainly by its greater ability to stimulate mental processes. Used as a vasoconstrictor for inhalation and local application in rhinology, to stimulate nervous centers in narcolepsy, postencephalitic parkinsonism and mental fatigue (instead of caffeine) also in various allergic conditions. May lead to addiction!
COBEFRIN 1-hydroxy-1-(3',4'-dihydroxyphenyl)-2-aminopropane <i>Syn</i> : Cobrasil	 CH ₃ HO-CH-CH-NH ₂	Has no material advantages of adrenaline, but is sometimes used as a substitute for the latter, especially in combination with cocaine derivatives, for infiltration anesthesia.

Name of Compound	Formula	Characteristics
EPHEDRINE 1-hydroxy-1-phenyl-2-methylamino-propane		Pharmacologically similar to adrenaline, but 1,000 times less active. Has advantage over hormone that it is orally active and exerts more prolonged effect. Unlike for the hormone, organs are not sensitized to it either by cocaine or by denervation, perhaps because it attacks the muscle directly. It is used as an analeptic, mydriatic with minimal effect on ocular tension, respiratory stimulant and in allergic conditions such as hay fever and asthma. <i>Pseudo-ephedrine</i> differs from ephedrine only in the steric position of the alcoholic hydroxyl. It has no notable pharmacologic advantages.
PAREDRENE 1-(4'-hydroxyphenyl)-2-amino-propane <i>Syn</i> : Neo-ephedrine		Differs from tyramine only in having an additional methyl group. Pharmacologically resembles adrenaline, but is practically devoid of actions on central nervous system. Used mainly as a mydriatic and for production of temporary cycloplegia.
PAREDRINOL 1-(4'-hydroxyphenyl)-2-methyl-aminopropane <i>Syn</i> : Veritol		Differs from paredrine only in possessing an additional methyl group on amino nitrogen. Orally active pressor agent used in various circulatory disturbances.
PROPADRINE 1-hydroxy-1-phenyl-2-aminopropane <i>Syn</i> : Nor-ephedrine Mydratine		Differs from ephedrine only in that it lacks methyl group on amino nitrogen. Pharmacologically similar to ephedrine.
SYMPATOL 1-hydroxy-1-(4'-hydroxyphenyl)-2-methylaminoethane		Differs from adrenaline only in the absence of the meta-phenolic hydroxyl group. 50 to 100 times less active than the hormone. Its pressor effect is more prolonged than that of adrenaline and unlike the latter, it is claimed not to increase the pressure in the auricles of the heart and is less likely to produce fibrillation. <i>SYNEPHRINE</i> is racemic sympatol which essentially shares the actions of the latter. <i>NEO-SYNEPHRINE</i> differs from synephrine only in that the phenolic hydroxyl is on C ₈ . It is used for local application to mucous membranes, in allergic conditions, for sustaining the blood pressure during spinal anesthesia. It can be given parenterally or orally and is less likely to cause cardiac arrhythmias than adrenaline.
TYRAMINE 1-(4'-hydroxyphenyl)-2-amino-ethane		Formed by action of bacteria upon protein as a result of tyrosine decarboxylation. Shares some of the effects of adrenaline (e.g., pressor action, stimulation of uterine muscle), but has no clinical applications as a sympathomimetic compound.

Chemistry of Cortical Hormones.
— All the cortical hormones, known up-to-date, are steroids. Their fundamen-

tal chemical characteristics have been described in the section on the steroids.

GENERAL PHARMACOLOGY OF THE ADRENAL HORMONES

STANDARDIZATION

Analytic Methods for the Detection of Adrenaline. — The direct gravimetric determination of adrenaline can rarely be employed because of the minute quantities present in body fluids and tissues. Hence, usually, colorimetric methods are used. Some of these are based on the property of adrenaline to form colored products when oxidized with various agents; others depend upon the formation of a colored reduction product from reagents exposed to the hormone; yet others on special reactions, due to the pyrocatechol ring in the adrenaline molecule. The most commonly used adrenaline reactions are the following :

(1) The FERRIC CHLORIDE REACTION (*Vulpian*, 1856). Neutral, or slightly acid, solutions of adrenaline give an intense, emerald-green reaction with ferric chloride, due to the phenolic hydroxyl of the adrenaline molecule. Pyrocatechol derivatives give the same reaction and hence interfere with its specificity. (Sensitivity about 1:10,000,000 under optimal conditions.)

(2) The IODINE REACTION (*Vulpian*, 1856). Free iodine causes a pink discoloration of adrenaline solutions due to partial oxidation of the hormone. (Sensitivity 1:2,000,000.) It is best to perform the iodine reaction in acid solutions and to read the coloration with Pulfrich's photometer. Adrenaline may then be detected by its green absorption band at $500\mu\mu$ (*v. Euler*, 1933).

(3) The IODATE REACTION (*Krauss*, 1909) is also based on the oxidation of adrenaline by free iodine, but here the halogen is freed from the iodate by the hormone. The sensitivity of the reaction may be increased, by the addition of sulphanilic acid, so that dilutions of 1:5,000,000 are still detectable.

(4) The SUBLIMATE REACTION (*Comessatti*, 1909) is based on the oxida-

tion of adrenaline by sublimate. (Sensitivity about 1:1,000,000.)

(5) The SUBLIMATE-IODATE-SULPHANILIC ACID REACTION (*Ruffmann*, 1922; *Bacq*, 1932; *Viale*, 1933) depends upon the successive sensitization of the iodate reaction by sulphanilic acid and of this reaction by sublimate (chain sensitization). (Sensitivity 1:400,000,000, under optimal conditions.)

(6) The PERSULPHATE REACTION (*Ewings*, 1910) has the advantage that it can be performed even in colored organ extracts since the persulphate decolorizes them. (Sensitivity about 1:500,000,000.)

(7) OTHER REACTIONS, BASED ON THE OXIDATION OF ADRENALINE, use gold chloride, manganese superoxide, potassium ferricyanide, bromine, chlorine, calcium chlorate, ammoniacal silver reagent, potassium bichromate, ammonium molybdate, sodium tungstate or diazo-benzenesulphonic acid, etc., all of which yield colored products on contact with adrenaline. Unfortunately, none of these color reactions have proven to be very specific or accurate, especially in the presence of contaminating substances.

(8) The SPECTROGRAPHIC DETERMINATION of adrenaline (*Handovsky and Reuss*, 1928) is based on the fact that the hormone shows an absorption band in the ultraviolet, with a maximum at $280\mu\mu$. The method gives encouraging results. It has also been used for the determination of sympathin, which exhibits an identical absorption spectrum.

(9) The green FLUORESCENCE of adrenaline solutions during ultraviolet irradiation, in the presence of alkali and oxygen (*Gaddum and Schild*, 1933; *Loewi*, 1936), has a sensitivity of 1:10⁸. This method can also be used for sympathin and gives results which check well with bioassays.

Among the analytic methods, the spectrographic technics tend to give the

more satisfactory results, but for most purposes bioassay methods are preferable, because they are more specific and in general also more sensitive. The substances likely to interfere with the colorimetric determination of adrenaline are, the less active optical isomer of the hormone, ascorbic acid and some other reducing substances.

Analytic Methods for the Detection of Corticoids. — While there are no satisfactory methods for the chemical determination of corticoids, rather encouraging results have been obtained with technics based upon the glucose-like reducing properties of their glycol side-chain (*Talbot, 1945; Sobel, 1945*).

Lowenstein et al. (1946), described a method for the determination of corticoids in the urine, based upon periodate oxidation of the primary alcohol group at C₁₇². This yields one mol of formaldehyde per mol of oxidized corticoid and the formaldehyde is determined. (The method is not yet practical.)

Bioassay of Adrenaline. — Among the many bioassay technics used for the estimation of adrenaline in its solutions, the following deserve special attention :

(1) **INTESTINAL SEGMENT OF RABBIT** (*Langley, 1901; Cannon and La Paz, 1911*). The peristaltic movements of a loop of rabbit intestine (suspended in oxygenated Ringer solution) are inhibited by adrenaline. By comparing the potency of unknown solutions, with those containing adrenaline in known concentrations, the hormone content of various fluids can be estimated with a fair degree of accuracy.

(2) **SEGMENT OF NON-PREGNANT RABBIT UTERUS** (*Frankel and Allers, 1909*). Adrenaline stimulates the contractions of such a preparation *in vitro*.

(3) **ISOLATED ARTERIAL RINGS** (*Friedmann, 1904; Rothlin, 1920*). A ring of a small artery (from cattle, pig or sheep) is suspended in Locke's solution and attached to a lever, which

registers its contractions on a kymograph. Adrenaline causes contractions of the arterial rings, when added to the suspension fluid.

(4) **THE DENERVATED EYE.** Removal of the sympathetic superior cervical ganglion greatly increases the adrenaline sensitivity of the pupil in the rabbit (dilatation) and of the nictitating membrane in the cat (contraction) (*Meltzer and Auer, 1904; Rosenblueth and Cannon, 1932*). The adrenaline concentration of fluids is tested by instilling them into the conjunctival sack of an animal whose pupil is thus denervated. The test may also be performed on the enucleated eye of the frog, which reacts to the hormone with mydriasis (*Lewandowsky, 1898*).

(5) **THE DENERVATED HEART** (*Cannon, 1922*). Following transection of the vagi, combined with removal of the stellate and second thoracic ganglia of the sympathetic chain, the heart is completely deprived of nervous control. Conditions which cause adrenaline liberation from the animal's own adrenal medulla, increase the pulse rate of the denervated heart. Detectable tachycardia is produced by as little as one part of adrenaline in fourteen hundred million parts of blood.

(6) **THE CAVAL POCKET** (*Stewart and Rogoff, 1916*). The inferior vena cava, of the dog or cat, is clamped below the entrance of the adrenal veins and just below the diaphragm. All veins entering this caval pocket are ligated, except those of the adrenals. The iris of the experimental animal is denervated as in test 4. After a certain period the upper clamp is removed. Dilatation of the pupil occurs if adrenaline has been secreted during the interval whilst the upper clamp was closed. If quantitative determinations are required, the blood collected in the caval pocket may be aspirated through a cannula and directly tested, *in vitro*, upon another

test object (such as the intestinal segment).

It will be noted that the denervated iris and the denervated heart are suitable for "internal bioassay technics," that is, the measurement of endogenous adrenaline.

(7) THE BLOOD PRESSURE OF THE CAT (*Elliot, 1905; Rosenblueth, 1932*)

is perhaps the most suitable indicator for routine use, since the rise in blood pressure is almost strictly proportional to the dose of adrenaline injected. Usually male cats weighing 3 to 4 Kg. are anesthetized with chloroform or ether, than tracheotomized and artificially ventilated. Following this the brain is removed, the spinal cord destroyed to the level of the 4th thoracic segment, and both vagi are cut. This operation causes a sudden decrease in the blood pressure to a level of 40-50 mm. of Hg, but it then remains quite constant, at this level. By comparing the effect of unknown and standardized solutions one can determine the adrenaline concentration, changing the dose of the unknown until the rise in blood pressure equals that caused by the standard of known concentration. With an arithmetic increase of the amount of adrenaline administered, the pressor effect increases logarithmically. This may be expressed by the formula

$$Kx = \frac{y}{A-y}$$

In this formula x = concentration and y = action as % of the maximal action A . $0.07y$ per Kg. suffices to produce a just perceptible rise in blood pressure, while $90y$ per Kg. causes a maximal increase.

Many other actions of adrenaline may be used as indicators in bioassays. Furthermore, even the tests described here may be varied, by using animal species other than those recommended above. For instance, the vasoconstrictor action may also be tested on the perfused hind-leg preparation of the frog (*Lawen, 1904; Trendelenburg, 1910*):

the improving effect of adrenaline upon the heart action may be studied on the perfused frog heart, rendered hypodynamic by aconitine, etc. A detailed description of all these bioassays would exceed the scope of this book, especially since most of them are too laborious to be useful in the clinical diagnosis of hyperadrenalinism.

Bioassay of Corticoids. — Bioassay of the cortical hormones gives far more satisfactory results than the chemical methods. Folliculoid, luteoid and testoid compounds of the adrenal cortex are assayed in the same manner as the corresponding gonadal hormones. (See: Ovary and Testis.) The corticoids themselves do not represent a pharmacologically uniform group and different bioassay technics have been worked out to estimate the different types of corticoid activity.

A. LIFE MAINTAINING ACTIVITY :

(1) IN THE DOG (*Swingle and Pfiffer, 1932*). This test is based upon the ability of corticoids to maintain adrenalectomized animals in good condition. The "dog unit" is defined as the minimum daily dose per Kg. of body weight, which, when given over a period of 7 days, maintains health and a normal blood urea level.

(2) IN THE RAT (*Kutz, 1931; Grollman and Firor, 1933*). This test is based upon the ability of corticoids to maintain immature, adrenalectomized animals alive and to permit their growth. Usually immature males of 40 to 50 gm. are adrenalectomized and given two daily subcutaneous injections of the unknown preparation, in oil solution. The unit is defined as the minimum amount necessary to maintain the health and growth, of such rats, during a period of at least 14 days. There are several variants of this test since some workers prefer slightly larger or smaller animals, while others recommend the use of castrates, because of the occa-

sional occurrence of accessory adrenal cortices in the gonads. The unit may also be defined in different ways, but now that pure crystalline corticoids are available, it is best to express the potency of the unknown preparation in comparison with that of a known standard substance, such as desoxy-corticosterone acetate, corticosterone or dehydrocorticosterone.

CARTLAND AND KUIZENGA (1936) define their rat unit as "the minimum daily dose of a substance which, administered by daily, single, subcutaneous injections, for 20 days, to four-week-old male rats (50 to 60 gm.), is enough to protect at least 80% of the rats and produce an average growth of at least 20 gm. per rat, per 20 days."

THE COLD TEST (*Selye and Schenker, 1938*), as many of its modifications, is based upon the fact that adrenalectomized rats are extremely sensitive to various types of non-specific damage, unless they are protected by life-maintaining corticoids. Instead of cold, which is used as a non-specific damage in this particular technic, other damaging agents (histamine, potassium, bacterial toxins, vaccines), may also be employed, but in our experience, cold gave the most uniform results. — Male or female adrenalectomized rats (45 gm. body weight) are placed in a refrigerator (temperature -5° to $+2^{\circ}$ C) 24 hours after adrenalectomy and henceforth given no food or water. They receive subcutaneous injections of the solution to be tested, at 0, 3, 4, 6 hours, during exposure to cold. The unit is defined as "the minimum amount which suffices to maintain alive 6 of 9 adrenalectomized rats, when 6 of 9 untreated, adrenalectomized controls are dead." Instead of the four injections, a single subcutaneous dose, at the beginning of exposure to cold, or even oral administration of the compound (by stomach tube) at that time, are recommended; the former for slowly

acting substances, the latter for solutions containing contaminants, which would not be tolerated parenterally. For greater reliability, larger groups of animals should be used, to compensate for individual variability in resistance to cold. The advantage of the technic is its great simplicity and sensitivity. It detects very active corticoids (e.g., corticosterone) in doses of 12-15 γ per animal.

WATER INTOXICATION TEST (*EverSOLE et al. 1940*). Male rats are fasted for a period of 12 hours, beginning 17 hours after adrenalectomy, then given distilled water in an amount corresponding to 6% of their body weight. This is administered by gavage, in five, hourly portions. Urine volumes are measured at intervals of 24 hours. The technic of hormone administration is variable but in general, it is best to express the results in comparison with a known standard. The criteria are, survival during this time and excretion of more than 90% of the administered water within 11 hours.

B. TESTS FOR GLUCO-CORTICOID ACTIVITY :

(1) DIABETOGENIC TEST ON INTACT RATS (*Ingle, 1941*). Rats are force-fed with high carbohydrate diets and injected with the substance to be tested. The criteria are the blood and urine glucose levels, urinary nitrogen excretion and tissue glycogen levels. Gluco-corticoid compounds cause glycogen deposition, hyperglycemia and glycosuria.

(2) DIABETOGENIC TESTS ON PARTIALLY PANCREATECTOMIZED RATS (*Ingle, 1941*). Daily injection of gluco-corticoids causes diabetes, even if the pancreatic remnant would otherwise suffice to maintain a normal metabolism.

(3) LIVER GLYCOGEN DEPOSITION TEST (*Reinecke and Kendall, 1942; Dorfman et al. 1946; Venning et al. 1946; Dobriner et al. 1946*). In the

various modifications of this test, adrenalectomized mice or rats are given corticoid compounds, during a period of fasting; the amount of liver glycogen deposited, or the maintenance of the initial glycogen levels act as indicators of gluco-corticoid activity. A high carbohydrate, low potassium diet and 0.9% NaCl as drinking fluid are given to the animals, during 4 days after adrenalectomy, prior to the test. The results are expressed in comparison with the potency of known extracts in order to obtain comparable results. Some of the modifications of this technique are so sensitive that they detect active gluco-corticoids in doses of about 10 γ per animal.

(4) ANTI-INSULIN TEST (*Jensen and Grattan, 1940*). Male mice (18-22 gm.) are divided into groups of 20 animals each. At the onset of a six hour fast they receive subcutaneous injections of the unknown preparation in 0.2-0.5 cc. of oil. At the end of the fasting period, each animal receives 1.5 or 2 units of insulin per Kg. of body weight and is maintained at a temperature of 34° C. The percentage of animals protected from insulin convulsions acts as a criterion of gluco-corticoid activity.

C. MUSCULAR ACTIVITY TESTS :

(1) SWIMMING TEST (*Gaarenstroom, Waterman and Laqueur, 1937*). Two days after adrenalectomy, rats (60 gm.) are given a single injection of the compound to be tested. The "swimming time" (period during which they can swim when placed in water) is recorded before the injection and on the following two days. The response is "positive" when the swimming time is doubled, "half positive" when it remains the same on the fourth as on the second day of treatment, and "negative" when it decreases.

(2) EVERSE AND DE FREMERY TEST (1932). Adrenalectomized rats are tied down under ether anesthesia, so

that only the left hind leg can move at the ankle joint. Short electrical stimuli are applied to this limb and the tetanic contractions of the gastrocnemius muscle are recorded. Improvement in the recovery of the fatigued muscle (due to four, daily injections given prior to the test), is used as a criterion and the unit is defined as the minimum daily effective dose.

(3) INGLE'S MUSCULAR ACTIVITY TESTS (1936-1942). Rats are subjected to work one hour after adrenalectomy. The gastrocnemius muscle is exposed under light phenobarbital anesthesia, weighted by 10 gm., and stimulated by repeated faradic stimuli for 24 hours or until fatigue or death ensues. There are several variants of this test but usually the unit is defined as the work equivalent of two 0.2 mg. doses of Kendall's cpd. "E".

D. TESTS FOR MINERALO-CORTICOID ACTIVITY :

It is known that corticoids decrease the blood potassium and cause sodium and chloride retention in adrenalectomized and, to a lesser extent, even in intact animals. Furthermore, prolonged administration of mineralo-corticoids causes nephrosclerosis in various animals; intact chicks, during the first few days of their lives being particularly sensitive to this activity. None of these reactions have been fully developed as indices for the accurate bioassay of mineralo-corticoid activity, but presumably they could be used for such purposes.

The increased resistance of adrenalectomized animals to potassium or even the above mentioned "water intoxication test" probably also depend, at least partly, upon mineralo-corticoid potency.

PHARMACOLOGY OF ADRENALINE DERIVATIVES

The most important pharmacologic properties of the principle adrenaline

derivatives have been mentioned on pp. 96-98.

MODE OF ADMINISTRATION

Adrenaline. — The official U.S.P. DESIGNATION for adrenaline is "epinephrine" while the B.P. designation is "adrenaline." Unfortunately, certain terms such as "adrenaline" and "su-prarenin," are commonly in use for the designation of the hormone, although they have been trade-marked by commercial companies.

The free hormone is rather insoluble in most solvents, hence the commonly employed PREPARATIONS contain it as water-soluble salts, especially the hydrochloride. Adrenaline is sensitive to the action of oxidizing agents, particularly in alkaline solution. Light enhances the oxidation of the hormone to pink, violet and eventually brown, inactive oxidation products; to prevent this it is preserved in dark, air-tight, bottles.

The dextrorotatory isomer is only about 1/18 as active as the levorotatory (natural) hormone. Hence, only the latter is employed for clinical purposes. The official U.S.P. "solution of epinephrine hydrochloride" or the B.P. "solution of adrenaline hydrochloride" are 1:1,000 sterile solutions of the compound in distilled water. They are nearly colorless and keep well if a preservative is added (for instance, 0.5% chlorbutanol or sodium bisulfite). The solution will stand a short period of boiling if necessary.

Usually it is administered INTRAMUSCULARLY or SUBCUTANEOUSLY as the standard aqueous (1:1,000) solution, in doses of 0.2-0.5 cc. Unfortunately, the resulting local ischemia makes the tissues ideal media for anaerobic, spore-bearing, microorganisms. Following subcutaneous injection of contaminated adrenaline solutions "epidemics" of tetanus have occurred in hospitals. Spores may also

be introduced, inadvertently, during injection. Such complications are less common after intramuscular than following subcutaneous injections, hence the former are preferred.

Adrenaline in oil (0.2%) solution can be given in doses of 0.75-1.5 cc. subcutaneously or intramuscularly, because of the slower absorption rate.

For delayed action, the comparatively insoluble, free base is distributed in the form of a suspension in oil (2.0 mg. per cc.) for intramuscular or subcutaneous injection. This is sometimes referred to as "slow epinephrine."

It is especially useful for instance, in inoperable hyperinsulinism and asthma.

Adrenaline is extraordinarily inactive BY MOUTH, presumably because most of the hormone is inactivated in the gastrointestinal tract, or during its passage through the liver.

INTRAVENOUS or INTRACARDIAC injection of the drug is dangerous and recommended only in cases of extreme emergency (e.g., for resuscitation). It is rarely necessary to give more than 0.25 mg. and even this should be injected very slowly.

A 1.0% solution of adrenaline hydrochloride in 0.9% sodium chloride is used for INHALATION, especially in asthmatic patients during an attack. For this purpose, it may also be given in a 1:1,000 glycerin solution. All-glass nebulizers should be used to avoid destruction of the hormone due to contact with metal.

Ointments (1:1,000 in petrolatum) and suppositories (1:1,000 in cacao butter) are used for the direct application of adrenaline to the SKIN or rectal MUCOUS MEMBRANE. The standard 1:1,000 aqueous solution is employed on other mucous membranes or wound surfaces, where the vasoconstrictor action of the hormone is desirable, to stop hemorrhage. This vasoconstrictor effect is particularly useful in com-

battling allergic rhinitis, sinusitis or simple, acute coryza. For intranasal application, oily solutions are preferable because of their delayed effects. Certain adrenaline derivatives (ephedrine, benzedrine) are often preferred for topical application, in respiratory and nasal disturbances, because (unlike adrenaline) their vasoconstrictor effect is not followed by a second phase of congestion and because their action is more prolonged. Yet it is claimed that adrenaline has the advantage of causing less local irritation.

By virtue of its vasoconstrictor action, adrenaline decreases the absorption of LOCAL ANESTHETICS and thus prolongs their effect, diminishes their systemic toxicity and decreases the likelihood of hemorrhage due to the surgical interventions, for which the local anesthetics are given. For this purpose the hormone is added to solutions of such compounds (e.g., procaine and its derivatives, which unlike cocaine, lack vasoconstrictor potency), in concentrations of 1:100,000 to 1:20,000.

Corticoids. — At the present time, only DESOXYCORTICOSTERONE ACETATE is commercially available in pure form for clinical use. It is distributed in oil solution, in sterile ampules for subcutaneous or intramuscular injection. It can also be administered in the form of sublingual drops of a propylene glycol solution, since it is effectively absorbed from the oral mucosa. This method of administration, as well as the subcutaneous implantation of compressed crystal-pellets (which are very slowly absorbed), save the patient the inconvenience of innumerable injections. The absorption rate is somewhat unpredictable, however, especially in the event of sublingual administration.

AQUEOUS EXTRACTS, or partially purified mixtures of the various cortical steroids, are prepared from cattle or pig adrenals and distributed in sterile

ampules for subcutaneous or intramuscular injections. These preparations have the advantage of possessing marked gluco-corticoid activity and are used if an especially rapid action is desired (e.g., addisonian crisis). Solutions of cortical extract concentrates in oil (for subcutaneous or intramuscular injection), are used when prolonged action is desirable. All these preparations are standardized in biologic units or in mg. equivalents of pure corticoids.

Additional pure corticoid steroids will probably soon be commercially available, especially DEHYDROCORTICOSTERONE, which can now be made by partial synthesis and has the advantage of gluco-corticoid potency as judged by animal experiments. It must be admitted however, that clinically, dehydrocorticosterone proved rather inactive. Perhaps higher doses or other gluco-corticoids will prove more satisfactory.

Corticoids are much less potent by mouth than parenterally, yet they can perhaps be given orally if sufficiently large quantities are made available. A CHARCOAL ADSORBATE of corticoids, prepared from impure extracts, has been advocated for oral use, since the intestinal juices elute the activity from the charcoal and make it available to the patient.

Intravenous administration of cortical extracts is necessary only in emergencies such as addisonian crises or shock.

ACTIVATION AND INACTIVATION

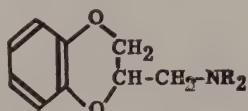
A number of drugs may influence the action of adrenaline by increasing or decreasing some, or all, of its activities.

Drugs which antagonize the actions of adrenaline and of sympathetic stimulation are generally referred to as "adrenolytic" and "sympatholytic" substances, respectively (*Bacq and Frede-ricq*).

ERGOTOXINE or ERGOTAMINE do not affect the inhibitory actions of the hormone (e.g., intestinal relaxation, vaso-dilatation), but inhibit the excitatory effects (e.g., motor and secretory stimulation). In this respect, the ergot alkaloids influence the actions of adrenaline, and those of sympathetic stimulation, in the same manner, that is, they are both adrenolytic and sympatholytic.

As a result of this selective inhibition of excitatory responses, some of the actions of the hormone appear to be inverted following pretreatment with ergot. This phenomenon is usually referred to as "ADRENALINE REVERSAL." Thus, the hormone causes a drop in blood pressure and expansion of amphibian melanophores following such pretreatment, while normally, the pressor and melanophore-contracting actions prevail. The hyperglycemic effect of the hormone is likewise abolished by ergotoxine.

It has also been possible to prepare a series of interesting sympatholytic DIOXANE DERIVATIVES of the general formula :



Among these, the most important are "F933" (2-piperidinomethyl-1,4-benzodioxane), which antagonizes the augmentor responses to adrenaline, but not to sympathetic stimulation; "F883" (2-diethylaminomethyl-1,4-benzodioxane), which counteracts the augmentor responses both to adrenaline and to sympathetic stimulation and "F1081" (2-methoxy-5-iodo-phenoxyethylidethylamine) which annuls the effect of inhibitory sympathetic stimulation.

In honor of *E. Fourneau*, whose work led to the synthesis of these compounds, they are generally designated by the letter "F" and the serial number which this author assigned to them.

It is of special interest that certain ADRENALINE DERIVATIVES (e.g., adrenoxine), inhibit or reverse the actions of subsequently administered adrenaline, in those organs which are normally excited by the hormone. Pretreatment with other adrenaline derivatives may, on the other hand, actually increase sensitivity to the hormone.

It is not within the scope of this book to discuss all the substances which may antagonize adrenaline actions; it is noteworthy, however, that many other compounds exhibit adrenolytic properties, for instance yohimbine, quinine and its derivatives, cotarnine, hydrastinine, emetine, corynanthine, chelidonine, lycorine, apocodeine and hordenine. Indeed, even a phenomenon of so-called "DOUBLE REVERSAL" has been observed following successive administration of ergotamine and corynanthine (both of which cause adrenaline reversal). That is to say, adrenaline increases the blood pressure in a dog pretreated with both of these drugs, just as it does in the non-pretreated animal, although separately, each of the drugs causes reversal of the pressor response to the hormone. The therapeutic possibilities of these combinations have not been adequately explored.

Other drugs antagonize adrenaline actions, merely because their own pharmacologic properties happen to be opposite to those of the hormone. Among these, we might mention HISTAMINE and ACETYLCHOLINE. Here, we do not speak of true adrenolytic properties but merely of drug antagonism.

Conversely, some compounds related to adrenaline (e.g., ephedrine, thyroid hormone), may INCREASE THE RESPONSIVENESS OF CERTAIN TARGET ORGANS TO SUBSEQUENT ADRENALINE STIMULATION. Cocaine and its derivatives enhance especially the vasoconstrictor, cardiac and pupillary reactions to adrenaline. Sympathetic denervation

of certain receptors (pupil, heart), causes a somewhat similar sensitization perhaps because, in the absence of constant nervous stimulation, the sympathomimetic substances tend to accumulate in the denervated or cocainized structures. It will be recalled that denervation also raises the sensitivity to acetylcholine, allegedly because its normal, continuous destruction by choline-esterase is deranged ("Law of denervation").

SENSITIZATION AND DESENSITIZATION

There is no convincing evidence to show that, upon repeated administration of either adrenaline or corticoids, the organism acquires any great increase or decrease in its sensitivity to these hormones. It must be kept in mind that adrenaline has a brief action and is rapidly destroyed or eliminated, hence even repeated daily injections will not result in a CUMULATIVE EFFECT. The corticoids on the other hand are generally more slowly acting and the effect of many daily, or even less frequent, injections may eventually result in a cumulative action (e.g., hypertension, kidney damage).

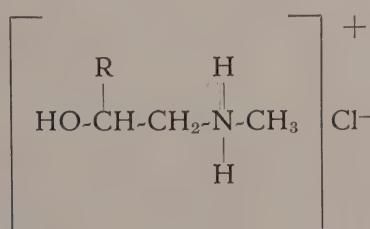
Some degree of ADAPTATION to adrenaline has been demonstrated, however, inasmuch as after repeated injections of small amounts, the resistance to subsequent fatal doses tends to increase. Similarly, it has been found that following repeated intraperitoneal administration of desoxycorticosterone acetate, the dose necessary to produce general anesthesia,

rises. The thymus atrophy of rats, chronically treated with subcutaneous injections of desoxycorticosterone, also tends to be transitory. Indeed, there appears to be a reversal of this effect after several weeks. These are not examples of true adaptation to the hormone itself, since the organism resistant to the anesthetic or the anti-thymus effect of the compound, indefinitely retains its sensitivity to most other effects (hypochloremia, compensatory adrenal atrophy, etc.). In such instances it is preferable to speak of selective "end-organ resistance." Bioassay of the blood of these animals has shown, furthermore, that the adaptation is not due to antihormone formation; indeed, there is little evidence of true anti-hormone production against any steroid compound.

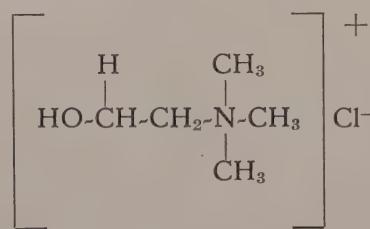
THEORIES CONCERNING THE ADRENAL HORMONES

Biogenesis of Adrenaline. — Attention has been called to the similarity between the chemical structure of choline and the side chain of adrenaline. This fact, and the presence of large quantities of choline in the adrenals, led to the supposition that the latter compound may play a rôle in the biogenesis of adrenaline.

This similarity is especially striking if choline chloride (the form in which choline exists in tissues) is compared with adrenaline hydrochloride, writing their formulae in accordance with current concepts concerning the valence of their nitrogen :



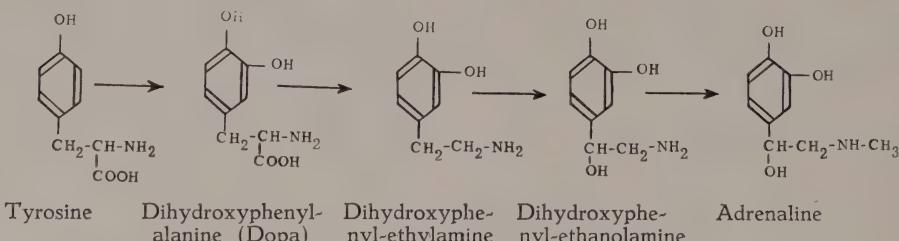
Adrenaline hydrochloride



Choline chloride

Most investigators agree, however, that the likeliest precursor of adrenaline is the amino-acid, tyrosine. The introduction of a second -OH, into the tyrosine molecule, has been shown to occur under the influence of the enzyme tyrosinase (found in plants but not in animals); some similar mechanism may

be effective in the adrenals. Thus, dihydroxyphenylalanine ("dopa") is formed from tyrosine, under the influence of ultraviolet light, in the presence of Fe^{++} ions or ascorbic acid. One possible pathway for the biogenesis of adrenaline would be the following :



It is also claimed that slices of kidney tissue can decarboxylate tyrosine and thus form tyramine in vitro. This tyramine can then be transformed, by adrenal-medullary tissue, in vitro, into a substance giving biologic and colorimetric adrenaline reactions.

Biogenesis of the Corticoids. — This problem has been discussed in the section on The Steroids, to which the reader is referred.

Fate of Adrenaline in the Body. — It is a well-known fact that the actions of adrenaline are extremely transitory; even following intravenous administration of large doses, the hormone soon disappears from the blood. Neither nephrectomy nor hepatectomy significantly influence the activity of adrenaline. From this it was concluded, that neither elimination through the urine nor hepatic detoxification play an important rôle in its inactivation. If adrenaline is injected into the portal vein or a peripheral artery (that is, if the hormone must pass through a capillary network before reaching the general circulation), its activity is diminished. Adrenaline is also inactivated, for instance, by repeated passage through a perfused frog-leg preparation. It appears that the hormone is

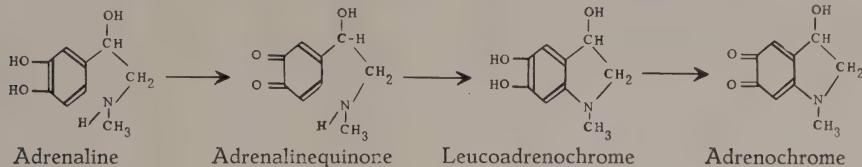
DETOXIFIED IN VARIOUS TISSUES, its inactivation not being limited to any one organ. This is hardly unexpected in view of the great lability of the hormone; however, urinary elimination of adrenaline metabolites, also plays an important rôle, especially if very large quantities are administered. Thus, it was shown that in rabbits much of the adrenaline injected reappears in the urine as protocatechuic acid. In man, adrenaline sulphate can be recovered from the urine following its ingestion (the yield being about 70% of the hormone given). This pharmacologically inert adrenaline ester can be reconverted into the active hormone, by hydrolysis.

Probably the most important method for the inactivation of adrenaline in the body is its oxidation. In vitro experiments indicate that AMINE-OXIDASE (tyraminase, adrenaline oxidase) can split the side chain at the amino group with the formation of an aromatic aldehyde having no pressor action (oxidative deamination).

Other enzymes, the POLYPHENOL OXIDASES (of potatoes and mushrooms, catechol oxidase, cytochrome-cytochrome system, peroxidase) are capable

of oxidizing the nucleus of adrenaline to an inactive red product "adreno-

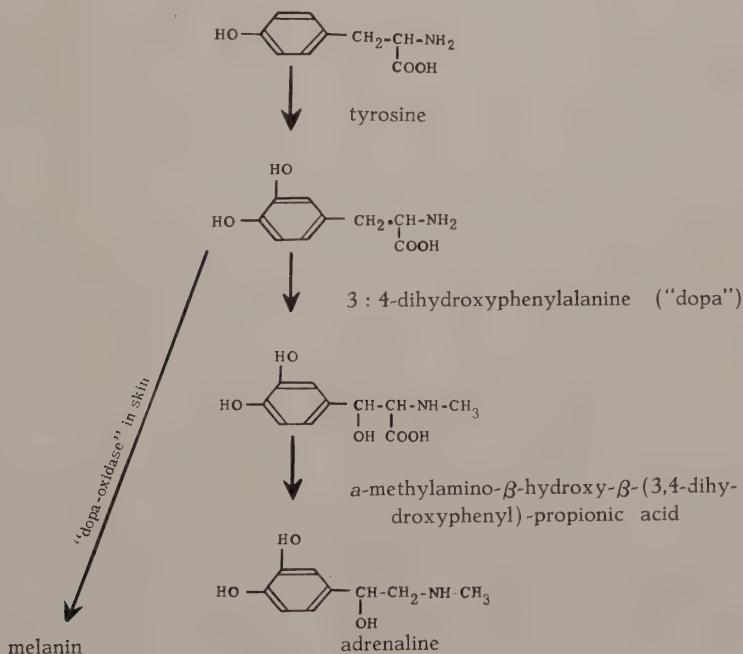
chrome," which is also obtained by the enzymatic oxidation of tyrosine:



This upon further oxidation is claimed to turn into "adrenoxine," a substance with acetylcholine-like negative ino- and chronotropic actions. Curiously, among organ extracts only those coming from structures inhibited by adrenaline (exception: small intestine) catalyze this reaction. Still further oxidation leads again to completely inactive products. It is doubtful what rôle, if any, is played by these reactions *in vivo*.

Numerous workers found that partial oxidation of adrenaline leads to melanin-like brown products and concluded that the hormone participates in *pigment formation*. This is of interest in connection with the skin pigmentation characteristic of Addison's disease. It

has been shown (Bloch) that slices of normal skin become deeply pigmented, when immersed in a solution of 3:4-dihydroxyphenylalanine (dopa), while albino skin remains unpigmented. Other allied compounds (e.g., tyrosine, tryptophane, pyrogallol), fail to cause pigmentation of skin sections. It was concluded that "dopa" is a precursor of melanin which can be transformed into the latter by the enzyme "dopa-oxydase," present in the skin. This was regarded as further evidence that dopa is an intermediary in the formation of adrenaline from tyrosine. Hypocorticoidism allegedly augments the formation of melanin, at the expense of adrenaline synthesis, from dopa.



Fate of Corticoids in the Body. — (See also : The Steroids.) It has been shown that partial hepatectomy greatly prolongs and increases the activity of various corticoid hormones, and that the latter are less active when administered through the portal circulation than when introduced into the systemic blood circuit. We may therefore conclude that the liver plays an important (though probably not the sole) part, in the inactivation of corticoids. Following administration of large doses of desoxycorticosterone acetate, there is a small increase in urinary pregnanediol elimination but this can only account for a minute fraction of the hormone destruction. Probably most of the exogenous or endogenous corticoids are completely oxidized or transformed into inactive steroid derivatives. In any case no significant amount of corticoids appear in the urine in the form in which they are present in the adrenals.

Mechanism of Adrenaline Action. — Since a striking similarity exists between the effects of adrenaline and of sympathetic nerve stimulation, it has been assumed that the hormone acts through SYMPATHETIC NERVE ENDINGS. This view was apparently corroborated by the observation that, unlike other vessels, those of the placenta, which are not innervated, do not respond to adrenaline. However, in chick embryos, adrenaline inhibits the rhythmic contractions of the amnion, which is not innervated; it also causes contraction of blood vessels, in chick embryos, before they are innervated; it accelerates the rate of the, not as yet innervated, heart in fish embryos or in nerveless tissue cultures of cardiac muscle cells. Denervated organs, far from being insensitive, become actually hypersensitive to the action of adrenaline. Topical application of the hormone, to sympathetic or sensory nerves or sympathetic ganglia, remains without effect; nico-

tine, which paralyzes sympathetic ganglion cells, does not inhibit the action of adrenaline. All these observations merely indicate, however, that the hormone does not necessarily act through the intermediary of nerves, without implying that it cannot influence the nervous system. The old concept, that adrenaline can migrate through the nerves (*neurocriny*), has not received confirmation.

As regards the CHEMICAL MECHANISM of adrenaline action, it has been found that calcium and potassium ions are indispensable for certain actions of the hormone, but their exact rôle has not been determined.

It will be kept in mind that adrenaline acts as a drug with gradient action ("Potenzialgift"), since it influences the heart and blood vessels, not only at the time when it enters into their substance, but also when it leaves them ("Auswaschphänomen").

Mechanism of Corticoid Hormone Action. — It is most probable that the GLUCO-CORTICOIDS owe most of their effects, primarily, to gluconeogenesis. These compounds facilitate the conversion of non-sugars (mainly proteins) into carbohydrates and thus replenish the glycogen stores of the body, even in the fasting animal.

The MINERALO-CORTICOIDS facilitate the elimination of potassium through the kidneys, but, at the same time, cause retention of sodium, chloride and water in the tissues. Since nephrectomy does not completely abolish the effect of mineralo-corticoids upon electrolyte and water distribution, it must be assumed that their action is not solely due to an influence upon urine secretion. Their marked effect upon capillary permeability and tissue affinity for water and electrolytes probably plays an important, though meanwhile cryptic, rôle in the mechanism of their action.

We are entirely ignorant of the mechanism through which the corticoids — like so many other steroids — exert their general ANESTHETIC EFFECT in acute experiments. It is reasonable to assume, however, that the periodic paralysis induced, especially in dogs, by chronic desoxycorticosterone acetate treatment, is the result of the replacement of muscle potassium by sodium. It has been shown that potassium administration and sodium withdrawal antagonize this action of desoxycorticosterone acetate and that a marked lowering of the blood and muscle potassium, together with an increase in muscle sodium, accompany the motor disturbances.

The LIFE-MAINTAINING ACTION of corticoids, in adrenal insufficiency, is probably due to a combination of effects. The diverse actions of the corticoids upon metabolism, circulation, heat regulation, etc., are all indispensable for the maintenance of life and especially for the acquisition of adaptation to changes in the external or internal environment of the body. It appears that the corticoids are particularly useful during the process of adaptation but become relatively dispensable for the performance of functions to which inurement has already been acquired. Thus, pretreatment with cold, drugs or exposure to muscular exercise, prior to adrenalectomy, endow the organism with a resistance, which is largely maintained even following subsequent removal of the adrenal glands. Any of these damaging agents is extremely noxious to unadapted adrenalectomized animals. This led to the concept that it is, not the actual performance of special functions, but, the acquisition of inurement, which requires specially large quantities of corticoid hormones. Since continuous adaptation to changes in our external and internal environment is the most characteristic feature of life,

the great biologic importance of corticoids, for all vital processes, is readily understandable.

Among the EARLIER THEORIES of corticoid action, the following deserve brief mention :

(1) *The Detoxification Theory* (Brown-Séquard, 1890; Langlois, 1893; Rimpl, 1938). According to this concept, the cells of the adrenals remove toxic substances from the blood that passes through the glands. In this original formulation, the theory has been abandoned, after it became known that adrenalectomized animals are maintained by extracts of the cortex.

A later modification of the same theory, postulates that the cortical hormones endow extra-adrenal cells with special detoxifying functions and that, in the absence of corticoids, the organism is poisoned by endogenous, toxic metabolites. This formulation of the detoxification theory is not incompatible with our views (e.g., the adaptation syndrome theory), but up to the present it has not been possible to demonstrate the accumulation of any hypothetic, toxic metabolites, in the tissues of adrenalectomized animals.

(2) *The Acidosis Theory* (Swingle, 1927), postulated that the manifestations of adrenal insufficiency are secondary to the development of a severe acidosis. This view has been abandoned.

(3) *The Respiratory Theory* (Bornstein and Holm, 1923). Hyperpnea is a rather characteristic manifestation in the terminal stages of adrenal insufficiency. The resulting decrease in the CO₂ tension of alveolar air and blood were regarded as the cause of the adrenal deficiency syndrome, but it was subsequently shown that animals kept in a high CO₂ atmosphere, in which a loss of blood CO₂ is impossible, still develop characteristic adrenal-insufficiency symptoms.

(4) *The Theory of Temperature Regulation* (Sajous, 1925), regarded the maintenance of body temperature as the main function of the adrenals. This view is incompatible with the fact that adrenal insufficiency develops even if the body temperature is artificially maintained.

(5) *The General Tissue Hormone Theory* (Hartman et al. 1932), regarded the corticoids as "general tissue hormones," necessary for the functions of all tissues. While this is undoubtedly correct, it is merely a statement of a fact rather than an explanation.

(6) *The Carbohydrate Metabolism Theory* (Britton and Silvette, 1932), regards all the manifestations of cortical insufficiency as secondary to hypoglycemia. However, adrenalectomized dogs may not show any decrease in blood sugar, nor be improved by glucose administration, at the time cortical insufficiency symptoms are manifest. Furthermore, pancreatectomy fails to counteract all the manifestations of cortical insufficiency, even if it raises the blood sugar concentration.

(7) *The Circulatory Theory* (Kellaway and Cowell, 1923), considers the hemoconcentration, and the increased capillary permeability, as the basic disturbances in cortical insufficiency. This would account for an inability to maintain consumed fluid in the circulation, which in turn would cause all other symptoms. However, prolonged withdrawal of food and water causes circulatory disturbances similar to those seen after adrenalectomy, without eliciting a typical adrenal insufficiency syndrome; conversely, continuous fluid infusions do not considerably prolong the life of adrenalectomized animals, even though the blood volume is restored.

(8) *The Potassium Intoxication Theory* (Zwemer and Truszkowski, 1936), was based upon the observation that the potassium concentration of the

plasma runs roughly parallel with the manifestations of adrenal-cortical deficiency, in suprarenalectomized animals. Toxic doses of potassium salts imitate certain manifestations of suprarenal insufficiency and adrenalectomized animals are extraordinarily sensitive to potassium ions. Corticoids restore the blood potassium to normal and raise the potassium resistance of adrenalectomized animals. It was concluded that a disturbance in potassium metabolism is the primary cause of adrenal insufficiency and that the changes in water distribution, sugar metabolism and the balance of sodium and chloride, are all secondary phenomena. It has been shown, however, that on certain diets, adrenalectomy causes no changes in serum potassium, yet it induces a typical deficiency syndrome. Furthermore, animals receiving large doses of potassium acquire a resistance to it, which persists even following subsequent adrenalectomy, although the blood potassium rises far above the level usually seen at death from cortical insufficiency.

(9) *The Histamine Intoxication Theory* (Lucas, 1926), assumed that endogenous intoxication with histamine is the cause of cortical insufficiency, but it has not been possible to demonstrate a constant increase in blood or tissue histamine following suprarenalectomy. It is true that the histamine resistance of adrenalectomized animals is decreased, but adrenaline appears to be even more potent than the corticoids in restoring histamine resistance to normal. In any case, a decreased resistance to histamine would not be significant since, in the absence of the adrenals, resistance to almost all toxic substances is diminished.

(10) *The Flavine Theory* (Verzár, 1936), was based on the claim that cortical extracts are unable to maintain adrenalectomized rats alive, if the diet is deprived of flavine — provitamin B₂.

It has also been stated that flavine-phosphate maintains the life of adrenalectomized animals just as well as cortical extracts, while lactoflavine is inactive. It was concluded that the function of the corticoids is to enable the organism to combine flavine with phosphate and that all symptoms of adrenal deficiency result from a breakdown of this mechanism. Subsequent work failed to confirm the observations upon which this theory was based.

(11) *The Sodium and Chloride Deficiency Theory* (*Harrop et al. 1933*) regards the derangement in NaCl metabolism, as the basic cause of the adrenal insufficiency syndrome. In the adrenalectomized dog, the hypochlоремia and hyponatremia run parallel with the increased loss of NaCl through the urine, since the loss of these electrolytes causes dehydration, due to the accompanying loss of extracellular tissue water. Administration of NaCl improves the condition of adrenalectomized animals, if it results in NaCl retention. It has also been found that, even in intact animals, sodium deficiency simulates the manifestations of adrenal insufficiency. However, in animals recovering from adrenal insufficiency, due to cortical extract administration, the chloremia and natremia may remain low, in spite of obvious clinical improvement, if electrolyte deficient diets are given. The disappearance of the symptoms is apparently due to a shift of fluids from the tissues into the blood and this shift may occur in spite of a low serum sodium and chloride concentration. The loss of NaCl through the urine is likewise not an essential prerequisite for the development of cortical insufficiency; adrenalectomized dogs can be maintained with cortical extracts, even on diets so poor in NaCl that subsequent withdrawal of the cortin causes no further loss of these electrolytes, through the urine.

It is obvious that any of the metabolic disturbances characteristic of adrenal insufficiency, if they are sufficiently severe, can be the immediate cause of death. Thus, hypoglycemia, (especially in animals fasted or kept on low carbohydrate intake), decrease in body temperature (especially in animals kept in cold surroundings), potassium intoxication (especially in animals receiving high doses of this ion), NaCl deprivation (especially on diets containing inadequate amounts of salt) can all be regarded as the most important disturbance in individual cases. Their fundamental rôle, in the insufficiency syndrome, is clearly demonstrated by the great improvement seen following administration of sugar, heat, sodium chloride, etc., under special circumstances. It is also evident that a severe disturbance, in any of these functions, secondarily aggravates the condition of adrenal insufficiency thus adversely influencing other functions, while correction of any one derangement exerts a beneficial action upon the organism as a whole. This does not mean that any one of the above mentioned changes is necessarily and always the cause of the entire deficiency syndrome, but merely that it can, under certain circumstances, become the weakest point, and hence the limiting factor, in the body's effort to survive.

Different Kinds of Adrenaline. — It has been claimed that adrenaline exists in the medulla partly in the form of a comparatively less active, side-chain-substitution product, designated as "PRO-ADRENALINE" or "VIRTUAL ADRENALINE." Even mere drying of the glands (in vacuo, over sulfuric acid) splits the substituent off the molecule and causes a marked increase in hormone activity.

Many other observations suggest that adrenaline may exist in the gland in a combined form attached to lipids.

ascorbic acid, or protein ("latent adrenaline") or lactic acid ("lactyladrenaline"). None of these adrenaline modifications have definitely been proven to play a physiologic rôle. Even if adrenaline were present in some esterified or otherwise masked form, there is no reason to believe, that this would cause a qualitative change in its pharmacologic properties, and thus result in a physiologically different kind of adrenaline.

Whether SYMPATHIN is essentially different from adrenaline is still much debated, although the liberation of an adrenergic substance by certain sympathetic nerves is no longer in doubt.

The action of some organs is inhibited (e.g., muscular relaxation), while that of others is stimulated (e.g., muscular contraction), by excitation of their adrenergic nerves. Cannon and Rosenblueth (1933) claimed that the sympathin liberated by excitatory adrenergic nerves, exerts an excitatory effect on distant organs, while the sympathin, produced by inhibitory adrenergic nerves, acts as an inhibitor upon the organs to which it is carried through the blood stream. These facts were interpreted as follows: The nervous stimulation liberates a substance similar to adrenaline, which has both inhibitory and excitatory effects (substance "M"). This compound is transformed into a pure excitatory substance "sympathin E" (E for excitatory) in organs in which it causes excitation and into a purely inhibitory substance "sympathin I" (I for inhibitory) in organs in which it causes inhibition. Both these substances are then carried to distant organs through the blood. Stimulation of the nerves of the liver, heart, extremities or tail, have marked excitatory effects but only very weak inhibitory actions on distant organs, hence they distinctly differ from adrenaline. Furthermore, in ergotoxin-treated cats, the pressor effects of adrenaline is reversed into a depressor action, while stimulation of the hepatic nerves still increases the blood pressure. It has also been shown that stimulation of hepatic nerves has no effect upon the denervated iris of the cat, while stimulation of the cardiac sympathetic causes mydriasis.

Tyrosinase or catechol-oxydase pretreatment changes the pharmacologic action of heart perfusates, obtained after sympathetic stimulation, so that instead of exerting an adrenaline-like effect, their action becomes purely inhibitory. These enzymes are also known to oxidize the

hormone to adrenoxine, a substance with acetylcholine-like negative ino- and chronotropic actions. This adrenoxine (or some similar adrenaline derivative) could be "sympathin I". In this connection, the resemblance between the side chain of adrenaline and choline (a depressor substance), should also be recalled. On the other hand, "sympathin E" has been considered to be identical with arterenol, which possesses the pharmacologic properties of the hypothetic excitatory substance.

In any case Cannon and Rosenblueth never actually proved the existence of a purely excitatory or inhibitory sympathin. They merely showed that during the humoral transmission of sympathetic impulses, more excitatory and less inhibitory effects are noted than after injection of adrenaline. Furthermore, contrary to what the theory postulates, the perfuse (or venous blood) of an inhibited organ fails to produce inhibitory responses in organs which are normally excited by sympathetic stimulation or vice versa.

It is probable that "sympathin-I" is adrenaline, "sympathin-E" is nor-adrenaline (arterenol). However, until this is definitely established, it may be best to use the designation "sympathin-N" ("N" for Nor-adrenaline) instead of "sympathin-E", and "sympathin-A" ("A" for Adrenaline) instead of "sympathin-I". This would emphasize, without definitely accepting, the probable identity of these two compounds with nor-adrenaline and adrenaline respectively (*v. Euler, Bacq*).

The above interpretation is mainly based upon the finding that extracts of various animal (mammalian) tissues, especially nerves, contain an amine which possesses nor-adrenaline-like actions, while other tissues (e.g., human coronary arteries and nerves) contain a compound with more adrenaline-like actions. Indeed, certain cells (e.g., those of the abdominal ganglion of annelids or the frog's heart) appear to produce adrenaline itself.

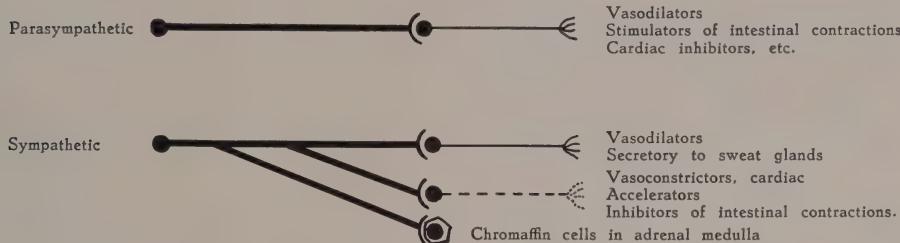
Perhaps many tissues can synthesize aminated derivatives of catechol and the synthesis of adrenaline goes through arterenol; in other words, the methylation of the nitrogen would be the last step in the synthesis. If this would fail

to occur in certain tissues, arterenol would be the end-product (*Bacq*).

In connection with the humoral transmission of adrenergic stimuli, it is also noteworthy that *acetylcholine* is the humoral agent responsible for the trans-

mission, not only of all preganglionic, and postganglionic parasympathetic impulses, but also, of all preganglionic, and some postganglionic, sympathetic stimuli, as indicated by the following diagram :

FIGURE 1



Schematic drawing illustrating distribution of cholinergic and adrenergic fibers.
Heavy lines, preganglionic; thin lines, postganglionic; interrupted lines, adrenergic;
solid lines, cholinergic.

Different Kinds of Corticoids. — The steroids isolated from the adrenal cortex have been discussed in the section dealing with "The Steroids." In addition to the fully characterized adrenal steroids, there are probably several others, whose chemical structure is not yet established. Thus, in the amorphous material, which remains after isolation of the known members from the adrenal steroid fraction, there appear to be compounds of high physiologic potency which exert glucocorticoid and mineralo-corticoid actions. Desoxycorticosterone does not appear to occur in the normal adrenal, except perhaps in traces.

Among the possible cortical hormones, whose existence is still in doubt, we might mention "CORTILACTIN" (*Hart-*

man et al. 1933), a compound supposedly present in adrenal extracts and necessary for the maintenance of lactation following adrenalectomy. It has also been stated that cortical extracts possess GONADOTROPHIC activity. It is possible that these effects are not due to separate hormones, but to some of the known adrenal steroids, or synergistic combinations of these. The existence of an ASCORBIC ACID SUBSTITUTED CORTICOID has also been postulated, but not definitely proven.

It is not yet known IN WHAT FORM CORTICOIDS CIRCULATE IN THE BLOOD. Since they are rather insoluble in aqueous media, it is probable that they form water-soluble esters or are bound to protein.

EXPERIMENTAL PHYSIOLOGY OF THE ADRENALS

EXPLANTATION OF THE ADRENALS

The cells of the adrenal cortex, unlike those of the medulla, grow quite well in tissue cultures. Perfusion experiments with whole adrenals also yield many interesting results, especially as regards the effect of various sub-

stances (which can be added to the perfusion fluid) upon adrenaline secretion, *in vitro*. It is noteworthy, furthermore, that the oxygen consumption of the perfused adrenal is extraordinarily high and is further increased by the addition of adrenaline to the perfusion

fluid, or by stimulation of the splanchnic nerves.

TRANSPLANTATION OF THE ADRENALS

Although heterotransplantation of adrenal-cortical tissue is rarely successful, homotransplants and especially autotransplants, take quite readily in various species, including man. Special care should be taken to free the cortex of all traces of medulla, since adrenaline causes necrosis, not only of the chromaffin cells, but, even of adjacent cortical tissue. A simple technic which permits transplantation of medullary tissue, is that in which the vascular pedicle of the gland is preserved and the organ is merely transposed. Otherwise the medulla takes only in rare cases.

That the medulla usually succumbs in adrenal grafts proves of value in studies concerning the physiology of pure cortical tissue. If the adrenals are removed, freed of visible medullary tissue and subsequently transplanted, it may be taken for granted that after some time the graft will consist of pure cortical tissue. Even if a few medullary cells had, accidentally, remained in the transplants they would have become necrotic.

TECHNIC OF ADRENALECTOMY

The previously mentioned fact, that in certain selachian FISH, the adrenal cortex forms an organ anatomically separate from the medulla, renders removal of this "interrenal organ" technically simple. The intervention results in pure cortical insufficiency, without damage to the adrenaline-producing system.

In most AMPHIBIA the adrenals are intimately connected with the anterior surface of the kidney and are best removed by cauterization.

In BIRDS, adrenalectomy is particularly difficult because of the close connections between the glands and the very friable vena cava. Here it is use-

ful to slit the capsules of the glands and to remove their parenchyme through a glass cannula, connected with a suction pump (similar to the cannula used for hypophysectomy). In order to prevent the continued growth of small, undetectable, cortical remnants, a cotton pad soaked with ferric chloride solution, may be applied to the inner surface of the adrenal capsules after the operation.

In most MAMMALS, including man, the removal of the adrenals is not attended with great technical difficulties. Adrenalectomy is best performed through two separate subcostal incisions, remaining in the retroperitoneal space as far as possible. The operation may be performed in one stage in the *laboratory rat* and *mouse*, which are comparatively resistant to adrenal insufficiency and in whom both adrenals are readily accessible. In the *guinea pig*, *rabbit*, *wild rat*, *cat*, *dog* and *monkey* it is preferable to remove the right adrenal (which, being adherent to the liver and vena cava, is more difficult to dissect) in a first stage, so that the animal still has functional adrenal tissue on the left side while recovering from the operation. One or two weeks later the comparatively free left adrenal can subsequently be removed without much surgical trauma. Before the second adrenal is removed, it is well to administer fairly high doses of corticoids, in order to prevent acute adrenal insufficiency during the post-operative period. In view of the rich nerve and blood supply of the adrenals, it is especially important that this operation be performed with impeccable surgical technic, using blunt dissection as far as possible.

SEPARATE REMOVAL OF THE MEDULLARY TISSUE is rather difficult. It can best be performed in small laboratory rodents (e.g., rat), in whom the cortex forms a regular, spheric envelope around the medulla. The medulla is

exposed by a deep incision and subsequently removed through a suction tube. Small remnants often persist, hence the most commonly employed procedure is to transplant the adrenal tissue, after freeing it of all visible medullary cells, taking advantage of the above mentioned fact that only cortical cells persist in grafts.

For many experimental purposes, simple DENERVATION of the adrenals suffices to inactivate the medulla, since adrenaline production ceases almost completely following destruction of the splanchnics.

EFFECTS OF ADRENALECTOMY AND TREATMENT WITH ADRENAL HORMONES

State. — ADRENALECTOMY causes the development of a rapidly fatal insufficiency syndrome, in most animal species. This is usually characterized by a rapid pulse, thirst, suppression of urine secretion, vomiting, diarrhea, muscular weakness, loss of appetite, a terminal fall in blood pressure and finally coma, death ensuing within a few days. Increased melanin deposition in the skin, so characteristic of adrenal insufficiency in man, is rarely seen in experimental animals and is apparently, largely dependent upon their diet.

In fish, amphibia, reptiles and certain mammals, especially the laboratory rat (not the wild rat, whose cortex is normally larger), the insufficiency syndrome takes a chronic course and death may not ensue until weeks after complete adrenalectomy.

The length of survival following adrenalectomy is largely dependent upon the conditions under which the animals are kept after the operation. High protein diets and exposure to any type of stress are especially damaging, since the corticoids play an essential rôle in gluconeogenesis from proteins, and in adaptation and resistance to diverse noxious agents. On

the other hand, high carbohydrate diets, and a rich supply of sodium, are most effective in prolonging the life-span of animals deprived of their adrenals. During hibernation animals are particularly resistant to adrenalectomy.

REMOVAL OF THE ADRENAL MEDULLA, or its inactivation by denervation, are not followed by any very obvious manifestations of insufficiency. Only in emergency situations is the lack of adrenaline secretion detectable. The existence of important extra-adrenal sources of adrenergic substances (sympathetic nerve endings, paraganglia) may explain why a syndrome of severe adrenal medullary insufficiency cannot be so produced.

In man, ADRENALINE given in toxic doses, may produce a feeling of anxiety, fear, tenseness, throbbing headache, tremor, weakness, dizziness, palpitation and difficulty of respiration. All these manifestations are usually transitory, yet quite alarming to the not forewarned patient. Still higher doses may produce an unduly sharp rise in blood pressure, accompanied by cardiac arrhythmias, anginal pain, and sometimes even cerebral hemorrhage or hemorrhagic lung edema. Ventricular arrhythmia, ventricular fibrillation and lung edema are the most common causes of death due to adrenaline overdosage. Patients with hypertension, arteriosclerosis (especially coronary disease) or hyperthyroidism are particularly sensitive to this hormone. Individual tolerance varies, but 2 mg. intravenously or 8 mg. subcutaneously, is usually fatal in normal man.

CORTICOIDS rarely produce any signs of acute overdosage. Upon prolonged administration, however, desoxycorticosterone acetate may cause a great increase in blood volume with cardiac dilatation, hypertension and proteinuria in man and in animals. Addi-

sonian individuals appear to be more than normally sensitive to desoxycorticosterone overdosage.

Animal experiments indicate that chronic overdosage with desoxycorticosterone causes sufficiently severe adrenal-cortical atrophy to produce a modified type of adrenal insufficiency, due to suppression of endogenous gluco-corticoid production. (See p. 119.)

Temperature. — ADRENALECTOMY causes a slight decrease in body temperature and a pronounced disturbance in thermoregulation, especially in animals exposed to cold. ADRENALINE tends to raise the body temperature slightly, while CORTICOIDS cause no noteworthy change.

Basal Metabolism. — ADRENALECTOMY causes a pronounced decrease in the metabolic rate of most animal species, to about 25-35% below normal. At the same time the R.Q. tends to fall. The rise in B.M.R., normally produced by hypophyseal extracts or thyroid hormone, is not inhibited by adrenalectomy. Hence, these hormones do not act merely through the intermediary of the adrenals. Administration of glucose raises the low B.M.R. and low R.Q. of suprarenalectomized animals. There is no reason to believe, therefore, that the adrenals are indispensable for the combustion of sugar.

ADRENALINE raises the B.M.R., without exerting a consistent effect upon the R.Q. CORTICOIDS and especially cortical extracts rich in gluco-corticoids, restore the low B.M.R. and low R.Q. of adrenalectomized animals to normal, but exert a rather inconstant influence upon the B.M.R. of intact animals.

Tissue Metabolism. — It has been shown repeatedly that the metabolism of tissues, taken from ADRENALECTOMIZED animals, is impaired. Their basal oxygen consumption is diminished and the increment in oxygen uptake, which follows the addition of

various amino-acids or the corresponding keto-acids, is subnormal. CORTICOID extracts correct these derangements.

The metabolism of various tissues is increased by ADRENALINE, especially if optimal amounts of oxygen are not available. All these findings indicate that the adrenals exert a direct influence upon the metabolism of tissues.

Carbohydrate Metabolism. — The fundamental factors regulating carbohydrate metabolism have been discussed in the section on the Pancreas (Experimental Physiology), to which the reader is referred.

THE BLOOD GLUCOSE concentration is decreased by adrenalectomy, especially if the experimental animal is deprived of exogenous sugar. Under optimal conditions of maintenance, that is, on a high carbohydrate, high sodium chloride intake, adrenalectomized animals (especially the rather resistant laboratory rat, in contrast to wild rats and most other species), may maintain a normal or even high blood glucose concentration. However, *fasting* or any *non-specific stress* which increases carbohydrate combustion, is likely to elicit a fatal attack of hypoglycemia in the adrenalectomized animal unless it receives corticoid hormone treatment. Presumably the corticoids are indispensable for rapid gluconeogenesis and especially for the formation of sugars from endogenous sources (body protein and fat).

The so-called *piqûre diabetes*, produced by surgical lesions to the floor of the fourth ventricle, is almost, but not entirely, prevented by adrenalectomy. This was interpreted to mean that in addition to the sympathetic nervous connections between the floor of the fourth ventricle and the adrenal medulla (which regulate adrenaline hyperglycemia), direct nervous connections exist between the fourth ventricle and the liver. The latter, perhaps

through the liberation of sympathin, may cause the slight residual hyperglycemia of adrenalectomized animals. Other experimental hyperglycemias and glycosurias, such as those produced by *splanchnic stimulation, theobromine, traumatic shock, etc.*, are likewise prevented by adrenalectomy; indeed the same agents which raise the blood sugar in the normal, may decrease it in the adrenalectomized animal. This is probably due to the inability of the latter to respond to stress, either with adrenaline or with gluco-corticoid secretion, which are indispensable for the production of the above-mentioned hyperglycemias and glycosurias.

Adrenaline causes hyperglycemia, both in the normal and the adrenalectomized animal. This effect depends, however, upon the presence of adequate hepatic glycogen stores, since it is due to the breakdown of liver glycogen into blood glucose. The degree and duration of adrenaline hyperglycemia is of diagnostic value, since it gives some indication of the amount of hepatic glycogen which can be mobilized by the patient.

The tendency to develop hypoglycemia after adrenalectomy can be counteracted by the administration of *gluco-corticoids* (e.g., corticosterone) while *mineralo-corticoids* (e.g., desoxycorticosterone acetate) have no beneficial effect, except, perhaps, by improving the general condition of the animals. Both in the adrenalectomized and in the intact animal, treatment with gluco-corticoids raise the fasting blood sugar level, presumably by transforming protein into sugar. Mineralo-corticoids (e.g., desoxycorticosterone) may cause such a pronounced compensatory involution of the adrenal cortex that a tendency to severe fasting-hypoglycemia ensues, presumably due to deficient endogenous gluco-corticoid formation.

Insulin causes a particularly pronounced and rapid hypoglycemia in the adrenalectomized animal. This great insulin hyper-sensitivity is probably due to the absence of gluco-corticoids, which normally antagonize it in the intact animal. There is also a mutual antagonism between the glycemic effects of adrenaline and insulin.

Thyroid hormone augments the hyperglycemic action of adrenaline unless the resulting hyperthyroidism is so severe as to deplete the hepatic glycogen stores.

The hyperglycemia and glycosuria characteristic of various types of diabetes, (e.g., that produced by *pancreatectomy*), is greatly alleviated by adrenalectomy. Conversely, even partial pancreatectomy suffices to sensitize experimental animals to the diabetogenic action of gluco-corticoids.

Neither adrenaline nor the gluco-corticoids influence the blood sugar after *hepatectomy*. This suggests that they do not materially affect glucose utilization or formation in extra-hepatic tissues.

GLUCOSE ABSORPTION from the alimentary tract is delayed by adrenalectomy; this deficiency is restored by gluco-corticoid hormone treatment. However, the intestinal absorption of all sugars is not equally impeded by adrenal insufficiency, there being apparently most pronounced interference with the normal, preferential, selective absorption of certain sugars.

STORAGE OF CARBOHYDRATE is likewise largely under adrenal control. Adrenalectomy decreases glycogen storage in the liver and muscles, while mineralo-corticoid administration raises the glycogen concentration, especially in the liver. Adrenaline depletes the hepatic glycogen reserves, since it transforms glycogen into blood glucose. There is also a simultaneous diminution of muscle glycogen. However, after a prolonged fast, and in

other conditions which cause a serious depletion of hepatic glycogen stores, adrenaline may actually raise the liver glycogen. This is due to the fact that the lactic acid, formed by the hormone from muscle glycogen, is reconverted into hepatic glycogen through the "Cori cycle." (See : The Pancreas.) Thus, adrenaline transforms muscle glycogen into lactic acid, the latter is converted into hepatic glycogen, which in turn is broken down into blood glucose by the same hormone.

The LACTIC ACID content of the blood and muscles tends to diminish after adrenalectomy, apparently because of decreased lactic acid formation. On the other hand, adrenaline increases the lactic acid content of the blood, due to the above-mentioned conversion of muscle glycogen into lactic acid.

Lipid Metabolism. — Only a few isolated facts are known concerning the effect of adrenal hormones upon lipid metabolism. The *fatty infiltration of the liver*, produced by a variety of experimental procedures (anterior-pituitary extracts, partial hepatectomy and fasting, pancreatectomy), is prevented by adrenalectomy, unless adequate corticoid treatment is administered. Certain steroids of the adrenal cortex allegedly produce heavy fatty infiltration of the liver and even massive *fat deposition in various other tissues*. Chronic desoxycorticosterone acetate overdosage tends to raise the blood *cholesterol* level, especially in the presence of renal insufficiency. All this work requires further elucidation, to be useful for the understanding of the underlying metabolic changes.

The *ketonemia*, and especially the *ketonuria*, of experimental diabetes (pancreatectomy, anterior-pituitary extract) and of fasting are diminished, or even abolished, by adrenalectomy. Treatment with corticoids restores the ketosis under such conditions, while adrenaline does not.

Protein Metabolism. — ADRENALECTOMY seriously interferes with protein metabolism, especially in young, growing animals. It prevents the normal apposition of new body protein, the regeneration of tissue protein, following partial excision of certain organs (e.g., liver, kidney), and eventually, it results in cachexia due to the predominance of protein-catabolic processes. Adrenal deprivation appears to affect especially the utilization of certain endogenous proteins for the formation of tissues. Thus, hepatic regeneration is inhibited in the fasting, adrenalectomized rat, although it proceeds rapidly in fasting, intact controls. This is additional evidence of the preëminent rôle played by the adrenal in adaptive processes, such as wound healing and regeneration.

CORTICOIDS restore the nitrogen metabolism of the adrenalectomized animal to normal, but we do not yet know which type of corticoid is responsible for this effect. In severe cortical insufficiency the high N.P.N. of the blood returns to normal when corticoid therapy is instituted. Even following bilateral nephrectomy, the rise in N.P.N. and death from uremia are delayed by corticoids. This suggests an extra-renal point of attack.

It is perhaps pertinent that adrenaline decreases the blood amino-acid concentration in the intact organism. It has also been stated that corticoids increase the γ -globulin content of the blood. These globulins are important for certain serologic defence reactions.

Salt and Water Metabolism. — ADRENALECTOMY causes dehydration of the tissues, probably with a decrease in extracellular, and an increase in intracellular water. This is accompanied by a sharp decrease in blood sodium and chlorides with an increase in blood potassium. Simultaneously, the urinary loss of sodium and chloride is increased, while that of potassium is di-

minished. Correction of these deficiencies is mainly dependent upon the administration of mineralo-corticoid hormones.

ADRENALINE induces a transitory rise in serum potassium, presumably due to discharge of K ions from various tissues (e.g., liver). This is followed by a more prolonged hypopotassemia. The fall in the inorganic phosphate content of the blood, produced by adrenaline, is apparently due to the phosphorolysis of glycogen, with the formation of glucose monophosphate.

Adrenaline may also cause marked diuresis, due to an increase of the filtration pressure in the malpighian corpuscles, occasioned by constriction of the efferent glomerular arterioles.

Desoxycorticosterone acetate and certain other CORTICOIDS cause diuresis, presumably because they interfere with the tubular reabsorption of Na and water. Under the influence of mineralo-corticoid overdosage, hypopotassemia develops and sodium tends to replace potassium in the muscles. This has been considered to be of importance in the pathogenesis of the muscular paralysis sometimes noted in dogs, monkeys and even patients, severely overdosed with desoxycorticosterone.

The effect of corticoids upon the mineral metabolism of animals, is subject to great species variations. Thus, marsupials (e.g., opossum) react to adrenalectomy with hyper-rather than with hypochloremia. Baby chicks appear to be especially sensitive to the edema-producing effect of desoxycorticosterone acetate and even short treatment, with relatively small doses of this steroid, suffice to produce excessive water accumulation in the connective tissue, as well as in the large body cavities.

Other Metabolites. — Numerous publications have dealt with the, allegedly important, influence of the adrenals upon the metabolism of GLUTA-

THIONE, ASCORBIC ACID, CHOLINE etc., but it is not yet possible to draw definite conclusions from the pertinent literature.

Growth and Bone Structure. — The somatic growth of the adrenalectomized animal is inhibited. It can be restored by corticoids but not by adrenaline.

Blood. — *Adrenalectomy* causes rather inconsistent changes in the BLOOD COUNT, these being largely dependent upon the degree of accompanying hemoconcentration. Often there is lymphocytosis.

Adrenaline administration elicits a marked increase in the number of circulating erythrocytes, and a less pronounced leucocytosis, due to the fact that it causes splenic contraction and thus evacuates blood corpuscles from the spleen into the blood stream. This is of importance in connection with the emergency function of the adrenals, since it augments the oxygen capacity of the blood and improves the chances of survival in the event of a hemorrhage. It is also of diagnostic value in certain diseases (e.g., malaria, lymphogranulomatosis, lymphosarcomatosis), since adrenaline may cause the appearance in the blood of abnormal cells otherwise hidden in the spleen. Adrenaline also increases the reticuloocyte count presumably by eliciting a discharge of immature red cells from the bone marrow.

Overdosage with corticoids causes lymphopenia, presumably due to the fact that numerous lymphocytes disintegrate as do the thymocytes within the thymic reticulum. It is claimed that, from the bodies of such disintegrating white cells, γ -globulin is freed for the manufacture of immune bodies, but this is doubtful.

Adrenalectomy does not markedly influence BLOOD COAGULATION but adrenaline administration accelerates it. This is also of importance in con-

nexion with the emergency function of the hormone; obviously, rapid coagulation of the blood is useful during emergencies, when wounds are likely to be contracted.

Cardiovascular System. — **ADRENALECTOMY** decreases the size of the heart and causes a fall in blood pressure. It also sensitizes animals to the hypotensive action of various drugs and damaging agents. This tendency towards a fall in blood pressure is effectively combatted by corticoids. Adrenaline produces only a temporary rise in the blood pressure of adrenalectomized animals, without really correcting the underlying deficiency. The hypertension normally elicited by constriction of the renal arteries is prevented by adrenalectomy unless adequate corticoid therapy is given.

ADRENALINE, administered in moderate doses, raises the blood pressure and strengthens the heart beat of the intact animal. At the same time the pulse rate is slowed, due to a vagal reflex through the pressoreceptors of the carotid sinus and aortic nerves. The bradycardia in this case is elicited only if the blood pressure rises, not if it is kept low by bleeding. If the vagi are previously transected, or eliminated by atropinization, adrenaline increases both the rate and the strength of the heart beat.

In man, adrenaline quickens the heart rate, even without atropinization, when it is given subcutaneously in small doses.

Certain drugs, such as chloroform, increase the sensitivity of the cardiac muscle to the production of ventricular fibrillation by adrenaline. It is claimed that this sensitization is due to a shortening, by the drug, of the ventricular refractory phase and accounts for many cases of sudden syncope and death in patients in whom adrenaline had been given, or suddenly discharged from the adrenal medulla, during chloroform anesthesia.

In approximately physiologic doses, adrenaline influences the various vascular territories in a different manner. The arterioles and capillaries of the mucous membranes, skin and splanchnic viscera (except the intestinal vessels), are constricted; simultaneously, the coronary vessels as well as those of the skeletal muscles and intestines are actively dilated. At these dose levels the vasoconstrictor effect predominates over the vasodilator action and a rise in blood pressure ensues. In coronary diseases, the dilatation of the coronary arteries (if it occurs) is overshadowed by the other effects, so that angina results. The arteries of the brain are passively distended through the general rise in blood pressure, while the lung vessels are not particularly affected unless large doses are given which constrict them. As a result of these changes, there is a redistribution of the blood, so that more becomes available to the skeletal and cardiac muscles, at the expense of the splanchnic area and skin.

Curiously, very small doses of adrenaline may cause a fall in blood pressure, presumably because the vasodilatation in the muscles overbalances the vasoconstriction in the skin and abdominal organs. However, if the blood pressure is already low, or the animal is kept under deep anesthesia, a pressor effect may be obtained, even with very small doses of the hormone. Conversely, following ergotoxin pre-treatment, even large doses of adrenaline have a depressor effect, owing to the previously-mentioned phenomenon of "adrenaline reversal." Presumably the ergot alkaloids paralyze the vasoconstrictor but not the vasodilator mechanism.

Long continued administration of adrenaline causes vascular sclerosis in certain animal species, especially the rabbit.

The oxygen consumption of the heart muscle is greatly increased by adrenaline.

Chronic overdosage with MINERALOCORTICOIDS (e.g., desoxycorticosterone) causes cardiac hypertrophy, a pronounced and prolonged rise in blood pressure, myocardial nodules (similar to those seen in rheumatic fever), as well as periarteritis nodosa, especially in the mesenteric, coronary, and brain arteries of various experimental animals. These vascular changes may cause death due to secondary thromboses and embolisms. Hypertension and focal areas of cardiac necrosis have also been observed in man, after desoxycorticosterone treatment. Diets rich in vitamins, carbohydrates and acidifying salts, but poor in protein and Na, tend to prevent all these changes.

Lymphatic System. — Most lymphatic organs, and especially the thymus, reach a physiologic maximum of development approximately at the time of puberty. In the adult man and animals, there is a certain "physiologic involution" of these organs. Adrenalectomy prevents this normal involution of the lymphatic tissue (especially of the thymus), without actually causing true hypertrophy or hyperplasia.

It is particularly noteworthy that the "accidental involution" of the thymus, produced by a variety of non-specific damaging agents (infections, intoxications, traumatic injuries, etc.), is completely prevented by adrenalectomy. Among the innumerable agents which cause thymus involution in the intact animal, only certain steroids can produce this effect following adrenalectomy. Folliculoids, testoids, corticoids and luteoids are active in this respect approximately in proportion to their folliculoid effect. (See The Steroids.) It is assumed that all other drugs and non-specific damaging agents act indirectly through the adrenals, by increasing their steroid

hormone production. (See: General-Adaptation-Syndrome.)

Adrenaline causes only a slight increase in the number of circulating leucocytes, presumably by discharging them from the spleen and bone marrow.

Respiratory System. — Following ADRENALINE administration, there is an initial period of *apnea* followed by an increase in the rate and depth of respiration. The apnea is probably the result of a reflex elicited by the rise in blood pressure through the pressoreceptors of the carotid sinus.

Adrenaline also exerts an inhibitory influence upon the plain musculature of the *bronchioles*. This action is of especial value in the therapy of asthma due to bronchiolar spasms.

When given in very high doses, adrenaline may cause fatal *lung edema* due to an excessive rise of pressure in the pulmonary circulation. The latter results mainly from back pressure consequent to the increased peripheral resistance in the systemic circulation.

ADRENALECTOMY exerts no specific effects upon the respiratory system; CORTICOID overdosage may, occasionally, cause pleurisy and even lung edema.

Muscles. — ADRENALECTOMY greatly diminishes muscular strength and causes ready fatigability. This can be combatted both by adrenaline and by corticoids, but the latter are much more effective.

ADRENALINE postpones fatigue and increases work capacity above normal even in intact animals. This is somewhat reminiscent of the so-called Orbely effect, that is, the postponement of fatigue in a muscle stimulated through its nerve, by simultaneous stimulation of the sympathetic fibers going to the muscle. Although a direct sympathetic innervation of muscles is not probable, it is believed that sympathetic liberation from vascular sympathetic fibers of muscles may be responsible for this action. The fatigue-combatting effect of adrenaline is not

sufficiently great, at non-toxic dose levels, to be of use in augmenting the muscular efficiency of man. Certain adrenaline derivatives are valuable analeptics however, because of their stimulating effect upon the central nervous system (see below).

It is doubtful whether, in intact animals, any of the CORTICOIDS can produce a significant increase in muscular strength. Overdosage with desoxycorticosterone may cause profound asthenia in patients, and hypokalemic paralysis in dogs and monkeys (see below). Severe overdosage with corticoid hormones may even produce foci of muscular degeneration in experimental animals.

Nervous System and Sense Organs.

— *Adrenalectomy* has no permanent direct effect upon the NERVOUS SYSTEM, although, perhaps partly through the resulting fatigue, depression and cachexia it diminishes the fighting instinct and libido of animals.

Adrenaline likewise fails to produce any clear-cut and specific direct effect upon peripheral nerves. It is useful in local anesthesia because, due to its local vasoconstrictor effect, it delays the absorption, and thus prolongs the action of admixed anesthetics (e.g., novocain). At the same time, it conveniently decreases bleeding. Certain *adrenaline derivatives* (e.g., benzedrine) are valuable central stimulants which temporarily combat mental fatigue, somewhat like caffeine does. Such stimulation is usually followed by a period of depression.

Corticoids elicit general anesthesia, if given in a manner to cause a rapid and pronounced increase in the hormone concentration of the circulating blood. In the event of chronic desoxycorticosterone overdosage, a periodic type of muscular paralysis ensues. This is accompanied by hypopotassemia and is curable by the administration of potassium. It bears a striking

resemblance to the so-called "familial paralysis" of clinical pathology. The occasional convulsive seizures of experimental animals, chronically treated with desoxycorticosterone, are usually due to the development of periarteritis nodosa in the brain, a lesion which may even cause fatal cerebral apoplexy.

In the EYE, adrenaline causes dilatation of the pupil, due to an excitation of the dilator muscle of the iris. This is of practical value in ophthalmology since adrenaline mydriasis facilitates ophthalmoscopic examinations and the readiness with which it is obtained has diagnostic significance in certain diseases of the eye. The effect is particularly pronounced following denervation of the iris by excision of the superior cervical ganglion. The hormone also causes protrusion of the eyeball in certain animal species. This is due to contraction of Mueller's muscles and retraction of the upper eyelid. The retraction of the nictitating membrane (e.g., in the cat) by adrenaline is likewise greatly increased by sympathetic denervation. The hormone may elicit lachrymation. Its purported value in the treatment of glaucoma is far from being reliable.

The marked vasoconstrictor effect of adrenaline, especially when locally applied to the mucous membranes of the NOSE is of practical value in combatting the nasal hyperemia of hay fever and rhinitis.

Digestive System. — Adrenaline inhibits both the tone and the peristalsis of the smooth musculature in STOMACH AND INTESTINES. This intestinal inhibition occurs *in vitro* even in concentrations of adrenaline as low as 1:400,000,000, and because of this great sensitivity it has been used as a basis for the bioassay of the hormone. (See : p. 100.) On the other hand, the pyloric, ileocolic and internal-anal sphincters are stimulated by adrenaline.

Adrenaline also stimulates SALIVARY SECRETION and (especially in the cat), elicits the production of a copious watery saliva. This is perhaps significant in connection with the emergency function of the gland, since in the cat, spitting (with an air of displeasure) is a normal defence reaction, calculated to inspire fear.

Adrenaline stimulates the GALL BLADDER musculature.

Adrenalectomy tends to cause some involution of the LIVER and PANCREAS; this may be accompanied by functional disturbances in these organs. The hormonal mechanism responsible for these actions has not been clarified, but to a large extent they can be explained on the basis of the malnutrition accompanying severe adrenal insufficiency.

Skin and Appendages. — Unlike in man, adrenalectomy usually causes no marked skin PIGMENTATION in experimental animals, although, under certain conditions, such effects have been seen.

Adrenaline contracts the melanophores of certain poikilothermic animals, for instance, the frog and horned toad. In the latter, melanophore contraction also occurs (presumably due to adrenaline liberation) when the animal is excited.

The CUTANEOUS VASOCONSTRICTION mentioned above, causes pallor of the skin following administration of adrenaline. This may also be regarded as a useful emergency reaction since it diminishes the danger of hemorrhage from possible surface wounds.

The ERECTOR MUSCLES of hair are also stimulated by adrenaline. This effect is responsible for the intimidating erection of the hair and feathers during emergencies in animals, as well as for the production of "goose flesh," in man.

In most mammals, including man, the SWEAT GLANDS are not excited by

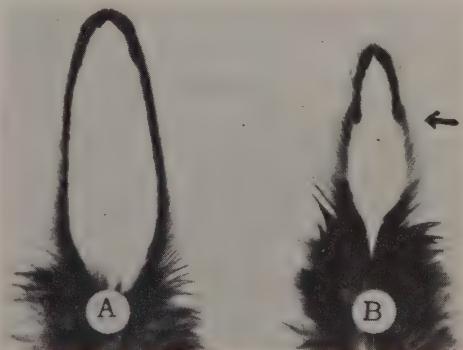
adrenaline, although they are innervated by the sympathetic nervous system. However, in certain species (e.g., horse, sheep), the hormone stimulates sweat secretion, and even in man, there is profuse sweating during the paroxysmal attacks caused by chromaffinomas.

In certain birds, adrenalectomy may cause serious disturbances in the development and especially in the pigmentation of the FEATHERS; in mammals, it leads to partial loss of HAIR. The hormonal mechanism responsible for these changes is not fully understood.

Urinary System. — ADRENALECTOMY causes no consistent morphologic changes in the urinary system, although it tends to produce a decrease in renal size and a diminution of urine production.

ADRENALINE increases diuresis due to a rise in the filtration pressure occasioned by a selective constriction of the efferent glomerular vessels. In very large doses, however, it causes anuria.

The smooth musculature of the urinary bladder is inhibited, but that of the trigonum and the ureters is excited by adrenaline.



Plumage changes after adrenalectomy. — A. Normal saddle hackles of a seabright bantam. — B. The part above the arrow grew immediately following sub-total adrenalectomy; after that regeneration occurred.



Nephrosclerosis produced by desoxycorticosterone in the cat. Compare granular surface of nephrosclerotic kidney of desoxycorticosterone-treated cat (top) with smooth surface of normal control kidney. The treated animal received 20 mg./day of desoxycorticosterone during six weeks and was sensitized by a high sodium diet.

CORTICOIDS (especially desoxycorticosterone) augment diuresis, probably through the inhibition of the tubular reabsorption of filtrate. Following chronic overdosage with desoxycorticosterone, marked glomerular lesions occur. These are similar to those of malignant nephrosclerosis in clinical medicine. There is hyalinization, especially of the afferent glomerular arterioles, and eventually complete hyalinization of the entire glomerular tuft. Increased permeability of the glomerular vessels to protein results and this leads to the formation of numerous hyaline casts, which occlude the lumina of the nephron and cause cystic dilatation of the segments prox-

imal to the casts. These changes are often accompanied by hypertension, pyelonephritis, nephritis and marked edema of the connective tissue surrounding the renal pelvis. Diets rich in Na aggravate these lesions, while a high intake of acidifying salts, carbohydrates and vitamins tend to prevent them.

Accessory Sex Organs. — ADRENALECTOMY causes some involution of the accessory genital organs in both sexes. Presumably this is partly due to lack of cortical steroid sex hormones, but it is difficult to estimate the extent to which it is merely the result of non-specific damage and the subsequent gonadal atrophy.

One of the most characteristic effects of ADRENALINE is the contraction of the pregnant or non-pregnant uterus in most animal species. In the cat, rat, mouse, guinea pig and man, the uterus is contracted by adrenaline only during pregnancy, while in the non-pregnant animal the hormone actually causes inhibition of the uterus. Thus, its actions differ from those of oxytocin. (See : Hypophysis.)

CORTICOIDS (e.g., desoxycorticosterone) possess both folliculoid and luteoid activities, the latter being, by far, more prominent. Thus, in the castrate female, suitably pretreated with folliculoids, the administration of desoxycorticosterone induces pregestational proliferation of the endometrium and even renders deciduoma formation possible.

None of the life-maintaining corticoids have yet been shown to exert any typical testoid effects. On the other hand, it will be kept in mind that pure testoids (e.g., dehydro-iso-androsterone, androsterone, adrenosterone) are also produced by the cells of the adrenal cortex. These latter compounds stimulate the growth of

male accessory sex organs, such as the seminal vesicles, prostate, epididymis, vas deferens and preputial glands of mammals, the comb of the capon, etc.

Various Other Effects. — REGENERATION AND WOUND HEALING are markedly impeded by adrenalectomy and restored towards normal by corticoids, but not by adrenaline.

The ESTRUS cycle is deranged and usually, permanent diestrus ensues following adrenalectomy in various animal species.

Adrenalectomy performed during PREGNANCY usually results in abortion, unless suitable corticoid therapy is instituted. Adrenaline is unable to

maintain gestation under such conditions.

Adrenalectomy causes almost immediate cessation of milk secretion, if performed during the period of LACTATION. Cortical hormones (but not adrenaline) may restore milk secretion, under such conditions. It has not been proven, although it is claimed by some, that a special cortical hormone, "cortilactin" is responsible for the maintenance of lactation. (See also p. 115.)

HIBERNATION may be temporarily interrupted by adrenaline injection in certain animals, perhaps because of the metabolism-stimulating effect of the hormone.

ADRENAL HORMONE CONTENT OF BODY FLUIDS AND TISSUES

ADRENALINE

It is still a debated question whether adrenaline is continuously secreted by the medulla or whether it is discharged only during emergencies. Since any type of stress causes a discharge of adrenaline, it is difficult to estimate its NORMAL BLOOD CONCENTRATION, because the excitement, incident to the determination, in itself causes adrenaline secretion in experimental animals. In any case, it is safe to conclude, that under resting conditions, the adrenaline concentration of the blood is so low (less than about 1 : 1,000,000,000), that it exerts no physiologic effects.

The emergency hyperadrenalinemia is due, firstly, to a discharge of hormone-saturated blood from the large venous sinuses of the medulla. The latter are evacuated during emergencies, owing to a relaxation of the venous sphincters. Secondly, the adrenaline granules stored in the chromaffin cells, are discharged and hence during stress the adrenaline content of the adrenal decreases parallel with the increase in blood adrenaline. This hyperadrenalinemia is characteristic of the first stage of the alarm reaction

and accounts for many of the manifestations seen in this initial phase of a general-adaptation-syndrome (hyperglycemia, loss of hepatic glycogen, transitory rise in blood pressure, tachycardia).

The ADRENALINE CONTENT OF THE RESTING ADRENAL is about 0.1% of its wet weight and the total hormone content of the two glands amounts to about 10 mg. in man. It is well to remember that the normal adrenal medulla actually contains more than the fatal dose (about 2 mg. intravenously or 8 mg. subcutaneously) of this hormone. This is especially noteworthy in connection with adrenal surgery, since mere massage of the gland is likely to cause a dangerous sudden discharge of adrenaline into the circulation.

REMOVAL OF THE VARIOUS ENDOCRINE GLANDS has comparatively little effect upon the adrenaline content of the blood and the adrenal medulla. This is particularly noteworthy as regards hypophsectomy. This operation causes a marked decline in the function of the adrenal cortex, but apparently remains without significant effect.

upon the hormone production of the medulla.

Similarly, TREATMENT WITH VARIOUS HORMONE PREPARATIONS exerts comparatively little effect upon the adrenaline content of the adrenals and the blood. Insulin represents a notable exception, since the hypoglycemia which it elicits causes a compensatory increase in the production of adrenaline. Oral administration of adrenaline itself results in the urinary elimination of inactive adrenaline sulfate but otherwise the hormone is not excreted by the kidney in appreciable quantities.

Unlike the function of most other endocrine glands, that of the adrenal medulla is primarily under NERVOUS control. Indeed, most stimuli, affecting the adrenaline production of the medulla, become inactive after denervation of the gland. Conversely, stimulation of the great splanchnics causes an immediate and pronounced adrenaline discharge, and a special center for adrenaline secretion appears to exist in the upper part of the floor of the fourth ventricle. It is through the intermediary of the preganglionic splanchnic nerves that impulses, originating from this center, reach the adrenal medulla, whose cells are excited by the liberation of acetylcholine at the nerve endings. It has been shown that injected acetylcholine stimulates adrenaline secretion, both in the intact and in the splanchnicotomized animal and that splanchnic stimulation elicits acetylcholine production in the medulla. Thus the nervous stimulus of splanchnic excitation is transmitted to the medullary cells through the same humoral (acetylcholine) mechanism by which preganglionic sympathetic fibers act upon peripheral ganglion cells. Indeed, even on embryologic grounds, the chromaffin elements may be regarded as modified peripheral sympathetic ganglia. (See p. 115.)

It is probable that there are higher centers of adrenaline secretion, even above that of the fourth ventricle. Stimulation of the hypothalamus and even emotional stimuli, originating in the cerebral cortex, can cause adrenaline secretion. It will be recalled that emotional hyperglycemia and tachycardia are mainly due to adrenaline discharge. Only to a small extent are these phenomena mediated by excitatory sympathin liberated at nerve endings. In experimental animals, the slight residual adrenergic manifestation which persists after ablation of the adrenal medulla, can be completely eliminated by denervation of the liver, the main source of excitatory sympathin.

An immediate loss of adrenaline from the chromaffin cells and a parallel rise in blood adrenaline is also induced within a few seconds by MUSCULAR EXERTION, COLD, VARIOUS ANESTHETICS, SURGICAL TRAUMA, HEMORRHAGE, BURNS, INTOXICATIONS, etc. It is most probable, however, that all these agents act merely by eliciting an ALARM REACTION, since none of them have been proven to act directly upon the medullary cells. During such emergencies, as following splanchnic stimulation, the adrenaline output of the medulla may rise, from a negligible amount, to as much as 0.004 mg./Kg. body weight/minute. But all these non-specific damaging agents fail to cause adrenaline liberation, following denervation of the gland.

CORTICOIDS

No accurate data are as yet available concerning the corticoid hormone content of the BLOOD and TISSUES, but bioassays indicate that normally the corticoid potency of peripheral blood is negligibly small. On the other hand, very large quantities of corticoids can be demonstrated in the venous blood

of the adrenal itself. According to one author, the average output of one suprarenal gland/minute/Kg. body weight, is equivalent to the activity that can be extracted from 0.6 gm. of ADRENAL TISSUE. Under certain conditions of stress, the corticoid activity of 1 cc. of suprarenal plasma may be 10 times as high as that extractable from 1 gm. of gland.

Noteworthy quantities of corticoid activity can also be extracted from the CORPUS LUTEUM, presumably because progesterone possesses corticoid potency. Other tissues have not been shown to possess any noteworthy degree of such activity.

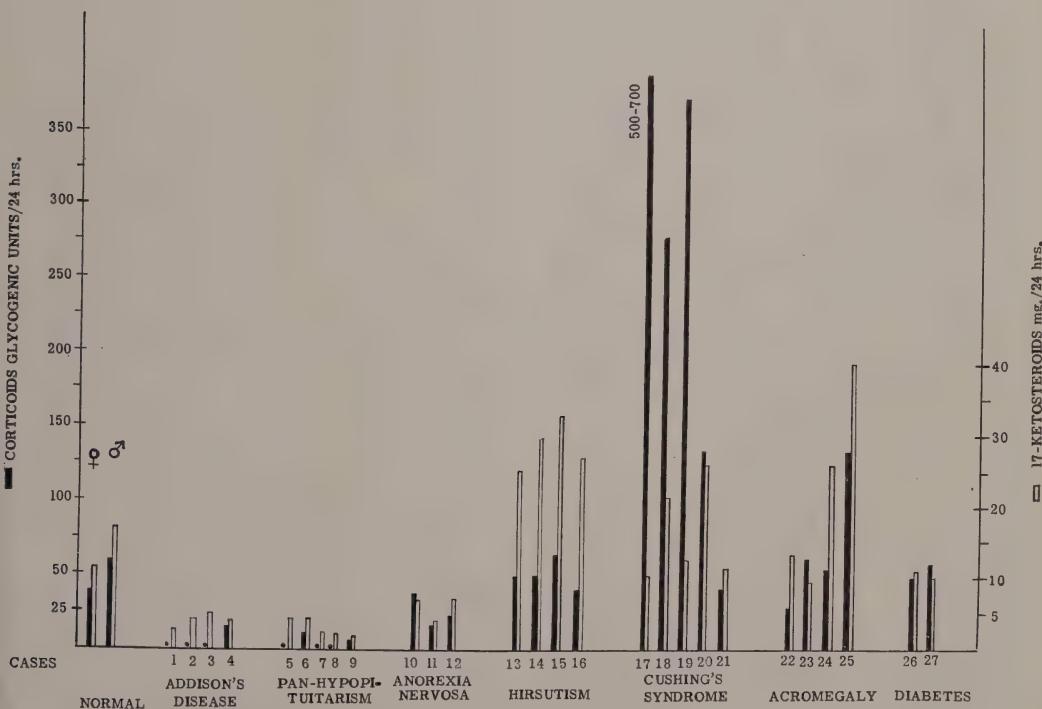
Normal human URINE contains just detectable quantities of corticoid material.

HYPOPHYSECTOMY causes a pronounced diminution of the corticoid

hormone content of the adrenals and SIMMONDS' disease diminishes, while CUSHING'S DISEASE increases the output of active corticoids through the urine. These observations are readily understandable if we remember that the function of the adrenal cortex is almost entirely under the control of hypophyseal corticotrophin. Active ADRENAL-CORTICAL TUMORS likewise raise the urinary corticoid titer. (For additional data see sections : The Steroids, Hypercorticoidism, Cushing's Disease, etc.)

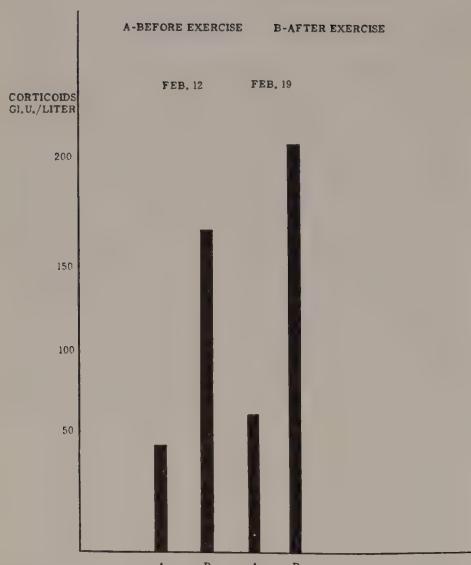
Adrenal denervation, or stimulation of the adrenal NERVES, causes no change in corticoid hormone production. This indicates, that unlike the medulla, the cortex is not markedly influenced by nervous stimuli.

Urinary elimination of corticoids rises significantly in patients a few



Excretion of gluco-corticoids and of 17-KS in various endocrine and other disorders

(Courtesy of Drs. E.-H. Venning and J.-S.-L. Browne.)



Effect of strenuous exercise on the excretion of corticoids in a group of 14 army recruits.

(After E.-H. Vennig and V. Kazmin:
Endocrinology 39, 131, 1946.)

hours after traumatic injuries, acute muscular exertion, burns, acute infections, etc., all of these, are conditions capable of eliciting an ALARM REACTION. The bulk of the corticoids produced are probably eliminated in the urine, after transformation into inactive metabolites. Hence, bioassays for the corticoid activity of urine can give, at best, only an approximation of the total quantity of cortical steroid production.

From the above data it appears that both adrenaline and corticoids are produced in large quantities under the influence of essentially the same types of non-specific damaging agents, the former somewhat more rapidly than the latter.

STIMULI INFLUENCING ADRENAL STRUCTURE

Extirpation of Endocrine Glands.

— PARTIAL ADRENALECTOMY induces marked compensatory hypertrophy of the remaining cortical cells, without any comparable enlargement of the residual medullary elements. This is perhaps due to the fact that only cortical cells are capable of producing significant amounts of corticoids, while sympathetic nerve endings and paranglia may substitute to some extent for the partial loss of chromaffin cells, even without compensatory proliferation of the medullary remnant. — It is well to remember that very minute cortical islets suffice to maintain adrenalectomized animals alive, and the compensatory hypertrophy of hardly visible cortical cell islets, or ectopic adrenal cortices, may explain the survival of supposedly completely adrenalectomized animals. The same applies to patients having destructive cortical lesions.

Treatment with large doses of glucocorticoid hormones pre-

vents compensatory hypertrophy after partial adrenalectomy.

HYPOPHYSECTOMY causes atrophy of the adrenal cortex, but does not influence the structure of the medulla significantly. In hypophysectomized animals, compensatory hypertrophy of cortical remnants does not occur after partial ablation of the gland, nor is there any cortical enlargement during the general-adaptation-syndrome. On the other hand, adrenocorticotrophic extracts of the anterior-pituitary stimulate the adrenal cortex, even in the hypophysectomized animal. It has been assumed therefore that the phenomena of "compensatory" and "damage hypertrophy" of the adrenal cortex are due to increased secretion of adrenocorticotrophic hormones, by the anterior lobe cells of partially adrenalectomized or damaged animals.

Important changes in the adrenal cortex of certain species, especially certain strains of mice, are induced by GONADECTOMY. In these strains the zona

reticularis, here designated as the "X-zone," is especially prominent and dark in adult females, but absent in males. Spaying (ovariectomy) has no effect, but castration (testis extirpation) causes the appearance of the X-zone in males, which normally do not possess it. The X-zone may be the source of adrenal testoids and the presence of a functional testis may cause its compensatory involution. The development of this zone in castrates could then be viewed as a compensatory effort to maintain some testoid hormone production. This view — though not proven — receives further support from the observation that administration of purified testoids (e.g., testosterone) causes the disappearance of the X-zone in castrate males, presumably owing to a phenomenon of compensatory involution.

In some animal species adrenal-cortical cancers develop following gonadectomy (e.g., mouse, guinea pig).

REMOVAL OF OTHER ENDOCRINE GLANDS exerts no important effect upon adrenal structure, outside of that which could be explained on the basis of the general-adaptation-syndrome elicited by such operations.

Hormones. — The ADRENOTROPHIC effect of pituitary extracts appears to be due entirely to the adrenotrophic hormone (or hormones) of the anterior lobe cells. Only extracts of the anterior lobe cause adrenal-cortical enlargement and hyperfunction, while those of the middle and posterior lobe have no such effect.

ADRENALINE causes no very striking compensatory atrophy of the adrenal medulla but, as mentioned above, both gluco- and mineralo-CORTICOIDS produce marked compensatory involution of the adrenal-cortical cells.

INSULIN causes degranulation of the chromaffin cells, since the hypoglycemia which it produces calls forth a compensatory secretion of adrenaline, to normalize the blood sugar level.

FOLLICULOIDS elicit a pronounced and rather specific type of cortical hypertrophy, much greater than could be accounted for by their non-specific damaging action.

TESTOIDS tend to decrease the size of the adrenal cortex, in all species so far examined. As previously stated, in certain strains of mice, the testoids prove particularly potent in causing the disappearance of the "X-zone" in females and castrate males, in which it is normally prominent. In other species (e.g., rat) testoids cause a peculiar type of vacuolization and atrophy of the adrenal cortex.

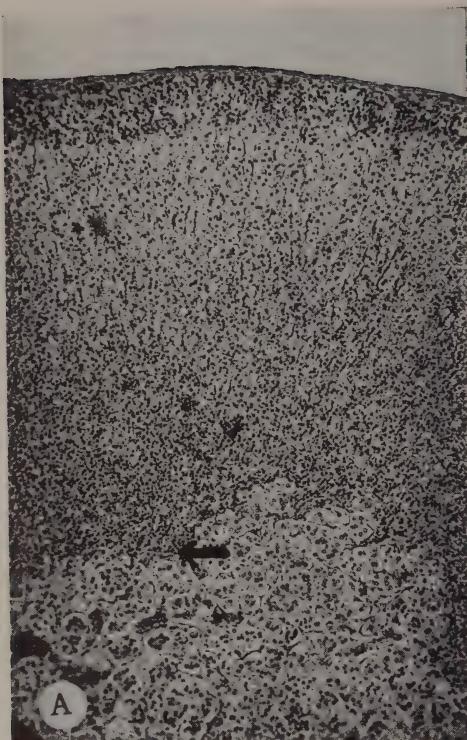
OTHER ENDOCRINE PREPARATIONS do not appear to have a very specific effect upon the adrenal, but tend to cause hypertrophy of the cortex in proportion to other manifestations of the general-adaptation-syndrome, which they elicit. It is not surprising, therefore, that heavy overdosage with *thyroxin*, which induces serious metabolic disturbances, is particularly effective in causing cortical hypertrophy.

None of these hormone preparations (with the possible exception of folliculoids), are effective in hypophysectomized animals and hence it is generally agreed that their effect is mediated by the anterior-hypophysis.

Diseases. — As may be expected, HYPOPITUITARISM causes involution, while HYPERPITUITARISM usually induces hyperplasia and hypertrophy of the adrenal cortex.

The adrenal lesions associated with various clinical forms of HYPO- and HYPERADRENALISM are discussed in the sections devoted to the latter.

In certain very acute INFECTIONS the adrenal stimulation is so pronounced that hemorrhages and necroses ensue in the suprarenals, with a secondary breakdown of their function. — *Waterhouse-Friedrichsen's syndrome*, usually elicited by acute meningococcic septicemia, is a typical case in point.



A



B



C



D



E



F

Effect of hypophysectomy and of various steroid hormones upon adrenal cortex. — A. Adrenal of a normal 3-month-old male rat. Note normal width of glomerulosa, fasciculata and reticularis zones. In this and following figures cortico-medullary junction is marked by arrow (adrenal weight 44 mg.). — B. Adrenal of similar rat 28 days after hypophysectomy. Note profound atrophy of the cortex, which affects all layers, but especially the fasciculata and reticularis. The medulla remains normal (adrenal weight 15 mg.). — C. Adrenal of similar rat receiving increasing doses (0.5 to 3 mg./day) of desoxycorticosterone acetate during 3 months. Note pronounced involution of cortex, while medullary cells remain normal (adrenal weight 23 mg.). — D. Adrenal of similar rat treated with estradiol (100 γ /day during 9 months). Note enlargement of entire cortex which takes up the whole visual field. There is marked hyperemia with great dilatation of cortical sinusoids. The glomerulosa, however, is inconspicuous (adrenal weight 122 mg.). — E. Adrenal of similar rat treated with small doses of testosterone (implantation of one 14 mg. pellet of methyl-testosterone one month prior to autopsy). Note extreme density of glomerulosa zone (adrenal weight 30 mg.). — F. Adrenal of similar rat receiving large doses of a testoid (10 mg. of methyl-testosterone/day during 3 months). Note density of glomerulosa and atrophy of cortex. The latter is studded with vacuolated "signet-ring cells" which are very characteristic of rats chronically overdosed with large amounts of testoids (adrenal weight 28 mg.).

OTHER DISEASES rarely cause any striking or specific changes in the adrenals, except the cortical hyperplasia and hypertrophy, which are almost constant accompaniments of any disease. (See : General-Adaptation-Syndrome.)

Diet. — Undernutrition, and especially deficiency in ascorbic acid or the vitamin-B complex, cause pronounced

enlargement of the adrenal cortex. Diets rich in protein are most, and diets high in carbohydrates least favorable for the development of cortical enlargement, during the general-adaptation-syndrome.

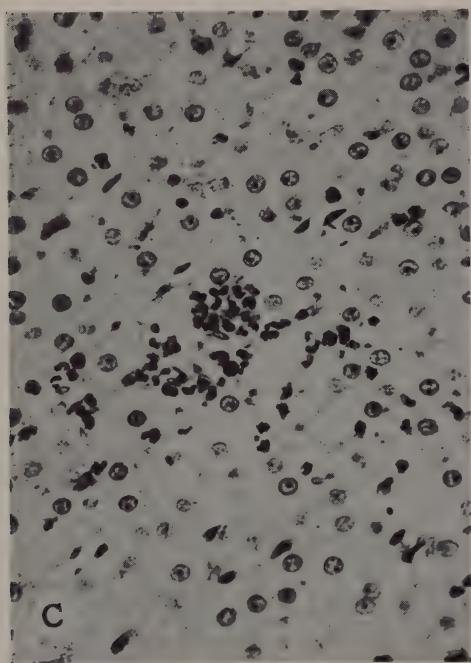
The ascorbic acid and cholesterol content of the adrenal cortex, partly depend upon the dietary intake of ascorbic acid and cholesterol respective-



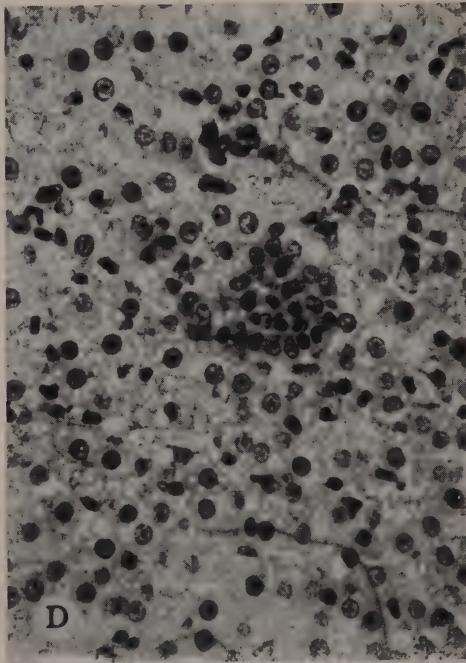
A



B



C



D

Effect of anterior-lobe extracts upon the adrenals. — A. Adrenal of an adult male rat receiving 20 mg. of lyophilized anterior-pituitary tissue daily during 30 days. The animal was sensitized to the corticotrophic action of the extract by castration, unilateral nephrectomy and a high-

ly. It is noteworthy that in the guinea pig, which is particularly sensitive to ascorbic acid deficiency, this avitaminosis elicits not only an almost complete loss of adrenal ascorbic acid but also severe degenerative and hemorrhagic lesions in the suprarenals.

Nervous Stimuli. — Denervation of the adrenals, or even mere transection of the splanchnic nerves, prevents the loss of chromaffin granules, otherwise occurring in the alarm reaction elicited by diverse non-specific agents. On the other hand, injection of acetylcholine, causes loss of chromaffinity, even following denervation, since this substance is the direct humoral transmitter of secretory stimuli to the adrenal medulla.

The appearance of the adrenal cortex, and its response to hormonal or noxious stimuli, remain practically unaltered by denervation of the gland. Stimulation of the adrenal nerves also fails to elicit any characteristic morphologic change in the cortex.

Age. — Soon after birth, the human adrenal cortex undergoes a process of so-called "*physiologic degeneration*." This is characterized by a great decrease in cortical width and the appearance of deep convolutions on the surface of the gland. The process begins during the first days after birth and is usually terminated by the end of the first to third month. In three-month-old babies, the adrenals weigh only about half as much as in the newborn.

Sex. — In general, the adrenals of female animals are considerably larger than those of males. The sexual di-

morphism of the mouse adrenal (the so-called "X-zone") has been mentioned in connection with the results of gonadectomy. (See p. 130.)

Estrus, Pregnancy and Lactation.

— Estrus, pregnancy and lactation exert no significant influence upon the adrenal medulla, but the cortex is somewhat enlarged during all these conditions.

Seasonal Changes and Hibernation. — In hibernating animals, both the cortex and the medulla undergo atrophy and there may be almost complete loss of chromaffinity in the medullary cells, during the winter season.

Other Conditions. — ANAPHYLAXIS, REDUCED ATMOSPHERIC PRESSURE, EXTREME HEAT OR COLD, SEVERE MUSCULAR WORK, HEMORRHAGE, BURNS, TRAUMATIC SHOCK AND A LARGE VARIETY OF DRUGS cause loss of chromaffinity from the adrenal medulla, loss of lipid and ascorbic acid granules with hypertrophy and hyperplasia of the adrenal cortical cells, that is, histologic signs of increased adrenaline and corticoid hormone secretion. All these phenomena run closely parallel with the general damaging effects of these agents and must be regarded as part of the general-adaptation-syndrome which they elicit. Their effect on the medulla is mediated by the sympathetic and prevented by adrenal denervation or splanchnicotomy; their influence upon the cortex is mediated by the anterior lobe and prevented by hypophysectomy.

sodium diet. Note great enlargement of the cortical cells, dilatation of the sinusoids and partial necrosis of the cortex (arrows) which is presumably due to adrenal periarteritis nodosa (adrenal weight 283 mg.). — B. Adrenal of a similarly treated rat. Here, the necrosis destroyed both adrenals almost completely, except for a thin subcapsular rim of cortical cells (arrows). These pictures show that anterior-pituitary extracts can not only stimulate the adrenal cortex, but, in the event of overstimulation (perhaps by producing adrenal periarteritis nodosa) cause necrosis with subsequent hypocorticism. — C. and D. High magnification of a region from the adrenal cortex of a similarly treated rat. Note "round-cell infiltration islets" consisting of small lymphocytes and polymorphonuclear leukocytes. There appear to occur transitional stages between these blood cells and the epithelial elements of the cortical cell columns. Similar infiltration islets are seen in rats bearing large, necrotic tumors or those injected with decomposing protein material.

Rays. — The adrenal glands (both normal and neoplastic) are comparatively resistant to the damaging effect of X-RAYS and, it has even been claimed that in small doses, X-ray treatment

may stimulate the function of adrenal cells. Heavy overdosage with X-rays causes degenerative changes, and even necrosis, both in the cortex and in the medulla.

DISEASES OF THE ADRENALS

MALFORMATIONS

It is doubtful whether complete APLASIA of the adrenals ever occurs. Supposedly pertinent cases have been reported in the literature but in these the possibility of a secondary destruction of the adrenals, during postnatal or even fetal life, must always be considered. It will also be kept in mind that complete aplasia of the adrenals is incompatible with the maintenance of postnatal life and could, therefore, never be observed in an adult.

Isolated aplasia of the adrenal medulla would theoretically be compatible with the maintenance of life, but allegedly relevant instances are also more probably due to secondary destruction by disease.

As with several other endocrine glands, we have no means of identifying a primary HYPOPLASIA of the adrenal cortex. The postnatal condition of the cortex is so largely under pituitary control, that any instance of insufficient development raises the suspicion of a secondary atrophy, due to anterior pituitary failure, from which it could not be distinguished on morphologic grounds. The literature contains descriptions of so-called cortical hypoplasia in new-born infants with *anencephaly*, *hydrocephalus* and other malformations of the brain, but here a concomitant primary failure in the development of the anterior lobe appears to give the most satisfactory explanation of the deficient adrenal-cortical development.

Similarly, cases of so-called cortical aplasia in one gland of a patient with a *contralateral cortical tumor*, are usual-

ly due to compensatory cortical atrophy, rather than true malformation.

In *status thymico-lymphaticus*, hypoplasia of both cortex and medulla have repeatedly been described, without there being any definite proof that the subnormal suprarenals are malformed, rather than involuted.

As far as we know, HYPERPLASIA of the adrenal cortex is always due to excess adrenotrophin secretion. It almost invariably gives rise to clinical manifestations of corticoid hormone overproduction and will be discussed in the corresponding section on hypercorticoidism.

Diffuse primary hyperplasia of the medulla has never been proven to occur, although adenomas and other tumors of the chromaffin cells are comparatively common.

ACCESSORY ADRENAL CORTICES are the most common malformations of the gland both in animals and in man. Their chief importance lies in the fact that they prevent the appearance of deficiency symptoms after complete destruction or surgical removal of the main glands. They are particularly common in some strains of rabbits, mice and rats, but much less frequent in the cat, dog, monkey and guinea pig. In man, accessory cortices tend to occur in the vicinity of the spermatic arteries, the testis and epididymis, the ovaries and their ligaments, the hepato-duodenal ligament, the mesentery and the kidney. Apparently, many accessory cortices disappear during postnatal life, since such structures are much more common in the testis region of boys, than in adults.

Extra-adrenal chromaffin tissue is so widely distributed in the normal organism that it would be difficult to identify ACCESSORY ADRENAL MEDULLAE, even if they did occur.

TRUE ACCESSORY ADRENALS, containing both cortical and medullary tissue, are extremely rare. They may be formed by the early embryonic separation of a portion from the main adrenal primordium or through the invasion of separate nodules of interrenal cortical tissue by adjacent sympathogonia.

Among OTHER MALFORMATIONS of the adrenals, only the very rare "butterfly adrenal" is worth mentioning. It results from the development of a connecting bridge between the two adrenals.

The diagnosis of adrenal malformations is hardly ever possible in the living patient, unless they are accidentally noted on the occasion of a laparotomy.

Clinical manifestations are rare, but if they occur, they are identical with hypo- or hyperadrenalinism due to other causes and must be treated in the same manner. (See : Hypocorticoidism, Hypercorticoidism.)

VASCULAR DISTURBANCES

HEMORRHAGES into the adrenal parenchyma are frequently observed in new-born infants, especially in premature babies. They are rarely sufficiently extensive, however, to cause severe insufficiency symptoms.

Small patches of necrosis and hemorrhage are common in the adrenals of experimental animals and man exposed to various types of acute non-specific damage, conducive to an alarm reaction (e.g., Waterhouse-Friedrichsen syndrome).

THROMBOSES of the adrenal veins sometimes occur in acute infections and intoxications. They may be bilateral and conducive to "apoplexy of the adrenals," with symptoms of acute fatal insufficiency.

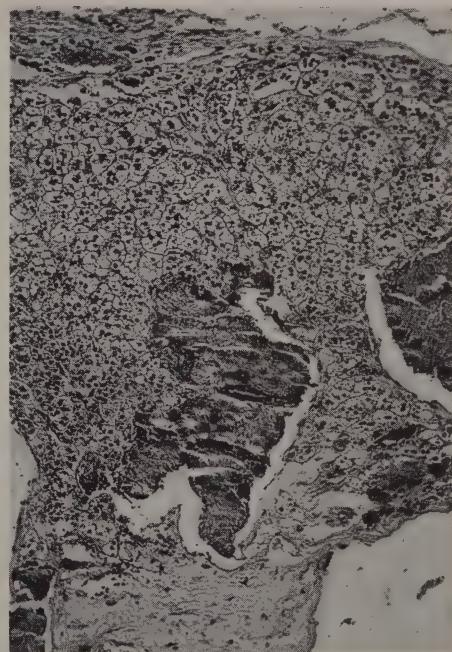
DEGENERATIONS

CALCIFICATION of the adrenals, sometimes accompanied by OSSIFICATION, may occur as sequelæ of local tuberculosis, degeneration and tissue necrosis.

AMYLOID degeneration of the adrenals is rarely pronounced except as part of generalized amyloidosis. It is sometimes associated with adrenal (and renal) vein thrombosis, but seldom with hypocorticoidism.

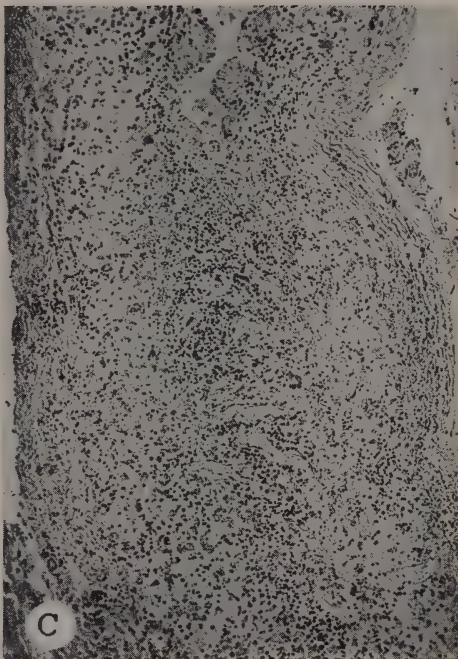
INFLAMMATIONS

ACUTE INFLAMMATION of the adrenals is rare, although, in generalized septicemia, foci of infection may become localized in one or both suprarenal glands.



Calcification and ossification of the adrenal. Note calcium deposit (dark) and beginning bone formation (light) among adrenal-cortical cell columns.

The so-called "PRIMARY CONTRACTED ADRENAL" is probably a special type of chronic inflammation, characterized by round cell infiltration and connective tissue stroma proliferation. It causes gradual destruction of the gland with addisonian symptoms.



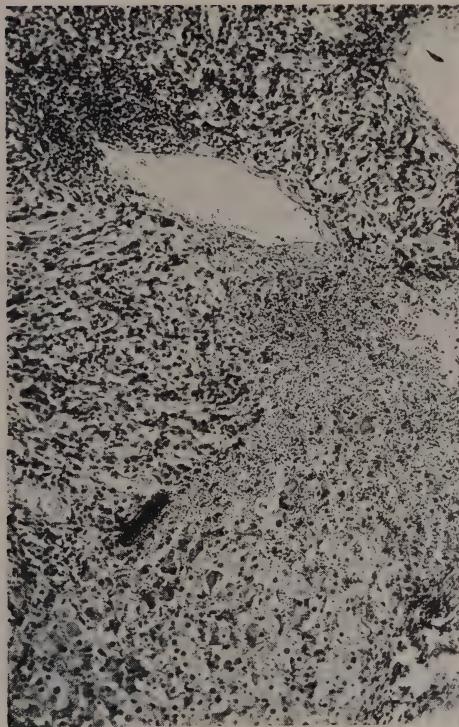
Primary contracted adrenal in Addison's Disease. (Courtesy of Dr. T. Waugh.) — A. Cross-section through normal human adrenal (low magn.). — B. Cross-section through primary contracted adrenal in Addison's disease (same magn. as A.). — C. Higher magnification of adrenal shown in fig. B. Note almost complete replacement of cortex and medulla by connective tissue and lymphocytes.



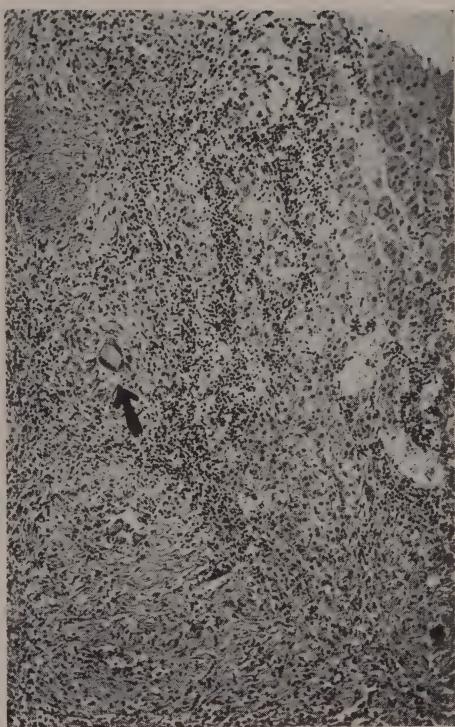
Amyloidosis of the adrenals. Hormogeneous amyloid deposits in adrenal cortex. Endocrine cells are compressed and atrophic.



Primary contracted adrenal. (Courtesy of Dr. W. Boyd.) Note that adrenal parenchyme is almost completely replaced by connective tissue and lymphocytic infiltration. The patient suffered from Addison's disease.



Primary contracted adrenal. So-called "primary contracted adrenal" with cortical atrophy in a patient who died from Addison's disease. Note intense lymphocytic infiltration especially around the large vein (top) while remaining cortical cells (bottom) are enlarged, due to compensatory hypertrophy. Small dots between cortical cells are erythrocytes, due to bleeding into hyperactive, hyperemic cortical remnant.



Tuberculosis of the adrenals. Caseous tuberculosis of the adrenals. Only few cortical cells (right) are still distinguishable. Note also marked lymphocytic infiltration and poly-nuclear giant-cell (arrow). Patient died from Addison's disease.



Adrenal tuberculosis. Note polynuclear giant cells (arrows), lymphocytic infiltration, epithelioid cells and caseation. The remaining cortical cells (top) appear to be enlarged, perhaps due to compensatory hypertrophy. The patient suffered from Addison's disease.

The most common inflammatory lesion in the adrenal is TUBERCULOSIS, which is frequently bilateral and conducive to Addison's disease. It may, or may not, be accompanied by widespread tuberculous lesions in other organs, and is, of course, always of hematogenous origin. It is not known why tuberculosis has such a predilection for localization in the suprarenales.

SYPHILIS likewise exhibits a special affinity for the adrenals. *Congenital syphilis*, tends to produce miliary necroses. Spirochaetes are very regularly present, in the adrenals of syphilitic

new-born, even if anatomic lesions are not observed.

Acquired syphilis may lead to diffuse gumma formation or to more or less generalized sclerosis of the adrenals, either of which may be the cause of Addison's disease. In certain cases, antisypilitic therapy has markedly improved the condition of addisonians whose endocrine deficiency was due to adrenal syphilis. It is also noteworthy that tertiary syphilis quite frequently causes amyloid degeneration of the adrenals.

HYPOCORTICOIDISM (ADDISON'S DISEASE)

(SYNOMYS : Morbus Addisonii, adreno-cortical deficiency, hypoadrenalinism, hypoadrenia, adrenal deficiency.)

DEFINITION

Addison's disease is a condition in which the hormone production of the adrenal cortex is sufficiently diminished to cause detectable deficiency manifestations. Although Addison described only one type of this disease, it is customary to use the term "Addison's disease" as synonymous with hypocorticoidism.

The condition is often designated simply as "adrenal deficiency," since hypofunction of the medulla hardly ever produces any manifestations of insufficiency, yet the term "hypocorticoidism" is more precise.

CLASSIFICATION

The clinical types of Addison's disease may be classified according to various points of view.

According to the MAIN MANIFESTATIONS, we may distinguish :

(A) ACUTE FORMS

(1) *The sudden, unexpected death of adrenal failure* usually appears without any warning, in patients in whom there was never any cause to suspect

adrenal insufficiency. Death may occur with epileptiform or angina pectoris-like symptoms. A slight trauma, child-birth, exhaustion or some other, often quite trivial, damage may be fatal. At autopsy, one finds almost complete destruction of the adrenals by disease or mere "adrenal hypoplasia." Most of these cases are closely related to the so-called "status thymico-lymphaticus."

(2) The *pseudoperitonitic form* is the most common acute type. In its symptomatology, it resembles acute appendicitis, peritonitis or the rupture of an ectopic pregnancy. There is intense abdominal pain, meteorism, facies peritonitica, vomiting, hiccup and a very rapid and weak pulse. Death ensues a few days, or even hours, after the onset of the symptoms. In such cases, autopsy often reveals complete destruction of the adrenals due to thrombosis or massive hemorrhage.

(3) The *choleriform or gastrointestinal type* resembles acute food poisoning or gastrointestinal septicemia. There is vomiting, persistent diarrhea, cold sweats, anuria, hypothermia and death ensues within a short time.

(4) The *apoplectiform type* simulates a stroke. The patient suddenly collapses and enters a deep coma, which

is but occasionally interrupted by convulsive spells or deliria. This form is also rapidly fatal and often due to massive bilateral adrenal hemorrhage.

(5) The *meningo-encephalitic form* imitates acute encephalitis or meningitis, particularly tuberculous meningitis, because the sugar in the spinal fluid may be low in either case.

(6) The *myocardial form*, described by the French school as "asystolie surénale," in which a fall in blood pressure and cardiac failure, often combined with arrhythmia, lead to death from cardiovascular failure, without any prominent anatomic change in the myocardium.

These acute forms are much less common than the chronic types of Addison's disease and there is usually considerable overlapping between the various types.

(B) CHRONIC FORMS

(1) *Typical chronic Addison's disease* is by far the most common type. Its main symptoms are: asthenia with dizziness and syncopal attacks, hypotension with decrease in blood volume and circulatory failure, loss of weight, pigmentation of the skin and mucous membranes, gastrointestinal disturbances with anorexia, nausea, diarrhea, hiccups and vomiting, a tendency towards hypoglycemia and great sensitivity to various types of strain (e.g., cold, muscular fatigue, intoxications and infections).

(2) The *chronic, nervous or solar type*, in which crises of solar neuralgia, with nausea and vomiting, alternate with periods of severe psychic disturbances, such as melancholia or spells of acute delirium with maniacal excitement. Occasionally, epileptiform convulsions are associated with a solar crisis. All these nervous disturbances are combined with typical signs of adrenal insufficiency, such as asthenia, pigmentation of the skin and mucous membranes.

In these chronic cases, the eliciting cause is about as frequently caseous adrenal tuberculosis as "primary contracted adrenal." Other etiologic lesions are rare.

According to the INTENSITY of the insufficiency syndrome, it is customary to recognize:

(1) *Mild cases* (also known as constitutional addisonism, temporary addisonism or "formes frustes"), which are generally due to a merely functional disturbance. It is debatable whether these should be included in the concept of Addison's disease.

(2) *Severe Addison's disease* which is the common result of extensive cortical destruction.

Obviously, it is also possible to classify pertinent cases according to their ETIOLOGY, thus distinguishing syphilitic, tuberculous, carcinomatous, etc., types of hypocorticoidism, since the various classifications overlap.

PATHOLOGIC ANATOMY

Addison's disease may be produced by a variety of adrenal lesions:

(1) "The PRIMARY CONTRACTED ADRENAL," as mentioned above, is the result of chronic inflammatory and degenerative lesions of unknown etiology. Usually both suprarenals are affected and after a period of connective tissue stroma proliferation and round cell infiltration, cortical atrophy ensues, due to extensive sclerosis of the parenchyma. It is often seen as a result of chronic intoxications and infections and may be related to the so-called "stage of exhaustion" of the general-adaptation-syndrome.

(2) *TUBERCULOSIS* of the adrenals used to be the most common anatomic change seen in patients with Addison's disease, but in recent years, adrenal atrophy is increasingly more often encountered. In the case of tuberculosis, the parenchyma is more or less completely destroyed by the granuloma and the caseous necrosis. It must be kept

in mind that usually, compensatory cortical hypertrophy, hyperplasia and adenoma formation tend to proceed simultaneously with the adrenal destruction. Hence, manifestations of hypocorticism remain latent until the regenerative power of the cortex is exhausted or a final hemorrhage destroys the residue of functional cortical tissue.

(3) SYPHILIS of the adrenals tends to produce Addison's disease only if widespread lesions, usually gummas, destroy most of the cortical parenchyme. This form is now comparatively rare, since tertiary syphilis has become less frequent, due to the advances in antisyphilitic therapy.

(4) Bilateral, non-functional, primary or, more often, secondary adrenal TUMORS cause Addison's disease, if they destroy the major part of the cortical tissue.

(5) THROMBOSIS of the adrenal blood vessels produces rapid destruction of the gland, which usually leads to the most acute types of Addison's disease.

(6) Adrenal AMYLOIDOSIS is rarely the cause of hypocorticism.

(7) TRAUMATIC INJURIES of the adrenal region are also very exceptional causes of hormonal insufficiency.

(8) POLYGLANDULAR INSUFFICIENCY may affect the adrenal cortex and cause functional failure, but it is very probable that most of the allegedly relevant cases, actually represent PITUITARY INSUFFICIENCY syndromes. It is noteworthy that, even severe pituitary failure rarely elicits the typical syndrome of Addison's disease with gastrointestinal crises, hyperpigmentation, etc., although a few pertinent cases have been authenticated by autopsy. Perhaps the simultaneous elimination of other metabolic functions is responsible for the fact that the hypopituitary patient (especially in the younger age groups) does comparatively well in

spite of cortical hypofunction. It is almost certain, furthermore, that some cortical activity persists, even in patients with complete anterior-lobe failure.

(9) PRIMARY HYPOPLASIA of the adrenal cortex has been held responsible for certain types of alleged cortical insufficiency, known as "status thymico-lymphaticus." However, the very existence of this latter syndrome is somewhat in doubt.

(10) REPLACEMENT OF CORTICOID-PRODUCING, BY TESTOID-SECRETING TISSUE is another possible, though uncommon, cause of adrenal insufficiency.

INCIDENCE

Although exact figures are difficult to obtain, the general incidence of frank Addison's disease is very low, according to the U. S. Bureau of Statistics : 0.3 per 100,000 and about 16 cases per 100,000 admissions in the Mayo Clinic. On the other hand, the various forms of "addisonism" are fairly common complications of infections, intoxications and other exhausting diseases.

The disease may occur at any AGE, but is probably most common during the third decade. Children are comparatively rarely affected and in them, the disease tends to take a chronic, benign course with predominance of gastrointestinal symptoms and pigmentation. Sometimes, however, acute forms of Addison's disease develop in the very young, as a result of adrenal hemorrhages, to which infants seem to be especially predisposed.

Addison's disease also occurs in very old patients, in whom the diagnosis is often difficult, because of the great resemblance of hypocorticism to senile marasmus.

The male SEX is more frequently affected by frank Addison's disease, especially among patients less than 45 years of age. Only the hypocorticoid-

ism due to primary pituitary failure is more common in women, who are generally more susceptible to hypophyseal disease. In large statistical series, in which the various types are considered conjointly, males usually predominate over females in the ratio of 2 to 1.

Temporary addisonism is frequently seen in the course of PREGNANCY, apparently because the corticoid hormone requirements rise during gestation. This also explains why, if an addisonian woman becomes pregnant (which is rarely the case), her condition tends to become more severe and artificial interruption of gestation may become necessary. The rare cases, in which an improvement was noted during the late stages of gestation, are explained by assuming that the fetal cortex gradually begins to compensate for the insufficiency of the mother's adrenal.

It has been claimed that RACIAL FACTORS are of importance and that Addison's disease is most common in highly pigmented southern populations (e.g., Spaniards), but this has not been confirmed; the malady is not infrequent among blond Scandinavians. On the other hand, it has also been stated, that the disease is rare in the negro, yet many authentic cases have been described as occurring in colored people. It is probable that Addison's disease is often not recognized in the negro patient because the most striking symptom, the pigmentation, is difficult to judge. Conversely, in the less markedly pigmented southern Europeans, the comparative darkness of the skin awakens a suspicion of Addison's disease, whenever the patient becomes debilitated from unknown causes. Definite proof of an important hereditary factor, in the pathogenesis of Addison's disease, is lacking.

PATHOGENESIS

Classical Addison's disease is due to a PRIMARY destruction of the adrenal cortex, by a local, directly acting, pa-

thogenic agent ("primary contracted adrenal," tuberculosis, syphilis, carcinoma, hemorrhage, etc.), within the suprarenal gland itself. Hypocorticoidism may, however, also be caused by SECONDARY adrenal-cortical involution, due to anterior-pituitary failure and perhaps even by excessive stimulation of the adrenals through adrenotrophic hormones, which eventually lead to exhaustion atrophy of the cortical cells. The most common causes of relative (in proportion to the requirements) or "functional" hypocorticoidism are cachexia, regional ileitis, ulcerative colitis, sprue and malaria.

The morphology of the adrenal changes, occurring in Addison's disease, have been discussed in the chapter on Pathologic Anatomy. The pathogenesis of the individual manifestations of adrenal-cortical insufficiency will be considered in the next chapter, entitled "Clinical Course," where these manifestations are described.

CLINICAL COURSE

State. — In typical cases, the course of Addison's disease is very characteristic. There is, firstly, an extraordinary debility and languor, usually following upon some infection of the upper respiratory tract; secondly, the full syndrome of chronic Addison's disease (see below) and thirdly, decrease in blood volume and fatal circulatory collapse ensue during a crisis.

The leading symptoms and signs of the fully developed syndrome are :

- (1) Gastrointestinal disturbances with loss of appetite, nausea with vomiting (almost a "sine qua non" in the diagnosis).
- (2) Loss of weight.
- (3) A typical brownish pigmentation of the skin and mucous membranes.
- (4) Marked muscular weakness with easy fatigability.
- (5) Arterial hypotension with spells of dizziness.

- (6) Decrease in blood volume.
- (7) Hypoglycemic attacks.
- (8) Disturbances in electrolyte metabolism.

(9) A great decrease in general resistance to any type of stress (e.g., intercurrent infections, intoxications, trauma, excessive muscular fatigue, extremes of temperature, malnutrition, administration of drugs, especially thyroid hormone, insulin, drastic laxatives, barbiturates, morphine).

It is important to remember that resistance and adaptation to stress, require much more cortical hormone than the mere maintenance of life. Hence, if addisonians are exposed to strain, a so-called "addisonian crisis" may be elicited and this is usually fatal, unless drastic therapeutic measures are rapidly introduced.

During the crisis, there is a severe aggravation of the symptoms and signs, especially of the muscular asthenia (which renders the patient bedridden), the gastrointestinal disturbances, nausea and vomiting, hypotension (which may lead to anuria), psychic and nervous disturbances (e.g., headaches, hallucinations, deliria, melancholia or mania, convulsions, photophobia, cutaneous hypersensitivity), dehydration, hypoglycemia (which is frequently the immediate cause of death), a drop in blood pressure, body temperature, blood sodium and chlorides, a rise in blood potassium and N.P.N.

Depending upon the relative prominence of one or the other of these manifestations during the crises, the evolution of the disease may take any of the forms described in the classification of the primarily acute types of Addison's disease (see : Classification).

Metabolism. — The BASAL METABOLIC RATE (B.M.R.) is, often but not always, subnormal and rarely falls to the extraordinarily low levels characteristic of hypothyroidism or severe pituitary failure.

The BODY TEMPERATURE may be a few degrees below normal, but only during severe crises is there any pronounced hypothermia. It is rather characteristic of addisonians that if they contract infectious diseases, which normally would produce marked fever, they merely react with a slight rise in temperature and, indeed, if the infection elicits a crisis, the body temperature may actually fall. On the other hand, the crisis may be associated with hyperthermia — which is a bad omen.

Addison's disease causes very prominent disturbances in CARBOHYDRATE METABOLISM. The fasting blood sugar level is rarely higher than 60-70 mg.%; even lower values are found during the crises.

Oral administration of glucose reveals an increased carbohydrate tolerance, the alimentary hyperglycemic curve being very flat and followed by, often dangerously steep, secondary hypoglycemia.

The sensitivity to *insulin* is greatly increased and since even moderate doses may cause fatal hypoglycemia, diagnostic insulin-blood-sugar curves should only be done with great precaution, after having prepared to administer glucose and adrenaline, if necessary.

Adrenaline generally fails to elicit marked hyperglycemia, because of the low hepatic glycogen reserves of addisonian patients.

Some of the corticoids, not only increase the fasting blood sugar, but, also prevent the hypoglycemic attacks, which are otherwise so readily provoked in addisonians by exposure to stress. Only gluco-corticoids and the adrenal-cortical extracts which contain them, are active in this respect, while desoxycorticosterone is not. This is presumably due to the fact that only gluco-corticoids exert a favorable influence upon gluconeogenesis from endogenous sources.

The blood LACTIC ACID content of addisonians rises markedly following, even mild, muscular exercise.

LIPID METABOLISM is less characteristically influenced by hypocorticoidism, although there is a great tendency to loose body fat and the blood cholesterol concentration tends to be low.

The most prominent changes in PROTEIN METABOLISM are, a tendency towards an increase in blood urea and total N.P.N., with creatinuria. Unless adequate corticoid therapy is administered, the nitrogen balance becomes negative.

A rise in the organic, non-protein sulfates of the blood, presumably glutathione, has also been noted in addisonians.

Among the disturbances in WATER AND SALT METABOLISM, the most prominent are, a decrease in serum sodium and chloride, accompanied by an increase in serum potassium. At the same time, there is a diminution in the urinary excretion of potassium, accompanied by a slight but continuous rise in the sodium chloride elimination.

The kidney, like the rest of the organism, loses its power of adaptation. Indeed, the outstanding renal defect is lack of adaptability to changes in intake of Na, Cl, K and water ("Fixed concentration"). High intake is followed by delayed excretion. Low intake is followed by comparatively high excretion of Na, Cl and probably also of water.

The negative NaCl balance is due to continuous losses. If large doses of NaCl are given (even without desoxycorticosterone) edema ensues.

Eventually there is a decrease in blood volume and hemoconcentration. These changes are finally accompanied by a diminished urine secretion, which terminally may become actual anuria.

All these derangements in water and salt metabolism respond rather well to treatment with sodium chloride, cor-

tical extracts or mineralo-corticoids such as desoxycorticosterone acetate. Excessive treatment with this steroid tends to produce edema, more readily in addisonians than in normal individuals.

The ALKALI RESERVE and the total base content of the blood are below normal in most patients suffering from Addison's disease.

It is noteworthy that many of the blood-chemical changes become severe only during the crises, while in the interim, the blood Na, K, Cl, CO₂-combining power, glucose and urea may remain within normal limits.

Growth and Bone Structure. — Contrary to expectations, if Addison's disease develops in children, the growth of the long bones is usually not very appreciably retarded. In adults, there is no great tendency to develop osteoporosis.

Blood Picture. — It is of historic interest that Addison himself, classified his disease among the ANEMIAS. This is perhaps not entirely justified, but a decreased red cell count and hemoglobin concentration is frequent in addisonians. It is perhaps partly due to the accompanying cachexia. Only if the hemoconcentration is very severe, does the loss of plasma fluid over-compensate for the diminution in red cell number.

Usually, there is LYMPHOCYTOSIS and this is not accounted for by accompanying tuberculosis; it occurs even if the underlying cause is adrenal sclerosis with atrophy. Yet sometimes the lymphocyte count is normal, or even subnormal, in Addison's disease.

Cardiovascular System. — The "remarkable feebleness of the HEART'S action," as described by Addison, is especially noticeable on auscultation. An accompanying diminution in cardiac size is noticeable in X-ray pictures and by percussion. Autopsy reports reveal that the smallness of the heart is large-

ly functional, and due to the decreased blood volume, but the actual weight of the heart is also diminished.

It is remarkable how rapidly the cardiac size of the addisonian increases under the influence of NaCl or desoxy-corticosterone acetate therapy, which restores the blood volume, and because the heart is flabby, tends to dilate it.

There is a great tendency towards HYPOTENSION, although, in patients who previously suffered from hypertension, the blood pressure may remain high until shortly before death. As a rule, systolic pressure varies between 90 and 100, diastolic between 60 and 70 mm. of mercury. This hypotension is largely responsible for the coolness of the body surface, the attacks of dizziness and many other characteristic manifestations of Addison's disease. The hypotension is often orthostatic, being evident mainly during erect posture. Not infrequently, addisonians suffer from mild RAYNAUD'S DISEASE.

Lymphatic Organs. — The THYMUS, LYMPH NODES, TONSILS, LYMPHATIC ELEMENTS IN PEYER'S PLAQUES AND SPLEEN, tend to be large in addisonians.

This is readily understandable if we remember that the physiologic and "accidental" involution of these tissues is inhibited by a lack of corticoids; yet it is striking to note their great development in emaciated addisonians, since with most other diseases which lead to loss of weight, the lymphatic organs are the first to involute.

The so-called, "STATUS THYMICO-LYMPHATICUS" is a condition which has been much discussed in the literature. Its main characteristics are : an excessive development of the thymus, tonsils and other lymphatic organs, with a concomitant "hypoplasia" of the adrenals. It is supposedly responsible for many cases of sudden death ("mors thymica") in apparently healthy, children or young adults exposed to some acute but minor stress, such as : a cold

bath, surgical trauma, anesthesia or psychic shock. Undoubtedly, the development of the thymico-lymphatic apparatus is enhanced by cortical hypo-function, which also greatly reduces resistance to sudden stress. However, many of the allegedly pertinent cases were not due to cortical insufficiency. The "hypoplastic adrenals" of many patients, described as belonging to this group, were actually normal in size, although much smaller than those of most other patients. It must be kept in mind, however, that if death is sudden, there is no time to develop the usual increase in adrenal size, which is part of the general-adaptation-syndrome elicited by most of the fatal diseases. Hence, the true normal size of the adrenals is much smaller than had been supposed, on the basis of measurements on average autopsy material. Great caution should be exercised therefore, before accepting a case as "status thymico-lymphaticus," without denying the possible existence of such a syndrome.

Respiratory Organs. — Even apart from the, often very mild (merely a Ghon tubercle), pulmonary tuberculosis, so frequent in addisonians, there appears to be a great sensitivity among them to various upper respiratory infections.

The respiratory rhythm is usually very irregular during the crises (Biot's type of respiration); there is dyspnea and in the terminal stages the respiration may become extremely slow. Sighing respirations, on the other hand, are more common in psychoneurotics.

The VOICE is occasionally also altered and the speech may become difficult to understand, perhaps due to debility of the muscles involved.

Muscles. — As already mentioned, asthenia, debility and languor are among the earliest symptoms of Addison's disease, although the histologic structure of the muscles is essentially normal. The muscular weakness de-

velops gradually. Early in the course of the disease, it is obvious only upon muscular effort but later, fatigue and exhaustion are evident, even after the slightest movement. Finally, muscular debility becomes the cardinal symptom. Like most of the manifestations of Addison's disease, the asthenia is subject to considerable fluctuation. In ambulatory patients, it is most pronounced in the early hours of the morning, perhaps because it is aggravated by orthostatic hypotension after rising. Objective ergographic examinations show that the weakness is not purely due to mental causes. They also reveal that the actual muscular strength is not very markedly impaired, in fully rested individuals, but declines rapidly after a few contractions. In this respect, the ergographic tracings are very similar to those obtained in adrenalectomized animals or patients with myasthenia gravis. Possible pathogenic relations with this latter disease are suggested by the frequency of excessive thymus growth in both conditions. (See : The Thymus.)

Nervous System. — Although nervous and mental symptoms may be lacking, they are often quite prominent. Frequently, there are SOLAR CRISSES or CELIAC NEUROSES. They manifest themselves by : acute pain in the celiac region, accompanied by vomiting, diarrhea, pain along the course of the aorta and its sympathetic plexuses. This may be so severe as to resemble that of coronary thrombosis. It has been claimed that pain in the abdomen is present in approximately half of the cases, particularly during crises. The pain has no special relationship to meals; it is usually dull and of paroxysmal character. It may be so intense that the patient screams with pain during the spells and the abdomen becomes rigid as in peritonitis. Often the pain is situated in the loins on both sides, and occasionally it is transmitted to the legs.

MENTAL SYMPTOMS. especially sudden psychic disturbances, such as melancholia or acute spells of maniac excitement, are quite frequent. The ability of concentration and thinking also suffer, as part of the general asthenia. In women, acute delirium with maniac excitement, often coincides with menstruation. Frequently there is a compulsive craving for salt.

SPELLS OF DIZZINESS are chiefly due to arterial hypotension, as stated above. That is probably why they are so common in the morning, when the patient gets out of bed and assumes the erect position, which predisposes to cerebral anemia. The fact that the blood sugar also tends to fall to its lowest levels during the night's fast, contributes, of course, to the morning malaise.

EPILEPTIFORM CONVULSIONS can be associated with the symptoms of a solar crisis or occur independently; they are possibly also of hypoglycemic origin.

THE TENDON AND PUPILLARY REFLEXES are usually normal, although, in some cases, there may be a positive Babinski or "labil" light reflexes of the pupil, especially during a crisis.

Among the **SENSORY** disturbances are : paresthesias, hyperesthesia of the skin, and photophobia. (See also : Sense Organs, below.)

MORPHOLOGIC LESIONS IN THE NERVOUS SYSTEM are uncommon in Addison's disease, although some investigators claim to have seen degenerative changes in the ganglia of the solar plexus.

Sense Organs. — The acuity of HEARING is often decreased and sometimes there is tinnitus.

VISION may become blurred and since this has been considered as one of the manifestations of the general asthenia, it has been described as "adynamy of vision." It is probably also the result of hypoglycemia.

Sometimes, addisonians suffer from a constant salty TASTE in the mouth,

which is probably due to atrophic glossitis. The resulting loss of appetite is largely responsible for the severe loss in body weight, so common in these patients.

Digestive System. — The prominent rôle played by the disturbances of the alimentary tract have already been mentioned in other connections. Usually there is nausea, sometimes accompanied by hiccups, gagging and occasionally intense diarrhea, but as a rule the patients tend more to constipation.

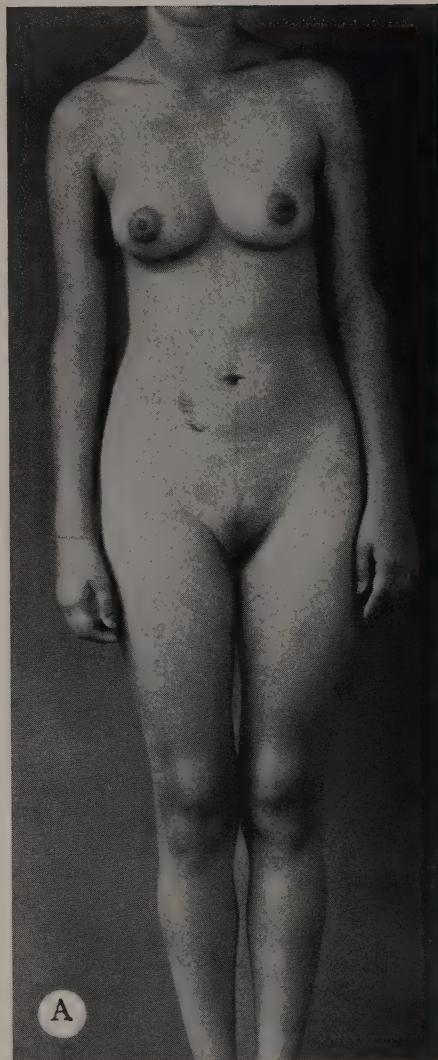
The symptoms of GASTRIC AND DUODENAL ULCERS may be super-imposed upon the ordinary signs of Addison's disease, although such ulcers usually occur only premortally.

GASTRIC ACIDITY fluctuates during the course of the disease, but tends to decrease whenever the condition of the patient becomes worse. Usually, the concentration of free HCl is reduced to 25-50% below normal and achlorhydria is common. This may be related to the disturbance in chloride metabolism, as mentioned above.

Function tests sometimes reveal slight disturbances in the activity of the LIVER, whose size is usually subnormal, but specific anatomic changes are rarely observed.

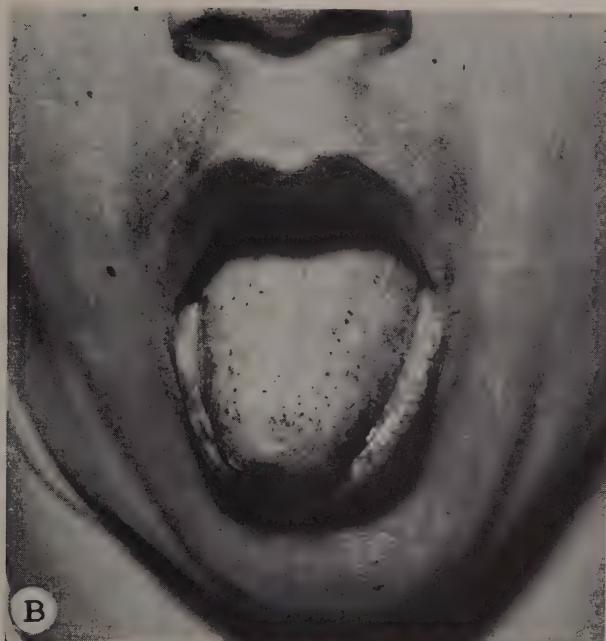
Skin. — The PIGMENTATION of the skin (melanoderma) and mucous membranes is one of the most characteristic symptoms of Addison's disease. It may develop very rapidly, but usually the acute cases of Addison's disease are conspicuous by the absence of excess pigmentation.

Melanoderma begins, and is usually most pronounced, in the wrinkles of the skin and in the parts exposed to light (e.g., hands, face) or pressure (e.g., under a tight garter, belt, collar, girdle, suspenders or studs). The tinge of the skin varies from that of a normal dark-skinned individual of the white race, to that of Arabians, Hindus



Pigmentation in Addison's disease.—A. Woman with Addison's disease. Absence of pubic hair. Pigmentation of areolæ, thighs, knees and legs. Patient usually had normal menses, occasionally interrupted by menorrhagias. Note deep pigmentation of appendectomy scar. (Cont'd.)

or even Negroes. Due to the accompanying anemia and hypotension the skin is rather grayish and dirty-looking. Usually, the discoloration is more or less diffuse, but jet-black lentigines and vitiligo-like, depigmented spots are also common. The pigmentation of the genital area and the nipples is



B. Spotty pigmentation of the lips and tongue. — C. Pigmentation of appendectomy scar and linea alba. (Cont'd.)

particularly marked. Although the melanoderma tends to become more pronounced as the disease progresses, it may improve during remissions.

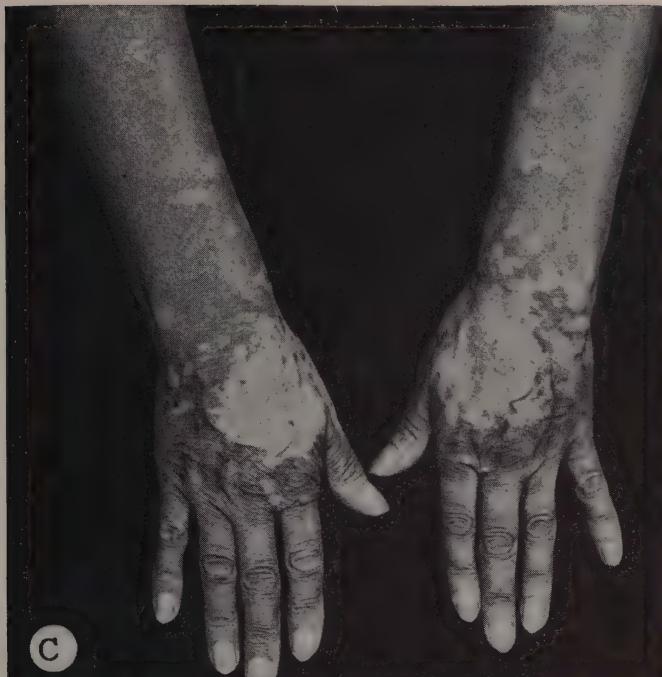
Patchy pigmentation of the mucous membrane within the mouth is not constant, but when present it is of great diagnostic value. It is seen on the lips,



D. Absence of axillary hair.



(For legend of A and B see p. 151)



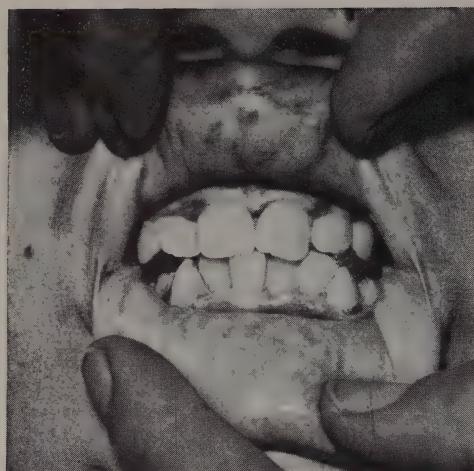
C



D

Pigmentation in Addison's disease.
A. Pronounced diffuse melanoderma of the hands and black lentigines in Addison's disease. — **B.** and **C.** Generalized pigmentation of the hands with vitiligo in Addison's disease. — **D.** Spotty pigmentation of the scrotum in Addison's disease.

(Courtesy of Dr. E.-J. Kepler).



Addison's disease

(After W. M. Yater: "Fundamentals of Internal Medicine" Appleton-Century, Publ., 1944).

Pigmentation of lips and gums.

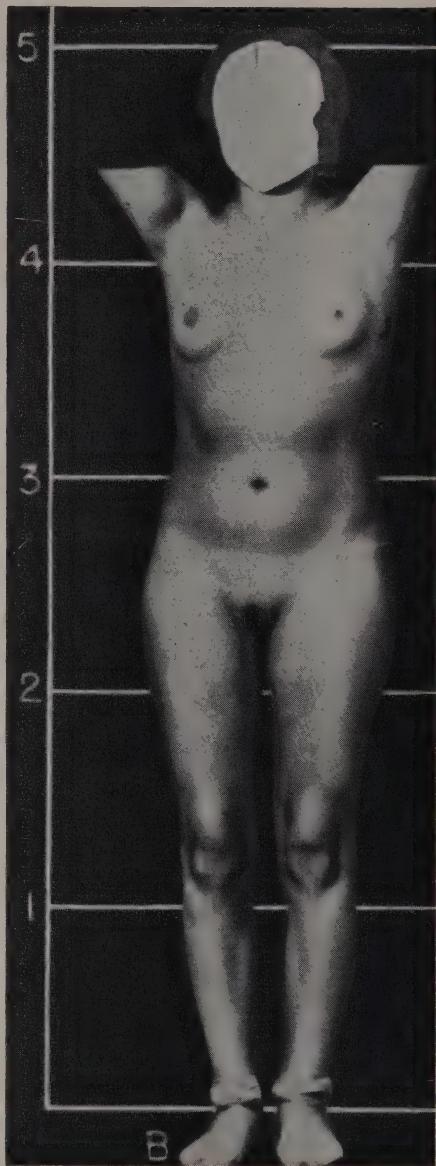
inner aspect of the cheeks, gums, hard palate and uvula. Even the margins of the eyelids, the conjunctiva and the limbus cornea may become pigmented. The mucous membranes of the rectum and vagina are rarely affected.

Histologically, the skin lesions are not diagnostic. As in sunburned patients, or those belonging to pigmented races, there is melanin deposition both in the epidermis and the cutis. Pigment granules are found throughout the epidermis, but are especially dense in the basal cells and in the tips of the rete ridges. The pigment deposition is chiefly intracellular. It is noteworthy that although corticoid treatment may improve the melanoderma, it rarely affects it as much as the other manifestations of cortical insufficiency.

The mechanism of pigment formation in Addison's disease has still not been clarified, but Bloch claims that a specific oxydase in the skin forms melanin from dihydroxyphenylalanine (dopa). (See p. 109.)

Scanty HAIR growth, and especially loss of axillary and pubic hair are rather characteristic.

Urinary System. — The KIDNEY rarely shows any characteristic structural changes, although nephrosis with tubular atrophy and renal tuberculosis are often found in addisonians. Renal



Addison's disease

(After Albright et al: Am. J. Med. Sc. 204, 625, 1942). Note complete absence of axillary and pubic hair, in spite of normal sexual development.

function is frequently deficient. The specific gravity of the urine is low and excretion of ingested water and salt is delayed. (See also page 145.) Albumin is usually present in the urine, although often only in small amounts. Sometimes there are pus cells and erythrocytes, but these are usually due to concomitant kidney tuberculosis. The rise in blood N.P.N. and the terminal anuria are chiefly due to the fall in blood pressure, not to any primary kidney failure.

Sex Organs. — The MENSTRUAL CYCLE is sometimes disturbed, although amenorrhea is rare and menorrhagias tend to occur only in patients approaching the menopause. The manifestations of Addison's disease are often exacerbated during menstruation.

In the male, a tendency to loss of libido and IMPOTENCE are not uncommon, but occasionally sex activity is maintained and patients of either sex may become the parents of normal children, in spite of manifest addisonian symptoms.

LACTATION is rarely possible in addisonian women.

COMPLICATIONS

Among the complications of Addison's disease, the manifestations of an underlying TUBERCULOSIS, (often pulmonary, renal, osseous or "healed" visceral), SYPHILIS or CARCINOMATOSIS are most important. As mentioned above, the underlying destruction of the adrenal, is frequently due to a local focus of these diseases. Of course, if the causative, primary, systemic malady progresses, its own manifestations can complicate suprarenal insufficiency.

The most important and dangerous complication of Addison's disease is the CRISIS, which may be elicited by a number of non-specific strains, enumerated above. Among these, complicating respiratory infections, such as broncho-

pneumonia, as well as tonsillitis and dental sepsis are especially common. The patients do not tend to be particularly susceptible to other (e.g., skin) infections, but any infection, once acquired, is poorly tolerated.

In addition to the complications of the disease, there are now appearing the complications of OVER-TREATMENT with desoxycorticosterone, such as : generalized edema, focal myocardial necrosis, arthralgias, arthropathies, pericarditis and possibly nephritis.

DIAGNOSIS

The diagnosis of Addison's disease is attended with great responsibilities, since it brands the patient as severely ill and permanently dependent upon expensive medical treatment. It is based upon :

- (1) Clinical manifestations of corticoid hormone deficiency.
- (2) Local signs of a lesion in the adrenal region.
- (3) Specific adrenal-cortical function tests.
- (4) Recognition of certain systemic diseases known frequently to affect the adrenals.
- (5) Differential diagnostic considerations.

(1) **Clinical Manifestations of Corticoid Hormone Deficiency.** — Typical and fully developed Addison's disease is readily recognized on the basis of the symptoms and signs as described under : Clinical Course. The progressive asthenia, accompanied by hyperpigmentation, gastrointestinal irritability and arterial hypotension, are of the greatest diagnostic value, especially if all of them are present. However, even in the most typical case, it is advisable to confirm the diagnosis, by the signs and tests listed below.

(2) **Local Signs of a Lesion in the Adrenal Region.** — Radiologic evidence of calcification in the adrenal

region (the angle between the ribs and the 12th thoracic or 1st lumbar vertebra), is comparatively common, especially in those cases of Addison's disease which are due to tuberculosis. Air insufflation into the stomach (in the case of the left adrenal), or peritoneum facilitates radiography of the adrenal region. Some authors recommend peri-renal air insufflation to produce a local emphysema, for the diagnosis of adrenal tumors, but none of these drastic procedures are permissible in the presence of untreated severe insufficiency. Large, palpable, destructive tumors, or signs of secondary invasion (e.g., hematuria) and displacement (detectable by palpation or radiography), with or without pyelography of the kidney, are comparatively rare. On the other hand, the rather common pain in the lumbar region is not very characteristic.

(3) Specific Adrenal-Cortical Function Tests. — These are based upon the inability of the hypocorticoid organism to respond normally to various types of stress.

The SALT DEPRIVATION TEST (*Hartop et al.* 1933), consists of putting the patient on a sodium chloride-poor diet for three days. If obvious symptoms of acute adrenal insufficiency ensue, it is very probable that we are dealing with Addison's disease. The test is not without dangers, however, and since it is evaluated on rather subjective clinical grounds, it does not enjoy great popularity.

The POTASSIUM RESISTANCE TEST (*Cutler, Power and Wilder*, 1938), is performed by placing the patient on a sodium chloride-poor and potassium-rich diet. Chloride concentration in the urine is determined during the last four hours; in Addison's disease it is above normal. While the test is rather reliable, it has the disadvantage of involving considerable danger (even deaths have been reported), because of the great potassium sensitivity of the

addisonians; false positives may occur in untreated diabetics and in certain types of renal disease.

The POTASSIUM TOLERANCE TEST (*Zwemer and Truszkowski*, 1936). This consists in the administration of potassium in the form of a function or tolerance test, with subsequent determination of the plasma potassium curve. A particularly steep hyperpotassemia is considered characteristic of Addison's disease. However, the test is dangerous and nephritis with edema, myasthenia gravis, malnutrition, neurasthenia and several other conditions may also decrease potassium tolerance sufficiently to give positive results.

The ROBINSON, POWER AND KEPLER TEST (1941), is regarded as a reliable function test and has the great advantage of not entailing any danger. It is based upon two facts :

(1) Following the rapid ingestion of a considerable amount of water, addisonians excrete the excess water much more slowly than normals, due to a diminished ability to produce a dilute urine.

(2) Addisonians tend to excrete excessive amounts of sodium and chloride, while retaining urea.

The test is sub-divided into two parts, to examine each of the above-mentioned disturbances separately. False positives are sometimes obtained in certain nephropathies, hyperthyroidism, and cachexia (relative hypocorticoidism?). Because of its practical value, we shall describe this test in detail, quoting its originators almost verbatim :

Procedure 1 (based on urine volume)
"The water test." — On the day before the test the patient eats three ordinary meals, but omits extra salt. He is requested not to eat or drink anything after 6 p.m. Until then, he may drink water as desired. At 10:30 p.m. he is requested to empty his bladder and discard the urine. All urine which is voided

from then on until and including 7:30 a.m., is collected. The volume of this urine is measured and the specimen saved for chemical analysis, if this should be necessary later. Breakfast is omitted. The patient is asked to void again at 8:30 a.m. and immediately thereafter he is given 20 cc. of water /Kg. of body weight (9 cc./pound). He is asked to drink this within the next forty-five minutes. At 9:30, 10:30, 11:30 a.m. and 12:30 p.m. he is requested to empty his bladder. In order to eliminate the effects of exercise and posture on urinary excretion, he is kept at rest in bed except when up to void. Each specimen is kept in a separate container. The volume of the largest one of these four specimens is measured.

Under these conditions some addisonians excrete so little urine that they are unable to void more than once or twice during the entire morning. In such instances, the amount of urine excreted per hour may be calculated; frequently, however, such calculations

are unnecessary because of the very low total urinary output.

If the volume of any single hourly specimen, voided during the morning, is greater than the volume of urine voided during the night, the test is negative, that is, it indicates absence of Addison's disease. If the volume of the largest hourly specimen voided during the morning, is less than the volume of urine voided during the night, the test is positive, but indicates only the possible existence of Addison's disease. In such instances, procedure 2 should be instituted to establish the diagnosis.

Procedure 2 (based on blood and urine chemistry). — To proceed with this test, blood is drawn (preferably under oil), while the patient is still fasting, and the urea and chloride contents are determined in the plasma and in the night urine specimen. From these four determinations and from the results obtained from procedure 1, the following equation is solved :

$$A = \frac{\text{Urea in urine (mg.\%)} \times \frac{\text{Chloride in plasma (mg.\%)} \times \text{Volume of day urine (cc.)}}{\text{Chloride in urine (mg.\%)} \times \text{Volume of night urine (cc.)}}}{\text{Urea in plasma (mg.\%)}}$$

The term "day urine" applies to the largest of the hourly specimens voided during the day; "night urine," to the entire amount which was voided from 10:30 p.m. to 7:30 a.m. It is immaterial how these values are expressed, provided that the same method be used throughout the equation. For example, if the concentration of plasma chloride is expressed as mg. of NaCl/100 cc. the concentration of urinary Cl, should be expressed in the same manner.

It is considered that if the value of "A" is greater in the above equation than 30, we are not dealing with Addison's disease. If this value is below 25, the existence of Addison's disease is very probable if nephritis has been

excluded. The test is most valuable if negative, since then it excludes Addison's disease with almost absolute certainty. A positive test nearly always means that the patient is severely ill but the hypocorticoidism may be relative rather than absolute (due to adrenal destruction). In doubtful cases a salt deprivation test is indicated.

THE THERAPEUTIC FUNCTION TEST is based upon the fact that administration of corticoid preparations is particularly beneficial if cortical function is deficient. A striking amelioration, in the clinical and laboratory manifestations of patients suspected of Addison's disease, following administration of corticoid therapy, may there-

fore also be used, as an innocuous and rather specific function test, among other diseases. Only secondary hypocorticism, due to anterior-pituitary deficiency, is likely to respond with an almost equally dramatic improvement. It is well to bear in mind, however, that purely subjective improvement is of no diagnostic value.

OTHER LABORATORY TESTS. Among other laboratory tests designed to reveal derangements, which are more or less specific of hypocorticism, the following are worth mentioning :

- (1) Severe fasting hypoglycemia.
- (2) Increased glucose tolerance, with severe secondary hypoglycemia after ingestion of sugar.
- (3) Extraordinarily severe hypoglycemia following insulin administration (dangerous!).
- (4) High plasma potassium values.
- (5) Low plasma sodium and chloride values.
- (6) Negative NaCl balance.
- (7) Markedly diminished 17-KS. elimination in the urine. (Very low in females, about half of normal in males.)
- (8) Increased blood N.P.N. and blood urea.
- (9) Achlorhydria.
- (10) A diminished B.M.R. in the absence of hypopituitarism or hypothyroidism.
- (11) Increased blood CO₂ capacity.
- (12) Hemoconcentration with decreased blood volume.

(4) Recognition of Certain Systemic Diseases Known Frequently to Affect the Adrenals. — As we have repeatedly said in other connections, tuberculosis is one of the most common causes of adrenal destruction and hence, the recognition of tuberculosis, in any organ, raises the suspicion of Addison's disease, if signs of probable hypocorticism are also present. The same is true, to a lesser degree, of syphilis, carcinomatosis, and some

exhaustive acute diseases (influenza, puerperal infections), which are likely to cause secondary exhaustion atrophy of the adrenals.

(5) Differential Diagnostic Considerations. — Confusion with melanoderma due to SOLAR OR X-RAY IRRADIATION, as well as that produced by exposure to heat, is readily eliminated by the past history of the patient. Pigmentation induced by various SKIN OINTMENTS AND LOTIONS (especially ointments containing mercury), is limited by the regions treated. RACIAL MELANODERMA may cause diagnostic difficulties, since it is sometimes not easy to estimate the degree of skin pigmentation, which could be called pathologic in pigmented races. Here again, a history of a sudden unexplained increase in the darkness of the skin, as well as the absence of the characteristic pigmentations and jet-black lentigines are significant. Metallic poisoning, especially with LEAD, ARSENIC, BISMUTH OR SILVER, must also be excluded, by an inquiry into the past history and a search for other manifestations of these intoxications. In ACANTHOSIS NIGRICANS, the pigmented areas are usually axillary and of velvety appearance, rather than of the satin-like type, typical of Addison's disease. HEMOCHROMATOSIS may be recognized by the demonstration of iron pigment in the skin, the presence (in the late stages) of sugar in the urine and an enlargement of the liver. The ordinary liver function tests, however, usually give normal values. SCLERODERMA is frequently accompanied by melanoderma, suggestive of Addison's disease, but its symptoms and signs are so characteristic that confusion will rarely occur. CAROTENEMIA AND JAUNDICE are recognized by the presence of the abnormal pigments in the serum. PREGNANCY may lead to hyperpigmentation. The same is true of "VAGABONDS' DISEASE" but here, the skin scratches due

to the itching, are also of diagnostic value.

In PERNICIOUS ANEMIA, the skin coloration is often dark and, in view of the accompanying anemia, the diagnosis may be difficult unless adequate hematologic studies are performed. It is of historic interest that Addison himself considered the two conditions as closely related to each other.

CHLOASMA UTERINUM is accompanied by menstrual disturbances and since the latter are fairly common in addisonians, this possibility should not be left unconsidered.

TUBERCULOSIS leads to diagnostic difficulties, firstly, because tuberculosis is so common among true addisonians and secondly, because, even if the adrenals are not involved, such signs as pigmentation, disturbances in the sex organs, arterial hypotension and severe asthenia with gastrointestinal manifestations, are often observed in tubercular individuals. Pain, or radiologically detectable calcification in the suprarenal region, is usually indicative of suprarenal involvement.

In MALARIA the antecedents, the characteristic fever, splenomegaly and the demonstration of the parasites in the blood will facilitate the diagnosis in spite of the pigmentation. It must be remembered, however, that a functional adrenal failure, with addisonian symptoms, is not uncommonly associated with malaria.

The cutaneous hyperpigmentation in SYPHILIS is usually restricted to certain regions, such as, the back of the neck and the shoulders. Furthermore, there are no other accompanying manifestations of Addison's disease and the serologic reactions facilitate the diagnosis, even if asthenia and arterial hypotension are present, as is frequently the case in chronic syphilitics. This differential diagnosis is of importance because of the frequent association of syphilis with true Addison's disease,

due to secondary involvement of the adrenals.

Among other conditions, likely to cause diagnostic difficulties, early LIVER CIRRHOSIS, HYPERTHYROIDISM with pigmentation, ANOREXIA NERVOSA and CHRONIC NERVOUS EXHAUSTION, should be considered.

Exploratory *skin biopsies* are of value in the recognition of certain types of melanodermas, for instance, those of acanthosis nigricans and metal poisonings, but they should not be performed, without previous corticoid treatment, since untreated addisonians are so very sensitive to any type of surgical intervention and the accompanying excitement.

PROGNOSIS

The prognosis of FRANK ADDISON'S DISEASE is always very grave. While the introduction of sodium chloride and especially of corticoid hormone therapy has greatly improved the chances of prolonged survival, it must be kept in mind that in a very large percentage of the cases, the adrenal destruction is merely one manifestation of such serious systemic diseases as tuberculosis, syphilis, carcinomatosis, etc. These, in themselves, are often fatal, if they reach a stage where secondary deposits destroy the adrenals. But even when Addison's disease is due to a primary adrenal failure, or if the basic disease is under control, the prognosis is poor, because it is extremely difficult to supply adequate therapy for all the contingencies of normal life. Any accidental interruption of the therapy, or any intercurrent stress, may be fatal unless immediately met by an appropriate increase in corticoid hormone administration. Hence, the average life span of the addisonian still rarely extends over more than a few years.

On the other hand, the so-called "FORMES FRUSTES," or cases of CONSTITUTIONAL ADDISONISM, due to tem-

porary, functional failure of the cortex, frequently end in complete recovery, even without therapy. In any event, their rational treatment is stimulation of the adrenal cortex (corticotrophins) rather than replacement therapy with corticoids, which tend to cause compensatory cortical atrophy and overdosage symptoms.

A large number of addisonians succumb due to overtreatment with desoxycorticosterone and NaCl, especially since the symptoms of this overdosage resemble those of the addisonian crisis (weakness, hypoglycemia), hence, the physician without adequate laboratory facilities tends to meet them by a further increase in desoxycorticosterone and NaCl therapy.

THERAPY

The treatment of Addison's disease may conveniently be discussed under the following headings:

(1) Treatment of the underlying disease responsible for the adrenal destruction.

(2) Treatment of the manifestations of hypocorticoidism.

(3) Treatment and prevention of the addisonian crisis.

(1) **Treatment of the Underlying Disease Responsible for the Adrenal Destruction.** — If TUBERCULOSIS occurs in combination with Addison's disease it is almost invariably the cause of the latter. In such instances, the systemic tuberculosis should be treated in accordance with generally accepted therapeutic principles. A detailed discussion of these would be beyond the scope of this book.

Essentially the same is true as regards SYPHILIS and MALARIA. In some pertinent cases adequate antisyphilitic or antimalarial therapy led to a permanent cure of hypocorticoidism.

If HYPOPITUITARISM is accompanied by especially severe manifestations of adrenal-cortical hypofunction, the ra-

tional therapy would consist in the administration of purified adrenocorticotrophic hormones. Such preparations are not as yet commercially available in adequate quantities and since hypocorticoidism of pituitary origin responds well to the usual corticoid and salt therapy, as discussed below, it is justified to treat these cases as if they were primarily due to cortical malfunction.

It should also be kept in mind that hypopituitary patients suffer from a complex hormonal disturbance and that even if the cortical deficiency is most prominent in the syndrome, the other hormonal derangements should also be treated. Hence, it is advisable to handle such patients in agreement with the principles enumerated in the section on the therapy of hypopituitarism.

(2) **Treatment of the Manifestations of Hypocorticoidism.** — Since purified natural corticoids are not yet commercially available in adequate quantities, the most effective practical therapeutic procedure is to administer ADRENAL-CORTICAL EXTRACTS several times a day subcutaneously, intramuscularly or, in the case of imminent danger, intravenously. The degree of purity and the potency of the various extracts is rather variable, but the average active preparation, now on the market, can be administered in almost any quantity without having to fear overdosage phenomena. Anywhere between 2 to 100 cc. per day may be necessary to keep the patient free of deficiency symptoms. The daily dose should be subdivided into several injections, given at regular intervals. Under basic conditions, two daily injections suffice but during exacerbations, injections may be necessary every six, or even every three hours, throughout the day and night. Fractionation of the dose is essential, because the effect is very transitory and a given amount is much more effective if ad-

ministered in small portions, than if given in a single large dose. Certain lipid-soluble extracts (e.g., Upjohn's lipocortical extract) are about five times as active as the usual aqueous solutions and require only one injection per day because of delayed absorption. Since for most patients, the cost of prolonged treatment with cortical extract is still prohibitive, there is a tendency to use an insufficient dosage.

Corticoid extracts or adsorbates of cortical material to charcoal given orally, are much less effective in clinical medicine, although in some animal species, corticoids are highly active by mouth.

Among the chemically pure cortical steroids, only DESOXYCORTICOSTERONE ACETATE is commercially available in adequate quantities at this time. Its chief advantage is its comparatively low price. Its main disadvantage is that, when given in high doses, it is toxic. It may cause a marked increase in blood volume, edema with anasarca, arterial hypertension, cardiac insufficiency, angina-like pain, nephrosclerosis with proteinuria, dyspnea, profound muscular weakness, arthralgias, and a dangerous degree of hypopotassemia. However, these toxic manifestations rarely occur with the minimum therapeutic doses and hence, under constant supervision, the hormone may be used to advantage. Unfortunately, desoxycorticosterone acetate is practically devoid of gluco-corticoid activity. Hence, patients otherwise adequately treated with this steroid may develop dangerous, sometimes fatal, addisonian hypoglycemia with asthenia, even if the other symptoms and signs of hypocorticoidism are completely prevented. Hypoglycemia may occur even in patients who simultaneously show marked signs of desoxycorticosterone intoxication.

In general, daily doses of 2 to 6 mg. of desoxycorticosterone acetate are suf-

ficient when given subcutaneously, intramuscularly or by the sublingual route. The latter mode of application is especially recommended. The subcutaneous implantation of compressed desoxycorticosterone acetate tablets is also effective; it is not recommended, however, since dosage cannot be adjusted to changing requirements. Many patients can be maintained for months, or even years, with no other hormone therapy but desoxycorticosterone acetate. They should constantly be checked, however, for danger signals of over-dosage or hypoglycemia. Their vigour and well-being may also be improved by simultaneous treatment with *testoids* (e.g., 30 mg. methyl-testosterone per day), in both sexes.

Since SODIUM CHLORIDE is particularly beneficial to hypocorticoid patients, their diet should contain a fair amount of salt. If corticoid preparations are unavailable, even mere salt therapy may suffice to control the manifestations of insufficiency. Conversely, no extra salt may be necessary if the patient can afford adequate doses of desoxycorticosterone. Na-citrate or bicarbonate are even more effective than NaCl, but usually salt therapy is employed in combination with cortical extracts or desoxycorticosterone. In the food, NaCl should be given only in quantities which are considered necessary to season it, otherwise, the already poor appetite of the addisonian may be adversely influenced. In addition to this, NaCl tablets containing 0.5 to 1.0 gm. each should be prescribed.

If these are badly tolerated, the following drink is recommended by some (*Del Castillo et al.*):

Sodium chloride	10 gm.
Sodium citrate	5 "
Glucose	160 "
Fruit juice (lemon)	80 cc.
Add water to bring total volume to one liter.	

Some patients prefer taking this drink (chilled), to the sodium chloride tablets, but it may

be poorly tolerated, although this is rarely the case in patients who really need salt therapy.

In times of danger, the subcutaneous, or even intravenous, administration of hypertonic sodium chloride solution may be necessary.

In patients receiving desoxycorticosterone, the necessary total daily dose of NaCl varies between 3 and 5 gm. in addition to what is taken with the diet. Somewhat higher doses are recommended in the absence of corticoid therapy (e.g., 10 gm. of NaCl and 5 gm. of Na-citrate).

In prescribing NaCl, it must be kept in mind that its chief advantage is to increase the efficacy of corticoids, but it also augments their toxicity.

In view of the great progress made in the extraction and synthesis of gluco-corticoids, it is most probable that these will be available for clinical use within a short time. This could hardly fail to improve the therapy of Addison's disease. At the present time, however, the combined salt and corticoid therapy, as described above, is still the best practical procedure.

In view of the great tendency of addisonians (even those treated with desoxycorticosterone acetate) to develop dangerous hypoglycemia, (especially before breakfast or during anorexia), it is well to administer a high CARBOHYDRATE diet in the form of foods rich in starch and sugar, supplemented by fruit juices, containing as much glucose as the patient can take without spoiling his appetite.

Since POTASSIUM salts are extremely toxic to addisonians, if tolerated, the diet should not contain more than 2 gm. of potassium, per day. This greatly increases the efficacy of salt or corticoid therapy but, in patients treated with desoxycorticosterone acetate, care must be taken not to reduce the dietary potassium too far, as this increases the danger of hypopotassemia. Some addisonians are extremely sensitive to, even small doses of, desoxycorticosterone acetate (0.5-1.0 mg.), perhaps

because in them the hormone tends to deplete the potassium stores without correcting the other manifestations of hypocorticoidism. Since low potassium diets are unpleasant, and reduction of dietary potassium is not necessary in the presence of adequate corticoid therapy, low K diets have been discarded in most clinics.

It is also important to ascertain that the patients continuously take CALORICALLY ADEQUATE AMOUNTS OF FOOD, especially that their evening meal be sufficiently substantial to prevent fasting hypoglycemia during the night and that they receive a minimum maintenance amount of PROTEIN and VITAMINS.

(3) Treatment and Prevention of the Addisonian Crisis. — Addisonians must take special care to AVOID STRESS OR STRAIN, such as: excessive muscular exercise, exposure to cold, exposure to the danger of intercurrent, respiratory infections. In view of the great sensitivity of addisonians to insulin, thyroid hormone, barbiturates and morphia, these drugs should be given only if absolutely necessary. The same is true of surgical interventions. If exposing an addisonian to one or the other of these agents is unavoidable, the corticoid therapy should be preventively increased. Thus, two or three days before an unavoidable surgical intervention, large doses of sodium chloride should be given in combination with much more than the normal maintenance dose of cortical extracts. Just before the surgical intervention, about two liters of a saline-glucose solution with 50 cc. of the usual potent commercial cortical hormone extract should be given slowly, intravenously. In adequately treated patients, Addison's disease is no longer a contraindication, even for severe surgical interventions if they are really necessary.

If a CRISIS ENSUES, the patient is immediately put to bed and kept warm. During the first day, it is advisable to administer one liter of 1.5% NaCl,

intravenously, as well as 1.5 liters of a 10% glucose solution. Each of these infusions should contain at least 25 cc. of cortical extract and in addition, 25 cc. may be given intramuscularly. If the patient is moribund 200-300 cc. of extract may be necessary. (In view of the comparatively slow activity, and dubious gluco-corticoid potency, of desoxycorticosterone acetate, it is not recommendable during the crisis.)

Unless the condition of the patient has advanced to an irreversible stage, improvement is almost instantaneous. However, intense sodium chloride, glucose and corticoid hormone therapy must be continued, together with the

administration of large amounts of fluid, until the patient has definitely passed the critical period. If these precautions are observed, it is often possible to save the patient even in advanced stages of a crisis, when therapeutic measures were invariably futile before the discovery of the corticoids. In very advanced cases, however, the condition becomes irreversible and cannot be improved by any known therapeutic procedure. Occasionally, recovery from a crisis is followed by permanent damage to the nervous system (loss of memory, mental deficiency etc.), reminiscent of that which tends to occur after insulin shock.

HYPERCORTICOIDISM

(SYNOMYS : hyperadrenalinism, hyperadrenia; when combined with pseudohermaphroditism : adrenogenital syndrome of Cooke-Apert-Gallais, suprarenal genital syndrome of Kraus, hirsutism of Apert, adrenal-hermaphroditism, adrenal virilism; when accompanied by precocious sexual development in children : macrogenitosomia precoox suprarenalis; when accompanied by glycosuria in women : diabetes of bearded women, Achard-Thiers syndrome.)

DEFINITION

Hypercorticoidism is a condition in which the hormone production of the adrenal cortex is sufficiently augmented to cause detectable overdosage manifestations.

It must be kept in mind that the adrenal cortex produces a number of hormones with qualitatively different properties. Since, under certain conditions, the excess production of one or the other of these substances may prevail, it is obvious that hypercorticoidism can manifest itself in a variety of clinical forms. We shall elaborate further on this point in the chapter on

Classification. It may appear somewhat artificial to group together, under the generic designation of hypercorticoidism, such a heterogeneous group of diseases as we shall find in this chapter. Yet, since they are all due to excessive function of adrenal-cortical cells, their conjoint discussion is justified.

CLASSIFICATION

It is rather difficult to find a satisfactory basis for the classification of clinical hypercorticoidism. Theoretically, there could be one clinical overdosage syndrome corresponding to each of the cortical steroids, which possess qualitatively different actions. Thus, there could be syndromes characterized by virilization (adrenal testoids), feminization (adrenal folliculooids and luteoids), diabetes (gluco-corticoids), hypertension and nephrosclerosis (mineralo-corticoids), and perhaps even syndromes specifically due to overdosage with "lipo-corticoid" (?) and anesthetic steroids. Actually, most cases of hypercorticoidism exhibit a mixed symptomatology. This is perhaps due, partly to the fact that usually several corticoids are simultaneously

produced in excess and partly, to the manifold biologic activities of individual steroids. The existence of pure, uncomplicated diabetes or renal hypertensive disease, secondary to hypercorticoidism, has not yet been definitely proven. Although there is a good deal of evidence indicating that such syndromes and a variety of "Diseases of Adaptation" (see corresponding section) occur as a result of hypercorticoidism, we shall limit ourselves here to a discussion of the so-called adrenogenital syndrome, whose dependence upon hypercorticoidism has been demonstrated beyond doubt.

According to the AGE OF ONSET we distinguish between :

- (1) Adrenogenital syndrome *in the fetus*, with pseudohermaphroditism in females.
- (2) Adrenogenital syndrome *in children* :
 - (a) Precocious puberty, with pseudohermaphroditism in females.
 - (b) Precocious puberty, without pseudohermaphroditism in females.
 - (c) Precocious puberty, without pseudohermaphroditism in males.
- (3) Adrenogenital syndrome *in adults* :
 - (a) With pseudohermaphroditism in women.
 - (b) With pseudohermaphroditism in men.

The pseudohermaphroditic development of the sex organs is much more common in females and most pronounced if the disturbance commences, at an early age, at least before the advent of puberty.

According to the INTENSITY of the clinical manifestations, we may distinguish :

- (1) Adrenal pseudohermaphroditism with marked heterosexual differentiation.

- (2) Simple virilization or feminization.

According to the UNDERLYING ADRENAL LESION, it is customary to distinguish :

- (1) Simple hyperplasia of the adrenal cortex.
- (2) Enlargement of accessory adrenal-cortical tissue.
- (3) Cortical adenomas.
- (4) Cortical carcinomas.

In all these types, symptoms of Cushing's syndrome (see : Diseases of the Hypophysis) may be superimposed.

It should be emphasized that, in the earlier literature, pseudohermaphroditism was regarded as a congenital malformation of the accessory sex organs, which caused them to assume characteristics of the sex opposite to that of the patients' gonads. These conditions are fulfilled, at least in the most precocious types of the adrenogenital syndrome. We know now that, both the embryonic and postnatal differentiation of the accessory sex organs is regulated by hormones and that these can direct it either in the normal or heterologous sense. No known facts indicate that genetically-conditioned malformations can accomplish this without the intermediary of hormones. Hence, it appears appropriate to include the heterologous differentiation of adrenal origin as one of the types of pseudohermaphroditism.

PATHOLOGIC ANATOMY

Some of the adrenal lesions, which can cause hypercorticoidism, such as cortical hyperplasia (see : Malformations on p. 136) and cortical tumors (see p. 190), have been discussed in other parts of this chapter. Hence, here we shall consider only those morphologic alterations which are most characteristic of the adrenogenital syndrome in particular.

Simple Hyperplasia of the Adrenal Cortex. — In many instances of adreno-

genital syndrome, simple hyperplasia of the cortex is the only detectable adrenal change. It has been claimed that in such cases, the cortex shows histologic characteristics typical of the suprarenogenital syndrome. Thus, *Broster and Vines* (1933) emphasized that in virilism certain cortical cells show a special affinity for ponceau fuchsin, even if no enlargement of the gland is observed. Others (*Goldzieher and Koster*, 1935), claim that hyperplasia and marked eosinophilia of the zona reticularis is a constant finding in adrenal virilism with adiposity, even if there is no marked increase in the total size of the cortex. As has been stated in other parts of this chapter, experimental work also suggests that this zone (which corresponds to the X-zone), is particularly concerned with the elaboration of adrenal testoids.

Enlargement of Accessory Adrenal Cortical Tissue. — This may take the

form of the so-called "hypernephroma of the ovary" or may be due to hyperplastic cortical cell nodules in the mesovarium, the spermatic cord or other ectopic sites.

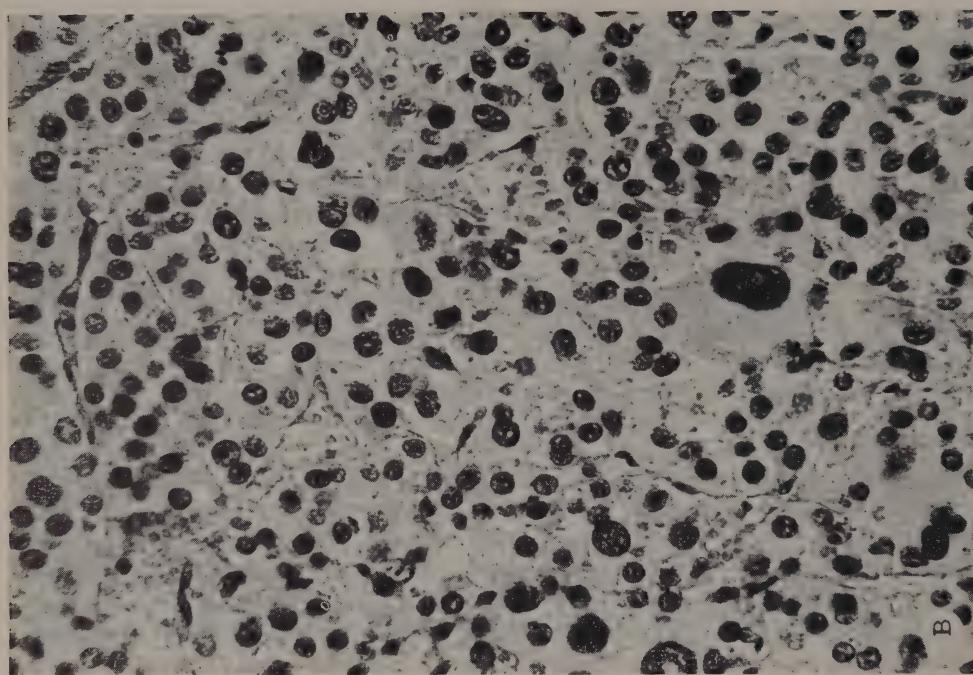
Cortical Adenomas. — The often multiple and minute, usually subcapsular, cortical adenomas, are frequently designated as nodular hyperplasia of the cortex. They rarely manifest signs of hypercorticoidism. However, single or multiple large adenomas are often conducive to the adrenogenital syndrome. The histologic structure of these neoplasms does not differ essentially from that of hormonally inactive cortical adenomas, or those merely associated with hypertension, without pseudohermaphroditism. (See also p. 190.)

Cortical Carcinomas. — The differentiation of the true adrenal-cortical carcinoma from the Grawitz' tumor or "hypernephroma" is discussed in connection with the adrenal tumors. Suf-

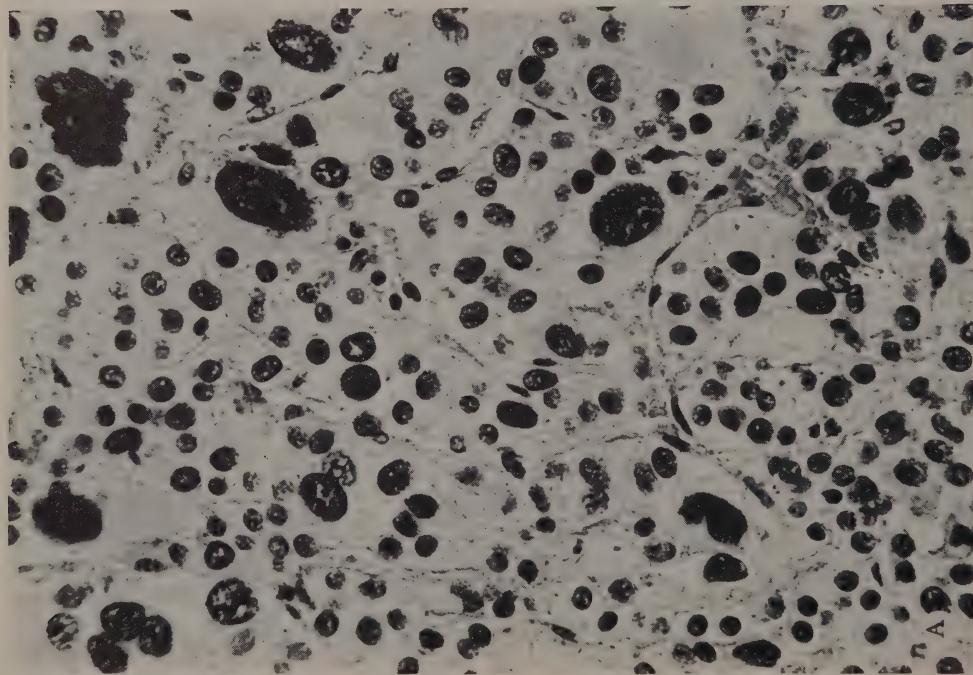


Adenoma of the adrenal cortex. Note typical appearance of light, vacuolated (lipid-containing) cortical cell trabeculae.

(Courtesy of Dr. P. Masson).



B



A

Carcinoma of the adrenal cortex. Girl, age 4 years, with metastasizing cortical carcinoma.—A. Primary tumor, with giant cells and very atypical polymorph cellular structure.—B. Section from a pulmonary metastasis of the same tumor. Note that the structure is very similar to that of the primary tumor. (Courtesy of Dr. P. Masson).

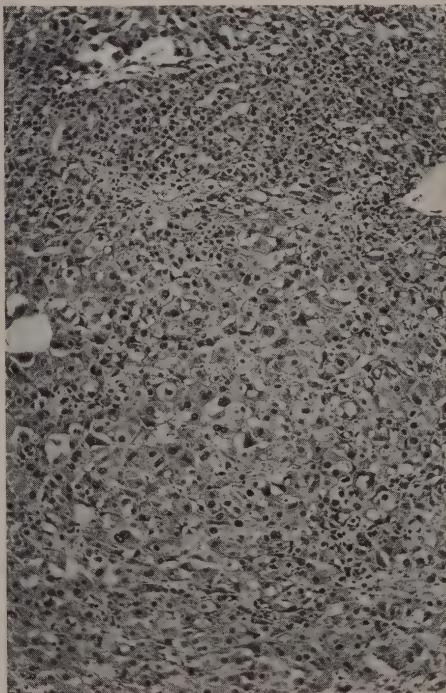
fice it to say here, that the more anaplastic the cortical carcinoma, the less likely it is to produce hormone over dosage symptoms. As in the case of the adenomas, those cortical carcinomas, which are conducive to hypercorticoidism, reveal no morphologic characteristics permitting the recognition of their functional activity.

In exceptional cases, an adrenal carcinoma, which originally caused hypercorticoidism, eventually destroys the adrenal and after metastasizing into the contralateral gland, results in the destruction of all functional tissue. In such instances, not only do the manifestations of hyperactivity disappear, but, secondary addisonism ensues. This phenomenon occurs more often as the result of hyperplasia of the testoid-producing, at the expense of corticoid-secreting adrenal tissue. In very young infants it may cause death, even before the sexual changes have had time to appear. In one case (*Kepler*), the child did not die because of replacement therapy and she subsequently showed signs of adrenal virilism. The clinical picture in these cases suggests pyloric stenosis.

It is of practical importance that a hyperactive, unilateral, cortical neoplasm, whether an adenoma or a carcinoma, usually causes severe compensatory atrophy of the contralateral gland. Hence, severe and often fatal adrenal insufficiency may ensue after extirpation of the tumor, unless the patient is preventively treated with adequate amounts of corticoid preparations. (See also p. 191.)

Other Diseases. — There is some disagreement regarding the adrenal changes in renal HYPERTENSION and NEPHROSCLEROSIS. Some investigators claim that signs of diffuse or nodular hyperplasia are more common in this disease than in the average autopsy material; others deny this. It is certain that renal hypertension is usually not accompanied by as marked cortical

lesions as the adrenogenital syndrome. Yet, in several clear-cut cases, cortical tumors or hyperplasia were accompanied by hypertension, nephrosclerosis or periarteritis nodosa and in some of these, removal of the proliferating cortical tissue had a curative effect. Experimental work (see pages 122 and 125), also suggests that the adrenal cortex plays an important part in the pathogenesis of renal hypertension. It is probable, therefore, that the lack of morphologic support for this interpretation is mainly due to our inability to recognize the histologic changes underlying certain types of function. It will be recalled that the fuchsinophilia of the reticularis in virilism likewise remained unrecognized until the discovery of its special staining qualities.



Carcinoma of the adrenal cortex. Very atypical carcinoma of the adrenal cortex, with numerous giant cells and highly irregular nuclei. Compare with the small cellular trabeculae of the normal adrenal cortex near the periphery of the field.

INCIDENCE

The GENERAL INCIDENCE of the fully developed adrenogenital syndrome is very low. Mild types of virilism, on the other hand, are so common among women that they may even be regarded as being on the borderline of the normal. In most of the mild cases, it is difficult to ascertain the adrenal origin of the manifestations and hence, statistical studies are almost impossible.

The general incidence of hypertensive disease and other "diseases of adaptation" is probably the highest of all maladies of man, but it is yet to be determined what percentage of the relevant cases are due to hypercorticoidism.

While the adrenogenital syndrome may develop at any AGE, the characteristic, severe cases are most common among children. This may be due, at least partly, to the greater ease with which pseudohermaphroditic changes develop at a time when the sex organs are still incompletely formed. Comparatively mild virilism (especially hirsutism), on the other hand, is very common among postmenopausal women, and there are good reasons to believe that this is of adrenal-cortical origin. It is frequently accompanied by hypertension and sometimes by increased urinary 17-KS excretion. The terms, "postmenopausal virilism" or "postmenopausal Cushing's syndrome" have been coined to designate relevant cases. Some authors think that after the menopause, the diminution of female sex hormone production may be conducive to an increased pituitary adrenotropic hormone secretion and secondarily to signs of hypercorticoidism. This theory is yet to be proven, especially since some of these changes may be due to anomalies of the end-organs, rather than to endocrine derangements. It is almost certain that such a mechanism is operative in the beardless Chinese and American Indian males.

The female SEX is about five times more frequently affected than the male.

HEREDITY also plays an important rôle in the pathogenesis of the disease. In many instances, the adrenogenital disturbances begin during embryonic life. The syndrome has repeatedly been seen in several members of the same family, frequently accompanied by other congenital malformations, such as spina bifida, harelip, or atresia of the anus. Some authors claim that it is most frequent among Jewish and Spanish people; mild degrees of hirsutism are very common among Mediterranean races.

PATHOGENESIS

The immediate cause of hypercorticoidism, that is, the underlying adrenal changes, have been discussed in the chapter on pathologic anatomy. In some instances, the adrenal hyperfunction is "idiopathic" or due to a suprarenal-cortical neoplasm, about whose origin we know as little as about that of any other true tumor. A pituitary adenoma (usually basophilic), may also be responsible for diffuse hyperplasia or adenomatosis of the cortex with secondary hypercorticoidism. In this case, our understanding merely stops one link higher in the chain of events, since we do not know the factors responsible for pituitary tumor formation.

If it is true that nephrosclerosis and renal hypertension are primarily hypercorticoid conditions, their original causative factor would be the adaptive reaction, elicited by exposure to some non-specific damaging agent.

In any event, the clinical symptoms and signs of the hypercorticoid syndromes are fully accounted for by the excess production of cortical steroids, a fact best illustrated by the curative effect of adrenalectomy. The virilization is presumably due to increased elaboration of adrenal testoids, the feminization to folliculoids, the diabe-

tes to gluco-corticoids and the hypertension to mineralo-corticoids. The existence of separate lipo-corticoids is not fully proven but if such exist, they could explain the fat deposition characteristic of the adrenogenital syndrome. As previously emphasized, most cortical steroids exhibit several biologic effects and hence, any one of these could cause several of the typical manifestations.

CLINICAL COURSE

State. — It is hardly possible to draw a characteristic picture of "typical," adrenogenital syndrome because the various types (see : Classification) differ so markedly from each other. The cardinal manifestations are, pseudohermaphroditic traits and, since the condition is much more frequent in women, these usually correspond to virilization.

In children, the development of heterosexual features, tends to be much more pronounced than in adults; it is usually accompanied by premature sexual and somatic development. There may be precocious uterine bleeding, abnormal or precocious libido with other signs of mental precocity, over-development of the musculature, breast development in boys (rare) and early precipitous growth in length, followed by premature closure of the epiphyses and subsequent dwarfism, not unlike that of achondroplasia (short extremities in comparison to length of trunk).

The condition is too rare in adult males to deserve a general description, but in adult females it is common and characterized by hirsutism, with beard and moustache growth, loss of scalp hair, deepening of the voice, atrophy of the breasts, amenorrhea, enlargement of the clitoris, and other signs of virilism.

In addition to sexual changes, hypertension, glycosuria and a great tendency to adiposity are very characteristic. The peculiar striation of the thighs and

abdomen is reminiscent of Cushing's disease. The fat deposition is often particularly striking in the upper half of the body and the face, but it may be very marked and in that case generalized. The characteristic, somewhat flushed, round "moon face," of patients suffering from adrenogenital syndrome, tends to render them somewhat similar to each other.

Metabolism. — The B.M.R. is usually normal. In the Achard-Thiers syndrome, there is HYPERGLYCEMIA AND GLYCOSURIA, accompanied by a marked decrease in glucose tolerance. However, glycosuria is comparatively rare among patients suffering from adrenogenital syndrome.

One of the most common manifestations of this syndrome is adiposity, but the fundamental nature of the LIPID METABOLISM disturbance has not yet been fully clarified. Sometimes, there is hypercholesterolemia.

Changes in ELECTROLYTE AND WATER METABOLISM rarely occur, but if present, are very characteristic. There may be hypokalemia, hypernatremia, hypochloremia and a rise in the CO_2 -combining power of the blood.

Growth and Bone Structure. — Usually children suffering from adiposogenital syndrome, grow very rapidly, at first. Frequently, however, there is premature ossification of the junction cartilages and this interferes with subsequent growth in length.

Cardiovascular System. — Hypertension is not uncommon in cases of adrenogenital syndrome, although it is by no means a constant finding. It tends to be accompanied by plethora and cardiac enlargement. Removal of the causative adrenal growth, restores the blood pressure to normal in many, though not in all, instances. Apparently, the pressure remains high, even in the absence of excess corticoids, if the anatomic cardiovascular changes have progressed too far.

Respiratory Organs. — Deepening of the voice is a very characteristic sign of the adrenogenital syndrome, especially in females. It is evident, not only in children, whose larynx is still in the process of growing, but, to a lesser extent, also in adult women. It is due to an actual anatomic transformation of the larynx from the female into the male type and rarely shows considerable improvement following ablation of the causative adrenal growth.

Muscles. — Muscular development and strength are often excessive in patients with the adrenogenital syndrome. This is particularly striking in women and children. The designation "enfants hercules" is often used to describe such children.

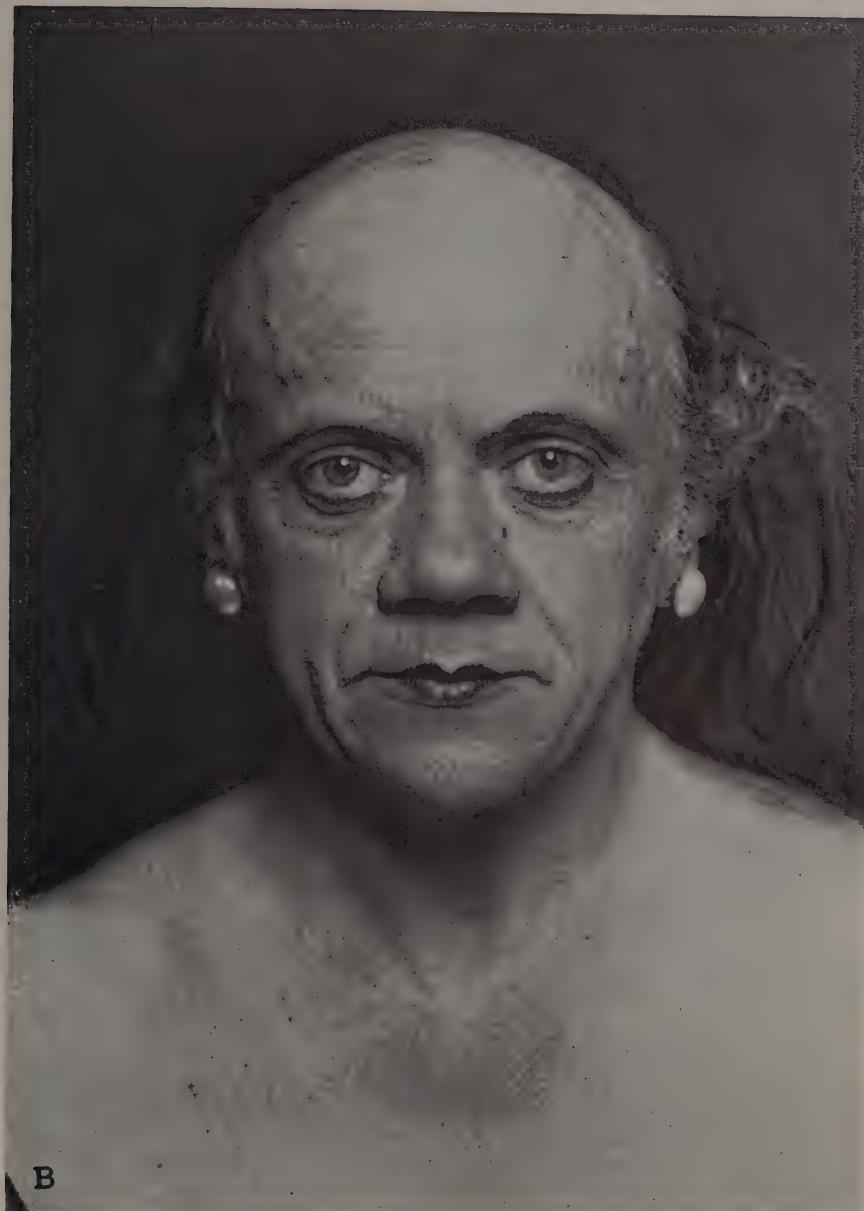
Nervous System. — Corresponding to the somatic development, precocious or homosexual libido and narcissism are frequently seen, although in some patients the mentality is not involved. When the sexual impulses become pathologic, due to an adrenal tumor, ablation of the latter may normalize the psyche. Women who merely lose their feminine libido, as a result of hypercorticoidism, may regain it after ablation of the growth and may subsequently even become pregnant. Usually, however, such refeminization is incomplete or absent if the derangement is of long duration.

Curiously, with the adrenogenital syndrome, homosexual tendencies are almost exclusively seen in women. Since in these, the development of the clitoris is particularly prominent, it is perhaps justified to consider the possibility that this somatic change, rather than a direct effect upon nervous centers, is responsible for the homosexual tendencies. The excessively developed, and highly sensitive, clitoris of these patients is constantly exposed to stimulation by rubbing against clothing or incidental touching, and the women soon learn that more intense

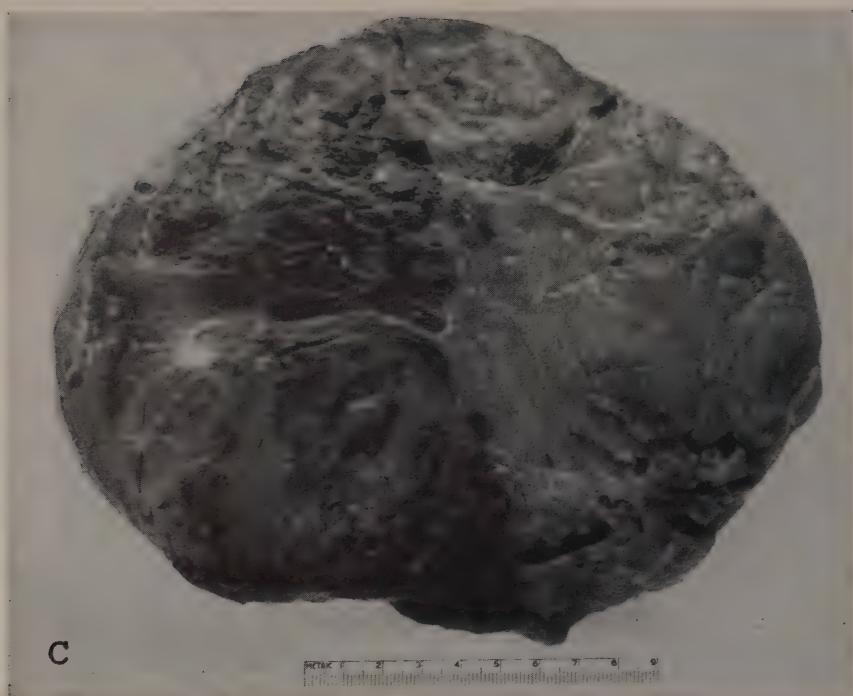


Adrenal virilism. — A. A 45 year-old woman with amenorrhea, hirsutism and obesity of about twenty years' duration. Note large nipples and imprints from clothing. (Cont'd.)

(Courtesy of Dr. E.-J. Kepler).

**B**

— B. Marked apical baldness and masculine facies. (Hirsutism of face not evident because of repeated electrolysis.) (Cont'd.)



— C. Macroscopic aspect of adrenal tumor which was successfully removed. Menses recurred three months after operation and became regular.

orgasm is obtained by the stimulation of this organ, than of their usually hypoplastic vagina. Thus, the patient gradually comes to seek means of satisfying her desire by using the clitoris as a penis, even without there being any fundamental change in her female libido.

The intellectual development of children with adrenogenital syndrome is rarely precocious but they tend to be very immodest and aggressive. The pertinent patients I have seen were all strikingly unattractive, arrogant brats. They present difficult social problems because they have the sexual urge of the adult without the adults' inhibitions.

Skin. — HIRSUTISM is one of the most characteristic signs; in very mild cases, it may be the only obvious manifestation of the disease. Growth of hair on the upper lip and chin, temporal or apical baldness, as well as the typically male pubic hair line, are manifestations of virilism characteristic of the adrenogenital syndrome in women. In both

sexes, there is also a great tendency to a particularly luxurious development of pubic and axillary hair and of hair on the thighs, calves and chest.

Follicular HYPERCERATOSIS, COMEDOS and ACNE are likewise common in such patients. Sometimes there is MELANODERMA, reminiscent of Addison's disease. This may be either generalized or patchy; it is not due to secondary destruction of the adrenals, since it can appear even in patients in whom only one adrenal is affected.

There was some controversy concerning the occurrence of CUTANEOUS STRIAE in the pure adrenogenital syndrome, due to a primary cortical tumor. Some investigators believed that these striations were characteristic of Cushing's disease with secondary adrenal involvement and represented a differential diagnostic criterion between this and the primarily adrenogenic hypercorticism. It has been definitely shown, however, that they can occur



A

"Feminizing" Adrenal-Cortical Carcinoma.

— A. Man, age 44 years, with carcinoma of the left suprarenal cortex and distinct gynecomastia. There was impotence and complete disappearance of libido. The breasts consisted of fibrous tissue, with scattered glands. The excretion of testoids and especially of folliculoids (up to several thousand M.U./day) in the urine was increased. It is doubtful whether one should speak of "feminization" in such cases, since breast enlargement of this same type is seen in patients chronically treated with excess testoids and those suffering from the Klinefelter syndrome (see: Diseases of the Testis). — B. Atrophic testis with almost completely hyalinized tubules and nearly complete absence of Leydig cells. — C. Part of the adrenal carcinoma showing marked polymorphism of cellular elements. In the center are groups of large cells with hyperchromatic nuclei.

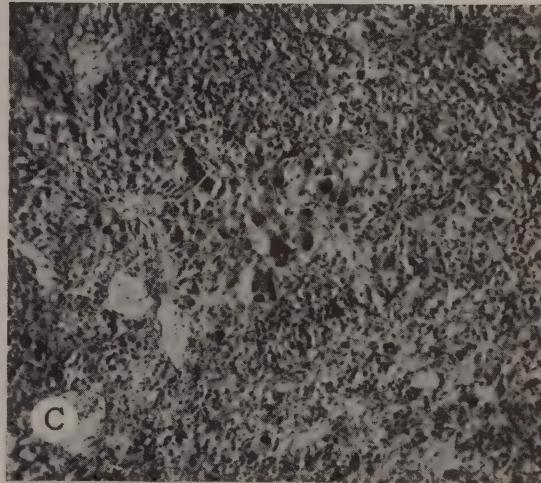
(Courtesy of Drs. K. Rohalm and G. Teilum).

both in primary and in secondary (hypophysogenic) cortical hyperfunction, indeed they disappear after ablation of the causative cortical neoplasm.

Urinary System. — Enlargement of the kidneys, sometimes with signs of nephrosclerosis, are mentioned in many autopsy reports of patients with adrenogenital syndrome. However, the pos-



B



C

sible relationship between cortical hyperfunction and nephrosclerosis has only recently been recognized and hence most pathologists paid no special attention to the condition of the kidneys.

Renal calculi are common in patients who develop osteoporosis as a result of hypercorticoidism.

Sex Organs.—In FEMALE children the precocious, and usually abnormal, development of the sex organs is a characteristic and constant sign of the adrenogenital syndrome. The *vulva* is greatly enlarged, pubic hair appears very early (frequently showing the male type of distribution), and the *clitoris* is greatly hypertrophied often resembling a hypospadiac penis. The female *prostate* and *seminal vesicles* may be well developed, sometimes communicating with the urethra through ejaculatory ducts. Hence, actual ejaculation is possible, although, of course, the seminal fluid contains no spermatozoa, since these pseudohermaphrodites do not possess testes. If the condition develops very early during embryonic life, the vagina may be extremely hypoplastic and the labia majora tend to unite, more or less completely, in the mid-line; under the clitoris, so that the genital organs become very similar to those of the male. In fact some of these patients can be mistaken for boys with hypospadias and undescended testes. Provided there is no tumor, it may be best not to operate, and to bring these children up as "males," since after successful surgery they lose their masculine characteristics, without showing any definite female differentiation and hence become even more ambisexual.

Several cases are known in which *uterine bleeding* occurred in children less than four years of age. There are no adequate histologic studies of the accompanying endometrial changes but it appears unlikely that these bleedings represent true menstruation, since the ovaries do not mature precociously and ovulation or corpus luteum formation does not occur in the adrenogenital syndrome of children. Yet, there is the possibility of luteoid formation by the adrenal cortex.

The *mammary glands* may develop precociously in children but their development, like the precocious uterine

hemorrhages, may cease at a later stage, when the heterosexual differentiation becomes more evident.

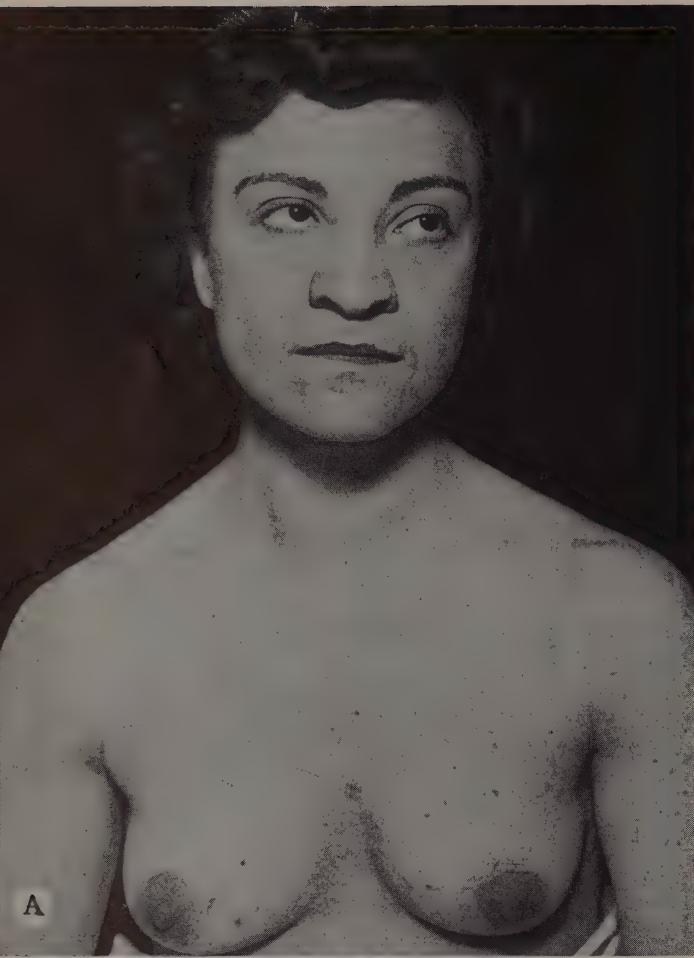


(For legend see p. 173)

**Adrenal carcinoma.**

A and **B**, 25-year-old woman with adrenal carcinoma of long standing which caused hirsutism, atrophy of the breasts, acne, purple striations, hypertension and finally death, due to metastases to the lungs. Note dropping eyelids and mouth, as well as characteristic short greasy hair, with recession of the hairline at the temples.

(After E.-G. Kepler and E.-H. Rynearson, *M. Clin. North America*, 24, 1035, 1940.)

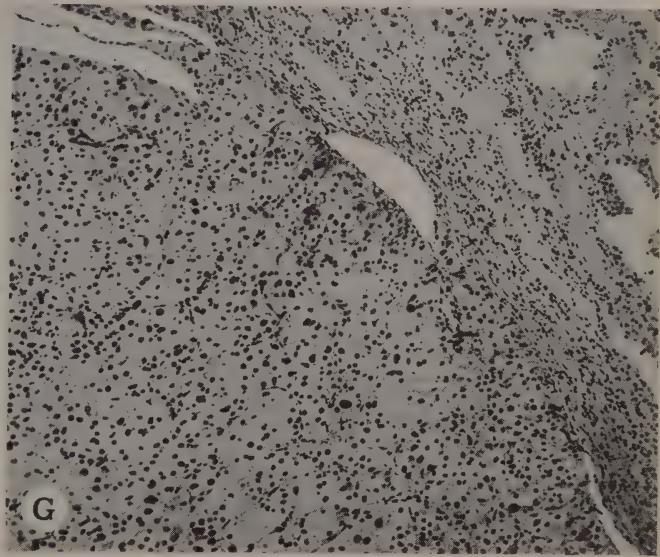




E



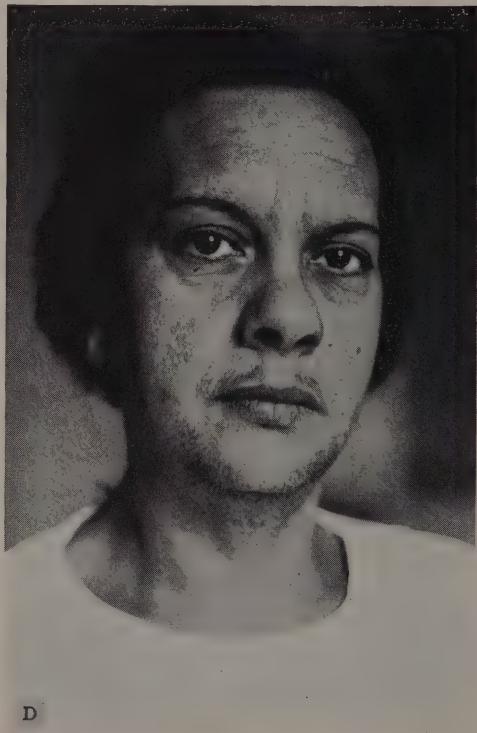
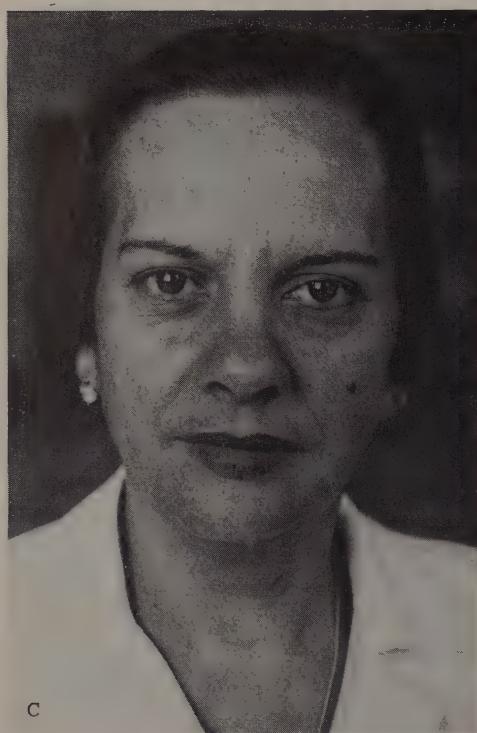
F



G

Metastasizing adrenal-cortical carcinoma. — A. and B. Preoperative appearance of woman, age 30 years, with virilizing adrenocortical carcinoma. Abnormalities included: amenorrhea, hypertrichosis of face (mild) and extremities (moderate), deepening of voice, masculine appearance of face, receding of temporal hair line, acne, arterial hypertension 170/100, polycythemia and deficiency of vaginal smear. Urinary 17-KS up to 123 mg./24 hrs.; also demonstrated in urine: Δ^5 -Androstene-3(β), 17(α)-diol and Δ^5 -Pregnene-3(β), 20(α)-diol. — C. Enlargement of clitoris. — D. Macroscopic appearance of adrenal neoplasm, which was surgically removed. — E. Patient died from recurrence and metastases 6 months after operation. Note gross appearance of huge, yellow hepatic metastasis at autopsy. On piece of gauze, lying over the intestines, is seen recurrent mass at site of removed adrenal carcinoma. There were additional metastases in several other organs. — F. Several round, circumscribed metastatic tumor nodules in the lung as seen under low magnification. — G. Borderline between lung and tumor tissue. Note great polymorphism of cellular pattern in the latter.

(Courtesy of Dr. E.-P. McCullagh).



Adrenogenital syndrome. — A. Patient at 17 years of age when she was obese, but otherwise normal. — B. Patient at 23 years of age, beginning hirsutism under the chin. — C. Patient at 41 years of age. Hirsutism extends to mustache and there is loss of weight. — D. Patient at 42 years of age, when hirsutism and loss of weight became aggravated. About 6 years prior to this, she developed insulin-resistant diabetes, muscular atrophy and osteoporosis. The urinary 17-KS is high (20 mg./24 hrs.). (Cont'd.)

(Courtesy of Dr. A.-B. de Ulhôa Cintra).



— E. Hypertrophy of clitoris. Removal of a cortical adenoma led to no significant improvement.



Juvenile adrenogenital syndrome. — A. and B. Apparently normal girl aged 16 months (A) and 1½ years (B) respectively. — C. Age 3 years 4 months, height 44", weight 58 lbs., muscles became large and strong. Voice excessively deep. Skeletal maturity approximately 11 years. Clitoris hypertrophy. Breast tissue palpable 4" diameter. Distinct evidence of pubertas praecox. Large palpable mass in right flank. Urinary 17-KS 1.2 mg./24 hrs. — D. 6 months post-operative; almost complete disappearance of pubic hair. Palpable breast tissue disappeared almost completely. Clitoris enlargement remains. (Same scale applies to C and D.). (Cont'd.)

(Courtesy of Dr. E.-P. McCullagh).



— E and F. Gross appearance of adrenal tumor.





B



C

Adrenogenital syndrome in a boy.
— A. 11-month-old infant with adrenal-cortical tumor. Note round "moon-face", excessive hair growth on forehead and drooping of the angles of the mouth ("sun-fish mouth" in Kepler's terminology). — B. Note stocky build and well-developed genitalia. — C. Same boy ten months after removal of the adrenal tumor. Note change in facies and body configuration as well as decrease in the size of the genitals. The boy did not grow during this period. — D. Macroscopic view of the removed neoplasm.

(Courtesy of Dr. E.-J. Kepler).



D



Adrenal virilism. — A. Appearance of woman with cortical carcinoma and virilism prior to operation. Note marked acne and roundish face. (Cont'd.)

(Courtesy of Dr. E.-J. Kepler).



— B. Same patient 2 years after removal of the adrenal tumor. Note disappearance of acne and change in facies.

In adults, the ovaries and female accessory sex organs (e.g., uterus, vagina, mammary gland) tend to be atrophic, and there is usually amenorrhea, probably as part of the pseudohermaphroditism.

In the MALE, the development of the sex organs show abnormalities which may be regarded as a counterpart, of what has just been said about the female. If the condition develops during early embryonic life, the external genitalia acquire a heterosexual character, so that one may easily mistake the atrophic and the hypospadic *penis* for a relatively large clitoris and vagina. In these very young pseudohermaphrodites, the *testes* are normal for the patient's age, but in adults they tend to be atrophic.

Excessive mammary gland development in pseudohermaphroditic males may sometimes be accompanied by actual lactation. In one such case, the enlarged mammary glands underwent rapid atrophy, following successful operation. It must be kept in mind, however, that testoids also cause mammary development and hence, breast growth in a male is not necessarily a true feminine characteristic, but may be merely due to excessive male hormone production.

COMPLICATIONS

Tumor METASTASES or adrenal hemorrhages, due to infiltrating cortical carcinomas may complicate the course of the adrenogenital syndrome. Fatal complications may also occur as a result of invasive growth into the adjacent kidney tissue.

An important "complication" of the adrenogenital syndrome in females is SUICIDE. The profound effect of heterosexual development upon the female mentality must not be underestimated. Even mere hirsutism may sufficiently disfigure the patient to cause serious disturbances in her mental equilibrium.

CARDIOVASCULAR COMPLICATIONS due to chronic hypertension, are comparatively rare in the adrenogenital syndrome. If renal hypertension of primarily adrenocortical origin should prove to be a common condition, the cardiovascular complications such as myocardial infarcts, cerebral hemorrhages, arteriosclerosis, periarteritis nodosa, etc., would assume importance.

DIAGNOSIS

The diagnosis of the adrenogenital syndrome is based upon :

- (1) The clinical manifestations of the disease.
- (2) Local signs of a lesion in the adrenal region.
- (3) Detection of an excessive cortical hormone production, by bioassay or chemical analysis of the urine.
- (4) Differential diagnostic considerations.

(1) **The Clinical Manifestations of the Adrenogenital Syndrome.** — In typical cases, the symptoms and signs, as described under "Clinical Course" are so characteristic that the diagnosis can be made, with a high degree of probability, without further investigation. Particularly valuable criteria are : the precocious and often heterosexual development in children and the pseudohermaphroditic traits in adults, especially if accompanied by hypertension, plethora and diabetes.

(2) **Local Signs of a Lesion in the Adrenal region.** — These are essentially the same as those described in connection with the diagnosis of Addison's disease; hence, we shall limit ourselves here to local signs particularly characteristic of hypercorticoidism as such.

Local manifestations in the adrenal region are most common in cases of adrenogenital syndrome due to cortical tumors. Here, a palpable or radio-



Aerograms of the adrenal. — A. Normal shadow of the adrenal. — B. Shadow of an enlarged adrenal.

(Courtesy of Drs. M. Malenchini, E.-B. del Castillo and J. Roca).

logically demonstrable displacement of the kidney or an increased area of density, detectable by percussion, may be of diagnostic value. Indeed, especially after air injections, the adrenal tumor itself may be radiologically visible. Sometimes, the tumors are so large that they deform the lumbar region and compress adjacent nerves or vessels, thus eliciting lumbar pain and additional local signs. In doubtful cases an exploratory laparotomy may be necessary.

(3) **Detection of an Excessive Cortical Hormone Production by Bioassay or Chemical Analysis of Urine or Blood.** — An increased excretion of 17-KS (especially dehydro-iso-androsterone) as judged by chemical tests, or of TESTOIDS, as judged by bioassays, is frequently, though not always, demonstrable in the adrenogenital syndrome. High $3(\beta)$ -hydroxy-17-KS are characteristic of cortical neoplasms, while normal levels usually, but not necessarily, exclude cortical tumors. (See also: The Steroids, The Testis.)

In a few instances, an increased urinary elimination of FOLLICULOIDS is noted

in the adrenogenital syndrome, both in women and in men, irrespective of the presence or absence of "feminization."

The urinary elimination of PREGNANEDIOL, and other biologically INACTIVE STEROID DERIVATIVES, is frequently augmented in the adrenogenital syndrome, but further studies concerning the metabolism of the cortical hormones will be necessary before the significance of such tests can be properly evaluated. Increased urinary excretion of LIFE-MAINTAINING CORTICOIDS, as judged by the cold resistance or glycogen deposition tests, is often very marked in hypercorticoidism of the Cushing syndrome type and may assume diagnostic significance.

(4) **Differential Diagnostic Considerations.** — Among the ovarian tumors the ARRHENOBLASTOMA may lead to virilism, similar to that seen in the adrenogenital syndrome. However, since this neoplasm consists of testicular elements, proliferating within the ovary, it causes a purer syndrome of virilization than the adrenal tumors and is unaccompanied by obesity, hypertension, striae, osteoporosis and diabetes. In doubtful cases, pelvic or rectal exam-

ination, or even exploratory laparotomy, may be necessary to ascertain the diagnosis.

The so-called "OVARIAN HYPERNEPHROMA" is indistinguishable by its hormonal manifestations, from the adrenogenital syndrome of adrenal origin. This is not unexpected since both tumors actually consist of the same type of adrenal-cortical tissue, orthotopic in one case, ectopic in the other. Only local manifestations of a tumor in the ovarian region are of differential diagnostic significance.

LUTEOMAS AND LEYDIG CELL TUMORS OF THE OVARY are also very difficult to identify. The former may be indistinguishable from the hypernephroma of the ovary, even histologically. The latter usually cause pure virilization, as does the arrhenoblastoma. As these neoplasms are extraordinarily rare, they are not of great diagnostic importance.

TRUE HERMAPHRODITISM is so rare that, for practical purposes, it hardly needs to be considered. It is always congenital and reveals only sexual disturbances, without the metabolic and cardiovascular manifestations of the adrenogenital syndrome. Usually the accompanying malformations of the sex organs are much more severe than with adrenal tumors.

TRUE OVARIAN PRECOCIOUS PUBERTY is also rare. It is characterized by a premature development of the ovary, with follicle maturation, ovulation and corpus luteum formation. This is accompanied by more or less regular menstrual cycles and may result in pregnancy in girls as early as the eighth year of life.

FOLLICULOMAS, or persistent FOLLICLE CYSTS, tend to cause precocious "pseudopuberty" in infants. They are unaccompanied by precocious ovulation or the heterosexual, metabolic and cardiovascular manifestations of the hypercorticoïd syndrome.

TUMORS OF THE HYPOTHALAMUS and adjacent areas, often present diagnostic difficulties because they may elicit heterosexual characteristics, accompanied by hypertension (in this case, due to increased intracranial pressure). The differential diagnosis between this condition and the adrenogenital syndrome is hardly ever possible unless local signs reveal the site of the tumor; that is, increased intracranial pressure signs or radiologic evidence of an intracranial neoplasm in one case, signs of a tumor in the adrenal region in the other.

PINEAL TUMORS usually develop in children under 12 years of age and also tend to cause precocious sexual maturity. They have never been observed in females nor have they been found to elicit pseudohermaphroditism. The somatic development is precocious but proportionate. The B.M.R. is frequently increased. In early stages there may be generalized adiposity but later the patients become cachectic and local signs of intracranial pressure develop.

CUSHING'S SYNDROME, resulting from a basophil adenoma of the anterior-lobe, (i.e., Cushing's Disease), presents the most serious differential diagnostic difficulties. In fact, there are many cases which may be considered to be intermediate types between Cushing's syndrome and the adrenogenital syndrome; at least they have features of both, inasmuch as basophil anterior-lobe adenomas and adrenal-cortical adenomas develop simultaneously in the same patient. In such instances, the entire symptomatology of the two syndromes is identical, except for the possible (rather rare) presence of local signs in the pituitary region (compression of the optic chiasma, increased intracranial pressure, radiologically detectable deformation of the sella). In characteristic cases of Cushing's disease, the increased red cell count, osteoporosis of the skull and spine and the metabolic manifestations tend to be

more prominent, while the sexual abnormalities are less conspicuous than in the adrenogenital syndrome. Yet there is no symptom or sign (not excluding the blood chemical changes) which could not occur in either malady with the possible exception of the clitoris enlargement; this apparently never occurs in Cushing's disease.

Certain THYMUS TUMORS may occur in conjunction with an otherwise typical Cushing's syndrome. In these, only local signs in the thymus region (dullness on percussion, X-ray shadow) permit the differential diagnosis.

PROGNOSIS

The prognosis of the adrenogenital syndrome depends largely on the causative adrenal lesions. In the case of simple hyperplasia, the course may extend over a whole lifetime, without causing any very serious disturbance. On the other hand, adenomas or carcinomas tend to progress.

Following early surgical ablation of adenomas or carcinomas, the prognosis is favorable if it is technically possible to remove all the tumor cells and if the patient survives the immediate shock of operation.

THERAPY

The indications for the SURGICAL TREATMENT of hypercorticoidism differ according to the nature of the underlying adrenal abnormality.

Malignant tumors of the adrenals should, of course, always be removed, whenever technically possible. Cortical carcinomas may cause great difficulties in this respect, owing to metastases or direct invasion into the renal vein, kidney and other adjacent tissues. Every effort should therefore be made to recognize malignant tumors, at an early stage, by surgical exploration of the adrenal region in all suspicious cases.

Heavy doses of cortical extract and desoxycorticosterone should be admin-

istered preventively several days before and after the operation, since the contralateral adrenal may be severely atrophic in the presence of a tumor in one gland. It is also important to explore the other adrenal, even if one gland has proven to be the bearer of the presumably causative tumor. In a few cases of adrenogenital syndrome there are bilateral, primary tumors, while in others, one adrenal may be completely destroyed by a growth which secondarily involves the other adrenal as well.

The prognosis of patients subjected to total bilateral adrenalectomy is extremely grave. Even if maximal doses of cortical extracts are administered, it is practically impossible to adjust the corticoid therapy to the varying requirements of everyday life. Very few patients have survived total bilateral adrenalectomy for more than a few weeks in spite of hormone therapy. Information, however, is limited almost entirely to the cases published by Huggins, who performed total adrenalectomy to alleviate cancer of the prostate.

Adrenal-cortical hyperplasia, with Cushing's syndrome, presents the problem of restoring the usually poor health of the patient. Reduction of the total mass of adrenal-cortical tissue by partial adrenalectomy (removal of one or partial resection of both glands), though logical, has rarely yielded very satisfactory results. Total bilateral adrenalectomy should not be tried until we learn more about substitution therapy.

Adrenal-cortical hyperplasia with postpuberally acquired pseudohermaphroditism does not tend to render the patient seriously ill and the external genitalia show only minor changes, such as enlargement of the clitoris. In these cases, extensive resection of adrenal tissue is usually inadvisable, because of the surgical risk and the probability of postoperative deterioration in the patient's health. Some in-

vestigators reported satisfactory results in similar instances, but usually the virilism (including hirsutism) tends to persist, although a transitory amelioration may occur.

Adrenal-cortical hyperplasia with prepuberally acquired pseudohermaphroditism, in its *mild forms*, is generally no indication for adrenal surgery, again because of the good health of these patients and the usually unsatisfactory correction of the virilism by partial adrenalectomy. In most pertinent cases, plastic surgical interventions, on the deformed external genitalia, give the most satisfactory results.

In the more or less *complete forms* of this type of pseudohermaphroditism, the patients develop into normal, vigorous "men," although legally and in the Catholic and Greek Orthodox Churches, they are regarded as women, if the female sex of the gonad is established by surgical exploration. Partial adrenalectomy or surgical attempts to make the external genitalia conform with the female sex of the gonads fail. Both physically and psychologically, these patients can make fairly normal "men," but invariably develop into very poor women. It must be remembered that these individuals are chemically and in most of their anatomic features, of the male sex. Plastic operations, psychotherapy and social arrangements facilitating their life as "men" are generally most satisfactory to the patient.

Hyperplasia of the adrenal cortex is frequently accompanied by the formation, usually multiple, of very benign adrenal adenomas. Since these have little or no tendency to undergo malignant transformation, they do not significantly alter the surgical indications as outlined above. On the other hand, if single, large adenomas are detected at exploratory laparotomy, they should be enucleated, as this is a simple operation and such neoplasms often have a

rapid growth rate and may become malignant.

Among other possible therapeutic measures, the administration of FOLICULOIDS may be mentioned, since these have been reported to be beneficial in some cases of virilism but their effect is usually very doubtful.

X-RAY TREATMENT of the hyperplastic adrenal may give beneficial results, but its value is so dubious that it cannot be recommended except for inoperable cases. Adrenal-cortical cancers are singularly resistant to X-rays.

The administration of high carbohydrate, low protein and low sodium DIETS, especially in combination with some acidifying salt such as ammonium chloride, would appear to be justified on the basis of experimental evidence (see : pages 122 and 126), since such diets decrease corticoid hormone production and antagonize the toxic effects of corticoids in animals. Up to the present, these dietary measures have not been subjected to adequate clinical trials.

As SYMPTOMATIC THERAPEUTIC MEASURES, electrolysis, X-ray treatment, diathermia or depilatory treatment of the hirsutism may be recommended in otherwise incurable cases and in those not sufficiently severe to justify a surgical intervention.

SPONTANEOUS HYPERCORTICOIDISM IN ANIMALS

Cases of pseudohermaphroditism, of presumably adrenal origin, have been noted for instance in the fowl. However, most cases of ambisexuality in animals are due to true bisexual development of the gonads (true hermaphroditism) or to ovarian and testicular tumors, while adrenal pseudohermaphroditism is extremely rare.

The spontaneous occurrence of adrenal tumors accompanied by cardiovascular lesions and periarteritis nodosa have also been described in animals.

HYPERADRENALINISM

(*Synonyms*: hyperepinephrinism, suprarenal sympathetic syndrome. — The terms, adrenal-medullary tumor, chromaffinoma, pheochromocytoma and paraganglioma of the adrenal are not truly synonymous with hyperadrenalinism. They merely designate the tumor itself, which usually elicits the syndrome.)

DEFINITION

Hyperadrenalinism is a condition in which the hormone production of the adrenal medulla is sufficiently increased to produce detectable symptoms of overdosage.

This disturbance is usually due to a benign adrenal-medullary tumor, developing from the chromaffin cells. Sometimes it may result from hyperplasia of the adrenal medulla and even from tumors or hyperplasia of the extra-adrenal, paraganglionic chromaffin tissue.

CLASSIFICATION

The only customary classification of hyperadrenalinism is according to the structure of the underlying ADRENAL LESION. Thus we distinguish :

- (1) Benign pheochromocytomas.
- (2) Diffuse hyperplasia of the adrenal medulla.
- (3) Malignant pheochromocytomas.
- (4) Paragangliomas outside the adrenal.

All but the first of these four types are great rarities.

INCIDENCE

Hyperadrenalinism is a rare disease, although many more cases have been observed than the approximately 150, which have been described in the literature. Among these, only very few were due to hyperplasia, medullary carcinomas or extra-adrenal paragangliomas.

As far as one can tell from the small series of published cases, hyperadrenalinism occurs with about equal frequency in both sexes. It can develop at any age, but is most common in middle-aged patients.

PATHOLOGIC ANATOMY

The morphologic characteristics of the underlying adrenal lesions will be discussed in the section on adrenal tumors and hence, need not be reconsidered here. (See p. 191.)

PATHOGENESIS

Nothing is known about the primary cause of hyperadrenalinism. When it is due to a tumor, its etiology is as cryptic as that of other neoplasms; when it results from diffuse hyperplasia we know even less about its ultimate pathogenesis than about that of most other endocrine hyperplasias, since the adrenal medulla is not stimulated by any specific trophic hormone which could act as a causative agent. Theoretically, it is possible that chronic excitation of the secretory nerves could lead to functional hyperadrenalinism, but this, if it occurs, must be rare, since almost all published instances of this disease were due to demonstrable chromaffin tumors.

Except for the cause of the chromaffin-cell-stimulation the pathogenesis of the syndrome is readily understandable, since all the symptoms and signs are manifestly due to increased adrenaline production.

That the spells often occur following exposure to diverse noxious agents, is not unexpected, since even the normal medullary cells discharge excess amounts of adrenaline in the first stage of the "alarm reaction." The principle characteristic of hyperadrenalinism is that the stores of adrenaline, and the hormone-producing ability of the

chromaffin cells, are very high, so that the normal emergency hyperadrenalinemia is greatly exaggerated.

CLINICAL COURSE

State. — The clinical picture of hyperadrenalinism of endogenous origin, is practically identical with that of exogenous adrenaline intoxication. There are: paroxysmal spells of extreme hypertension, severe cardiac palpitation, emotional disturbances, sweating, nausea, headaches, abdominal pain, mydriasis, intense pallor, hyperglycemia with glycosuria and, sometimes, lung edema. It is particularly characteristic that even if the condition is relatively mild, severe paroxysms are readily elicited by any physical or psychic stress likely to cause adrenaline liberation. Even mere palpation of the abdomen or straining to empty the bladder and especially histamine or insulin injections can induce severe spells.

Metabolism. — Spells of HYPERTHERMIA are frequent during the paroxysms of hyperadrenalinism. In one patient, the practically normal basal temperature rose to 107° F. as a result of excitement. This rise in body temperature is accompanied by a corresponding increase in the B.M.R.

Other important and frequent signs of this disease, are GLYCOSURIA, HYPERGLYCEMIA and mild ALBUMINURIA. The latter may be due to vascular disturbances within the kidney.

Blood Picture. — There may be some erythrocytosis but this is inconstant.

Cardiovascular System. — Spells of HYPERTENSION are among the most constant clinical manifestations of hyperadrenalinism. They are usually transitory and appear in combination with other symptoms of adrenaline intoxication, such as pallor, hyperthermia, faintness, lung edema, mydriasis, etc. Occasionally, however, a more

permanent increase in blood pressure is observed, which sometimes is as high as 260 to 300 mm. of mercury (systolic). In these cases there is often hypertensive retinitis. The B.M.R. remains permanently very high (up to +70 to +100%), while spells no longer occur.

The pallor is due to the sudden constriction of the minute cutaneous BLOOD VESSELS, readily demonstrable with the capillaroscope. Generalized arteriosclerosis has been observed in several cases; this is particularly striking in children who otherwise rarely show any such vascular lesions. The phenomenon is apparently the clinical counterpart of the adrenaline sclerosis produced in animals by continuous overdosage with the hormone.

Hypertrophy of the HEART is an almost constant finding and probably a result of the hypertension. The pulse rate increases rapidly during the spells, just as it does following exogenous adrenaline administration. In a few cases, however, marked bradycardia has been noted during the attack. This is understandable since adrenaline may also elicit either tachycardia or bradycardia, according to dosage and the responsiveness of the individual. These changes in cardiac rhythm frequently elicit painful sensations and palpitation in the chest.

Respiratory Organs. — Acute, hemorrhagic LUNG EDEMA is a very characteristic sign of experimental adrenaline intoxication and is also encountered during the spells of spontaneous hyperadrenalinism. It is frequently the immediate cause of death. The lung edema may be so profuse that blood-stained foam fills the trachea and mouth and the patient actually suffocates in his own lung edema fluid.

A marked increase in the RATE OF RESPIRATION is frequently noted during the spells, even in the absence of lung edema.

Other Changes. — Among other manifestations of hyperadrenalinism, multiple small HEMORRHAGES into the mucosa of the gastrointestinal tract, the liver and the kidneys are worthy of mention. PIGMENTATION OF THE SKIN is rare, although it has been observed occasionally, especially in the (comparatively common instances) in which hyperadrenalinism is combined with NEUROFIBROMATOSIS. Great EMOTIONAL INSTABILITY or profuse SWEATING has also been frequently noted during the attack and this may be accompanied by MYDRIASIS, excessive SALIVATION and ANURIA.

COMPLICATIONS

The complications of hyperadrenalinism usually occur during the spells. They consist of fatal lung edema, the rupture of a blood vessel or sudden death due to cardiac irregularities.

DIAGNOSIS

The very characteristic CLINICAL SYMPTOMATOLOGY of hyperadrenalinism renders it readily recognizable if the physician remembers to think of this rare disease as a possibility.

In suspicious cases we must search for LOCAL SIGNS indicative of an adrenal tumor. The pertinent diagnostic technics are discussed in the chapter on Addison's disease. It is especially important to establish the side on which the tumor is located, in order to decide upon a correct surgical approach. Great care should be taken not to precipitate a fatal adrenaline discharge during the palpation of the adrenal region. It is true that a mild adrenaline discharge, thus produced, may help the diagnosis by provoking a spell, but this is a rather dangerous procedure.

The DIFFERENTIAL DIAGNOSIS must first consider the possibility of other adrenal tumors. Those of the cortex are almost invariably accompanied by sexual disturbances which are absent in hyperadrenalinism; other adrenal

blastomas (e.g., ganglioneuromas, sarcomas) do not give rise to any clinical manifestations of a hormonal disturbance, unless they destroy both adrenals and produce addisonism. Since even malignant tumors of the chromaffin tissue itself rarely, if ever, give rise to hyperadrenalinism, the identification of the syndrome practically limits the diagnosis to benign chromaffinomas and the (exceptional) diffuse hyperplasia of the medulla.

Other conditions, conducive to spells of hypertension, such as eclampsia, lead poisoning, certain infections, cranial traumas, etc., also have to be considered in formulating the definite diagnosis. In doubtful cases the precipitation of an adrenaline discharge by histamine injection may aid the diagnosis, but this is dangerous. Some ascribe diagnostic importance to the possibility of inhibiting the spells with sympatholytics (e.g., benzodioxanes).

PROGNOSIS

The speed, with which the disease develops, is extremely variable but spontaneous cures hardly ever occur. In the case of surgical therapy, performed with the precautions recommended in the following chapter, the prognosis is favorable. If the tumor is completely removed recurrences do not occur, since the causative neoplasms are almost invariably benign. In the absence of suitable therapy, death usually ensues during one of the spells as a result of cardiac complications, fatal lung edema or a fatal hemorrhage.

THERAPY

The logical therapy of hyperadrenalinism is the SURGICAL REMOVAL of the excessive medullary tissue. This has to be performed with extraordinary care because of the great shock-sensitivity of these patients. It is well to keep in mind that adrenal chromaffinomas may contain several thousand

times the lethal dose of the hormone and a sudden discharge of even a small percentage of this, may cause death. (See: Adrenaline Content of the Adrenals, on page 127.)

It is best not to tell the patient exactly when the operation will be performed and to pretreat with sedatives, so as to avoid any emotional upset. It is imperative to handle the adrenals as little as possible and to ligate their larger vessels before removing the gland, to avoid the sudden outflow of stored adrenaline. Most patients, who succumbed after the operation, did so as a result of the direct adrenaline overdosage precipitated by the oper-

ative intervention. Indeed, the sensitivity of the patients is so great that in many cases, death (usually from lung edema) ensued after minor incidental surgical interventions for other reasons, such as spinal anesthesia, the removal of hemorrhoids, extraction of a tooth or a normal delivery.

X-RAY TREATMENT has been attempted in several cases but is hardly justified unless the patient refuses operation. There is a great danger of provoking fatal adrenaline liberation by the breakdown of the tumor; in the few patients who benefited from X-ray therapy, the improvement was usually only transient.

TUMORS OF THE ADRENALS

DEFINITION

With the adrenals, as in the case of several other endocrine glands, the distinction between hyperplasia and true tumor formation is somewhat artificial because of the many transitional types.

The classical cortical and medullary adenomas and carcinomas, conducive to hyperadrenalinism, are discussed in conjunction with the resulting clinical syndromes in the chapters on hyperadrenalinism and hypercorticoidism, respectively. In this section we shall only attempt a purely morphologic characterization of the adrenal neoplasms.

CLASSIFICATION

The tumors of the adrenals may be classified as follows:

- (A) TUMORS OF THE ADRENAL CORTEX.
 - (1) Adenomas.
 - (2) Carcinomas.
- (B) TUMORS OF THE ADRENAL MEDULLA.
 - (1) Chromaffin tumors.
 - (2) Neuroblastomas.
 - (3) Ganglioneuromas.

(C) TUMORS OF THE ADRENAL STROMA.

- (1) Sarcomas.
- (2) Lipomas.
- (3) Hemangiomas.
- (4) Lymphangiomas.

(D) SECONDARY (OR METASTATIC) TUMORS OF THE ADRENALS.

PATHOLOGIC ANATOMY

(A) TUMORS OF THE ADRENAL CORTEX.

— Presumably, most of the cortical cell tumors originate as adenomas but some of them subsequently become malignant.

(1) SMALL ADENOMAS OF THE ADRENAL CORTEX are so common that it is questionable whether they should be considered as pathologic. It is estimated that one out of every three adults has at least some evidence of cortical adenoma formation. Usually these neoplasms are only a few mm. in diameter and are localized in the outer layer of the cortex. Multiple, minute adenomas are indistinguishable from the so-called "nodular hyperplasia" of the cortex. They rarely produce any manifestations of hypercorticoidism unless they reach a considerable size

and large adenomas are rare. (See p. 163.)

(2) CARCINOMAS OF THE ADRENAL CORTEX are usually devoid of lipid and glycogen. They consist of cells having a distinctly epithelial character, which form strands and bands (like the fasciculata), or acini (like the glomerulosa). Sometimes the cells are arranged more or less irregularly around blood vessels in a "perithelioma-like" fashion. Often the tumors contain many giant cells and bear no resemblance to normal cortical tissue.

Macroscopically, these tumors are grey or greyish-yellow, soft and prone to hemorrhage. They are very malignant and tend to metastasize early, both through lymphatic and blood vessels into lymph nodes, liver, lung, brain and the contralateral adrenal. They often spread into the kidney, frequently through the adrenal and renal veins.

These neoplasms should be distinguished from the so-called "Grawitz tumors," often referred to as "*hypernephromas of the kidney*." The latter are light-yellowish, lipid-containing neoplasms situated within the kidney tissue. Their vacuolized cells bear a striking resemblance to those of the normal adrenal cortex, hence, their discoverer, Grawitz, thought they were adrenal-cortical carcinomas originating from embryologically misplaced cortical primordia, within the kidney tissue. A Grawitz tumor often contains papillae which resemble renal papillomas. It has a marked tendency to invade the kidney veins but causes no hypercorticoidism.

Since the term hypernephroma has been used indiscriminately to designate both renal carcinomas and the cancers of the adrenal cortex, it is preferable to dispense with this designation and to refer to the former as Grawitz tumors and to the latter as adrenal-cortical carcinomas. (See also p. 163.)

(B) TUMORS OF THE ADRENAL MEDULLA. — These develop from endocrine or nervous elements.

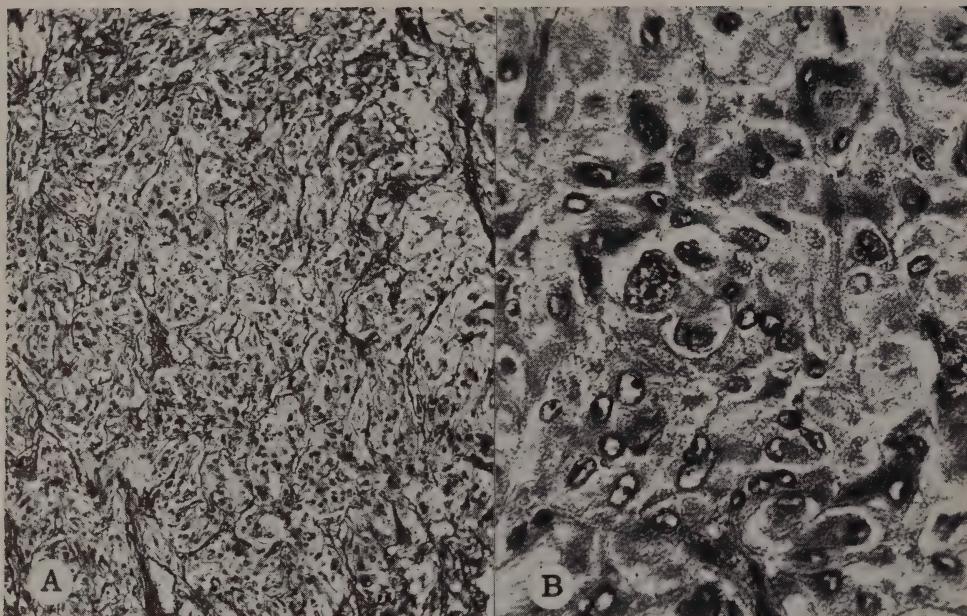
(1) The CHROMAFFIN TUMORS (paragangliomas, chromaffinomas, pheochromocytomas) are rare, benign, epithelial neoplasms (adenomas), characterized by numerous cytoplasmic chromaffin granules. Their cells resemble those of the normal adrenal medulla and they are usually conducive to hyperadrenalinism (see: Hyperadrenalinism). Sometimes they remain asymptomatic and are found accidentally at autopsy.

The name "paraganglioma" has been given to these tumors because they occur, not only in the medulla of the adrenals, but also in extra-adrenal chromaffin tissue such as abdominal paranganglia, Zuckerkandl's organ or the carotid body. Frequently they contain follicle-like formations filled with cell debris and colloid ("struma



Pheochromocytoma. Benign pheochromocytoma which gave rise to typical signs of hyperadrenalinism. Note the uniform, mature aspect of the cells. (Low magnification.)

(Courtesy of Dr. P. Masson).



Pheochromocytoma. Benign pheochromocytoma which gave rise to typical signs of hyperadrenalinism. Note the rather polymorph appearance of the cells which are much more atypical than in the tumor shown in previous figure. — A, low, and — B, high magnification. Note thick capsule separating tumor from normal cortex.

(Courtesy of Dr. P. Masson).



Pheochromocytoma of the adrenal. Large cellular pheochromocytoma of the adrenal medulla. Patient suffered from hyperadrenalinism.

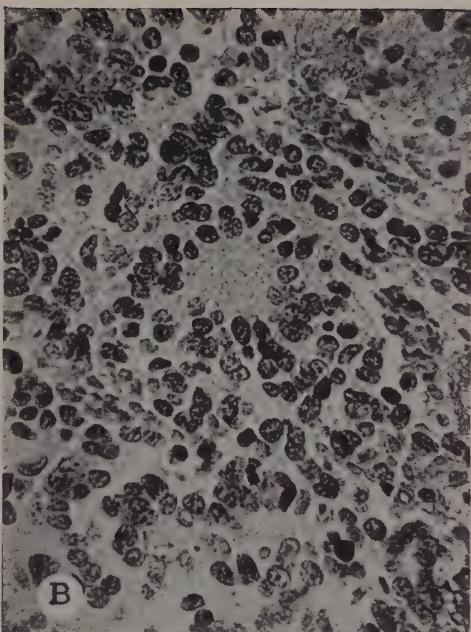
of the suprarenal medulla"). Usually the cells are arranged in the form of trabeculae and solid cell-nests without follicle formation.

Chromaffin tumors of the adrenals are frequently found in patients suffering from v. Recklinghausen's neurofibromatosis. This led to the statement that this neoplasm is "a naevus of the sympathetic nervous system."

(2) NEUROBLASTOMAS (neuroblastoma sympathicum embryonale, immature neurocytoma, sympathogonia, sympathoblastoma) are more immature types of medullary neoplasms, usually found in children or fetuses, hardly ever in adults. Since both the chromaffin cells and the sympathetic ganglion cells develop from the same embryonic sympathogonia, it is understandable that, depending upon the stage of their maturity, the tumors of the medulla may either develop into chromaffin tumors or consist of the less differentiated sympathogonia and sym-



A

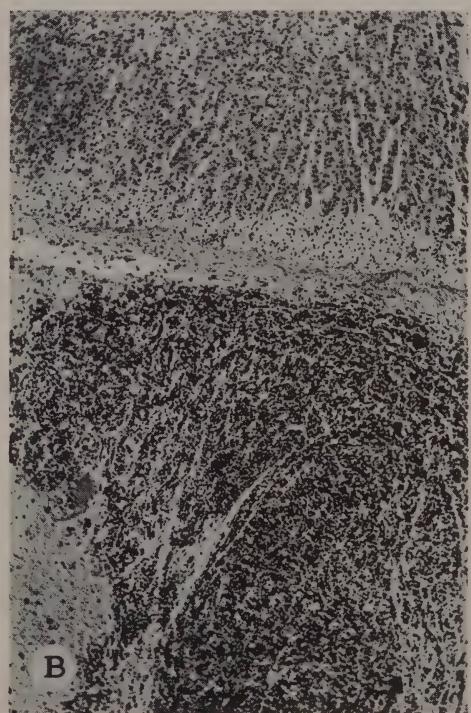


B

Neuroblastoma of adrenal. — A. Typical small cellular neuroblastoma of adrenal medulla. Note rosettes (with light centers). The neoplasm is entirely limited to the medulla; the surrounding cortex is intact. — B. One of the "rosettes" under high power.



A



B

Neuroblastoma of the adrenals. — A. Neuroblastoma of adrenal medulla surrounded by well-preserved, normal cortex. (Very low magn.) — B. Small, dark cells of very undifferentiated embryonic neuroblastoma are clearly separated by a thick connective tissue membrane from the larger, light cells of the adrenal cortex. (Medium magn.)



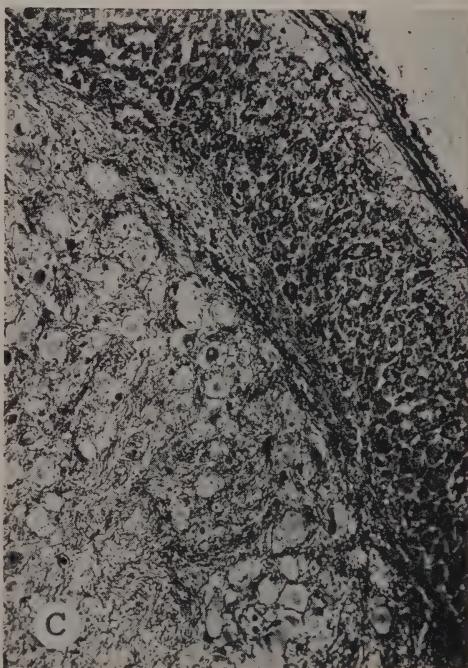
A



B

pathoblasts (see : Embryology, on page 94). They have often been confused with round-cell or lympho-sarcomas. Their round cells are often arranged in rosettes surrounding indistinct fibrils. The neuroblastomas of the medulla are malignant metastasizing neoplasms similar to the corresponding tumors which originate from nervous elements in other regions.

(3) **GANGLIONEUROMAS** are much more mature, typical nerve-tissue neoplasms, consisting of a network of non-medullated nerve fibers, cells of Schwann's sheath and typical ganglion cells. The latter are often arranged in groups resembling ganglia. Ganglioneuromas are usually benign, hard, fibrillar, greyish tumors, found accidentally at autopsy in patients who have died of other diseases. Occasionally, however, they develop in the form of malignant, infiltrating and metastasizing neoplasms. Although these are primary tumors of the adrenals,



C

Ganglioneuroma of the adrenal medulla. — A. 4-year-old girl who died of rhabdomyosarcoma of the pelvis. The ganglioneuroma, limited to the adrenal medulla, was accidentally discovered at autopsy. — B. Higher magnification of cortico-medullary border zone (H. & E.) showing small cortical-cells (right) and large ganglion-cells of the tumor. — C. Section from the same region (Bielschowsky stain) to show fine nerve filaments (dark lines around ganglion cells).

(Courtesy of Dr. P. Masson).

they are essentially similar to neoplasms developing from nervous primordia in other organs. Ganglioneuromas may occur at any age.

(C) TUMORS OF THE ADRENAL STROMA. — SARCOMAS of the adrenals are extremely rare and probably most of the so-called round-cell and lymphosarcomas of the old literature were actually immature types of neuroblastomas (see above).

Much has been written about the usually bilateral MELANOSARCOMAS of the adrenals, but it is difficult to prove whether these actually originated in the suprarenals. In many carefully investigated cases, minute extra-adrenal primary melanosarcomas were found in other organs (e.g., eye, a melanotic naevus).

LIPOMAS, LYMPHANGIOMAS and OTHER NEOPLASMS of the adrenals are rarities.

(D) SECONDARY (METASTATIC) TUMORS OF THE ADRENALS. — It is a curious fact that carcinomas originating in other organs (especially the breast and lung), frequently metastasize into the adrenals and often form deposits simultaneously in both glands. The metastatic adrenal carcinomas may be



Metastatic carcinoma of the adrenal. Note squamous cell carcinoma islet within adrenal-cortical tissue. The cancerous tissue contains cornified "pearls".

the immediate cause of death due to the hypocorticoidism which they produce.

REFERENCES

ADDISON, T.: *On constitutional and local effects of disease of suprarenal capsules.* M. Classics 2, 244 (1937).

ADDISON, T.: "Anemia : disease of suprarenal capsules." M. Classics 2, 239 (1937).

Reproduction of the two classical monographs of Thomas Addison, in which the disease, now bearing his name, was first described. These writings, of one of the fathers of endocrinology, make most interesting and instructive reading, not only from a historic point of view, but also because they show how much can be accomplished, without any of the modern clinical and laboratory facilities, by keen observation of the living and dead patient. They are prominent examples of concise and descriptive medical writing; as such, they are highly recommended to investigators, physicians and students alike.

BROSTER, L. R. AND H. W. C. VINES: "*The Adrenal Cortex. A Surgical and Pathological Study.*" London, H. K. Lewis & Co. Ltd. (1933).

A monograph (94 pages, 4 colored illustrations, and 12 references), mainly concerned with the results of surgical therapy, and the interpretation of adrenal fuchsinophilia, in the adrenogenital syndrome.

CAHILL, GEORGE F., MEYER M. MELICOW AND H. H. DARBY: "*Adrenal cortical tumors. The types of Nonhormonal and Hormonal Tumors.*" Surgery, Gynecology and Obstetrics 74, 281 (1942).

A survey (25 pages, 30 illustrations, 74 references) of personal observations concerning patients with adrenal-cortical tumors. Chief emphasis is laid upon the underlying histologic changes, the value of surgical therapy and diagnosis.

DIETRICH, A. AND H. SIEGMUND: "Die Nebenniere und das Chromaffine System (Paraganglien, Karotisdrüse, Steissdrüse)." Henke und Lubarsch's Handbuch der speziellen Pathologischen Anatomie und Histologie. Drüsen mit innerer Sekretion 8, 951 (1926).

This is one of the best sources of data concerning the pathologic anatomy of the adrenals and paraganglia (138 pages, 46 excellent illustrations, many of them in color, almost the entire pertinent literature up to about 1925). Highly recommended reading for all those interested in adrenal morphology. (In German.)

GOLDZIEHER, MAX A.: "The Adrenal Glands in Health and Disease." F. A. Davis Company Publ. Philadelphia (1944).

A treatise (727 pages, 81 illustrations, numerous references) surveying the morphology, physiology and diseases of the adrenal glands. The greatest emphasis is laid upon the clinical section.

HEARD, R. D. H.: *Chemistry and Metabolism of the Adrenal Cortical Hormones*. In: *The Hormones*. Pincus, G. and K.V. Thimann (Ed.). Academic Press Inc., Publ., New York (1948).

An up-to-date review (80 pages, many tables and charts, 271 references) compiled mainly from the chemist's point of view.

KOHN, ALFRED: "Die Paraganglien." Archiv für mikroskopische Anatomie und Entwicklungsgeschichte 62, 263 (1903).

A monograph on the morphology of the paraganglia (102 pages, 27 illustrations, 74 references) by one of the best observers of these structures. In spite of the date of this paper, it is highly recommended to those interested in this field. (In German.)

LEIBOVICI, RAYMOND: "Chirurgie des surrénales." Laffont: Encyclopédie Médico-Chirurgicale Traité de Médecine et de Chirurgie sur fascicules mobiles constamment tenus à jour. 18, Rue Séguier, Paris (6^e), 10016 (1936).

Synopsis (7 pages, 3 illustrations, no references) of the main indications and surgical techniques of adrenalectomy. (In French.)

LOEB, ROBERT F.: "Adrenal Insufficiency." Bull. New York Acad. Med. 16, 347 (1940).

A brief (20 pages, 9 illustrations, 31 references), but very authoritative summary of adrenal insufficiency from the clinician's point of view.

MONNET, ROBERT: "Contribution à l'étude de la Physiologie normale et Pathologique de la Cortico-surrénale." Imprimerie A. Joyeux, Alger (1941).

A monograph (346 pages, 2 drawings, 1118 references), mainly planned to act as a guide to literature on the experimental physiology of the adrenal cortex. (In French.)

ROWNTREE, LEONARD, G. AND A. M. SNELL: "A clinical study of Addison's disease." Mayo Clinic Monographs, W. B. Saunders Co., Philadelphia (1931).

An extensive, yet very readable, monograph, summarizing most of the important facts concerning Addison's disease, which were known up to the time of the discovery of active corticoid extracts.

SILVER, SOLOMON: "The Cushing Syndrome; neoplasms of the adrenal gland." Bull. New York Acad. Med. 16, 368 (1940).

A stimulating graduate lecture (12 pages, 10 illustrations, 21 references) on personal observations concerning the clinical manifestations of Cushing's disease, with an attempt to elucidate their pathogenesis.

SOFFER, LOUIS J.: "Diseases of the Adrenals." Lea & Febiger, Philadelphia (1946).

A monograph (304 pages, 42 excellent illustrations, some in color, numerous references) describing the morphology, physiology and diseases of the adrenal glands. The main emphasis is laid upon clinical problems.

SWINGLE, W. W. AND J. W. REMINGTON: "The Role of the Adrenal Cortex in Physiological Processes." Physiological Reviews 24, 89 (1944).

A critical review (38 pages, no illustrations, 531 references) of the physiology of the adrenal cortex, written mainly as a guide to the pertinent modern literature.

THADDEA, S.: ... "Die Nebenniereninsuffizienz und ihr Formenkreis." Stuttgart: Ferdinand Enke (1941), Mit einem Geleitwort von G. v. Bergmann.

Monographic treatise (232 pages) of Addison's disease and associated syndromes. (In German.)

VACCAREZZA, AMERICO J.: "Histofisiologie de la Corticosuprarrenal." Medicina 5, 3 (1945).

Review (48 pages, 22 illustrations, 270 references) of the normal and pathologic histology of the adrenal cortex. (In Spanish.)

YOUNG, HUGH HAMPTON: "Genital Abnormalities, Hermaphroditism. Related Adrenal Diseases." The Williams & Wilkins Company, Baltimore (1937).

An excellent and detailed treatise (649 pages, 380 illustrations, numerous references) on hermaphroditism and pseudohermaphroditism. This is perhaps the best contemporary monograph on the subject.

III

THE HYPOPHYSIS

HISTORIC INTRODUCTION

The rôle of the pituitary was completely misunderstood by early physicians; indeed the name "pituitary" (from *pituita*=nasal mucus) was given to the gland by *Vesalius* (1543) in the erroneous belief that its function is to secrete fluid into the nose. Later, the anatomist *Willis* (1684) thought that the gland secreted cerebro-spinal fluid; conversely, *Magendie* (1847) assumed that the cerebro-spinal fluid is taken up by the gland and secreted into the blood.

It was the great merit of the French neurologist, *Pierre Marie* (1886), to recognize an interrelationship between the hypophysis and a type of GIGANTISM, "acromegaly." However, his interpretation was entirely erroneous. Having found that in acromegalic patients, the hypophysis is replaced by tumor tissue, he felt justified in concluding that the normal function of the gland is to inhibit somatic growth and that gigantism is a result of a lack of this inhibition due to destruction of the gland. This historic error clearly shows that even erroneous theories may be very valuable in directing investigations. Although Marie's assumption is exactly the opposite of the truth, yet he correctly recognized that the little gland, at the base of the brain, is a regulator of growth and should be investigated from this viewpoint.

Although the earlier literature contains many case reports of destructive hypophyseal lesions accompanied by DWARFISM and INFANTILISM, *Lorain* (1871) and *Erdheim* (1916) were the first to consider a causal relationship between pituitary damage and the in-

hibition of somatic development. The suspicion was raised that hypo- and not hyperpituitarism impedes growth. Later *Simmonds* (1914) in Vienna, described a peculiar type of cachexia following pituitary disease in man. This called attention to the metabolic functions of the gland.

Well-controlled experimental work was necessary, however, to clarify the intricate physiologic rôle played by the hypophysis. There were some instructive early reports on the results of hypophysectomy in the dog (*Paulesco*, 1908; *Cushing et al.* 1910; *B. Aschner*, 1912; *Camus and Roussy*, 1913) and even the possibility of a substitution therapy had been established by experiments in lower vertebrates, such as tadpoles (*P. E. Smith*, 1916; *B. M. Allen*, 1919). However, the function of the gland in mammals was very incompletely understood until 1921, when *H. M. Evans* conclusively proved that the anterior-lobe of the hypophysis contains a GROWTH-PROMOTING-HORMONE. This was demonstrated by injecting crushed bovine anterior-lobe tissue into rats, whose somatic growth was greatly stimulated by this procedure. Subsequently, it was shown that hypophysectomy inhibits somatic growth, as well as the development of the gonads, thyroids and adrenal cortex, and that hypophyseal extracts and implants restore all these changes to normal; indeed, they can even cause gigantism in hypophysectomized animals such as the rat (*P. E. Smith*, 1930). This work culminated in the preparation of crystalline growth-hormone by *Li et al.* (1948).

It is of historic interest that, in their earliest publications, *Evans and Long* (1921) came to the conclusion that while their anterior-hypophyseal extracts cause accelerated growth, "at the same time, the effect of the anterior-lobe has been to repress sexual development by delaying sexual maturity and lengthening the estrus cycles, in some cases estrus being entirely inhibited." Other investigators interpreted these findings as indicating that growth hormone inhibits sex development, especially since during the period of maximal growth in childhood, sexual development is dormant, while after cessation of growth, puberty ensues. *Evans and Long*, however, soon noted that while their experimental rats were almost continuously diestrous, the ovaries of these animals showed signs of excessive, abnormal luteinization and there was no parallelism between the degree of growth stimulation and the extent of luteinization. Hence, they correctly concluded that the gonadotrophic activity is probably independent of the growth hormone and we know now that the diestrus was merely due to the preponderance of luteinization over follicle stimulation, not to an actual inhibition of ovarian development.

The independent nature of the gonadotrophic activity soon received even more convincing support. In 1926, *P. E. Smith*, in America, and *B. Zondek*, in Germany, almost simultaneously observed that pituitary implants, introduced into sexually immature female rodents, produced precocious ovarian development, due to the presence of GONADOTROPHIN a sex-stimulating-principle of the anterior-lobe. Subsequently, it was found that there are actually two gonadotrophins, one stimulating follicle maturation (FSH) and the other, luteinization of existing follicles (LH). Gonadotrophins had also been detected in large quantities in the urine of pregnant women; this was used in developing the Aschheim-Zondek test

for pregnancy. They are also abundant in the placenta, probably because this organ is the source of the increased gonadotrophin production during gestation.

Experimental investigations of this type eventually led to the isolation of pure LUTEINIZING HORMONE or LH (*Li et al.* 1940), PROLACTIN OR LUTEOTROPHIN (*White et al.* 1937) and CORTICOTROPHIN (*Li et al.* 1942; *Sayers et al.* 1943). The FOLLICLE STIMULATING hormone or FSH, and THYROTROPHIN have not yet been isolated, but very pure preparations of them are available and their existence as separate hormonal principles is no longer in doubt.

All these observations centered interest upon the anterior-lobe of the hypophysis as an important hormone-producing organ and a regulator of the activity of other endocrine glands. Probably several additional hormones are produced by the anterior-lobe and through them, it influences metabolism and the development of many organs. However, the chemical identity of these latter hormones has not yet been fully clarified.

In 1932, *Harvey Cushing*, a surgeon of Boston, described the syndrome now known as CUSHING'S DISEASE, which is characterized by proliferation of the basophil cells in the anterior-lobe, adrenal-cortical hypertrophy, hypertension, as well as glycosuria and other metabolic disturbances. This discovery further emphasized the important action of the hypophysis upon metabolism.

Although many old case reports mentioned excessive diuresis in patients with pituitary disease, *E. Frank* (1912) was the first to clearly express the view that the hypophyseal lesion is probably the cause of the polyuria in DIABETES INSIPIDUS.

In 1894, *Oliver and Schäfer* in England, found that posterior-pituitary extracts exert a vasopressor action. This suggested that the posterior-lobe, which, histologically, does not resemble

endocrine tissue, nevertheless produces hormone-like substances. Subsequently in the United States, Kamm (1928) succeeded in separating the crude posterior-lobe extract (known as "pituitrin") into a VASOPRESSOR and an OXYTOCIC fraction. Later work suggested that the posterior-lobe also produces an ANTI-DIURETIC principle, the deficiency of which is probably the main factor responsible for diabetes insipidus. The relationship of this substance to the other two posterior-lobe hormones has still not been completely clarified, but it is probably identical with vasoressin.

In 1901, the Viennese physician, Alfred Fröhlich, described the ADIPOS-

GENITAL SYNDROME, which now bears his name. In this disease, deficient sexual development is combined with adiposity, due to hypothalamic lesions.

Recent investigations have brought forth increasingly more evidence that one of the most important physiologic rôles of the anterior-lobe is concerned with ADAPTATION to various types of non-specific stress and that many of the most common diseases of mankind (hypertension, nephrosclerosis, as well as certain cardiovascular and "rheumatic" diseases) may be "DISEASES OF ADAPTATION," due to a derangement of the pituitary response to stress. (See: General-Adaptation-Syndrome and the Diseases of Adaptation.)

NORMAL MORPHOLOGY

ANATOMY

In man, the hypophysis cerebri (pituitary/body, pituitary gland) is attached to the base of the brain by a thin stalk, emerging from the tuber cinereum. The hypophysis is enclosed in the sella turcica or hypophyseal fossa of the sphenoid bone, which protects it on all sides and on its base. Its upper surface is covered by a circular fold of the dura mater, the diaphragma sellæ, which is perforated by the stalk. The subdural space does not extend around the pituitary body.

The gland consists of three main parts, namely :

(1) The ANTERIOR-LOBE (pars anterior, pars glandularis, anterior hypophysis), which consists of the *pars distalis* and continues anteriorly in the form of the *pars tuberalis*, a partly supra-diaphragmatic prolongation of the anterior-lobe attached to the stalk and tuber cinereum.

(2) The INTERMEDIATE-LOBE (pars intermedia), which is absent or vestigial in man but well developed in many animal species; it is sometimes regarded as part of the posterior-lobe although its structure is glandular.

(3) The POSTERIOR-LOBE (pars nervosa, neurohypophysis, neural lobe).

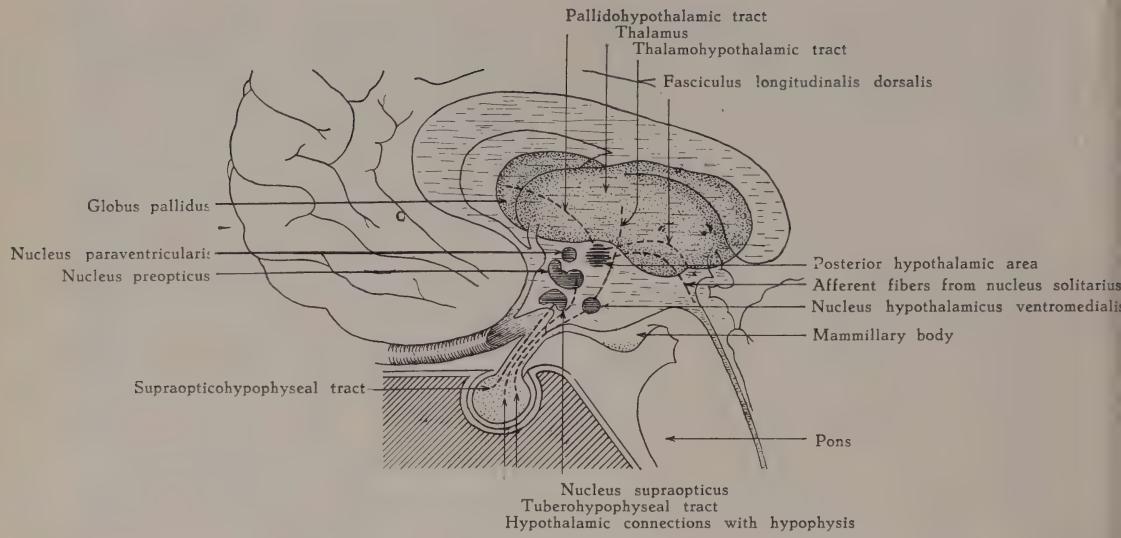
The above nomenclature is currently employed in medical literature, but anatomists prefer the more elaborate one given in the table on p. 200.

The hypophysis measures about 1.2-1.5 cm. transversely, 1.0 cm. antero-posteriorly and 0.5 cm. vertically. It weighs about 0.5 to 0.6 gm. in the adult, being larger in women, especially in multiparae, in whom it may exceed 1.0 gm.

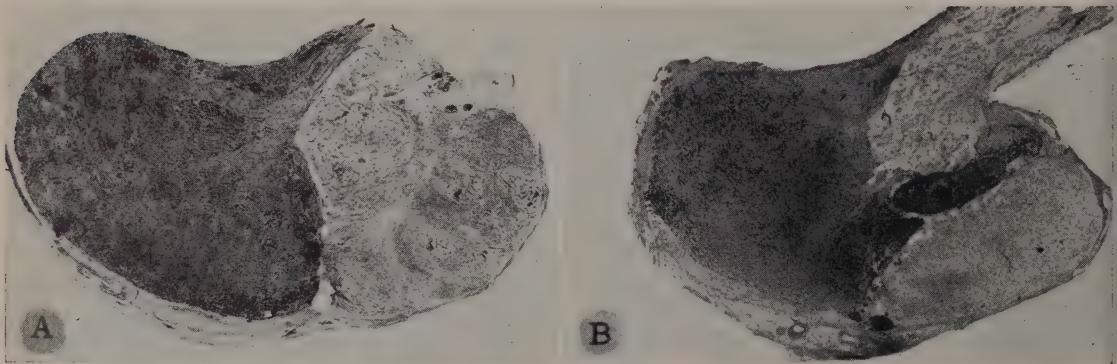
The anterior-lobe is pinkish-gray, the posterior-lobe is pearly gray and translucent, resembling nervous tissue. The anterior-lobe continues to grow until the 4th decade; after that it undergoes a gradual involution. The posterior-lobe is less likely to show changes in weight with progressing age.

The PHARYNGEAL HYPOPHYSIS is a small vestigial remnant of Rathke's pouch which is often detectable between the pharyngeal mucosa and the sphenoid bone in man. It resembles atrophic anterior-lobe tissue, but assumes importance only if it gives rise to tumor formation.

THE HYPOPHYSIS



Schematic drawing illustrating interrelation between hypothalamic area and pituitary



Normal hypophysis (Man) — A. Very low magnification of a sagittal section through the pituitary. Dark anterior-lobe, light posterior-lobe and barely visible cystlets corresponding to "intermediate-lobe" between the two. The pars tuberalis extends upwards from the anterior-lobe towards the hypothalamus. — **B.** Another normal human hypophysis (courtesy of Dr. W. Bonin) in which stalk and "intermediate-lobe cysts" are more evident.

Terminology of the mammalian hypophysis recommended by the International Commission on Anatomic Nomenclature

	MAJOR DIVISIONS	SUBDIVISIONS
ADENOHYPOPHYSIS	{ Lobus glandularis	{ 1. Pars distalis 2. Pars tuberalis 3. Pars intermedia } Anterior-lobe
	{ Lobus Nervosus (neural lobe)	{ 1. Processus infundibuli } Posterior-lobe
NEUROHYPOPHYSIS	{ Infundibulum (neural stalk)	{ 1. Pediculus infundibularis (stem) 2. Bulbus infundibularis (bulb) 3. Labrum infundibularis (rim) or median eminence of the tuber cinereum

HISTOLOGY

Anterior-Lobe. — The anterior-lobe consists of epithelial cell nests and columns supported by a delicate connective tissue reticulum. Between the epithelial cells are wide sinusoids, whose walls contain sessile reticulo-endothelial macrophages. Only rarely do the epithelial cells form acinus-like structures, whose lumina include serous or colloid-like material, and which are reminiscent of thyroid follicles.

The epithelial cells of the anterior-lobe are of three main types :

(1) The ACIDOPHIL CELLS (oxyphil or alpha cells) represent about 37% of the total epithelial cell count and are characterized by numerous acidophilic granules within their cytoplasm. In some animal species they possess a characteristic Golgi net, which surrounds the nucleus like a cap.

(2) The BASOPHIL CELLS (beta cells) represent about 11% of the total cell count and are characterized by basophilic cytoplasmic granules. In certain animal species they possess a distinctive, spherical Golgi net, which lies beside the nucleus. Although the Golgi apparatus itself is not visible without special stains, a "negative Golgi image" is often readily distinguishable on ordinary hematoxylin-eosin sections. In the basophil cells, this negative Golgi image is seen in the form of a pale ring-shaped hiatus in the cell body, with some cytoplasm in its center.

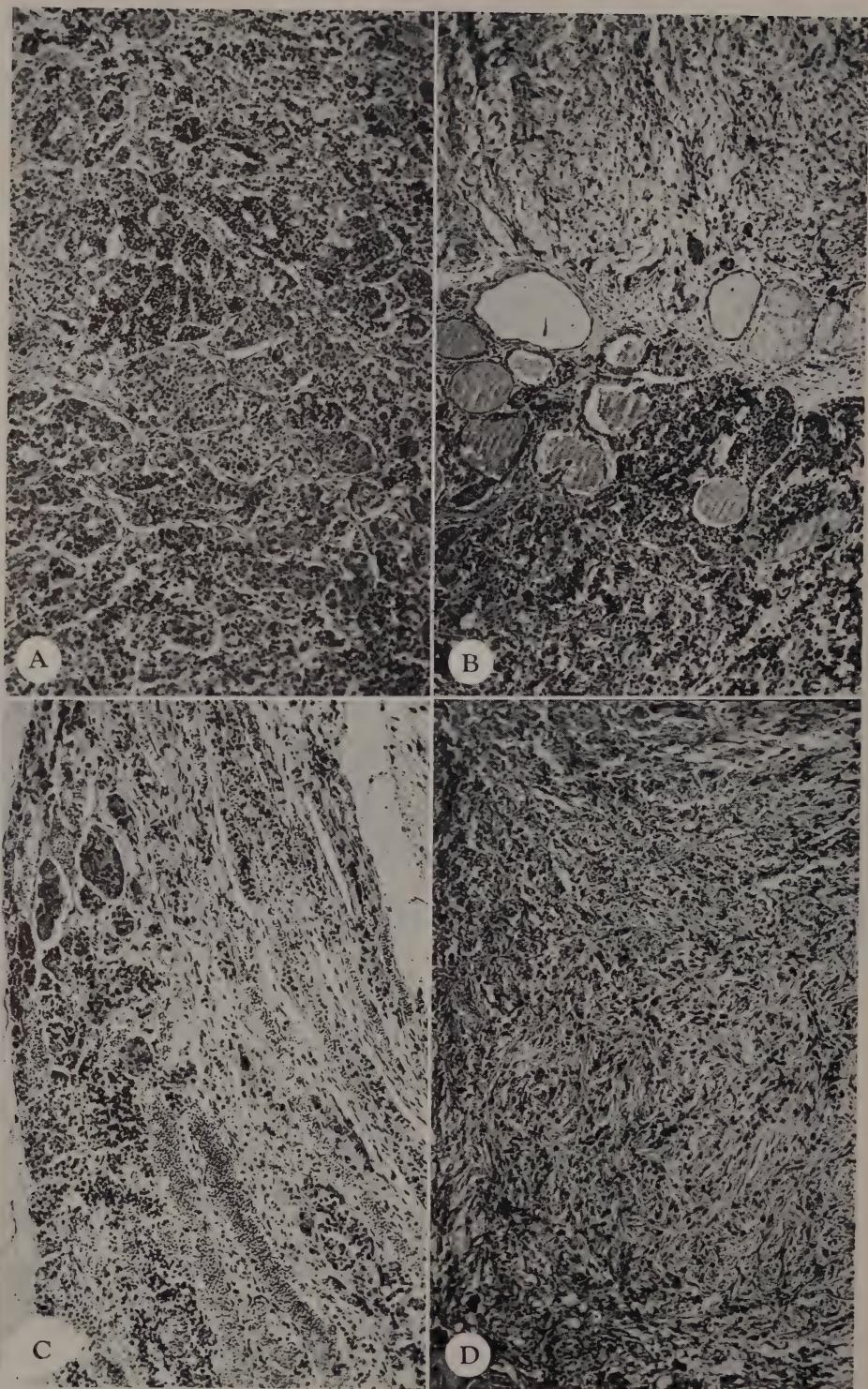
(3) The CHROMOPHOBEC CELLS (chief cells, principal cells, reserve cells) represent the remaining approximately 50% of the epithelial cells in the anterior-lobe. They have no special affinity either to basic or acid dyes and hence, their comparatively scarce cytoplasm is pale on histologic sections. They are presumably the younger forms, the precursors of the basophilic and acidophilic cell types, respectively. Most of the chromophobe cells have a characteristic Golgi apparatus, corresponding either to the acidophilic or basophilic type.

Some workers distinguish a fourth cell type, the CARMINE CELL, which occurs in the anterior-lobe of certain species (e.g., dog, cat, rabbit). It has a special affinity for azo-carmine and erythrosin.

The Pars Tuberalis. — This is anatomically a prolongation of the anterior-lobe, which it resembles even histologically. Its cells do not contain chromophil granules however, but possess a rather diffusely basophilic cytoplasm. These cells may form irregular colloid-filled cavities, which generally appear only after birth. Scattered islets of squamous epithelial cells are also found in the pars tuberalis of man. They are vestigial rudiments of the embryonic buccal ectoderm from which the anterior-lobe is derived. Their chief importance lies in their tendency to give rise to tumor formation (craniopharyngiomas).

Intermediate-Lobe. — In man only a few, vestigial, cystic formations are found between the anterior- and posterior-lobes, but in most animals the intermediate-lobe is well developed, in the form of a more or less regular disc between the anterior- and posterior-hypophysis. Its cells contain no chromophil granules, but are rather diffusely basophilic. They are closely packed, with only a fine reticular stroma between them.

Posterior-Lobe. — In man, this part is composed almost entirely of irregular, usually spindle-shaped cells, with fine processes. It resembles the glia-containing portions of nervous tissue. However, the specific posterior-lobe cells or "PITUICYTES" are not modified nerve cells. They contain no Nissl substance and must be regarded as of ependymal origin. Numerous HYALINE BODIES (bodies of Herring) are seen between the fibers of the pars nervosa, especially near the floor of the third ventricle, under the ependyma. Their origin and function are not known. There are also many NERVE FIBERS in the posterior-



Normal hypophysis (Man) — A. Anterior-lobe showing islets of large eosinophils (dark) and basophils (light), as well as many undifferentiated, small, almost lymphocyte-like chief-cells. — B. So-called "middle-lobe", which in the human hypophysis, consists merely of vestigial colloid filled cysts, separating the anterior-lobe (bottom) from the posterior-lobe. — C. Pars tuberalis consisting of cells somewhat similar to those of anterior-lobe but attached to the nervous tissue of the tuber cinereum. — D. Posterior-lobe with typical nerve-tissue-like cell structure.

lobe; they come from the brain through the stalk.

BASOPHIL CELLS are almost invariably found in the pars nervosa of man. It is believed that these invade the tissue from the anterior- lobe and subsequently decompose.

The Stroma. — The stroma of the hypophysis consists of a delicate connective tissue network.

The gland receives the superior hypophyseal ARTERIES in the form of several branches, which arise from the internal carotid and posterior communicating arteries. These anastomose with each other, as well as with those of the opposite side, on the infundibulum and stalk. Arterial branches enter the stalk and break up into sinusoids within its substance. Other small arteries proceed directly to the anterior-lobe and split up into sinusoids there. The inferior hypophyseal arteries are two paired branches of the internal carotid, which pass through the cavernous sinus and, after anastomosing on the dorsal side of the gland, supply the pars tuberalis and the posterior-lobe.

In the hypophysis, we find both portal and systemic VEINS. The portal veins come from the capillaries of the stalk and tuber region. After collecting into larger venules, they split up into a second capillary network within the hypophysis. Direct observation in amphibia proved that the blood flow in these portal veins is caudally directed, from the hypothalamus to the pars distalis. The systemic veins are represented by the lateral hypophyseal veins draining from the anterior-lobe and the infundibular process into the cavernous or intercavernous sinuses.

The physiologic importance of the portal circulation in the hypophysis is not yet understood, but it is interesting to speculate upon the fact that all three organs which produce active pressor substances: the adrenal medulla, the renal tubules and the posterior-hypophysis, possess a purely or almost purely venous circulation. Perhaps a low intravascular pressure or anaerobic conditions of metabolism are favorable for the production of pressor substances.

A thick tract of unmyelinated NERVE fibers originates in the vicinity of the supra-optic nucleus, just above the optic chiasm, and descends through the stalk to the pars nervosa. A few fibers of the tract continue into the pars intermedia. Some of these have branched endings which may possess large end-bulbs. The anterior-lobe receives unmyelinated fibers from the paraventricular nuclei and the carotid plexus. These terminate between the epithelial cells.

COMPARATIVE MORPHOLOGY

It is questionable whether there is any organ corresponding to the hypophysis in INVERTEBRATES, although the "X organ" in the eye-stalk of some crustaceans and the corpora cardiaca and allata of *periplaneta* have been suspected to be precursors of the pituitary.

In *AMPHioxus*, there is a glandular organ, corresponding to the anterior-lobe of the hypophysis, but no infundibulum.

In *CYCLOSTOMATA*, (e.g., *petromyzon fluvialis*), the hypophysis is a flat organ attached to the base of the brain. It consists of an anterior-lobe, which contains epithelial tubules, a distinct intermediate-lobe and a thin posterior-lobe.

In HIGHER VERTEBRATES, the pituitary is always well developed and essentially of the same structure as in man. However, in some (fowl, armadillo, manatee, whale) the intermediate-lobe is missing, while in others the anterior- and posterior-lobes are almost entirely separate, there being only a cylindrical stalk-like connection between the two (e.g., whale). A remnant of the lumen of the originally hollow pars distalis primordium, the "residual lumen," persists throughout adult life between the anterior and intermediate-lobes in most animal species (rodents, cat, etc.) but not in man.

It is interesting that although in man there is a certain proportionality between the weight of the pituitary and the size of the individual, such does not

exist if we compare different species. For instance, in certain whales (*Balaenoptera Sibbaldii*) weighing 100,000-150,000 Kg., the average weight of the anterior-lobe is only 32.5 gm., that of the posterior lobe 1.4 gm.

EMBRYOLOGY

The glandular portions of the hypophysis develop from an ectodermal pocket, originally located in front of the pharyngeal membrane. From here an invagination (Rathke's pouch) emerges in the form of a shallow sack in embryos about 3 mm. in length. It gradually comes into contact with a similar, glove-finger-like invagination of the infundibulum, which grows in the opposite direction and represents the primordium of the posterior-lobe. After the two primordia meet, Rathke's pouch loses its connections with the oral ectoderm at the end of the second month of embryonic development. The lumen of the pouch becomes the "residual lumen" of the adult gland. It persists during postnatal life in most animal species, although in man it is reduced to isolated cystic sacs.

The characteristic permanent structure of the hypophysis is attained at about the 3rd and 4th month, at which time the rostral wall of Rathke's pouch proliferates and differentiates into the ANTERIOR-LOBE. The portion between the original lumen and the posterior-lobe remains less developed in all animal species. It may involute or differentiate into the PARS INTERMEDIA. The so-called "PARS TUBERALIS" stretches forward along the infundibulum and is eventually partly situated above the dia-phragma sellæ. It is also a derivative of Rathke's pouch and essentially of the same origin as the anterior-lobe.

The POSTERIOR-LOBE likewise tends to lose its lumen, a prolongation of the third ventricle, although a small recess persists in man, and deep invaginations are retained during postnatal life in many animal species.

THEORIES CONCERNING THE HISTOPHYSIOLOGY OF THE HYPOPHYSIS

It is difficult to understand why three apparently independent endocrine glands, the three lobes of the hypophysis, are joined into a single organ and why they are placed into such intimate contact with the hypothalamic centers. Could it be that the hormones of one lobe are manufactured from precursors furnished by another? Is the special type of circulation in the pituitary a common prerequisite for the production of hormones by the several lobes? Does the anatomic union of the three parts — and their location in the particular position which they occupy — occur because of a common dependence upon direct nervous stimuli from the hypothalamic region? Could the three lobes secrete their products directly into the adjacent vegetative centers of the brain? All these are intriguing questions, but only some of them lend themselves to an analysis by the methods known to us at the present time.

Interdependence of the Various Pituitary Cell Types and the Adjacent Brain Centers. — Transplants of pure anterior-lobe tissue take quite readily in hypophysectomized animals and continue to produce somatotrophic, thyrotrophic, gonadotrophic, adrenotrophic and other hormones, as judged by the fact that the usual consequences of hypophysectomy are prevented by these grafts. This does not mean that the endocrine functions of the anterior-lobe are independent of the other lobes or of direct nervous stimuli reaching it through the stalk. It is quite possible that the hormone production of the transplants is not entirely normal, but we may conclude that, in principle, anterior-lobe cells are capable of continuing their hormone production when transposed into an abnormal location.

Similarly, it may be said, on the basis of partial hypophysectomy experiments, that isolated removal of the anterior-

lobe does not prevent the continued function of the posterior or middle-lobes, and vice versa.

Interrelations Between the Various Cell Types of the Anterior-lobe. — In the past it had been thought that basophils can be transformed into acidophils and vice versa, since certain investigators believed to have seen "TRANSITIONAL CELLS," containing both basophilic and acidophilic granules. It has been shown, however, that the supposedly acidophilic granules in basophils, were actually mitochondria and that true transitional types do not occur. When discussing the histology of the anterior-lobe, we mentioned that the Golgi apparatus has a different shape in the acidophils and basophils and that even among the chromophobes, some have one and some the other type of Golgi net. This led to the theory that the nature of a certain chromophobe cell is predetermined before granules develop in it, so that certain chromophobes can only develop into basophils, others only into acidophils. If later chromophils lose their granules due to exhaustive incration, they revert into chromophobes, which still retain their original, either eosinophilic or basophilic character, as regards the Golgi net (*Severinghaus*).

It has been claimed (*Collin*) that anterior-lobe cells may multiply by "ENDOCYTOGENESIS," that is, by the formation of a new cell within the cytoplasm of another. It has subsequently been demonstrated, however, that histologic pictures suggesting a cell within another cell are artefacts, due to the invagination of a small cell into the cytoplasm of a larger one. It is now generally agreed that the cells of the anterior-lobe, like those of other organs, multiply almost exclusively by mitotic and, to a negligible extent, by amitotic division.

Theories Concerning the Pathways of Secretion. — There has been much discussion concerning the pathways through which the hormones of the pi-

tuitary reach the target organs upon which they act. The fundamental basis of all morphologic studies concerning the pathways of hormone secretion is the assumption that the hormone precursors, or the hormones themselves, are visible in histologic sections. This has never been proven, but it is tacitly assumed by most morphologists that the tingible granules are hormone carriers, precursors, or (less likely) the hormones themselves, and hence, that by following the discharge of these granules from the cytoplasm we may gain knowledge concerning the mechanism of hormone secretion. In the case of the adrenal medulla, there is good evidence indicating that the chromaffin granules are actually adrenaline or its precursor, but, in the case of the anterior-lobe, the hormonal nature of the tingible granules is much more hypothetic. It is assumed, however, that cells rich in granules, are storing their secretion, while degranulated cells have discharged it.

The simplest and probably best proven type of pituitary secretion is the DIRECT DISCHARGE OF HORMONES INTO THE BLOOD VESSELS, or "hemocrin" (*Collin*) as it occurs in other endocrine glands. On histologic sections, granules similar to those seen in the cytoplasm of the anterior-lobe cells, can often be distinguished within the lumina of the sinusoids.

Allegedly there is another mode of secretion in the pituitary, the DIRECT DISCHARGE INTO THE HYPOTHALAMUS by way of the posterior-lobe and stalk. This process has been designated as "neurocrin" (*Collin*). On histologic sections, it is possible to detect colloid globules, which apparently come from the pars anterior and invade the posterior-lobe, subsequently ascending through the stalk into the hypothalamus. The existence of such granules, the so-called "Herring bodies" has been established beyond doubt, but it is difficult to prove that they are derived from epithelial portions of the anterior

or middle-lobe, which then enter into the nervous system. It has been pointed out that certain secreting neurons in the hypothalamic region, especially in the supraoptic and paraventricular nucleus of many vertebrates, are capable of forming colloid and apparently represent an endocrine structure, the "mid-brain gland" (Scharrer, 1932). Consequently, the mere presence of colloid granules in these neurons does not necessarily prove that they come from the pituitary.

There is increasingly more evidence to show that posterior and intermediate-lobe hormones are present in considerable quantities within the tissue of the tuber cinereum. It has not been possible to demonstrate, however, whether these substances are formed locally or come from the hypophysis through the stalk. Anterior-lobe hormones are present only in traces in the nervous tissue adjacent to the anterior-pituitary.

On histologic sections we sometimes see the hyaline colloid bodies (supposedly derived from the pituitary) entering the cerebrospinal fluid through the ependyma of the third ventricle. This has been interpreted as indicating a DIRECT SECRETION OF PITUITARY HORMONES INTO THE CEREBROSPINAL FLUID or "hydrencephalocriny" (Collin). While the existence of such histologic pictures is undeniable, it is difficult to interpret them. Vasopressor and oxytocic substances have been demonstrated in the cerebrospinal fluid of the third ventricle in sufficiently high concentrations to suggest (Cushing and Goetsch, 1910) that these principles are, at least partly, discharged directly into the cerebrospinal fluid. On the other hand, only insignificant traces of anterior-lobe hormones are found in the cerebrospinal fluid and hence, the latter are probably not discharged through the same mechanism.

The discovery of the pituitary portal vein system led to the assumption of yet another secretory pathway, the

SECRETION OF HORMONES INTO THE PITUITARY "PORTAL" CAPILLARIES, with subsequent migration through the portal veins to the hypothalamic centers, in which these vessels form a second capillary network. This has been termed "hemoneurocriny" (Collin). Recent investigations suggest, however, that the direction of the blood flow in the pituitary portal circulation is from the hypothalamus to the pituitary, so that if this vascular system were of importance in the direct transfer of hormones, the latter would have to originate in the hypothalamic region and act upon the pituitary, rather than vice versa.

Since complete transection of the pituitary stalk results in no pronounced deficiency symptoms, it appears that neurocrine, hydrencephalocrine, hemo-neurocrine secretion, and even the direct nervous connections between the pituitary and the hypothalamus are not indispensable. Yet, if pituitary hormones acted directly upon vegetative centers, smaller concentrations would suffice if they reached the centers directly, than if they were diluted by the entire blood volume after entering the general circulation. Furthermore, the nervous regulation of pituitary hormone production may aid in the accurate adjustment of endocrine secretion to functional needs. In any case the anatomic continuity, between the pituitary and the adjacent vegetative centers, is indispensable for the function of the posterior-lobe.

Which Cell Produces Which Hormone? — There are only three clearly distinguishable cell types in the anterior-lobe. We do not know how many hormones the pars distalis produces, but, since it certainly elaborates more than three clinically different principles, it is evident that several hormones must originate from the same cell type.

The GROWTH HORMONE is presumably produced by the eosinophils, since eosinophil adenomas are commonly asso-

ciated with acromegaly. Furthermore, in some animals (e.g., pigeon) eosinophils are especially plentiful in the anterior-lobe during the period of maximal growth, and in certain strains of dwarf mice, eosinophils are entirely absent from the hypophysis. Since these mice grow up to normal size if treated with implants of normal pituitaries, it was thought that the stunting of their growth results from the deficient development of the anterior-lobe eosinophils. It should be emphasized, however, that both the stunted growth and the absence of eosinophils are recessive hereditary characteristics and that the individuals in which these features are manifest, are sterile because of gonadal atrophy. The animals are not congenitally irresponsible to the pituitary hormones, since hypophyseal extracts restore both the growth-rate and the atrophic gonads. Hence it would appear that not only the somatotrophic but also the gonadotrophic activity is to some extent linked with the eosinophils.

GONADOTROPHINS are allegedly elaborated mainly by the basophils, since castration causes a pronounced increase in the gonadotrophin content of the anterior-lobe and urine (e.g., man), simultaneously with a marked hypertrophy of the anterior-lobe basophils (e.g., rat). Furthermore, cattle pituitary is practically free of gonadotrophic hormones, except along the borderline of the middle-lobe where basophils are plentiful. On the other hand, extracts of both eosinophilic and basophilic adenomas of human pituitaries can cause precocious sexual maturity in rodents, and castration causes proliferation of eosinophils in the anterior-lobe of man and most animal species other than the rat. It is not improbable, therefore, that the eosinophils also participate in gonadotrophin production. There is some evidence that the basophils produce FSH, the eosinophils LH.

The CORTICOTROPHINS appear to be chiefly elaborated by the basophils, since in Cushing's disease, basophilic adenomas of the anterior-lobe are accompanied by proliferation of adreno-cortical tissue and an increased corticotrophin production.

It has been claimed that PROLACTIN is secreted chiefly by the basophils, since these reach a maximal development in pigeons when the crop gland is fully functional.

It must be admitted, however, that all this evidence concerning the probable origin of the various anterior-lobe hormones is far from convincing, and even less is known about the cell-type responsible for the elaboration of OTHER ANTERIOR-LOBE PRINCIPLES.

INTERMEDIN, the chromatophore dilating hormone, arises in the intermediate-lobe in those animal species in which such a structure is present. This is shown by experiments indicating that only extirpation of the intermediate-lobe causes deficiency, and implantation of intermediate-lobe tissue results in symptoms of overdosage with this hormone. However, it must be admitted that in man, in whom a distinct intermediate-lobe does not develop, intermedin is demonstrable in the anterior-lobe, and to a lesser extent in the posterior-lobe tissue. Perhaps, in man, other cell elements have taken over the functions of the intermediate-lobe.

The vasopressor and oxytocic POSTERIOR-LOBE HORMONES are apparently both elaborated by the pituicytes, since other potentially endocrine cells are not demonstrable in the pars nervosa. In any event, it is certain that some elements in the posterior-lobe, and perhaps also the adjacent hypothalamic region, are the source of these hormones, because the anterior and intermediate-lobes are virtually free of them, and anterior or intermediate-lobe removal causes no deficiency manifestation in this respect.

CHEMISTRY OF THE HYPOPHYSIS

CHEMICAL COMPOSITION OF THE GLAND

Since the chemistry of the various hypophyseal hormones will be discussed in the next chapter, it suffices here to give a brief account of the other chemical constituents which make up the tissue of the pituitary body.

The CARBOHYDRATE content of the hypophysis is low. In cattle only about 1.5% (of the dry weight) of the anterior-lobe and 1.1% of the posterior-lobe, is extractable sugar.

The LIPID content of cattle pituitary has been estimated to be about 14% (of the dry weight) in the anterior, and 20% in the posterior-lobe tissue. The cholesterol content of the anterior-hypophysis (1.65%) is also lower than that of posterior-lobe (2.12%). It has been noted, in several species, that the lipid content of the pituitary tends to decrease during the breeding season.

The PROTEIN content of cattle anterior-lobe is about 78%, and that of the posterior-lobe 66% of the dry weight.

Among the INORGANIC constituents of the hypophysis, the iodine content received special attention. Dried cattle anterior-lobe powder has an iodine content of about 0.13-0.14 mg./gm. The iodine content of one human pituitary is estimated to be about 13 γ ; it tends to diminish under the influence of a variety of diseases. As judged by experiments with radioactive iodine, it appears that iodides (but not thyroxin-iodine) tend to accumulate in the pituitary following their introduction into the body. Yet the iodine affinity of the hypophysis is far below that of the thyroid.

The bromine content of fresh pituitary tissue varies between 0.4 to 0.8 mg.% and — contrary to earlier reports in the literature — it is not significantly higher than in other tissues.

The zinc content of human pituitary tissue is claimed to be very high (112

mg./100 gm. of dry weight), but this requires confirmation.

Among other inorganic constituents, calcium, phosphate, sulphur, manganese, iron and even traces of arsenic have been demonstrated in pituitary tissue; their concentration here is approximately the same as in most other tissues.

The WATER content of the pituitary varies between 74-80% in the various species. It is claimed that the posterior-lobe contains slightly more water than the anterior-hypophysis.

The ASCORBIC ACID content of cattle anterior-lobe (1.95 mg./gm. of fresh tissue) is significantly higher than in the posterior-lobe (0.46 mg./gm.). It is noteworthy that the hypophysis (like the adrenal cortex) is one of the organs containing the highest concentration of ascorbic acid; it loses this substance under the influence of a variety of non-specific damaging agents.

CHOLINE, ACETYLCHOLINE AND HISTAMINE have also been demonstrated in pituitary tissue; because of certain similarities in the pharmacologic actions of these compounds and those of posterior-lobe hormones, earlier investigators believed that the former are largely responsible for the biologic actions of posterior-lobe preparations. This has been disproven, but the biologic significance of these compounds in the pituitary is not yet known.

Among other compounds, whose presence has been demonstrated in the hypophysis are: GLUTATHIONE, a variety of VITAMINS, ENZYMES, and CAROTENE. It is interesting that the posterior-lobe, which is comparatively rich in lipids, also contains more lipase than the anterior-hypophysis.

CLASSIFICATION AND CHEMISTRY OF THE HYPOPHYSEAL HORMONES

Classification of the Anterior-Lobe Hormones. — The anterior-pituitary produces a large number of hormones,

only five of which (the follicle-stimulating luteotrophic, somatotropic, adrenocorticotropic and luteinizing hormones) have been isolated in pure form. There is convincing evidence that the thyrotrophic activity of anterior-lobe extracts is likewise due to a separate hormone.

Since the purification of these principles has only recently been accomplished, it has not yet been possible to examine to what extent they and their combinations can duplicate the manifold effects obtainable with impure anterior-lobe extracts. The latter produce a variety of biologic effects, which may be due to their content in additional hormones which have hitherto not been isolated; however, they could also be due to one of the above-mentioned six authenticated hormones, or to combinations of the same.

In a discussion of the anterior-lobe hormones, it is well to keep in mind that, depending upon dosage, route of administration, simultaneous treatment with other substances, etc., the actions of a hormone can be most significantly modified, hence great care should be exercised before we postulate the existence of new principles.

It may help orientation in this complex field to enumerate the AUTHENTICATED ANTERIOR-LOBE HORMONES, together with their abbreviated names (in brackets) and synonyms, as well as a few of their chief characteristics :

(1) FOLLICLE-STIMULATING HORMONE (FSH). Synonyms : follicle stimulator, thylakentrin. This substance has recently been isolated (*Li et al. 1949*) in pure form as shown by electrophoresis, ultracentrifuge and diffusion studies. Its isoelectric point is at pH 4.5 and its molecular weight is 60,000. Highly purified preparations are characterized by their ability to stimulate the growth of the granulosa cells in the ovaries of hypophysectomized animals, without preventing the atrophy of the theca cells and without eliciting folliculoid hormone secretion by the ovaries. The hormone also stimulates the seminiferous epithelium in the testes of intact or hypophysectomized animals. It is especially abundant in : the anterior-lobe, the urine of castrates, and pregnant mare serum (PMS).

(2) LUTEINIZING HORMONE (LH). *Synonyms* : interstitial-cell-stimulating-hormone (ICSH), chorionic gonadotrophin, metakentrin. It transforms mature ovarian follicles into corpora lutea and stimulates the growth and folliculoid hormone secretion of the theca cells, even in the hypophysectomized animal. It probably also enhances folliculoid secretion by the granulosa and corpus luteum, but does not prevent the atrophy of preexisting corpora lutea, nor does it cause them to secrete luteoid hormones. It stimulates the development and testoid hormone secretion of the Leydig cells, both in intact and in hypophysectomized males. It is particularly abundant in: anterior-lobe tissue, human placenta and human pregnancy urine (PU). Electrophoretically pure, crystalline human "chorionic gonadotrophin" has been prepared. 6-8000 I.U./mg. causes ovulation in women (*Claisson et al. 1948*).

(3) LUTEOTROPHIC HORMONE (LTH). *Synonyms* : luteotrophin, mammatrophin, prolactin, galactin, lactogenic hormone. The chief characteristics of this principle are that it maintains fully formed corpora lutea in the ovaries of intact or hypophysectomized animals and causes them to secrete luteoid hormone. It also stimulates milk secretion, but only if the mammary glands have previously been brought to full development. In the pigeon, it causes growth and secretion of the crop glands. It occurs in significant quantities only in anterior-lobe tissue.

The above-mentioned three hormones are collectively referred to as *gonadotrophins* or *gonadotrophic hormones*, since they are the chief regulators of gonadal activity.

(4) CORTICOTROPHIC HORMONES (ACTH). *Synonyms* : Adreno-corticotropic hormone, adrenotrophin, corticotrophin, adrenotrophic hormone, corticotrophic hormone. This substance has been isolated in pure form. It stimulates the growth and hormone produc-

tion of the adrenal cortex, both in intact and in hypophysectomized animals. It tends to deplete the cortex of its lipid and ascorbic acid content. It has no effect upon the adrenal medulla. Only anterior-lobe tissue contains significant amounts of this principle. (See p. 212.)

(5) THYROTROPHIC HORMONE (TTH). *Synonyms*: thyrotrophin, thyreotrophic hormone. This hormone has not been isolated. Its most characteristic action is to stimulate the growth and thyroid hormone secretion of the thyroid gland, in intact and hypophysectomized animals. Significant amounts of it occur only in anterior-lobe tissue.

(6) SOMATOTROPHIC HORMONE (STH). *Synonyms*: somatotrophin, growth hormone. This principle has been crystallized. Its chief characteristic is to stimulate somatic growth in general, both in intact and in hypophysectomized animals. As long as the epiphyseal junction cartilages are still open, it causes the skeleton to grow both in length and in thickness; it also promotes the development of soft tissues. After ossification is completed, it can no longer stimulate growth in length, but retains its other effects. It does not cause selective growth of any one organ. This principle occurs in significant amounts only in anterior-lobe tissue.

A number of actions of impure anterior-hypophyseal extracts have not been proven to be caused by special hormones. — Some of these have been, or still are, alleged to be produced by principles distinct from the above-mentioned six authenticated anterior-lobe hormones; for others, no such definite claim has been made. We shall enumerate the most important actions of this type, in order to avoid possible confusion arising from unjustified claims in the earlier literature; the list will also call attention to the potential existence of additional anterior-lobe hormones:

(1) THE OVULATION-INDUCING ACTION. — FSH causes maturation of the follicles, while LH transforms them into

corpora lutea. Under ordinary conditions of experimentation, this results in the formation of atretic corpora lutea, with enclosed ova, due to lack of ovulation. Certain gonadotrophic preparations, however, elicit ovulation and this led to the belief that they contain a special "ovulation-inducing hormone." It is now almost generally agreed, however, that under optimal experimental conditions, LH itself can produce ovulation following pretreatment with FSH, in several animal species. Other species (e.g., man) are singularly refractory to this effect, but they also fail to respond to impure extracts, allegedly containing the "ovulation-inducing hormone." Hence, it is unnecessary to postulate the existence of a special principle to explain this action.

(2) THE SYNERGISTIC ACTION. It has been claimed that, in females, the gonadotrophic effect of LH preparations can be greatly augmented by certain anterior-lobe extracts, which do not contain demonstrable amounts of gonadotrophins. This was attributed to a hypothetic "synergist" or "augmenting factor," presumed to be a hormone. It is now agreed, however, that this synergistic effect was due to sub-threshold amounts of FSH in the anterior-pituitary preparations used.

(3) THE ANTAGONISTIC ACTION. Certain anterior-pituitary extracts, when given intraperitoneally, antagonize the gonadotrophic action of simultaneously, subcutaneously administered gonadotrophic extracts. This effect has been attributed to the presence, in the anterior-lobe extracts, of an "antagonist" or "atresin"; a hormone which causes follicular atresia and inhibits the action of gonadotrophins. It has since been demonstrated, however, that even purified FSH or LH preparations, when given by the intraperitoneal route, antagonize the trophic effect of subcutaneously administered gonadotrophic extracts. Hence, it is no longer necessary to invoke the existence of this principle.

(4) THE ANTILUTEOGENIC ACTION. After hypophysectomy, the involution of corpora lutea is slow, especially in certain animal species (e.g., rat). It can be accelerated, however, by impure anterior-pituitary extracts. Hence, it has been postulated that the latter contain a hypothetic "antiluteogenic hormone" which accelerates the regression of corpora lutea. It has since been demonstrated, however, that both purified FSH and LH preparations have a similar effect, especially if administered by the intraperitoneal route.

(5) THE THYMOTROPHIC ACTION. Gonadotrophins and ACTH cause thymus involution because they liberate steroids (from the gonads and adrenal cortex respectively) which possess anti-thymic effects. Certain crude anterior-lobe extracts, however, tend to increase the size of the thymus. This led to the belief that a special "thymotrophic hormone" may be secreted by the anterior-lobe. The thymotrophic effect has now been duplicated with the pure somatotrophin preparations and it is doubtful whether it should be ascribed to a special hormone. It is known, furthermore, that short-term treatment with very high doses of desoxycorticosterone acetate causes thymus involution, while prolonged administration of the same compound in small doses actually increases thymic weight. This may be due to the resulting compensatory atrophy of the adrenal cortex and the consequent reduction in the elaboration of the more potent anti-thymic cortical hormones. Some such mechanism may well be responsible for the thymotrophic effect of anterior-pituitary extracts, hence the existence of the special "thymotrophic hormone" remains to be demonstrated.

(6) THE RENOTROPHIC ACTION. Certain impure anterior-pituitary extracts enlarge the kidney disproportionately, that is to say, more markedly than could be expected on the basis of their somatotrophin content. Although the existence of a special renotrophic hor-

mone has never been postulated, this possibility suggests itself, especially since pure somatotrophin causes no disproportionate renal growth. It must be kept in mind, however, that the most active renotrophic anterior-pituitary preparations also exert definite hepatotropic and other splanchnotrophic actions, so that the effect is not entirely specific. In thyroidectomized animals, all these actions are greatly diminished, but not abolished. This proves that they are not merely due to the thyrotrophic-hormone content of the extracts and the resulting increased thyroid-hormone secretion. Gonadectomy and adrenalectomy likewise fail to prevent the kidney-stimulating effect of anterior-lobe extracts, hence, this action cannot be ascribed to renotrophic, gonadal or adrenal hormones (e.g., testoids). Further work with purified anterior-pituitary hormones will be necessary to establish whether the renotrophic, hepatotropic and other splanchnotrophic actions of impure extracts are due to special splanchnotrophic hormones or to combinations of several of the already authenticated anterior-lobe principles (e.g., a combined effect of somatotrophic plus thyrotrophic homones).

(7) THE NEPHROSCLEROTIC ACTION. Certain impure anterior-pituitary extracts cause nephrosclerosis and hypertension. The renal lesions so produced are strikingly similar to those elicited by overdosage with desoxycorticosterone acetate, except that they are complicated by the simultaneous production of the renotrophic effect, presumably due to the impurity of the anterior-pituitary preparations employed. The existence of a special nephrosclerotic pituitary-hormone has never been postulated. The bulk of evidence suggests that anterior-pituitary extracts cause nephrosclerosis through the intermediary of the adrenal, by virtue of their corticotrophic-hormone content. The possibility must also be considered that several anterior-pituitary hormones may coöperate in the production of nephro-

sclerosis, especially because previous thyroidectomy diminishes, while thyroid hormone treatment augments the nephrosclerotic action of the impure anterior-lobe extracts. Even other metabolic hormones of the anterior-lobe may be involved in this effect, particularly since it has been established that high-sodium, high-protein diets augment, while acidifying salts and certain sugars antagonize the nephrosclerotic properties of the anterior-pituitary extracts.

(8) THE GLUCO-CORTICOTROPHIC, MINERALO-CORTICOTROPHIC, LIPOCORTICOTROPHIC AND TESTO-CORTICOTROPHIC ACTIONS. It is known that the adrenal cortex produces gluco-corticoids (e.g., corticosterone), mineralo-corticoids (e.g., the "sodium factor," which resembles desoxycorticosterone in its actions), testoids (e.g., dehydro-iso-androsterone, adrenosterone) and perhaps even fat deposition-stimulating lipocorticoids. Clinical experience indicates that all these hormones are not necessarily produced in set proportions, but that under varying conditions, one or the other type may predominate. It is known, furthermore, that testoids can cause selective atrophy of the reticularis (the testoid-producing zone) in females, while experiments on male castrates revealed that stimulation of the adrenal cortex, by folliculoids or alarming stimuli, causes a pronounced increase in the production of life-maintaining corticoids, without the slightest increase in endogenous testoid production. Since the function of the adrenal cortex is under the influence of the anterior-pituitary, it is tempting to assume that the latter can produce various corticotrophins, which selectively stimulate one or the other function of the adrenal cortex. It must be admitted, however, that the adrenocorticotrophic hormone (ACTH) now available, exhibits all the characteristics of a pure protein and the existence of different types of corticotrophins has not been proven. It is possible that the same

pituitary trophic principle exerts different actions, depending upon the simultaneous presence of other hormones (e.g., steroids). Further experimentation with pure preparations will be necessary to elucidate this point, but it is well to keep in mind the possibility of several corticotrophins and the certainty of several corticotrophic effects.

(9) THE ADRENOMEDULLOTROPHIC ACTION. It has been claimed that certain anterior-pituitary extracts selectively stimulate the growth and hormone production of the adrenal medulla. These claims have not been substantiated.

(10) THE PARATHYROTROPHIC ACTION. Following the administration of impure anterior-pituitary extracts, the parathyroids tend to enlarge in certain animal species. This effect is very inconstant, and contrary to earlier claims, hypophysectomy causes no significant atrophy of the parathyroids. Although some investigators postulated the existence of a special "parathyrotrophic hormone," this is hardly justified on the basis of available evidence. The enlargement of the parathyroids, occasionally noticed following anterior-pituitary extract treatment, could well be secondary to the metabolic changes induced by these preparations. It is known that renal lesions, rachitogenic diets and bone destruction due to a variety of other causes, produce secondary parathyroid enlargement, not unlike that caused by "parathyrotrophic" anterior-pituitary extracts.

(11) THE MAMMOGENIC ACTION. Impure anterior-pituitary extracts stimulate the growth of the mammary glands. This effect is distinctly different from that of the luteotropic (or "mammatrophic") hormone, which merely causes secretion, but not development of the breast. Since folliculoid, testoid and luteoid steroids cause marked breast growth only in the presence of the pituitary, it is tempting to assume that the anterior-lobe produces a special mamrogenic principle, whose elaboration

would be increased by the above-mentioned steroids. It must be kept in mind, however, that certain steroids can slightly stimulate breast growth, even in the absence of the anterior-lobe; hence, the mammogenic effect may merely be due to a potentiation of the mammogenic action of steroids, by a pituitary principle. None of the pure anterior-lobe hormones have been shown to possess such a mammogenic effect, but the existence of a special, "mammogenic hormone" has not been proven.

(12) **THE GLYCOTROPIC OR ANTI-INSULIN ACTION.** Impure anterior-pituitary extracts induce an insensitivity to the action of insulin, both in intact and in hypophysectomized animals. This effect is not seen after adrenalectomy, and similar actions can be elicited by ACTH and gluco-corticoids; hence it is highly probable that the anti-insulin effect of anterior-lobe extracts is mediated by the gluco-corticoids, which increase the glycogen stores in the liver and thus provide more sugar to antagonize insulin-hypoglycemia.

(13) **THE GLYCOSTATIC ACTION.** Under certain conditions, anterior-pituitary extracts restore the ability of fasting hypophysectomized animals to preserve their glycogen stores. Since muscle glycogen can thus be maintained even following adrenalectomy, the effect is not mediated by the gluco-corticotropic principle. It has been ascribed to a special "glycostatic factor," as this effect is not associated with any of the authenticated anterior-lobe compounds. However, corticoids greatly potentiate this action and it is not impossible that the effect is due to one of the known anterior-lobe hormones or to synergistic combinations of these.

(14) **THE PANCREATOTROPHIC ACTION.** It has been found that repeated treatment with impure anterior-pituitary extracts can cause an increase in the size, number and insulin content of the Langerhans islets in the rat. It has therefore been assumed that the anterior-pituitary produces a trophic substance,

which augments the morphologic development and insulin production of the islet tissue. It is noteworthy, however, that hypophysectomy does not cause any significant involution of the Langerhans islets. The possibility must be kept in mind that the stimulation of the islets may merely be a secondary, compensatory reaction, such as is noted for instance on high carbohydrate diets or following partial ablation of the pancreas. Further work will be necessary to establish whether there is a special pancreaticotropic hormone or whether the pancreaticotropic effect merely represents a compensatory reaction to the metabolic derangements caused by anterior-pituitary-hormone overdosage.

(15) **THE DIABETOGENIC ACTION.** Sometimes following prolonged treatment with impure anterior-pituitary extracts, a state of permanent diabetes develops in experimental animals (e.g., dog). The Langerhans islets are at first stimulated (pancreatotropic effect?), but later destroyed, by repeated injections of the active material. Simultaneously, the insulin content of the pancreas gradually diminishes to a negligible quantity. The diabetic state so produced is not intensified by complete pancreatectomy. Unlike the depancreatized dogs, those in which the diabetes is produced by diabetogenic, hypophyseal extracts can survive for long periods without insulin treatment.

The diabetogenic activity of anterior-lobe extracts is associated with the globulin and pseudoglobulin fractions, but isolation of the active material has not yet been accomplished. It appears that the effect depends upon an "exhaustion" of the islet tissue, since the destruction of the pancreatic islets is preceded by a period of hyperplasia and hypertrophy; insulin counteracts this, while over-feeding (especially with carbohydrate diets) and partial pancreatectomy sensitize to the diabetogenic effect. Further experiments will have to show whether this action is merely a result of metabolic derangements

(which cause a secondary breakdown of the Langerhans islets due to their inability to compensate indefinitely), or whether we are dealing with a direct hormone action upon the islet tissue. It is also questionable whether the diabetogenic action is caused by a single hormone or by combinations of several known metabolic principles.

(16) THE ANTI-DIABETIC EFFECT OF ORALLY ADMINISTERED ANTERIOR-LOBE PREPARATIONS. Earlier claims that certain pituitary extracts elicit an anti-diabetogenic effect in pancreatectomized animals and in diabetic patients when given by mouth, have not been substantiated.

(17) THE CONTRA-INSULAR ACTION. Impure anterior-pituitary extracts cause hyperglycemia in the dog, especially when injected directly into the cerebrospinal fluid. From this it was concluded that a special "Kontrainsuläres Hormon" is elaborated by the anterior-lobe and acts directly upon the "carbohydrate-metabolism centers" of the brain. The evidence supporting this concept is unconvincing and it appears very probable that the effect is due to direct nervous irritation by toxic substances in the extract with a consequent discharge of adrenaline.

(18) THE KETOGENIC ACTION. Impure anterior-pituitary extracts increase ketonemia in experimental animals. It is highly doubtful whether this is due to a special "ketogenic" or "fat-metabolism hormone" ("Fettstoffwechsel Hormon"), rather than to the diabetogenic effect, perhaps in conjunction with other metabolic hormones. The dependence of the ketonemia upon the adrenal cortex suggests the possibility that ACTH may also be involved.

(19) THE FATTY-LIVER-PRODUCING ACTION. Hypophysectomy prevents fat deposition in the liver, under various experimental conditions (e.g., partial hepatectomy and fasting, certain diets) which normally cause fat migration from the tissues into the hepatic cells. Treat-

ment of hypophysectomized animals with impure anterior-lobe extracts restores their ability to form fatty livers, and overdosage with such pituitary preparations may, in itself, suffice to cause fat infiltration in the liver. The available evidence does not suffice to postulate the existence of a special, fatty-liver-producing pituitary principle. It is especially noteworthy that adrenalectomy also prevents the production of fatty livers under similar conditions, hence the ACTH probably plays an important part in conditioning this action of anterior-lobe preparations.

(20) THE PREPUTIAL GLAND-STIMULATING ACTION. Impure anterior-pituitary extracts stimulate the growth of the preputial glands, especially in rodents. This effect is not mediated by the gonads, as it can be produced in castrates, yet certain steroids, not necessarily testoids (e.g., pregnenolone), potentiate the preputial gland-stimulating action of the anterior-lobe preparations. Since the preputial gland is merely a modified (and readily measurable) sebaceous gland, the effect may be related to the sebaceous gland-stimulation and acne of hyperpituitarism. We do not know which hormone(s) is responsible for this effect.

(21) OTHER ACTIONS ALLEGEDLY DUE TO SPECIAL HORMONES. A number of other actions have been ascribed to special anterior-lobe hormones, on the basis of very insufficient evidence. Among these are: the *restropic* (reticuloendothelial-system stimulating), the *hemopoiesis-stimulating action*, the *ketonemia-decreasing*, the *blood-pressure-depressing*, the *protein-metabolism-stimulating*, the *deaminizing* and the *thyroid-depressing* hormones. The latter action is allegedly elicited only by the pituitaries of rats fed with thyroid hormone.

Chemistry of Anterior-Lobe Hormones.—A detailed discussion of the extremely complex chemistry of the anterior-lobe hormones would be far beyond the scope of this textbook. Suffice it to say, that chemically, the six authenticated anterior-lobe principles

fall into two groups : the glycoproteins (thyrotrophin, FSH and LH) and the simple proteins or polypeptides (somatotrophin, corticotrophin and luteotrophin). Generally speaking, somatotrophin and luteotrophin are water-insoluble proteins, while FSH, LH and thy-

rotrophin are highly soluble in water; ACTH is intermediate.

The most outstanding physico-chemical and analytic data of pure preparations of LH, corticotrophin, luteotrophin and somatotrophin are summarized in the two tables below :

Physico-chemical properties of LH (ICSH), corticotrophin (ACTH), luteotrophin (lactogenic hormone) and somatotrophin (growth hormone)
(after C.-H. Li, Ann. Rev. Biochem. 1947)

Determination	ICSH		ACTH		Lactogenic Hormone		Growth Hormone (Ox)
	Sheep	Swine	Sheep	Swine	Sheep	Ox	
N, %	14.2	14.93	15.65	15.47	15.86	16.50	15.65
S, %			2.3	2.33	1.79	2.0	1.3
Cystine, %				7.19	3.11	3.4	2.25
Methionine, %				1.93	4.31		3.06
Tryptophane, %	1.0	3.8			1.25	1.3	0.84
Tyrosine, %	4.5				4.53	5.7	4.3
Molecular Weight, M							
Osmotic Pressure	40,000				26,500	26,500	44,250
Sedimentation		100,000	20,000	20,000		32,000	
Oiffusion constant, $D_{10} \times 10^7$			10.4		9.0	7.5	7.15
Sedimentation constant as Svedberg units, S	3.6	5.4	2.08	2.04-	2.11	2.65	
Isoelectric Point, pH	4.6	7.45	4.65-	4.70-	5.73	5.73	6.85
Partial specific volume, V_1			4.70	4.80			
Relative viscosity					0.721		0.760
Dissymmetry constant, f/f_0				1.1	6.65		7.64
					1.29		1.31

The amino-acid composition of the various anterior-lobe hormones appears to be essentially similar to that of other tissue proteins. As far as we know, these hormones are not characterized by the presence of any amino-

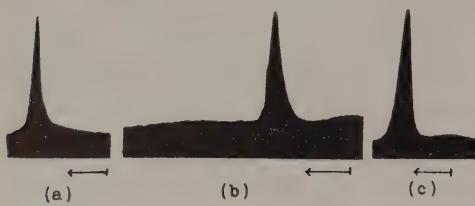
acid which would not occur in other tissues in approximately similar quantities. As an example we cite the composition of somatotrophin in the table below:

Composition of Somatotrophin
(after C.-H. Li, Ann. Rev. Biochem. 1947)

Constituent	N as % Protein N	Amount per 100 gm. protein	Minimal Mol. Wt.	Assumed number of residues	Calculated Mol. Wt.
Amide-N (Ammonia)	6.3	1.2	1420	30	(42,600)*
Arginine	18.8	9.1	1910	25	47,700
Aspartic-Acid	6.1	9.0	1480	32	47,300
Cystine	1.7	2.25	10700	4	(42,808)*
Glutamic Acid	7.9	13.0	1130	42	47,400
Glycine	4.5	3.8	1980	24	47,400
Histidine	4.6	2.65	5850	8	46,800
Isoleucine	2.7	4.0	3280	14	46,000
Leucine	8.3	12.1	1080	44	47,700
Lysine	8.7	7.1	2060	23	47,400
Methionine	1.7	2.9	5140	9	46,200
Phenylalanine	4.3	7.9	2090	23	48,000
Threonine	6.8	9.0	1320	36	47,600
Tryptophane	0.7	0.84	24500	2	48,700
Tyrosine	2.1	4.3	4220	11	46,400
Valine	3.0	3.9	3000	16	48,000
Total found	88.2	93.04		343	
Mean \pm Standard deviation					47,300 \pm 600

* Values in bracket are omitted from the mean.

Since electrophoresis is frequently used as a method for the determination of homogeneity in protein hormone preparations (see : General Endocrinology, p. 27) we show the electrophoresis patterns of pure somatotrophin as an example :



Electrophoresis patterns of the ascending boundary of somatotrophin preparations
(Courtesy of Dr. C.-H. Li)

- (a) acetate buffer of pH 4.0, 120 minutes electrolysis;
- (b) acetate buffer of pH 4.95, 540 minutes electrophoresis;
- (c) barbiturate buffer of pH 9.60. All buffers of 0.10 ionic strength; 1.5°C.



Somatotrophin (the growth hormone) as it appears at a magnification of 150.
(Courtesy of Dr. C.-H. Li.)

It will be noted that the hormone migrates as a single component in all three buffer solutions. If the protein in the three sections of the electrophoresis cell is separately recovered, no difference in growth potency of the three fractions is noted. The homogeneity of the preparation with regard to its electro-chemical properties is thus demonstrated by the impossibility to achieve separation by electrophoresis.

Somatotrophin has recently been crystallized from a dilute solution of 15% ethanol at 2°C. The crystals appeared as thin plates and were highly soluble at room temperature. They are shown on this page.

As regards the RESISTANCE OF HYPOPHYSEAL HORMONES TO VARIOUS AGENTS, the following facts are noteworthy :

(1) FSH and LH. — PROTEIN PRECIPITANTS (picric acid, picrolonic acid, flavianic acid) are claimed to inactivate FSH, in concentrations which do not affect LH. Allegedly 2.5% trichloroacetic acid completely precipitates LH, but not FSH. Further work with pure hormone preparations will be necessary, however, since these claims have not been uniformly confirmed.

CYSTEINE, which reduces -S-S- linkages in proteins, does not readily inactivate FSH and LH preparations since the disulphide-bonds of these protein-hormones are less reactive in this respect than those of many other proteins. However, upon long continued reaction with cysteine, this type of inactivation does occur.

KETENE is a mild acetylating agent, which attacks the amino, phenolic hydroxyl and sulphydryl group in aqueous protein solutions. FSH is comparatively resistant, while LH is rapidly inactivated during the process of ketene acetylation. It is assumed that this inactivation is due to the acetylation of the free amino-groups, and that the amino-groups are essential for activity, but this has not yet been definitely proven.

Among the ENZYMES, crystalline pure trypsin destroys the activity of LH more readily than that of FSH, while pepsin tends to destroy FSH more readily than LH, but only upon prolonged reaction. Ptyalin selectively abolishes FSH activity, LH being much more resistant.

(2) LUTEOTROPHIN. UREA greatly increases the viscosity of luteotrophin without permanently inactivating it, since, if the urea is removed by dialysis, the original viscosity and activity return to normal. DETERGENTS of the sodium alkylarylsulfonate-type (Nacconol) likewise reversibly increase the relative viscosity and diminish the activity of the hormone.

Luteotrophin is destroyed by certain ENZYMES (pepsin and trypsin). In the absence of salt, at pH 8.0 luteotrophin shows little loss in potency after boiling for one hour; it is more resistant to heat in acid than in alkaline solution.

(3) CORTICOTROPHIN. KETENE acetylates both the phenolic and the amino-groups of corticotrophin; this diminishes its hormonal activity. Here again, it is assumed that the biologic action is, at least partly, dependent upon the free amino-groups. That free amino-groups are essential for the activity of corticotrophin is also shown by other observations. Thus, compounds which attack these groups (e.g., NITROUS ACID and FORMALDEHYDE) readily destroy the potency of corticotrophin.

The IODINE uptake of the hormone corresponds roughly to its tyrosine content; since during iodination, the biologic potency is diminished, tyrosine groups appear to be essential for the corticotrophin action.

Partial digestion of corticotrophin with ENZYMES yielded most interesting results. With trypsin, no significant diminution in corticotrophin activity is observed until about 18% and with pepsin until about 50% of the hormone is hydrolyzed. This indicates that the products of hydrolysis whose molecular

weight is lower than that of the hormone (presumably polypeptides), still possess corticotrophic activity.

(4) SOMATOTROPHIN. Denaturation of somatotrophin by treatment with UREA does not influence its growth-promoting potency. From the loss of activity following treatment with NITROUS ACID and KETENE, it was concluded that here again free, primary amino-groups are essential for the hormonal potency. Proteolytic ENZYMES, such as trypsin or pepsin, greatly diminish the potency of somatotrophin upon incubation at 37° C. Somatotrophin is rather heat-sensitive. Activity is completely lost at pH 4.0-8.9 following boiling for 10 minutes. It is somewhat more stable in alkaline than in acid solutions.

Chemistry and Biologic Characteristics of Intermedin. — INTERMEDIN (*Synonyms*: middle-lobe hormone, chromatophorotropic hormone, melanophore-expanding principle, B-hormone), is a protein-like substance found in the middle or intermediate-lobe of the pituitary, in animals in which there is an anatomically distinct structure of this type. Similarly, tissue cultures of intermediate-lobe cells are rich in this substance, while those of the anterior or posterior-pituitary are free of it, or contain only traces which may have entered it by diffusion from the intermediate-lobe. Such secondary diffusion is probably also responsible for the presence of intermedin in the simple posterior-lobe extracts (e.g., "pituitrin") commonly in clinical use. In those species which possess no distinct pars intermedia (e.g., porpoise, whale, fowl), intermedin is found only in extracts of the anterior-lobe, not in those of the posterior-pituitary. On the basis of all these observations, it has been assumed that intermedin is a distinct hormone, elaborated by the intermediate-lobe cells or, in species in which there is no middle-lobe, by the anterior-lobe cells.

In poikilothermic animals (e.g., toad, frog), whose skin contains contractile

pigment cells (chromophores or chromatophores), the function of this hormone appears to be clearly established. Injection of intermedin causes the pigment granules in the melanophores to become dispersed throughout the bodies and branches of these cells, thus causing a darkening of the skin. For instance, in the frog, whose chromatophores ("melanophores") contain black granules of melanin, intermedin causes the skin to become coal black. In other species, in which the chromatophores contain red ("erythrophores") or yellow ("xanthophores") pigment granules, the hormone produces a red or yellow coloration respectively. Since in these same species, hypophysectomy causes blanching of the skin, while intermedin restores its color to normal, the substance appears to represent a physiologic hormonal principle regulating skin coloration. Not only the distribution, but even the production of melanin in the skin of the frog is allegedly stimulated by intermedin. It has also been claimed to accelerate the oxidation of the pigment producing tyrosine-tyrosinase system *in vitro*.

The rôle of intermedin in mammals, especially its influence upon cutaneous pigmentation in man, is still a debated problem.

Treatment with alkali greatly increases the potency of intermedin. The most purified preparations cause definite darkening of the frog's skin at a dose level of 0.005γ and are 73 times as active as the standard posterior-pituitary powder. They are practically free of oxytocin and vasopressin. They contain tyrosine, arginine and cysteine.

Chemistry and Biologic Characteristics of the Posterior-Lobe Hormones. — Posterior-lobe extracts exert three chief activities : they raise the blood pressure (due to their vasoconstrictor effect), they cause uterine contractions and they diminish diuresis. Separation of the oxytocic and vasoconstrictor actions by chemical means is

possible, but whether the anti-diuretic activity can be separated from vasoconstrictor extracts is very doubtful. It is generally agreed therefore that there are at least two distinct posterior-lobe principles, the oxytocic and the vasoconstrictor hormones respectively; the latter probably being responsible for the anti-diuretic action. Some investigators believe that these two fractions are artefacts and due to the breakdown in the course of the extraction procedure of an originally single principle, but this view has not been proven. The compounds appear to be polypeptides, with a comparatively small molecular weight of about 2,000. The usual commercially available "posterior-pituitary injection" or "pituitrin" preparations are simple extracts containing all three activities.

It is also debatable whether the posterior-lobe principles are true hormones, in the sense of the definition given under "General Endocrinology" (p. 11). Some investigators regard them as pharmacologically active tissue extracts, which do not perform any physiologic, hormonal functions. This view is mainly based upon the comparatively vague deficiency syndrome, following extirpation of the posterior-lobe and the apparently non-glandular nature of the neuro-hypophysis. Yet, the pituicytes may well be the producers of true hormones and the absence of pronounced posterior-lobe-deficiency symptoms following complete hypophysectomy is probably due to the simultaneous ablation of the anterior-lobe, whose presence appears to aggravate posterior-lobe deficiency. Furthermore, cells of the tuber cinereum and of the adjacent hypothalamic region can perhaps partly compensate for the lack of the pars nervosa.

It is regrettable that many of the scientific terms used for the designation of crude pituitary extracts or purified posterior-lobe fractions, have frequently been employed to designate specific products sold by various pharmaceutical

companies. Consequently, a clear-cut distinction between trade names and scientific names is somewhat difficult.

(1) **VASOPRESSIN.** *Synonyms:* vasopressor principle, postlobin-V, pitressin, vasopressor-anti-diuretic principle. The hormone has not been isolated, but some preparations contain as much as 200 pressor units (and only 10 oxytocic units) per mg. Such highly purified pressor preparations have been proven to contain arginine, proline, isoleucine, cysteine and tyrosine. Histidine, hydroxyproline and glycine were shown to be absent, while tryptophane is present only in traces, if at all. The activity is readily destroyed by hydrolysis with dilute acid or alkali, active reducing agents and the enzymes of the gastrointestinal tract.

The "anti-diuretic hormone" of the posterior-pituitary is probably identical with vasopressin, since claims concerning the separation of these two activities have not been confirmed.

(2) **OXYTOCIN.** *Synonyms:* oxytocic hormone, oxytocic posterior-lobe principle, postlobin-O, pitocin. The chemical and physical properties of oxytocin are very similar to those of vasopressin. Hence the separation of the two hormones is difficult. Oxytocin has not yet been isolated, but highly purified preparations contain as much as 250 oxytocic units (and only 5 pressor units) per mg. The amino-acid composition of these highly purified preparations is almost the same as that of vasopressin,

except that they appear to contain leucine instead of isoleucine. Oxytocin is readily destroyed by an enzyme ("pitocinase") contained in human blood. Since the blood-concentration of this enzyme rises rapidly during the first weeks of gestation, it may even act as an indicator in the diagnosis of pregnancy.

It is noteworthy that simple posterior-pituitary extracts inhibit diuresis only when the osmotic pressure of the urine is low. The same preparations exert a diuretic action, at times when urine with a high osmotic pressure is excreted. Both diuretic and anti-diuretic actions are accompanied by increased urinary elimination of certain salts. This led to the assumption that the diuretic action is dependent upon the salt output. If posterior-lobe extract is given when the salt concentration in the urine is already high, the increase in salt excretion necessitates a further rise in water output. If rats are given much water by stomach tube, 0.0001 oxytocic units per 100 gm. of body weight of a highly purified oxytocin preparation, have a definite diuretic action. This quantity of the substance weighs only 0.0000004 mg. It seems very improbable that the diuretic action is due to a contamination in this quantity of material. Probably it is oxytocin which causes diuresis in hydrated animals, while vasopressin elicits merely "salt-diuresis" after dehydration, and only at low dose levels.

GENERAL PHARMACOLOGY OF THE HYPOPHYSEAL HORMONES

STANDARDIZATION

Analytic Methods for the Detection of Hypophyseal Hormones. — There are no satisfactory analytic methods for the determination of any of the anterior, middle or posterior-lobe hormones. All these principles are estimated by bioassay.

Bioassay of the Follicle-Stimulating Hormone. — The INTERNATIONAL UNIT

(I.U.) of FSH is defined as the specific gonadotrophic activity of 250 γ of a standard pregnant-mare-serum preparation kept at the Department of Biological Standards, National Institute for Medical Research, London, England. It is recommended only for the assay of gonadotrophic preparations of the serum of pregnant mares. Recent progress in the purification of FSH will necessitate

the establishment of a revised and more generally applicable I.U.

The most satisfactory test-object for the bioassay of FSH is the HYPOPHYSECTOMIZED RAT. In females deprived of their pituitaries, pure FSH preparations cause only follicle maturation without luteinization or stimulation of the degenerated theca cells ("wheel cells"). One rat unit is defined as the minimal total amount which, given in three, daily, subcutaneous injections to hypophysectomized rats (26-28-day-old at operation and 6-8 days postoperative when injections are commenced), causes appearance of normal, non-atretic, antrum-bearing follicles 72 hours after the first injection (*Evans et al. 1939*). If the thecal "wheel-cells," which are characteristic of hypophyseal deficiency in rats, disappear during the assay, the preparation can be regarded as contaminated with LH.

In male hypophysectomized rats (21-day-old at operation, used two days postoperatively) after four daily injections, autopsy on the 5th day reveals a testis enlargement proportionate to the FSH concentration of the preparation. If the preparation is free of LH this is unaccompanied by any stimulation of the accessory sex organs or of the Leydig cells, the enlargement being due exclusively to stimulation of the seminiferous tubules (*Greep et al. 1940*). This technic is unreliable in the presence of LH, since the latter also causes some testis enlargement.

Other methods for the bioassay of FSH use intact female rats as test objects and are based upon the AUGMENTATION OF THE OVARY-STIMULATING EFFECT CAUSED BY SIMULTANEOUSLY GIVEN LH, the RAPID STIMULATION OF FOLLICLE MATURATION, the INCREASE IN UTERINE WEIGHT or the PRODUCTION OF CORNIFIED VAGINAL SMEARS. All these technics are less reliable, since compensatory gonadotrophin secretion by the animal's own pituitary tends to blur the picture; indeed the response elicited in

some of these tests (e.g., estrus changes) are due to secondary LH-production and not to the injected FSH itself.

Bioassay of the Luteinizing Hormone. — The INTERNATIONAL UNIT (I.U.) of LH is defined as the specific gonadotrophic activity of 100 γ of the standard human pregnancy urine preparation kept at the Department of Standards, National Institute of Medical Research, London. It is recommended only for the assay of LH from human pregnancy urine. This I.U. is approximately equivalent to most of the rat units (R.U.) currently in use. Recent progress in the purification of LH will necessitate the establishment of a revised and more generally applicable I.U.

In HYPOPHYSECTOMIZED FEMALE RATS, pure preparations of LH cause restoration of the involuted interstitial cells ("wheel-cells"). Immature rats (26-28-day-old, injections commenced 6-8 days postoperatively) are given three, daily, intraperitoneal injections and are killed 72 hours after the first injection. The minimum amount sufficient to cause just detectable restoration of the "wheel-cells" is considered one unit (*Simpson et al. 1942*). It should be kept in mind that the hormone is about five times less active by the subcutaneous than by the intraperitoneal route.

LH can also be assayed in HYPOPHYSECTOMIZED MALE RATS (21-day-old at operation, injections commencing 2 days postoperatively), given 4, daily, subcutaneous injections with autopsy on the 5th day. In this test the increase in the weight of the *ventral prostate* acts as an indicator of activity. The unit is defined as the dose necessary to cause a 100% increase in prostatic weight, as compared with untreated controls (*Greep et al. 1941*). The sensitivity of the method can be increased by intraperitoneal administration of the hormone. This technic has the advantage that FSH does not potentiate the above-

mentioned action of LH and hence, contamination with FSH does not interfere with the results.

Another bioassay technic is based upon the morphologically detectable stimulation of the Leydig cells by LH in hypophysectomized male rats.

The increase in the OVARIAN WEIGHT, LEYDIG-CELL DEVELOPMENT or SEMINAL VESICLE WEIGHT IN INTACT IMMATURE RATS, the MELANIN DEPOSITION IN THE BREAST FEATHERS of the African weaver finch, etc., have also been recommended as indicators; however, these technics are less specific than the assay on hypophysectomized animals, mainly because the endogenous production of gonadotrophins by the pituitary can obscure the results.

Bioassay of Comparatively Impure Gonadotrophin Preparations. — Most of the above-mentioned technics are too complicated or time-taking for clinical purposes; hence, several simpler bioassay methods have been developed for the routine estimation of gonadotrophins in blood and urine. Although these are not sufficiently specific or sensitive for the assay of purified FSH or LH, they are useful in the study of clinical problems. The following are of special value :

(1) THE ASCHHEIM-ZONDEK TEST (1929), as originally described, is performed on a group of five, 3 to 4-week-old female, white mice, each weighing 6-8 gm. To 25-30 cc. of a morning sample of urine, one drop of tricresol is added as a preservative, and then it is slightly acidified with a few drops of 10% acetic acid. The urine thus prepared is injected subcutaneously twice daily on three consecutive days, each mouse receiving a total of 6 injections. The individual doses, each given 6 times to one of the 5 animals, are : 0.2, 0.25, 0.3, 0.35 and 0.4 cc. Autopsy is performed 96 hours after the first injection. If time permits, the ovaries are examined histologically, but usually, examination with a magnifying lens suf-

fices. The test is considered positive if there is a single hemorrhagic follicle, the so-called "Blutpunkt" (anterior-pituitary reaction or "APR" II) or corpus luteum (APR III) in any of the test animals. The finding of mature, but unruptured and not luteinized follicles (APR I), indicates the presence of FSH. The test is especially useful in the early diagnosis of pregnancy, choriocarcinomas and hydatidiform moles, all of which cause a positive LH reaction (APR II or III), while the urine of castrates tends to elicit only follicle maturation (APR I) due to its high FSH content. (See also pp. 377, 378.)

The major objections to this test, as originally devised, are that it is rather time-taking and that the urine is sometimes so toxic that it kills the experimental mice. Hence, many modifications have been developed (see below) in an effort to partially purify the gonadotrophic preparations used for assay and to improve experimental conditions, in order to obtain more rapid results.

(2) The OVARIAN HYPEREMIA TEST is based upon the fact that LH causes marked hyperemia of the ovary, as early as 2-6 hours after injection. This reaches a maximum after 24 hours and disappears in 48 hours. Since this hyperemia precedes the formation of follicles or corpora lutea it can act as an early indicator of LH activity. The hyperemia is apparently due to LH, although FSH seems to act as an augmenting factor. The test is particularly useful in the rapid, early detection of pregnancy (e.g., in cases suspected of ectopic gestation, where an immediate decision must be made about the necessity of a surgical intervention). Several technics are in use. Zondek *et al.* (1945) designated as "one hyperemia unit that amount of gonadotrophic hormone which induces hyperemia of the whole ovary in an infantile female rat within 24 hours."

For this modification, the infantile rat proved preferable to the mouse. A

total dose of 4 cc. of pregnancy urine is given to prepubertal rats subcutaneously in two injections of 2 cc., at an interval of one hour. If the test is read two hours after the injection, it is extremely unreliable, but after six hours, it gives positive results during gestation in 92.2% of the cases, and after 24 hours, in 100% of the cases. False positives in non-pregnant women are usually due to LH-producing tumors. Even readings after two hours are reliable, if the test is positive.

(3) The FRIEDMAN TEST (1929) is also rapid and simple. It is performed on adult female rabbits, which have been isolated from the male for at least three weeks prior to the test (in order to avoid pseudopregnancy). The morning urine is filtered and 10 cc. of it are injected into the marginal vein of an ear. For preservation, two drops of cresol are added to each ounce of the remaining urine and the specimen is kept in a refrigerator for 24 hours, when a second similar injection is made. 48 hours after the first injection, the rabbit is killed and the ovaries are examined. The macroscopically detectable hemorrhagic follicles are indicative of a positive pregnancy test.

If the urine proves to be toxic, 30 cc. of filtered urine are shaken for 3-5 minutes, with 90 cc. of ether. The excess is poured off and the residual ether is removed from the urine by an electric fan. 0.9 gm. of glucose is added to the remnant and then this detoxified specimen is injected in the usual manner. This test gives very reliable results, but only animals weighing more than 800 gm. can be used and the necessity for maintaining a stock of isolated females renders the assay somewhat tedious.

(4) The XENOPUS TEST (*Shapiro and Zwarenstein, 1933; Weisman et al. 1942, etc.*) is based upon the fact that following injection of detoxified pregnancy-urine extracts into the dorsal lymph-sac of female, South-African,

clawed frogs (*Xenopus laevis*), extrusion of ova occurs within 24 hours. The eggs are readily detected either in the water in which the animals lay, or following autopsy, within the oviducts. This test is alleged to give 98.6% correct results in the diagnosis of pregnancy. At present, in most countries, the difficulty of obtaining these frogs interferes with the general applicability of this, apparently highly satisfactory test.

(5) The FROG TEST (*Galli-Maini, 1947*) takes as a criterion the extrusion of spermatozoa by the male frog, which is readily elicited by pregnancy urine. It appears to be a simple and reliable technic.

(6) A number of OTHER TESTS have been developed in which chemical purification of the gonadotrophins permits the administration of amounts which would not be tolerated by experimental animals, if the original body fluids were injected as such. The biologic assay of the concentrates is otherwise essentially the same as in the previously-mentioned tests.

Bioassay of Luteotrophin. — The INTERNATIONAL UNIT (I.U.) of luteotrophin (prolactin) has been defined as the specific activity contained in 100 γ of a standard preparation, made from anterior-pituitary tissue and kept at the Department of Biological Standards, National Institute for Medical Research, London. The use of this unit was recommended "for recording the activity of all crop-gland stimulating preparations."

(1) The CROP-SAC-WEIGHT METHOD (*Riddle et al. 1933*) is based upon the observation that the combined weight of the two excised crop-sacs of pigeons is proportional to the amount of luteotrophin injected. 6-10-week-old pigeons are injected intramuscularly once daily on 4 days and autopsied about 96 hours after the first injection. Under these conditions, the weight of the crop sacs is a linear function of the log of the dose administered. It is advisable always to compare results with those obtained under equal conditions with the standard preparation, since num-

erous factors (body weight, breed, season, light, temperature, etc.) influence the sensitivity of the experimental animals. Subcutaneous injections are most effective and intraperitoneal administration is least efficacious.

(2) The MINIMUM CROP-SAC-STIMULATION METHOD is based upon the fact that minute doses of luteotrophin cause definite crop-gland proliferation detectable if the dissected glands are examined against light (*McShane and Turner, 1936*). The pigeon unit is defined as "the total amount of hormone injected during a period of 4 days, which causes a minimal, but definite proliferation of the crop-glands of $50 \pm 11\%$ in 20 common pigeons weighing 300 ± 40 gm." *Li (1947)* uses Silver King pigeons, 4 to 5 weeks of age and weighing 400-550 gm., which are injected subcutaneously once daily for 4 days, with 0.5 cc. of the hormone-containing solution. 24 hours after the last injection, the crop-gland is dissected and examined against light for a positive reaction. The unit is defined as the minimum amount necessary to produce a positive response in two out of three birds. Three birds per group are reported to suffice for an assay.

(3) The LOCAL INTRADERMAL CROP-SAC TEST OR MICRO METHOD (*Lyons and Page, 1935*) is so sensitive that it detects $1/10,000$ of a unit as determined by the "minimum crop-sac-stimulation test." The hormone solution (0.1 cc.) is injected intradermally into the skin covering one of the crop-sacs on 4 consecutive days. On the 5th day, the birds are killed and the crop-sacs dissected, stimulation being estimated by comparing the transparency to light of the crop-sac on the injected side with the contralateral one.

(4) OTHER TECHNICS are based upon the milk secretion-stimulating-action of luteotrophin in the suitably pretreated rat or guinea pig, or its ability to maintain a functional corpus luteum in hypophysectomized animals.

These methods are less accurate and more complicated than the pigeon tests, mentioned above.

Bioassay of Corticotrophin. — No INTERNATIONAL UNIT of corticotrophic activity has as yet been accepted.

(1) The REPAIR TEST (*Simpson et al. 1943; Sayers et al. 1943*) is usually performed on 26-28-day-old hypophysectomized female rats, which are used 14 days postoperatively when the adrenals have already undergone pronounced involution. The hormone is administered once daily on 4 consecutive days and the animals are sacrificed 96 hours after the first injection. The adrenals are fixed in formalin and frozen sections are stained with Sudan Orange. The lowest dose which causes recognizable REDISTRIBUTION OF THE CORTICAL LIPIDS is considered as the unit; the adrenal weight is not significantly increased even after 100 such units, hence the histologic control is far more sensitive than that based on a weight increase.

(2) The MAINTENANCE TEST (*Simpson et al. 1943*) is based upon the ability of corticotrophic preparations to maintain the weight of the adrenals in animals in which injections are commenced immediately after hypophysectomy. 40-day-old male rats are hypophysectomized and injected intraperitoneally once daily during two weeks. By the end of this period, the adrenals of non-treated hypophysectomized rats have regressed from the normal average of 26 mg. to a constant weight of approximately 12 mg. The amount of corticotrophin necessary to maintain the adrenals at the 26 mg. level is defined as one "Maintenance Unit." The sensitivity of this test is greatly augmented if the daily dose is divided into 2 or more injections. Since the strain and body-weight of the test rats influences sensitivity, it is well to compare the results with those obtained using a standard preparation under the same conditions and to express the adrenal

weight increase per 100 gm. of body-weight.

(3) OTHER METHODS OF ASSAY are based upon the *weight increase produced by corticotrophin in the remaining adrenal of a hypophysectomized animal*, whose other adrenal was removed after hypophysectomy had caused significant involution. The effect of corticotrophin on the *ascorbic acid or cholesterol content of the adrenals* may also be used as an indicator of activity in hypophysectomized rats.

The assay of corticotrophic preparations in intact animals is not to be recommended, since numerous toxic extracts cause a discharge of corticotrophin from the animal's own pituitary, due to the resulting general-adaptation-syndrome. The activity of such endogenous corticotrophins can naturally not be differentiated from that of the injected hormone and this seriously interferes with accurate bioassay. There are no generally accepted methods for the assay of gluco-corticotrophic, mineralo-corticotrophic and other possible specific stimulants regulating the selective production of certain steroids by the adrenal.

Bioassay of Thyrotrophin. — The INTERNATIONAL UNIT (I.U.) of thyrotrophin has been defined as the specific thyrotrophic activity, equivalent to 250γ of a standard pituitary preparation, kept at the Department of Biological Standards, National Institute for Medical Research, London. The third International Conference on the Standardization of Hormones (1938) agreed that only those tests can be considered as safe, which are based on the actual observation of thyroid stimulation, since other, indirect effects may be due to non-specific impurities.

(1) In the RAT (*Anderson and Collip, 1934*) thyrotrophic hormone can be assayed on the basis of the increase in B.M.R. or the histologically-detectable thyroid stimulation which it produces when given after hypophysectomy. In

view of the fact that pituitary principles other than the thyrotrophic hormone may also raise the B.M.R., the latter indicator is more reliable.

(2) The GUINEA-PIG is the most commonly employed test-object for thyrotrophin, because of its great sensitivity to the hormone. *Rowlands and Parkes (1934)* adopted as a unit "the thyrotrophic activity contained in an amount of extract which, given daily for 5 days, will cause the thyroid of the 200 gm. guinea pig to attain a weight of 600 mg., i.e., about double the normal." Essentially similar assay technics are used by several investigators, although some prefer to measure the average height of the follicular epithelium rather than the weight of the gland.

Heyl and Laqueur (1935) worked out a scale of 6 different stages based on histologic signs of activity; they designated these by the letters p-u. A "border-line dose" is defined as the amount, which given in two intraperitoneal injections on two consecutive days, will cause (within 48 hours) in 2/3 of the treated 150-200 gm. guinea-pigs a reaction "s" in the middle-part of the thyroid. This reaction "s" consists of a thickening of the cells in which the nucleus became round and the cytoplasm developed on the distal cell-pole equals the diameter of the nucleus. In order to keep this unit as close as possible to those generally in use, one "cavia unit" is defined as 1/4 of the "border-line-dose."

Several modifications of these tests are in common use at present.

(3) IMMATURE CHICKS are also very sensitive to thyrotrophin (*Stimmel et al. 1936; Smelser, 1937*). *Bergman and Turner (1939)* employed the one-day-old white Leghorn chick, emphasizing that males are more sensitive than females. They defined the unit of thyrotrophic activity as the total amount of hormone administered subcutaneously once daily during 4 days, causing a mean increase of 50% (about

5.4 ± 0.26 mg.) in the thyroid weight of 20 chicks whose body weight averages 55 ± 10 gm.

Since the thyroid is not as sensitive to the effect of non-specific damaging agents as the adrenal cortex, it is not always necessary to perform thyrotrophin assays on hypophysectomized animals. Nevertheless, in view of the great sensitivity of the gland to certain dietary constituents, it is well to keep the test animals on "resting diets." Some authors even recommend pre-treatment with iodine or diiodotyrosine before the test, to assure that the gland shows no signs of excess activity before the injections are started. The rationale of this procedure is open to question however, since iodine compounds diminish the responsiveness of the thyroid to thyrotrophin and thus decrease the sensitivity of the test.

(4) Various OTHER TESTS are based upon the decrease in the *iodine content of the thyroid* induced by thyrotrophin in the rat or guinea pig; the *restoration of the atrophic thyroid epithelium in the hypophysectomized rat or pigeon*, the *stimulation of the thyroid in the intact grass snake (tropidonotus natrix)*, in which the gland is continuously in the resting condition, unless stimulated by exogenous thyrotrophin, etc.

Bioassay of Somatotrophin. — An INTERNATIONAL UNIT of somatotrophin activity has not yet been established.

(1) The BODY GROWTH IN NORMAL INTACT FEMALE RATS about 5-6 months of age and weighing 220-280 gm. is a good indicator of somatotrophin activity, since such animals are "plateaued," that is to say, their spontaneous growth rate is practically at a standstill (*Evans and Simpson, 1931*). Usually the hormone preparation is injected daily, intraperitoneally or subcutaneously, in a group of at least 10 normal "plateaued" female rats for 20 days. The dose which causes an increase in body weight of 40-60 gm. during this period, lends itself best for such assays. The unit

is defined as the daily dose required to produce a total body weight increase of 40 gm. in 20 days. Since the slope of the growth line increases towards the end of the injection period, a shorter course of injections is not recommended.

(2) The BODY GROWTH OF HYPOPHYSECTOMIZED FEMALE RATS is a more sensitive test-object for somatotrophin. Since the weight, age, sex, strain and general condition of the animals influence the results obtained, it is best to use a standard preparation for comparison under identical experimental conditions. Only animals in which the completeness of the hypophysectomy is established by such criteria as cessation of growth, decreased muscular tonus, and maintenance of the fluffy infantile lanugo fur, should be used for the test; the sella must be carefully examined at autopsy, in order to eliminate animals with remnants of anterior-lobe tissue. A unit is defined as the daily dose which causes an average weight gain of 10 gm. in a group of 10 hypophysectomized female rats, 28-30 days of age at operation, and used 10-14 days postoperatively, at which time a course of 9 daily, intraperitoneal injections is commenced and the animals are sacrificed on the 10th day. For greater accuracy, a 15-day injection period is recommended. A straight line is obtained if the gain in weight is plotted against the log of the dose given. Animals should not be used for such tests more than once since some adaptation (anti-hormone formation?) to somatotrophin occurs in time.

(3) HISTOLOGIC CHANGES IN THE BONES (e.g., TIBIA) OF HYPOPHYSECTOMIZED RATS likewise furnish a useful basis for bioassay purposes. (*Freud et al. 1939; Ray et al. 1941; Evans et al. 1943*). 26-28-day-old hypophysectomized female rats are used for the test 12-13 days postoperatively; the hormone is administered intraperitoneally, once daily on 4 successive days.

Autopsy is performed 24 hours after the last injection and the right tibia of each animal is dissected, split with a razor blade and fixed in neutral formalin. The calcified part of the bone is stained with AgNO_3 and $\text{Na}_2\text{S}_2\text{O}_3$. The uncalcified portion of the epiphysis can then be measured under the microscope with a calibrated eye-piece micrometer. The width of the uncalcified cartilage, plotted against the log of the dose of hormone injected, gives a straight line relationship. The test is based upon the fact that after hypophysectomy, the width of the epiphyseal cartilage-plate decreases rapidly, although cartilaginous growth and bone formation may continue for a short time in young animals. This decrease in thickness is due to a disturbance in the normal equilibrium between cartilage and bone formation. Administration of somatotrophin rapidly restores the thickness of the epiphyseal cartilage plate by first stimulating chondrogenesis and then osteogenesis, until the normal equilibrium is re-established. This test is approximately 3 times as sensitive as that based upon body growth.

Bioassay of Other Hypophyseal Hormones. — No generally accepted bioassay procedures have as yet been devised for the estimation of other possible anterior-lobe hormones. This is not unexpected, since the very existence of additional anterior-pituitary hormones is still in doubt. The divers actions of impure anterior-hypophyseal extracts are customarily studied under the optimum conditions for the manifestation of their effects, (see above: Classification and Chemistry of the Hypophyseal Hormones). Thus the nephrosclerosis-producing action of anterior-lobe extracts is best demonstrated on unilaterally nephrectomized rats, maintained on a high-sodium and high-protein diet; the diabetogenic action by the resulting destruction of the Langerhans islets, or the hyperglycemia and

glycosuria in the dog or partially pancreatectomized rat, etc.

Bioassay of Intermedin. — (1) In the HYPOPHYSECTOMIZED FROG (*Rana pipiens*) weighing 30-40 gm. (Teague, 1939; Calloway *et al.* 1942) the potency of intermedin may be assayed about 24 hours after the operation, when melanophore contraction becomes maximal. The same animals may be used repeatedly so long as 24 hours elapse between complete contraction of the melanophores and re-injection. Injections are made into the ventral lymph sac, through the floor of the mouth. It is important to control the pH of the injected specimen and always to compare results with those given by a known specimen at the same pH and at the same temperature. Reading is done under a binocular dissecting microscope, using as a criterium of activity the length of time required for the melanophores to return to full contraction after injection. A minimum of six animals should be used for each unknown and six for the reference standard. The melanophore-hormone unit is defined as the activity equivalent to that of one γ of alkali-treated U.S.P. XIII Posterior-Pituitary Reference Standard.

(2) In the SOUTH AFRICAN CLAWED FROG (*Xenopus laevis*), intermedin may be determined with a probable error of about 10% (Langrebe and Waring, 1941). The maximum melanophore contraction attained after injections of extracts into the dorsal lymph-sacs of fully blanched intact or hypophysectomized animals is used as an indicator. In the intact frog, depigmentation is achieved by keeping the animals on a white background.

It has been suggested that the international unit of melanophore activity in this test be defined as the amount contained in 0.5 mg. of the international standard powder. However, this international unit has not yet been officially accepted.

(3) Among OTHER BIOASSAY techniques, the melanophore-dispersing action in hypophysectomized reptiles (*anolis*) *Kleinholtz and Rahn* (1940), or the erythrophore-dispersing action in fish (*phoxinus*) *Zondek and Krohn* (1932) have also been recommended. Some investigators believe, however, that the latter test is not specific, because the erythrophores respond to a hormone different from that influencing the melanophores.

Bioassay of Vasopressin. — There are no generally accepted convenient and reliable methods for the bioassay of vasopressin. The official (U.S. Pharmacopeia) posterior-lobe preparations are standardized by their oxytocic potency, which, in most commercially available extracts, runs roughly parallel with the vasopressor activity. Unofficial preparations are available, however, which contain the vasopressor principle in a highly purified state. These are usually assayed for their pressor activity in anesthetized dogs, one unit of pressor action representing that exhibited by 0.5 mg. of the U.S.P. XIII reference standard powder (see: "Bioassay of Oxytocin," below).

Bioassay of Oxytocin. — The most generally accepted tests for the bioassay of oxytocin are based on a comparison of the unknown preparations with the U.S.P. XIII POSTERIOR-PITUITARY REFERENCE STANDARD. 0.5 mg. of this powder is defined as 1 U.S.P. POSTERIOR-PITUITARY UNIT. According to the prescriptions of the U.S. Pharmacopeia XIII, "Posterior-Pituitary Injection" is a sterile, aqueous solution of the water-soluble principles from the posterior-lobe of the pituitary of healthy domestic animals used for food by man. The pituitary must be removed immediately after killing the animals, then dried and either extracted at once or kept in a frozen state until extracted. The potency of Posterior-Pituitary Injection must be 1 U.S.P. posterior-pituitary unit per 0.1 cc. The U.S.P.

reference standard is prepared under the supervision of the U.S. Pharmacopeia Committee of Revision and is distributed through the office of the Chairman.

Oxytocin is usually assayed on the GUINEA PIG uterus (U.S.P. XIII). The test-animals must weigh 175-350 gm. and should neither have been pregnant nor be in heat. After killing the animal (by a blow on the head or decapitation), the uterus is immediately dissected and one horn is suspended in a bath containing not less than 100 cc. of oxygenated Locke-Ringer solution and kept at a temperature of 37-38°C. One end of the uterine horn is attached to the muscle lever of a suitable kymograph and the assay is commenced 15-30 minutes later, when the uterus is completely relaxed. Appropriate quantities of the preparation to be assayed and of the reference standard are weighed and diluted with isotonic NaCl solution. First, one determines the quantity of the diluted standard-solution and of the preparation to be assayed, which, when alternately administered, elicit a series of four, approximately equally intense, contractions; two with the standard and two with the unknown. After this, a third dose of the diluted standard-solution is given, which is 25% larger than the two preceding doses of the standard. The height of each of 5 contractions is measured. The first four contractions are considered submaximal and equivalent; they constitute an adequate assay, if the difference in height between the highest and lowest of these four contractions is less than half the difference in height, between the lowest of the four and that elicited by the increased dose of the standard. The potency is calculated from the quantities required to produce the four equivalent contractions and is expressed in U.S.P. Posterior-Pituitary Units. — Because of the inherent errors of the method, assays 20% above or below the potency of the

standard are generally considered acceptable.

Several OTHER METHODS for the bioassay of oxytocin have been recommended, but none of these are commonly in use.

MODE OF ADMINISTRATION

None of the pituitary hormones are active when given by the ORAL route, presumably because these proteins and polypeptides are hydrolyzed by the gastrointestinal enzymes.

All hypophyseal hormones are usually administered SUBCUTANEOUSLY or INTRAMUSCULARLY. Chemically pure anterior-pituitary preparations are not yet commercially available. They are usually distributed in the form of partially purified extracts in aqueous solution, the potency being expressed in various biologic units. Highly purified *LH* (from human pregnancy urine, human placenta or animal pituitaries) are available in ampules containing 100 to 5,000 I.U./cc. *FSH* (from pregnant mare serum) or mixtures of *FSH* and *LH* (since the two hormones mutually potentiate each other), as well as aqueous solutions of *luteotrophin*, *thyrotrophin*, *somatotrophin* and *corticotrophin* are distributed in ampules. Their potency is also expressed in various biologic units.

Clinical experience with these extracts does not yet warrant detailed discussion of recommendable doses. Meanwhile, in determining the optimum dose for any one case, the physician must largely depend upon trial and error, relying partly upon the recommendations of the pharmaceutic companies which distribute these preparations.

The use of *luteotrophin* (prolactin) has been recommended to increase subnormal milk secretion, but so far, results in lactating women are not particularly striking.

Somatotrophin is used in the treatment of dwarfism, especially the type due to anterior-lobe failure. However,

the danger of causing premature closure of the epiphyses must be kept in mind.

Thyrotrophin is recommended for the treatment of hypothyroidism, especially that due to pituitary failure.

Corticotrophin would appear to be the logical therapy of hypocorticoidism, especially in cases of anterior-pituitary failure, but at present, an adequate supply of potent corticotrophin is not yet commercially available.

Other anterior-lobe principles and *intermedin* are not yet used in clinical medicine.

The usual commercial *posterior-pituitary preparations* are the U.S.P. Posterior Pituitary Solution, standardized to contain one U.S.P. Posterior-Pituitary Unit/0.1 cc. *Pituitrin* (N.N.R.) is merely a brand of posterior-pituitary solution. U.S.P. *pitressin* (N.N.R.) contains 10 units of pressor activity and less than one unit of oxytocic potency per cc., while *pitocin* (N.N.R.) contains 10 units of oxytocic activity and less than 0.5 unit of pressor potency.

It is customary to distinguish the so-called "surgical" posterior-pituitary solution, which possesses twice the potency of the official Posterior-Pituitary Solution, from "obstetrical" Posterior-Pituitary Solutions which are of standard strength (1 unit/0.1 cc.).

LH is considerably more active when given INTRAPERITONEALLY than by the subcutaneous or intramuscular route, a fact which has already been mentioned in connection with its application to bioassays. — It is noteworthy, however, that the gonadotrophic effect of crude anterior-pituitary preparations is much more pronounced following subcutaneous than after intraperitoneal injection. In fact when given intraperitoneally they may even inhibit the gonad-stimulating effect of simultaneously, subcutaneously injected *LH*. — The mechanism of this action is not yet clear (greater intraperitoneal activity of contaminating anti-gonadotrophic substances? Greater endogenous anti-

hormone formation in case of intraperitoneal injection?).

Various hormones of the pituitary have been proven to act directly upon their target organs, following topical application. Thus, for instance LH acts directly upon the OVARY, since in the rabbit, injections into individual mature follicles can cause their selective luteinization while the untreated follicles remain unchanged. In amphibia, gonadotrophin treatment of the isolated ovary can even elicit ovulation *in vitro*.

Prolactin stimulates the CROP-GLAND of the pigeon directly, as indicated by the high activity of local administration (see : Bioassays, pp. 222, 223).

Thyrotrophin acts even when directly applied to the THYROID, a fact which can be demonstrated *in vitro*. However, this route of administration is not clinically applicable.

It is highly probable that other trophic actions upon endocrine glands, e.g., that of corticotrophin on the ADRENAL CORTEX and that of the gonadotrophins on the TESTIS, are likewise direct, but this has not yet been clearly proven by experimentation.

Intermedin has a direct effect upon CHROMATOPHORES in poikilothermic animals as shown by its local application to certain areas of the skin. This hormone has no clear-cut clinical indication as yet, although it has been claimed to be effective in certain cutaneous pigment anomalies (vitiligo, etc.).

Both posterior-pituitary principles are usually administered by the subcutaneous or intramuscular route, although they are also absorbed from MUCOUS SURFACES (e.g., vagina, nose, etc.).

Since oxytocin causes contractions of the UTERUS and vasopressin contractions of ARTERIAL SEGMENTS *in vitro*, it is evident that both these principles act directly upon their respective peripheral target-organ.

LH is readily absorbed by the TRANSPLACENTAL ROUTE as shown by the high LH concentration of the blood and urine in women during pregnancy.

SENSITIZATION AND DESENSITIZATION

Anterior-Pituitary Hormones. — There is no clear-cut evidence showing that upon continued administration true sensitization to ANTERIOR-PITUITARY HORMONES can occur. On the contrary — probably as a result of antihormone formation — chronic treatment with FSH, LH, prolactin, somatotrophin, thyrotrophin and corticotrophin causes a considerable degree of desensitization, often culminating in complete lack of responsiveness (see : Antihormones).

Intermedin. — Upon continued treatment with intermedin, the organism does not become abnormally sensitive or resistant to it, as long as a period of about one day is allowed between two injections.

Posterior-Lobe Hormones. — Oxytocin and vasopressin likewise fail to cause resistance, as a result of chronic pretreatment. However, if two injections of the vasopressin are administered in rapid succession, the second dose often proves inactive or at least less active. This phenomenon has variously been referred to as "tachyphylaxis" or "skeptophylaxis."

ACTIVATION AND INACTIVATION

FSH and LH. — The fact that LH AND FSH MUTUALLY ACTIVATE EACH OTHER, in the sense of a synergistic (potentiating) effect, has already been mentioned.

Prolonged treatment with FOLLICULOIDS (resulting in ovarian atrophy) slightly counteracts the action of FSH or LH, but apparently only because the ovary is subnormal in size at the onset. Conversely, sudden heavy overdosage with folliculooids (presumably through peripheral synergism with circulating hypophyseal luteotrophin) tends to increase the efficacy of FSH and LH preparations in intact animals, inasmuch as it permits the formation of the large pregnancy-type corpora lutea.

A number of protein-precipitants and other AGENTS CAUSING LOCAL IRRITA-

TION (e.g., salts of mercury or zinc, urine extracts, various proteins, trypan blue, tannic acid, etc.) also tend to augment the action of FSH and LH, but this is probably only due to a delay in their absorption rate. The effect is somewhat comparable to the augmentation of effect obtainable when steroid hormones are given as their esters or as crystal pellets, rather than as readily absorbable, oily solution of the free compounds. This interpretation is based mainly upon the fact that when the gonadotrophin and the irritating substances are injected at different sites, the latter exert no potentiating action.

Luteotrophin. — The effect of luteotrophin upon the ovary — unlike that upon milk secretion — is greatly augmented by folliculoids. This is apparently due to a peripheral synergism since the production, by impure anterior-pituitary extracts, of pregnancy type corpora lutea, is enhanced by folliculoids even in the hypophysectomized rat.

Intermedin and the Posterior-Lobe Hormones. — There are no noteworthy data concerning the activation and inactivation of INTERMEDIN, except its activation (*in vitro*) by alkali (p. 218).

The actions of OXYTOCIN and VASOPRESSIN are not significantly influenced by simultaneous treatment with other drugs. However, substances which delay the absorption (e.g., tannic acid) of these hormones tend to increase and prolong their effect, while conjoint treatment with oxytocin and other oxytocics or vasopressin and other vasopressor substances may lead to the phenomena of synergism.

THEORIES CONCERNING THE HYPOPHYSEAL HORMONES

Biogenesis and Metabolism of the Hypophyseal Hormones. — There are no reliable data concerning the PRECURSORS from which the various hypophyseal hormones are synthesized in the body. It is evident, however, that the

molecules of these protein-hormones must be made from the constituent amino-acids or polypeptides.

The fate of the anterior-lobe hormones, and the pathways through which they are metabolized, are likewise incompletely understood. HEPATIC DETOXIFICATION does not appear to play an important rôle in the inactivation of any hypophyseal hormone. If very large quantities are administered, or endogenously produced, they tend to be ELIMINATED IN THE URINE; presumably because of their comparatively low molecular weight. This is particularly obvious in the case of the gonadotrophins FSH and LH, of which very large quantities are excreted in the urine, especially during pregnancy and following gonadectomy. Traces of the anterior-lobe hormones, as well as intermedin and the posterior-lobe hormones, have likewise been claimed to be demonstrable in the urine, but only on the basis of indirect evidence.

As outlined in the Introduction, hormones are not likely to be "UTILIZED" by their respective target organs while they exert their effects. This has been shown in an especially convincing manner for the gonadotrophic, thyrotrophic and corticotrophic hormones whose activity appears to be entirely independent of the amount of receptive (target organ) tissue present in the organism.

Mechanism of Hypophyseal Hormone Actions. — FSH, LH, luteotrophin, corticotrophin, thyrotrophin, somatotrophin, intermedin, vasopressin and oxytocin are all believed to act directly upon their respective target organs as judged by the results of topical application *in vitro* and *in vivo*. (See : Mode of Admin. pp. 228-229.) The intimate mechanism through which they influence the receptive cells is not known, but it is probable that they act through their effect upon the enzyme mechanisms regulating organ growth and function.

It has been claimed that thyrotrophin has certain actions even in the absence

of the thyroid (e.g., exophthalmos), but the experiments upon which this conclusion was based will have to be repeated with entirely pure hormone preparations. There is no evidence to indicate that FSH, LH or corticotrophin exert any direct effect upon tissues other than the receptive cells in the gonads and adrenal cortex respectively.

The available data concerning the actions of impure anterior-hypophyseal extracts upon the pancreas, kidney,

thymus, metabolism, etc., are not sufficient to express any opinion concerning the underlying mechanism of action.

Different Kinds of Hypophyseal Hormones. — Unlike in other sections of this book, the possibility of additional, hitherto unproven, hormones of the hypophysis will not be considered here, since it has been discussed previously under the heading : "Classification and Chemistry of the Hypophyseal Hormones," (see : pp. 210-214).

EXPERIMENTAL PHYSIOLOGY OF THE HYPOPHYSIS

EXPLANTATION OF THE HYPOPHYSIS

The anterior-lobe of various animal species grows quite well in TISSUE CULTURE. The hypophyses of lower vertebrates lend themselves better to this type of experiment than those of the higher mammals, but rat and guinea-pig anterior-lobe tissue can also be maintained and may even proliferate in vitro. Parker's method (which involves the use of a fluid medium and a high oxygen tension) is especially recommended. Posterior-lobe tissue, on the other hand, is singularly unsuited for in vitro cultures.

ORGAN CULTURES of anterior-lobe tissue are rarely successful, because of the complex blood-vessel system, which does not lend itself well to perfusion with a pump. However, for many types of experiments it suffices to isolate and perfuse the entire head (e.g., of a dog), comparing the composition (e.g., hormone concentration) of the perfusion fluid before and after removal of the hypophysis from the preparation.

TRANSPLANTATION OF THE HYPOPHYSIS

Anterior-lobe tissue and to some extent, even intermediate-lobe tissue lends itself to grafting. Autotransplantation and to a lesser extent, homotransplantation of anterior-lobe tissue is often successful, but heterotransplantation has never been proven to give permanently surviving grafts.

TECHNIC OF HYPOPHYSECTOMY

The technic of hypophysectomy is different in the various species, depending mainly upon the anatomic configuration of the skull and the sella turcica. Since this is a rather delicate intervention frequently used in endocrine research we shall describe it in some detail:

It is especially simple in AMPHIBIA. In the frog, for instance, it suffices to tie the animal to a board on its back and then, under ether anesthesia, the mucosa can be incised along the midline of the palate, so as to expose the pararsphenoid. The mouth is washed with an antiseptic solution and, with a dental burr, a hole is made at the place where the median line crosses a transverse line which goes through the lateral processes of the pararsphenoid. The hole should not go all the way through to the pituitary, the last remnant of cartilage being removed with a cataract-needle. After this, the hypophysis is extracted by suction through a glass pipette.

In the anura, permanent contraction of the chromatophores (due to lack of intermedin) and great muscular weakness are the most outstanding characteristics of hypophyseal deficiency.

In BIRDS, either the trans-buccal or the trans-orbital approach may be used. Since the orbits of birds are very large, and separated from the hypophysis merely by a thin membrane of bone, it

is easy to reach the pituitary after ablation of an eye-ball.

In the DOG, we usually employ the following technic: The dog (preferably a young animal weighing 6-10 Kg.) is tied to the table on his back under light ether anesthesia. Since hypophysectomy greatly decreases resistance to injection anesthetics, ether-saturated air is introduced into the trachea through a rubber catheter during the entire intervention.

The table is slightly inclined, so that the head is at the lowest level. This prevents aspiration of mucus and blood during the operation. Two vertical steel bars are fastened to that edge of the table where the head lies. Two horizontal cross rods slide up and down along the vertical bars. The two horizontal bars are placed between the teeth of the dog and then separated as far as possible, in order to open the mouth maximally. The prominent canine teeth keep the bars from sliding out of the oral cavity. The whole table is then covered with sterile cloth, leaving only the oral cavity exposed; this is washed with an antiseptic solution (e.g., trypaflavine). A longitudinal incision is made in the midline of the soft-palate, beginning about one cm. behind the caudal margin of the hard-palate and extending 3-4 cm. backwards. The wound is opened wide by two lead sutures passed through the palate, one on each side, attached to the vertical bars after maximal lateral traction. The posterior wall of the pharynx thus becomes clearly visible when illuminated by a strong beam of light from a lamp attached to the surgeon's forehead. Then the pharyngeal mucosa is washed with antiseptic solution.

At this time, the gloves and all instruments are changed, since the first part of the operation is septic. The pharyngeal muco-periosteum is now strongly compressed with a cotton-pad, so as to render it anemic. Then a cross-incision is made, the transverse branch of which is at the level of the two ptery-

goid processes. Along the margins of the incision, the muco-periosteum is now separated and reflected from the bone, thereupon, in young dogs, the sphenoooccipital synchondrosis becomes visible as a bluish, transverse cartilage line; in old animals, a transverse crista may take its place, or it may be entirely absent. Just rostral from this line, a small emissary vein comes out of the sphenoid and since this bleeds, it serves as a readily detectable landmark, designating the place where the drill hole should be made to gain access to the hypophysis.

The drill used should be somewhat smaller than the pituitary itself. As soon as the dura is reached, the circular sinus surrounding the pituitary becomes faintly visible as a blue circle in the middle of which is the pink anterior-hypophysis. The dura is incised longitudinally with a fine lancet, upon which the pituitary protrudes as a result of intracranial pressure. At this point the gland is first loosened by means of a blunt probe and then extracted through a glass cannula which is connected with a suction pump. This is followed by the loss of several cc. of clear cerebro-spinal fluid. To ascertain that no pituitary tissue is left behind, it is advisable to scrape the whole interior of the, now vacant, sella with a small, blunt curette. Finally, the pars tuberalis is removed from the stalk with a very fine suction cannula.

It is not necessary to close the drill hole, since soon after operation a clot is formed in the sella and this prevents infection of the meninges. The soft palate is closed by two or three stitches and the mouth rinsed with antiseptic solution.

Essentially the same approach may be used for hypophysectomy in the CAT, RABBIT and MONKEY.

In the RAT, as in many other rodents, the para-pharyngeal approach is preferred. The operation is performed as follows:

(1) The ether-anesthetized rat (preferably 40-120 gm. body-weight) is

fastened to a board on its back and placed on a narrow table. The operator sits near the tail, the assistant, near the head end of the animal. A longitudinal incision of about 2 cm. is made from the submental papilla downwards.

(2) The operator introduces two bent forceps just below the thyroid artery, to the spheno-occipital synchondrosis. Since this artery is much more caudally situated than the synchondrosis, the direction in which one has to advance the forceps is at the same time ventro-dorsal and caudo-cranial. If this approach is used, it is not necessary to cut the digastric or any other muscle, since one can advance all the way through in the intermuscular spaces. When the forceps touch the sphenoid, they are handed over to the assistant without moving their tips. These forceps keep soft tissues out of the way and expose the sphenoid bone. If a cannula has been inserted into the trachea (to facilitate respiration), the forceps need not be moved until the end of the operation. With some practice, it is possible, however, to perform the operation without the use of a tracheal cannula. In this event, the forceps compressing the trachea have to be removed from time to time in order to allow the animal to breathe. Since a great deal of practice is needed to perform the operation in the latter way, beginners are advised to use the tracheal-cannula-method.

(3) After the sphenoid is exposed, the bone is cleaned, rostrad from the synchondrosis, with the help of a piece of cotton pushed with the end of a hard probe.

(4) In animals weighing 100 gm. or more, the bone is perforated by means of a No. 10 dental burr. For smaller animals, smaller burrs are used.

(5) After the bone has been removed, the dura is torn with the help of a small dental pick and the gland is aspirated. It usually comes out in one piece, if the suction is applied very gently at first, so as to lift the hypophysis out of its normal position before removing it with

strong suction. If the gland should break, one may remove the remainder from behind the bone, with the help of the dental pick.

(6) The assistant removes the forceps and the skin is sutured.

Since the intervention should not take more than three minutes, it suffices to have the animal well anesthetized at the beginning of the operation. If the tracheal cannula plugs up with mucus and the animal becomes asphyctic, one may apply artificial respiration through the cannula itself. The width of this tube should not exceed 1/4 of the width of the trachea.

With but minor modifications, to adjust the operation to the varying anatomy of the skull, this same approach is recommended for hypophysectomy in the GUINEA PIG, MOUSE, FERRET, HEDGEHOG, and most other mammals with elongated crania.

In MAN, the *transnasal* or *transsphenoidal* approach is usually selected for the removal of pituitary tumors, which tend to grow into the sphenoid towards the nasal cavity. In general, however, the *transfrontal* approach is preferred in human surgery. The procedures used by different surgeons vary, but the main feature is to make an osteoplastic flap, so that the frontal lobe, preferably the right, can easily be retracted and the chiasmal region approached. Two types of skin incisions are used, the first runs up from the temporal region along the coronary suture to the midline, then turns down sharply to the root of the nose and after this, turns back along the upper margin of the orbit. This leaves almost no scar as the two parallel incisions, the orbital and the temporal, are covered with the eyebrow and hair respectively. The only scar which shows is a straight line running down on the forehead and this is almost invisible, if the edges of the skin wound are carefully adapted. A second type of incision follows the parietal suture from one temporal region to the other and the skin flap is reflected towards the

face, so as to expose the frontal and parietal bones. The bone flap is made from these with a temporal basis in the same manner as if the first type of incision had been used. With this technique, no visible scar remains. The operative mortality is about 5%.

EFFECTS OF HYPOPHYSECTOMY AND HYPOPHYSEAL HORMONE TREATMENT

State. — The appearance of experimental animals following HYPOPHYSECTOMY resembles that of hypopituitary patients (see : Hypopituitarism). If the operation is performed early in life, the animals fail to grow and retain their lanugo-like puppy fur. If, on the other hand, the pituitary is removed during later life, the fur is merely scanty. In either case, the muscular tonus and strength are far below normal and the sex organs are deficiently developed.

Resistance to various types of non-specific damaging agents (infections, intoxications, trauma, etc.) is greatly diminished by hypophysectomy, mainly because in the absence of the anterior-lobe, animals are incapable of responding to damage with the normal increase in corticoid hormone secretion.

All these deficiency manifestations can be almost completely eliminated by adequate *anterior-lobe hormone* treatment. Resistance to non-specific damage can also be improved by *corticoid therapy*.

Treatment of intact animals with excessive doses of ANTERIOR-LOBE HORMONES rarely produces any acute signs of overdosage, but following several days of treatment, the specific biologic effects of the various hormone principles become evident (stimulation of the gonads, thyroid, adrenals, somatic growth, etc.).

INTERMEDIN does not appear to be toxic or to cause any change in the appearance of mammals, although in lower vertebrates, it elicits the well-known changes in skin color. (See : pp. 217 and 243.)

In man, toxic doses of POSTERIOR-LOBE EXTRACTS cause marked pallor of the face as a result of vasoconstriction, but a rise in blood pressure is rarely observed. Stimulation of intestinal motility is likely to elicit nausea, belching, intestinal cramps and a desire to defecate. Women often complain of dysmenorrhea-like uterine cramps. Because of possible vascular complications, the use of posterior-pituitary extracts is contraindicated in individuals suffering from cardiovascular and especially coronary disease.

Temperature. — The body temperature of HYPOPHYSECTOMIZED animals is usually slightly below normal. Crude anterior-lobe extracts, thyroxin and corticoids tend to raise it towards normal. However, the hypothalamic centers are much more important for temperature control and incidental lesions of these nuclei may well be responsible for some of the changes observed after hypophysectomy.

In intact animals, overdosage with ANTERIOR-LOBE EXTRACTS (unless they contain large amounts of thyrotrophin) rarely causes any significant change in body temperature.

POSTERIOR-LOBE EXTRACTS tend to raise the body temperature, especially in animals whose temperature-regulating mechanism is deranged due to destruction of the thalamus.

Basal Metabolism. — HYPOPHYSECTOMY causes a considerable decrease in the B.M.R.; in the rat, for instance, B.M.R. measurements of -45% have been recorded after hypophysectomy.

The rise in B.M.R. normally elicited by *pancreatectomy* in the dog is not seen in previously hypophysectomized animals.

Thyroidectomy causes a further decrease in the already low metabolic rate of the hypophysectomized animal.

Anterior-lobe extracts (especially thyrotrophin) and *thyroid hormone* are highly effective in restoring the low B.M.R. of hypophysectomized animals to normal, while *posterior-lobe extracts*

are ineffective. It is probable, however, that thyrotrophin is not the only anterior-lobe hormone responsible for the maintenance of the normal metabolic rate.

The specific dynamic action of *proteins* is diminished by hypophysectomy and restored to normal by suitable anterior-pituitary extract therapy.

In intact animals, excessive doses of ANTERIOR-LOBE EXTRACTS raise the B.M.R. above normal, again mainly, (but perhaps not exclusively) due to their thyrotrophin content. *Iodides* inhibit the B.M.R. raising effect of thyrotrophin, presumably due to their ability to counteract the action of this hormone upon the thyroid itself.

POSTERIOR-LOBE EXTRACTS likewise tend to raise the B.M.R. in various animal species including man, vasopressor preparations being generally more active than the oxytocic. Some investigators claim to have prepared a "specific metabolic principle" from the posterior-lobe. It allegedly causes an immediate rise in B.M.R., accompanied by a fall in R.Q., these changes being more rapid than those obtained with other pituitary preparations.

Carbohydrate Metabolism. — (1) **BLOOD SUGAR.** While the blood sugar of well-fed HYPOPHYSECTOMIZED animals tends to remain within normal limits, fasting or exposure to almost any type of non-specific damage elicits an unusually pronounced, often fatal, hypoglycemia. This is generally ascribed to deficient gluconeogenesis, which renders hypophysectomized animals incapable of mobilizing sugar from non-carbohydrate stores.

Gluco-corticoids and *anterior-pituitary extracts* restore the fasting blood sugar to normal, but further work, with highly purified anterior-lobe principles, will have to be performed in order to identify the particular hormone (or hormones) responsible for this effect; *posterior-lobe extracts* are ineffective.

Pancreatectomy fails to elicit the customary marked hyperglycemia and

glycosuria, in the absence of the pituitary. Thus, for instance, dogs which have been both hypophysectomized and pancreatectomized ("Houssay dog") develop only a very mild diabetes and may survive almost indefinitely without insulin therapy. Apparently the absence of diabetogenic and metabolism-stimulating (e.g., thyrotrophic) anterior-lobe hormones partly compensates for the loss of the Langerhans islets.—(Cf. hexokinase theory, p. 494.)

Conversely, *adrenalectomy* or *insulin* administration, tend to cause a particularly pronounced and rapid hypoglycemia following hypophysectomy. As for the pronounced fasting hypoglycemia, a deficient gluconeogenesis is principally responsible for this reaction.

The hyperglycemic action of *adrenaline* is decreased in hypophysectomized animals, even if the hepatic glycogen stores are adequate. This is not due to a delay in the absorption (diminution of hyperglycemic action manifest even if adrenaline is injected intravenously), nor is it due to increased sugar utilization (intravenous glucose still produces marked hyperglycemia).

After feeding of certain carbohydrates (e.g., starch), the rise in blood sugar is not quite as marked in hypophysectomized as in intact rats. However, this may be merely due to a decreased intestinal absorption rate. The hyperglycemia caused by intravenous administration of glucose lasts longer in hypophysectomized than in normal animals, perhaps because in the absence of the pituitary, glycogen deposition in the liver is diminished.

After hypophysectomy *phlorhizin* causes hypoglycemia and a subnormal degree of glycosuria. Simple anterior-lobe extracts suffice, however, to normalize the response of hypophysectomized animals to phlorhizin.

In intact animals impure ANTERIOR-LOBE EXTRACTS raise the blood sugar. It is reasonable to assume that the glycotropic, glycostatic, and diabetogenic actions (see: Classification of the

Hypophyseal Hormones, p. 213) are responsible for this effect (see also effect on "Glycogen stores," below). However, only experiments with pure anterior-lobe hormones will permit elucidation of the underlying mechanisms.

While pure INTERMEDIN has no effect on the blood sugar, POSTERIOR-LOBE EXTRACTS cause marked hyperglycemia, due to their vasopressin content. However, the blood sugar rise can only be elicited with toxic doses and is accompanied by such marked vasomotor phenomena that it can hardly be regarded as a specific hormone-action. A secondary liberation of adrenaline and the non-specific glycogenolytic effect of such toxic doses suffice to explain the observed results.

(2) GLYCOGEN STORES. The glycogen stores of fed HYPOPHYSECTOMIZED animals remain approximately normal. However, after a comparatively short period of fasting, the liver-glycogen and, to a lesser degree, the muscle-glycogen values decrease much more rapidly in the hypophysectomized than in the intact animal. This has been ascribed to a lack of the "glycostatic factor," since treatment with suitable anterior-lobe extracts restores the power of the fasting hypophysectomized animal to maintain its glycogen stores.

Anterior-lobe extracts maintain the muscle-glycogen values of hypophysectomized rats much more readily than the liver-glycogen stores. This effect is manifest even in adrenalectomized rats and is not due to corticotrophin. Several other observations also support the view that following hypophysectomy, the derangement in glycogen storage is not merely due to a secondary deficiency in gluco-corticoid secretion. Thus, hypophysectomized rats are unable to maintain their fasting muscle-glycogen values immediately after the operation, while adrenalectomized animals develop this deficiency only gradually. Furthermore, in adrenalectomized rats, corticoid or salt therapy restores the gly-

cogen values towards normal much more readily than after hypophysectomy. However, large doses of *glucocorticoids* also tend to replenish the liver and muscle-glycogen stores in hypophysectomized animals. Hence, corticotrophins probably also influence this reaction.

It is noteworthy that although separately both *hypophysectomy* and *pancreatectomy* decrease the muscle and liver-glycogen concentration, this effect is not seen if both glands are removed from the same animal (e.g., dog, cat).

The glycogenolytic effect of *adrenaline* is inhibited by hypophysectomy, but only if the hormone is administered subcutaneously, not in the event of intravenous infusion. It is concluded that the inefficacy of subcutaneously administered adrenaline is merely due to a deficient hormone absorption.

In intact animals (e.g., rabbit, rat), impure ANTERIOR-PITUITARY EXTRACTS increase the liver glycogen stores, while POSTERIOR-LOBE EXTRACTS (especially vasopressin) deplete them in proportion to the resulting hyperglycemia.

(3) CARBOHYDRATE ABSORPTION. Following oral administration of glucose, the deposition of liver and muscle glycogen in previously fasted, hypophysectomized rats proceeds at an almost normal rate. Thus there is no evidence of any significant decrease in glucose absorption, although the uptake of complex carbohydrates (e.g., starch) is delayed.

(4) LACTIC ACID. The blood-lactic-acid content does not show very pronounced variations, either after hypophysectomy or after anterior-pituitary extract treatment.

Vasopressin (but not oxytocin) raises the lactic acid content (e.g., dog).

(5) HEXOSEMONOPHOSPHATE. The hexosemonophosphate content of muscles does not show any significant change after hypophysectomy in the rat. Adrenaline, which increases the hexosemonophosphate concentration in

the muscles, retains this effect even after hypophysectomy.

Lipid Metabolism. — HYPOPHYSECTOMY slightly decreases the fatty acid and total lipid concentration of the blood and markedly inhibits the lipemia otherwise produced by pancreatectomy.

The production of fatty livers (by partial hepatectomy and fasting, carbon tetrachloride, phosphorus, etc.) is inhibited by hypophysectomy, but restored following anterior-pituitary extract treatment. Adrenalectomy likewise prevents fat-deposition in the liver under similar conditions, while corticoids restore the ability of adrenalectomized or hypophysectomized animals to develop fatty livers; hence it is highly probable that corticotrophin is important for this response.

The tendency of hypophysectomized animals (especially dogs) to become very adipose is partly due to the polyphagia caused by accompanying hypothalamic lesions and partly to the aversion of these animals to perform muscular exercise. Bilateral lesions, interrupting the nerve fibers which originate from the caudal portion of the paraventricular nuclei and descend to the brain stem, cause polyphagia and adiposity irrespective of the presence or absence of the hypophysis.

In intact animals, the changes in blood-lipids obtained with ANTERIOR AND POSTERIOR-LOBE EXTRACTS are rather variable; since no systematic work has been done with purified hormones, they do not deserve a detailed discussion here.

It is noteworthy, however, that various impure anterior-lobe extracts cause marked *ketonuria* and *ketonemia*, although the assumption that a special "ketogenic hormone" exists has not been proven. Extensive partial-hepatectomy inhibits the ketogenic effect of anterior-lobe extracts, because the liver is the source of ketone body formation.

Nitrogen Metabolism. — HYPOPHYSECTOMY causes no very specific

change in nitrogen metabolism, although protein anabolism is obviously impeded (mainly due to lack of somatotrophin). The fact that fasting hypophysectomized animals tend to eliminate less endogenous nitrogen than normals can be explained by the comparatively low protein reserves of the former.

Phlorhizin causes a less pronounced loss of nitrogen and a lower urinary D/N (dextrose per nitrogen) ratio in hypophysectomized than in normal, or even in thyroidectomized, animals. This may likewise be partly due to subnormal nitrogen reserves and perhaps also to the inhibition of gluconeogenesis from protein during anterior-lobe failure.

ANTERIOR-LOBE EXTRACTS (as well as pure somatotrophin) increase nitrogen anabolism and tissue-growth both in hypophysectomized and in intact animals. POSTERIOR-LOBE HORMONES exert no specific effect upon protein metabolism.

Endogenous creatinine excretion and the phosphocreatine concentration of the muscles decrease following hypophysectomy, but are restored towards normal by anterior-pituitary extracts. It has been stated that a derangement in creatine metabolism may be partly responsible for the muscular asthenia, so characteristic of anterior-lobe deficiency.

Salt and Water Metabolism. — Hypophysectomy causes transient POLYURIA in most animal species, but this operation in itself does not produce permanent diabetes insipidus. To cause marked, persistent polyuria some anterior-lobe tissue must be preserved and the lesion must either destroy the posterior-lobe or sever the connection between the supraoptic nuclei and the hypophysis; the latter operation is followed by atrophy of the pars nervosa, whose endocrine secretion is apparently under the nervous control of these nuclei. Unlike the permanent phase of diabetes insipidus, the initial, transitory

Polyuria is not affected by the NaCl intake or by injection of posterior-pituitary extract; it is abolished by thyroidectomy and is independent of the presence of anterior-lobe tissue.

It is now generally accepted that the anti-diuretic action of posterior-pituitary extracts is due to stimulation of the water re-absorption in the descending loop of Henle within the kidney. In vitro, in a heart-lung-kidney preparation (dog) there is profuse polyuria which can be inhibited by introducing into the perfusion system a head containing the hypothalamus and hypophysis. No such anti-diuretic effect is achieved by introduction of a head deprived of the hypophyseal-hypothalamic system.

Data indicating that the anti-diuretic hormone is probably identical with vasopressin has been enumerated in the chemical section. (See : p. 219.)

Several observations suggest that administration of water inhibits the secretion of anti-diuretic pituitary hormone, while dehydration increases it. Through this mechanism, the posterior-lobe could participate in the normal regulation of diuresis. Nevertheless, even in hypophysectomized animals whose hypothalamus is simultaneously destroyed, the administration of water causes transitory diuresis, indicating that the hypophyseal-hypothalamic mechanism is not the only regulator of this activity.

The polyuria elicited by posterior-lobe destruction precedes the polydipsia and excessive diuresis continues for some time, even if water is withheld. Furthermore, complete nephrectomy prevents the abnormally high fluid intake caused by posterior-lobe destruction. Hence, increased urine formation appears to be the primary phenomenon in this type of polyuria.

In amphibia (e.g., frog) posterior-lobe extracts (presumably vasopressin) increase the total water content of the body, primarily due to an increase in the water intake through the skin.

Treatment with impure anterior-lobe extracts, corticoids or thyroid hormone increases diuresis even long after complete hypophysectomy.

Administration of sodium chloride causes more pronounced polyuria in an-

imals with hypothalamic diabetes insipidus than in intact controls.

Normally, water reabsorption (such as is characteristic of the anti-diuretic hormone) is usually enhanced by an increase in the concentration of serum solutes, perhaps specifically Na (Verney, 1946). According to Peters (1948) it can also be inferred by the comparison of the two types of experimental diabetes insipidus, as outlined below:

Posterior-lobe destruction	Desoxycorticosterone overdosage
Initial inhibition of water reabsorption	Initial increase of Na reabsorption
Primary diuresis	Primary thirst
Deficient body water	Excess body water
Deficient body Na	Excess body Na

CHLORIDE elimination through the urine is greatly diminished in dog kidneys perfused by means of a heart-lung preparation. However, the addition into the circulation of the head of a dog (containing the pituitary), or addition of posterior-pituitary extract to the perfusion fluid, results in a decreased diuresis due to better chloride concentration.

Destruction of the posterior-lobe, and to a lesser degree, incomplete hypophysectomy, decreases both the absolute amount and especially the concentration of chloride eliminated in the urine; this effect is also prevented by posterior-lobe extract.

In the intact organism, purified vasopressin likewise increases chloride elimination in the urine in spite of its anti-diuretic action; oxytocin, however, raises the total Cl output (only due to its diuretic action), without changing the urinary Cl concentration.

Data are somewhat contradictory concerning the serum-chloride concentration after selective removal of the posterior-lobe or complete hypophysectomy; sometimes the serum-chloride level remains normal, while in other instances, hyper- or hypochloremia is observed. For reasons not yet clearly understood, the hypochloremic effect

of desoxycorticosterone acetate is inhibited by hypophysectomy.

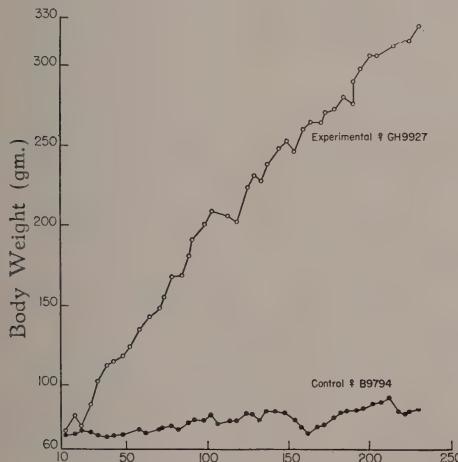
The urinary SODIUM elimination tends to run parallel with the chloride excretion following various types of hypophyseal and hypothalamic lesions. The blood sodium concentration shows less consistent changes.

POTASSIUM and CALCIUM metabolism are not very significantly influenced by hypophysectomy or the administration of pituitary extracts. It is noteworthy, however, that hypokalemia (similar to that produced with desoxycorticosterone) can be elicited with certain corticotrophic anterior-lobe extracts.

The inorganic PHOSPHATE content of the blood and muscles tends to decrease after hypophysectomy in several animal species, and can be restored towards normal by treatment with crude anterior-lobe extracts. Excessive doses of such preparations may even raise the serum-phosphate concentration above normal.

Both anterior and posterior-lobe extracts counteract the hypophosphatemia normally elicited by insulin or glucose administration.

Hypophysectomy decreases the blood IODINE concentration while subsequent treatment with thyrotrophin raises it to or even above normal. This reaction is apparently mediated by the thyroid, since it does not occur following thyroidectomy.



Growth curves of hypophysectomized female rats

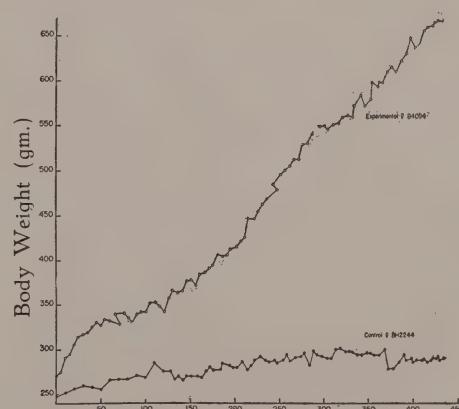
(Courtesy of Dr. C.-H. Li)

The experimental animal received 0.10 mg. of somatotrophin/day during first 140 days; the daily dose was then increased to 0.20 mg.

Other Metabolites. — The decrease in metabolism and growth-rate occasioned by hypophysectomy and the stimulation of these processes by anterior-pituitary extracts, produces secondary changes in the requirements of the body for essential amino-acids, vitamins, minerals, etc. However, neither hypophysectomy nor administration of pituitary extracts has been shown to have any consistent and significant specific effect upon the metabolism of other body constituents.

Growth and Bone Structure. — In all mammals and even in most other vertebrates, hypophysectomy causes an immediate cessation of growth in length, except during embryonic and very early (first few weeks) postnatal life. Curiously, in certain lower vertebrates (e.g., amphibia, reptiles) and in very young mammals, growth in length is largely independent of the hypophysis. This is also true of tissue cultures in vitro where mammalian cells can proliferate in the absence of somatotrophin.

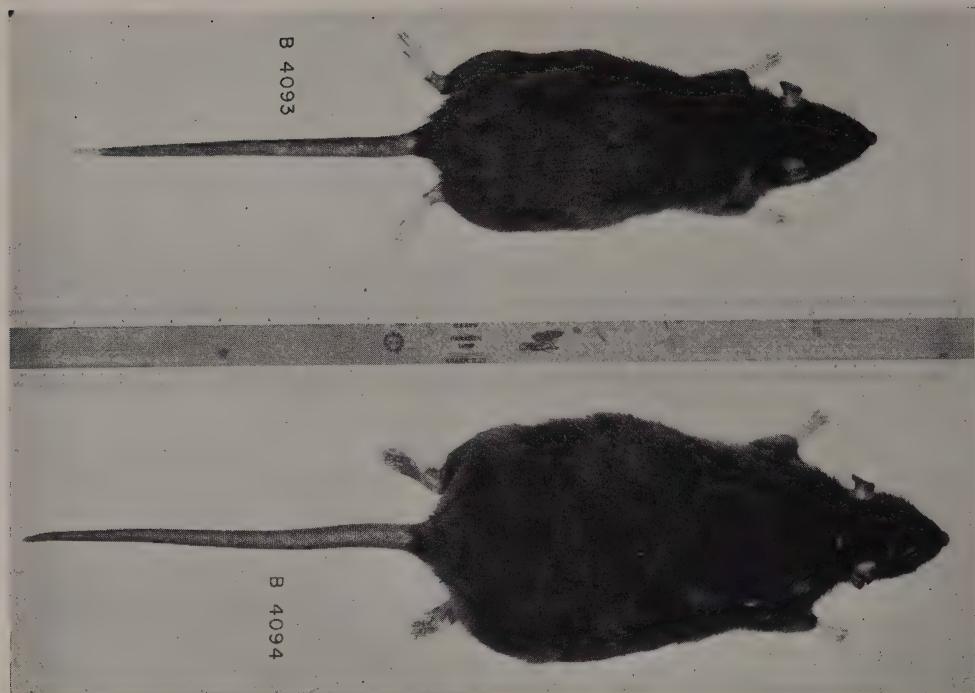
Very young rats, for instance, continue to grow, irrespective of their size at the time of the operation, until they reach a weight of approximately 75 gm.



Growth curves of normal "plateau" female rats

(Courtesy of Dr. C.-H. Li)

The experimental animal received 0.40 mg. of somatotrophin/day during the first 23 days, this dose being gradually raised to 2.0 mg./day.



Effect of somatotrophin on intact rat. Typical photograph of a normal "plateau" rat, that is, one having reached a weight at which normal growth is almost arrested (top) and similar animal after 432 days of treatment with somatotrophin.

(Courtesy of Dr. C.-H. Li.)

On the other hand, in rats hypophysectomized after they have reached the weight of 75 gm., growth ceases immediately upon ablation of the hypophysis. It is not known why the lowest stages, both in the ontogenetic and phylogenetic scale of development, are independent of pituitary growth-hormone.

It has been ascertained that the growth-promoting effect of the pituitary is due to a hormone of the anterior-lobe, since growth in length is re-initiated in hypophysectomized animals by anterior, but not by posterior or middle-lobe extracts. As outlined in the section on the chemistry of the anterior-lobe hormones, somatotrophin is a chemically distinct hormonal principle which has been prepared in pure form. (See : pp. 210, 215, 216.)

In intact animals, somatotrophin causes fairly proportionate growth of all tissues and thus produces gigantism. Its effect upon the longitudinal

growth of bones is principally due to a specific action upon the junction cartilage plates in which it promotes cell proliferation and subsequent ossification. However, it also stimulates subperiosteal growth in width and proliferation of soft tissues. It exerts no specific effect upon cells controlled by special trophic hormones (e.g., thyroid, adrenal cortex, gonads).

Corticotrophin inhibits growth-stimulation by somatotrophin.

Blood-count. — HYPOPHSECTOMY does not significantly influence the blood-count, although it tends to cause some degree of anemia and a fall in blood-volume.

Administration of CRUDE ANTERIOR-LOBE EXTRACTS, on the other hand, tends to raise the red-cell-count, as well as the blood volume. This action may be involved in the polycythemia and plethora of certain hyperpituitary syndromes such as Cushing's disease.



Effect of hypophysectomy in the dog. Two littermate female dogs, the one on the right was hypophysectomized two months before this picture was taken. Their initial body weight was equal (3 Kg.) while now the control dog weighs 7 Kg. and the hypophysectomized animal (right) 3.0 Kg. Note also typical fuzzy "puppy fur" of hypophysectomized animal.

CORTICOTROPHIN normally causes a decrease in the lymphocyte-count, but in adrenalectomized animals and addisonian patients this effect cannot be elicited. Apparently, the effect of corticotrophin is mediated by the adrenal cortex whose corticoids are responsible for the lymphocyte-destroying effect.

An increase in the red-cell-count and a change in the differential white-cell-count have also been observed in animals following isolated hypothalamic lesions.

Cardiovascular System.— In HYPOPHYSECTOMIZED animals, the heart and blood vessels are atrophic and the pulse rate and blood pressure are low. Conversely, treatment of hypophysectomized or intact animals with crude ANTERIOR-LOBE EXTRACTS causes marked hypertrophy of the heart and blood

vessels with an increase in blood pressure and pulse rate. These effects are partly mediated through the thyroid and can be duplicated by the administration of pure thyrotrophin or of thyroid-hormone itself. However, even in thyroidectomized animals some increase in heart weight can be obtained by anterior-lobe preparations, so that the effect cannot be regarded as solely mediated by the thyroid. It is not yet known, which of the other hypophyseal principles participates in this effect.

Upon continued treatment with large doses of crude anterior-lobe extracts, severe hypertension, periarteritis nodosa, myocardial (Aschoff?) nodules and even coronary infarcts have been produced. All these changes are especially readily obtained in rats, sensitized to these toxic effects by unilateral nephrec-

tomy and high-sodium, high-protein diets (see: General-Adaptation-Syndrome). These latter effects appear to be mediated by the adrenal cortex, since they cannot be elicited in adrenalectomized animals. However, as they have not yet been produced with pure corticotrophin, it is possible that other principles in the crude anterior-lobe extracts are also essential.

VASOPRESSIN causes constriction of the coronary arteries with rather characteristic changes in the E.C.G. and in the cardiac output. The latter is greatly increased following an initial transitory decrease.

Only under certain conditions, especially in anesthetized animals (e.g. in the ether-anesthetized cat or dog), does vasopressin cause a marked rise in blood-pressure; in non-anesthetized animals and man, the pressor effect is minimal or replaced by an actual drop in blood pressure with bradycardia. Not even all anesthetics permit the production of increased blood pressure by posterior-pituitary extract. The pressor effect is not mediated by the nervous system, since it can occur after destruction of the brain or spinal cord and after paralysis of the vasomotor ganglia by nicotine. Probably the pressor effect depends largely upon an increase in the capillary tonus, and to a lesser degree upon constriction of larger arteries and arterioles. To some extent it may even act by increasing the production of renal pressor substances since it causes marked constriction of the renal arteries, and this is known to stimulate the pressor-hormone production of the kidney.

Vasopressin has been recommended to combat hypotension in various types of shock, but because of the variability of the pressor response and the danger of further diminishing the already low blood pressure, it should not be administered in such cases.

It is questionable whether vasopressin plays any important part in the main-

tenance of the normal blood pressure, since removal of the posterior-lobe does not cause hypotension.

Lymphatic System. — In HYPOPHYSECTOMIZED animals, the thymus and other lymphatic organs are usually of moderate size. They are not completely atrophic nor do they show the excessive development observed after adrenalectomy. However, after hypophysectomy, as after adrenalectomy, exposure to various types of stress fails to elicit the usual alarm reaction type of acute thymus and lymphatic organ involution. This is apparently due to a lack of defensive corticotrophic hormone secretion.

Hypophysectomy inhibits while hypophyseal extracts stimulate erythrocyte production in the bone marrow. The size of the spleen is markedly diminished after hypophysectomy and increased by crude anterior-lobe extracts.

CORTICOTROPHIN injections cause speedy involution of the thymus — and to a lesser extent of other lymphatic organs — both in hypophysectomized and in intact animals.

SOMATOTROPHIN is definitely "thyrotrophic" and certain impure anterior-lobe extracts even prevent thymus atrophy during the alarm reaction.

HYROTROPHIN also stimulates the growth of lymphatic organs (as does thyroid hormone), hence the effect of impure anterior-lobe extracts is variable, depending upon the relative proportions of corticotrophin, somatotrophin and thyrotrophin which they contain.

Muscles. — Anterior-lobe removal and complete hypophysectomy decrease, while anterior-lobe implants increase the contracture of the rectus abdominis muscle (frog) normally elicited by acetylcholine. Myograms of somatotrophin-treated rats revealed no improvement in muscular strength in spite of greater size.

Nervous System and Sense Organs. — HYPOPHYSECTOMY causes no clear-cut changes in the nervous system or

the sense organs. On the other hand, lesions to the hypothalamic nuclei may cause hypersomnia, and narcolepsy (sometimes with cataplexy). It is very doubtful whether there is a "sleep center" in the hypothalamus, but it is well established that this entire region plays an important part in the mechanism of sleep.

It has been claimed that INTERMEDIN is responsible for the migration of melanin granules in the retina, which is essential for dark-adaptation of the eye. This is very controversial, however, since it is known that hypophysectomy does not interfere with dark-adaptation.

POSTERIOR-LOBE EXTRACTS may cause profound, generalized depression, if administered in very large doses. It is of diagnostic importance that vasopressin — especially when given after abundant water ingestion — causes convulsions in epileptics, but not in normal individuals. Vasopressin (but not oxytocin) injected directly into the cerebral ventricles of monkeys and man causes pronounced, generalized vasodilatation, allegedly due to direct stimulation of the adjacent autonomic centers.

Digestive System. — HYPOPHYSECTOMY causes profound atrophy of the entire gastrointestinal system, including the liver and the exocrine pancreas. Furthermore, hypophysectomized animals have a great tendency to develop the acute gastric ulcers characteristic of the alarm reaction. Ulceration of the esophagus, stomach and duodenum, as well as disturbances in intestinal motility are also frequently observed following hypothalamic lesions.

Crude ANTERIOR-LOBE EXTRACTS cause an increase in the thickness and length of the gastrointestinal tract; this is accompanied by a marked hypertrophy and hyperplasia of the liver and the acinous tissue of the pancreas. (The effect of pituitary extract upon the Langerhans islets has been discussed in the section on the pancreas.) Certain impure anterior-pituitary extracts cause

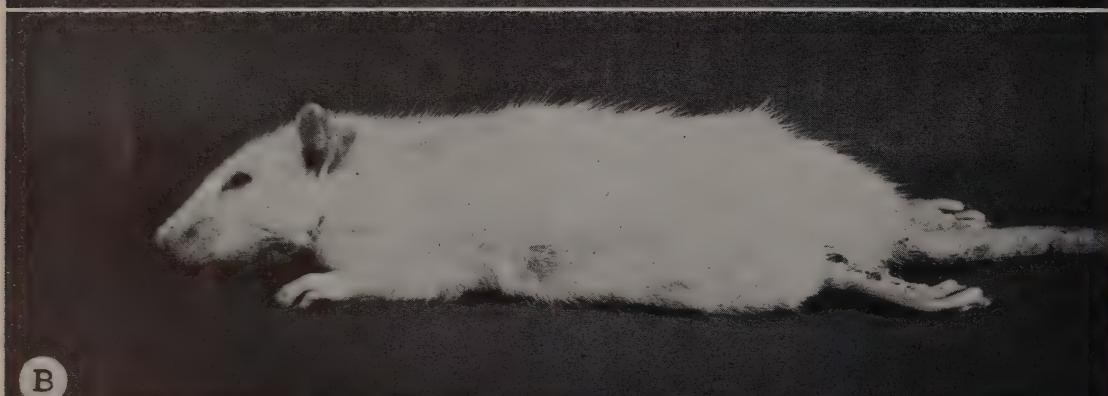
fat deposition in the liver, an effect which has sometimes been ascribed to a special "ketogenic or fat-metabolism hormone." Since adrenalectomy prevents this fat deposition, the latter is more probably due to corticotrophin. After partial hepatectomy, the compensatory hypertrophy of the hepatic remnant is greatly inhibited by hypophysectomy and accelerated by anterior-lobe extracts.

POSTERIOR-LOBE EXTRACTS have no specific effect upon the morphologic development of the digestive system. The influence of posterior-pituitary extracts on the motility of the intestine is variable. It depends upon the animal species, the type of anesthetic used and other experimental conditions. In human subjects with enterostomies (artificially placed fistulae between intestine and skin), it has been found that pituitary-extract (probably because of its vasopressin content) markedly stimulates peristalsis, both in the colon and in the ileum. This increase in motility is unaccompanied by any effect upon the constant tonus. If moderate doses (not exceeding 20 units) of posterior-pituitary extract are used, the action rarely lasts longer than 90 minutes. Nevertheless, vasopressin is often useful in the relief of intestinal paresis and distension after abdominal operations and febrile infections; at certain dose-levels it can also cause contraction of the biliary passages.

Skin and Appendages. — In fish, amphibia and reptiles HYPOPHYSECTOMY causes blanching of the skin, due to persistent contraction of the cutaneous melanophores. This may be accompanied by a decrease in the number of melanin granules and an expansion of the light xantholeukophores. Consequently, the animals become unable to adapt their coloration to their background. The change is apparently due to intermediate-lobe deficiency, since isolated removal of the intermediate-lobe alone has a similar effect, while selective anterior



A



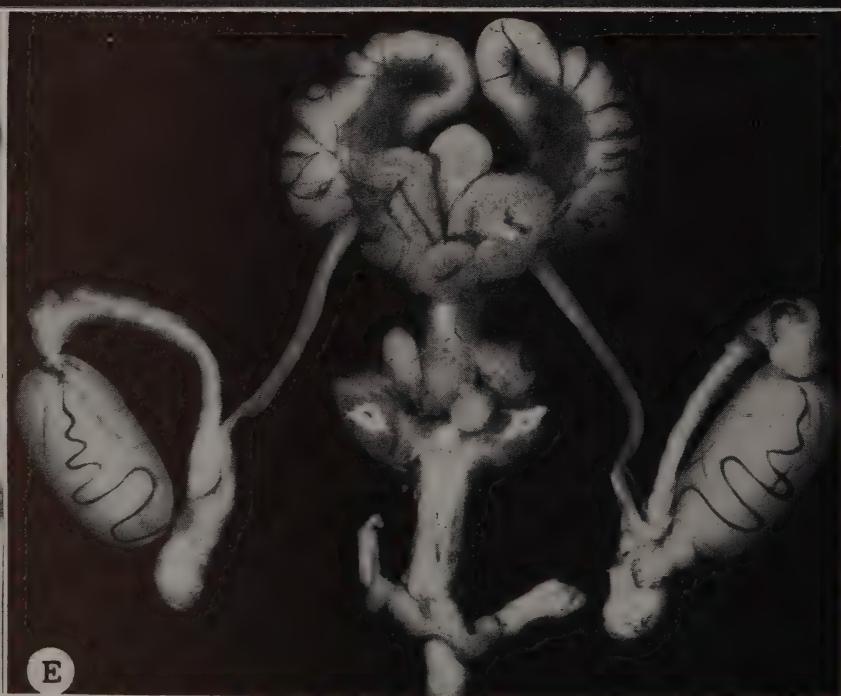
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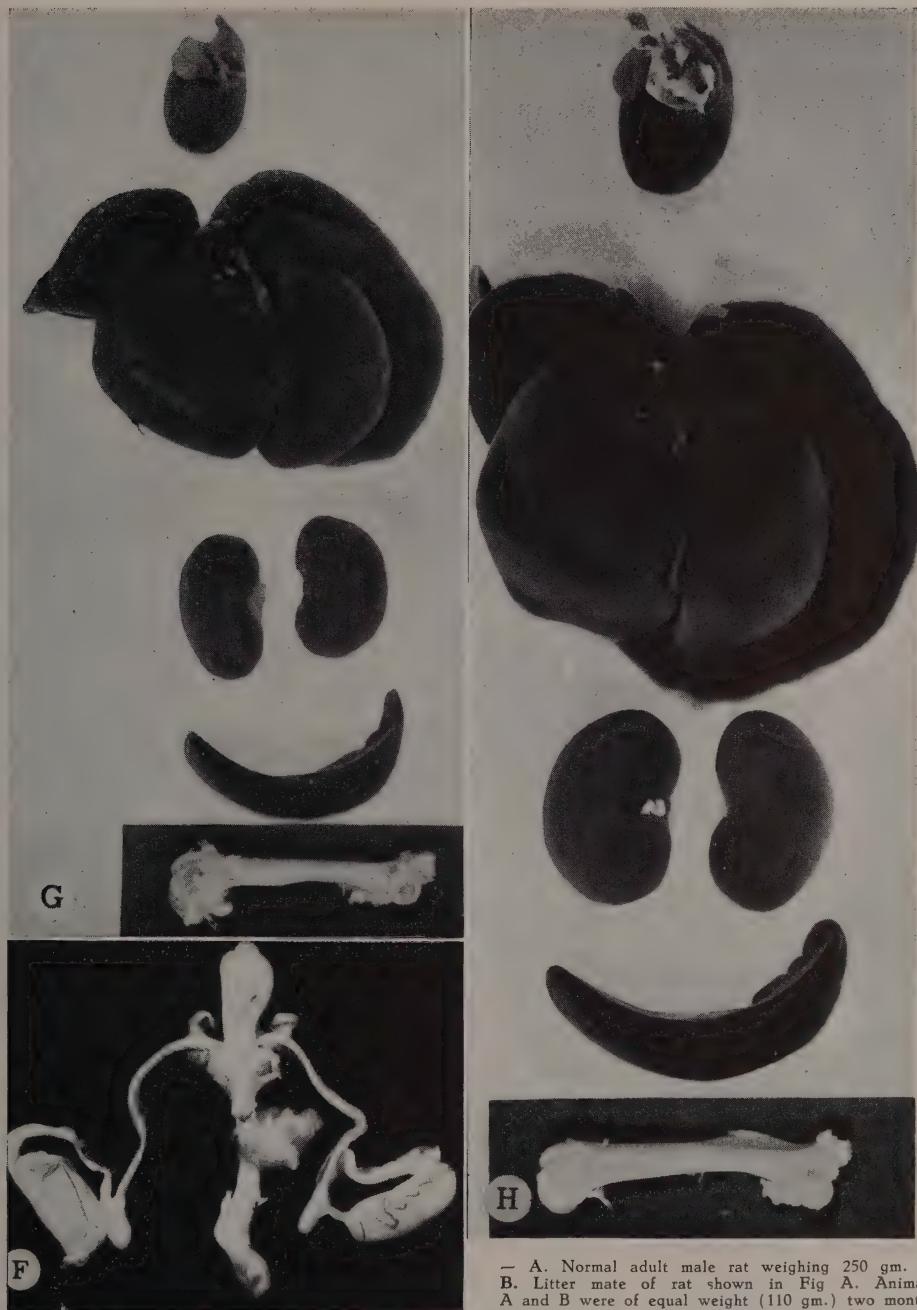


D



E

Effect of hypophysectomy on the organs of the rat. (Cont'd.)



— A. Normal adult male rat weighing 250 gm. — B. Litter mate of rat shown in Fig. A. Animals A and B were of equal weight (110 gm.) two months earlier when B was hypophysectomized. Note great retardation of growth. At the time of autopsy this

rat weighed 86 gm. — C. Base of the skull (with pituitary), thyroid (20 mg.), adrenals (30 mg.) of intact rat shown in Fig. A. — D. Base of the skull (note absence of pituitary), thyroids (4 mg.), adrenals (6 mg.) of hypophysectomized rat shown in Fig. B. — E. Sex organs of intact rat (6.8 gm.). — F. Sex organs of hypophysectomized rat (666 mg.). — G. Heart (398 mg.), liver (4.8 gm.), kidneys (835 mg.), spleen (310 mg.), and tibia (320 mg.) of hypophysectomized rat shown in Fig. B. — H. Heart (962 mg.), liver (12.6 gm.), kidneys (2.7 gm.), spleen (1.0 gm.), and tibia (675 mg.) of normal rat shown in Fig. A. — Note that the weight of the body as a whole and that of most organs is considerably smaller in the hypophysectomized than in the intact animal, but atrophy of endocrine and sex organs is even more pronounced, while atrophy of bones is proportionately less marked.

or posterior-lobe removal does not produce blanching of the skin.

In certain snakes, hypophysectomy causes almost continuous moultling of the skin at irregular intervals. This is apparently due to lack of thyrotrophin, since thyroidectomy exerts a similar effect.

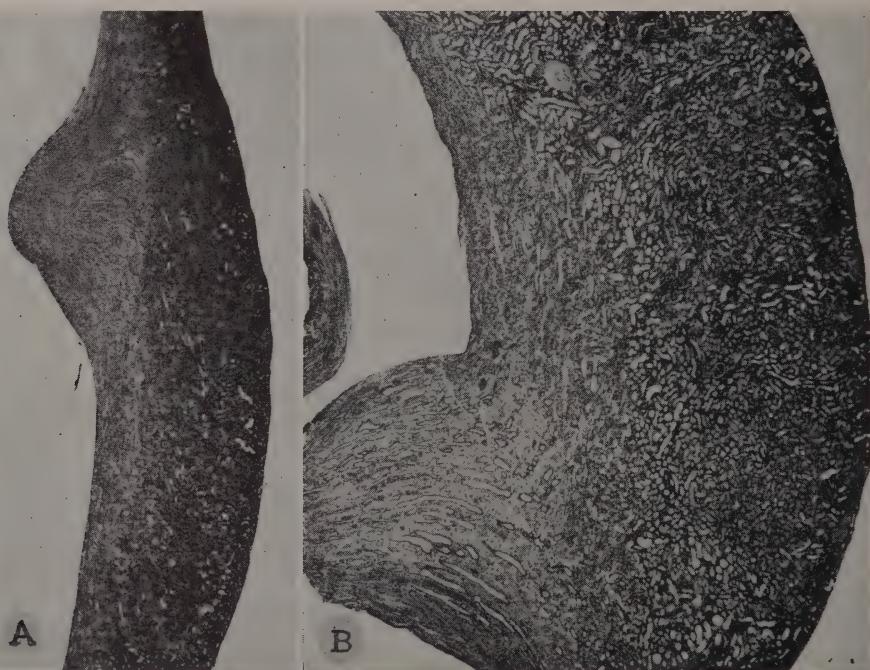
In mammals, the cutaneous changes after hypophysectomy are less obvious; there is some degree of skin atrophy, often accompanied by loss of hair. Hypophysectomy in immature animals causes permanent retention of the very fine "puppy fur." (See : p. 241.)

In birds with sex-specific plumage types (e.g., fowl) hypophysectomy causes plumage changes reminiscent of those produced by gonadectomy. However, hypophysectomy also eliminates the plumage-influencing effect of the thyroid and hence the resulting changes are complicated by those of thyroid-deficiency.

Treatment with crude ANTERIOR-PITUITARY EXTRACTS results in excessive development of the sebaceous glands. The intense acne often seen in hyperpituitary syndromes (e.g., Cushing's disease) is probably related to this effect.

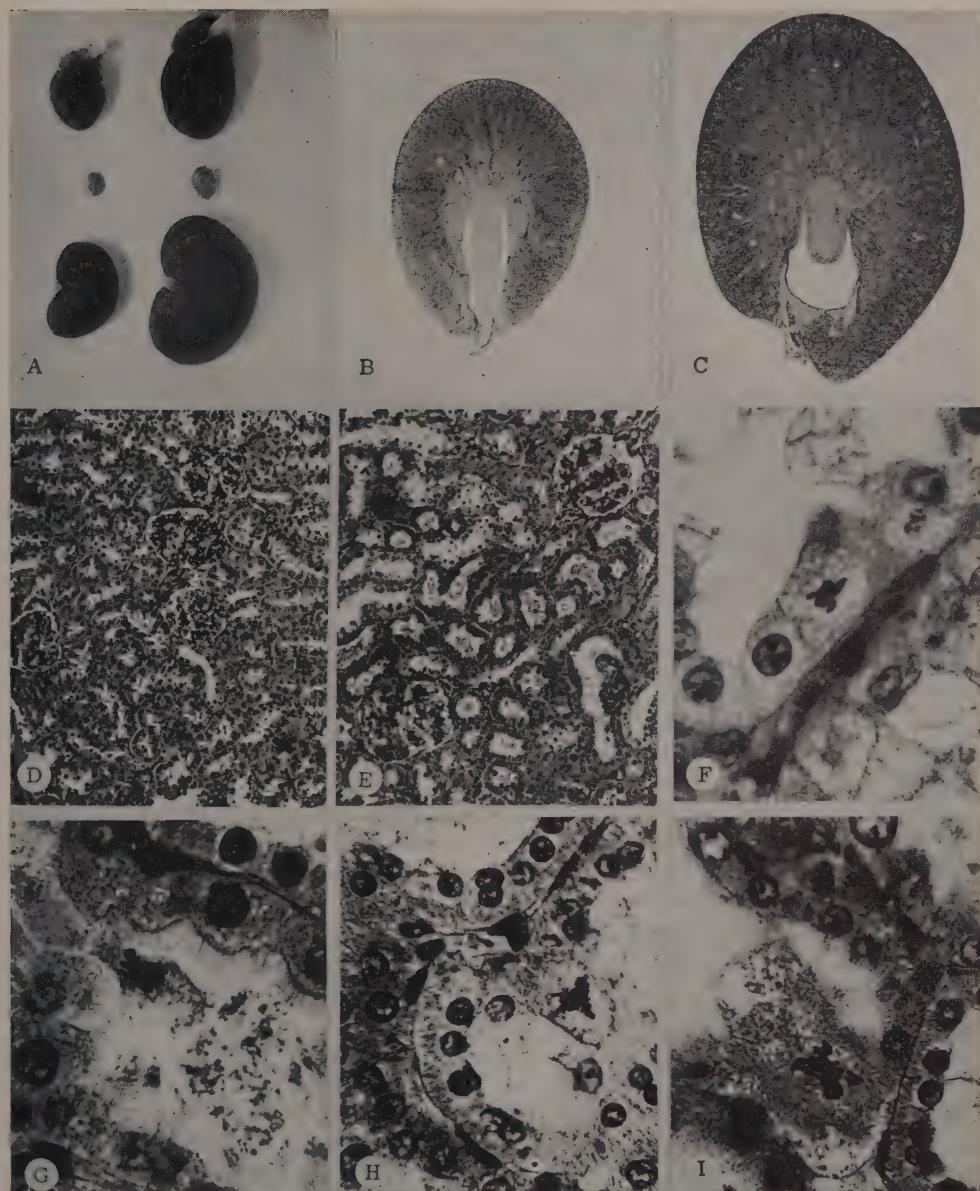
Administration of INTERMEDIN causes expansion of the melanophores in poikilothermic animals and thus leads to darkening of the skin. VASOPRESSIN elicits profound pallor, even in mammals, presumably due to constriction of the cutaneous capillaries.

Urinary System. — HYPOPHYSECTOMY causes pronounced atrophy of the kidney in all animal species. It also interferes with the compensatory hypertrophy of the remaining kidney, in the event of unilateral nephrectomy. It has not yet been established whether there is a special "renotrophic anterior-lobe hormone," but it is certain that the action upon the kidney is due to the anterior-lobe.



Renotrophic effect of anterior-pituitary extract. — A. Low magnification of a cross-section through a rat kidney 20 days after ureter ligation. Note marked pressure atrophy of kidney tissue, which is reduced to a very thin layer. — B. Cross-section through a similar rat kidney 20 days after ureter ligation. In this animal renal atrophy was inhibited by daily treatment with renotrophic anterior-pituitary extract.

(After H. Selye and C. Hollett: J. Urol. 53, 498. 1945.)



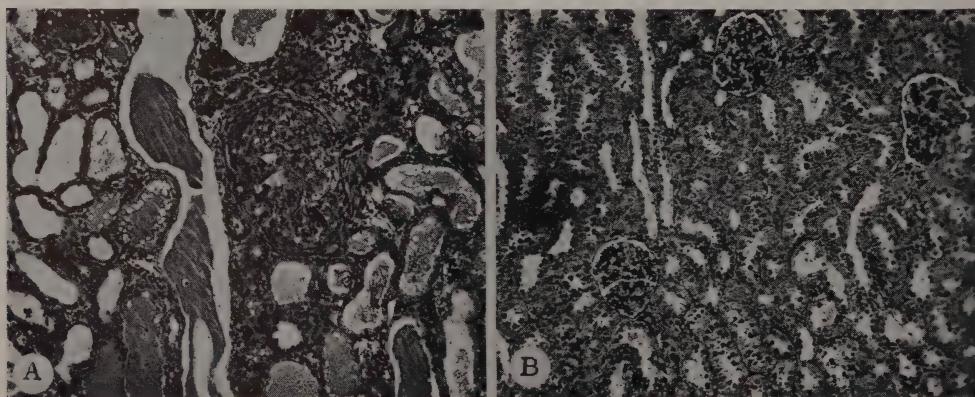
Renotrophic effect of combined treatment with anterior-pituitary extract and thyroxin. — A. Macroscopic aspect of heart, adrenal and kidney of a control (left) and an experimental animal; the latter was treated with lyophilized hypophyseal tissue and thyroxin for 20 days. — B. and C. Low magnification of cross-sections through the kidney of a control (B) and anterior-pituitary plus thyroxin treated (C) rat. Note great enlargement, especially of the renal cortex, in the latter. — D. and E. Medium magnification of the renal cortex of a control (D) and an hypophyseal extract plus thyroxin treated rat (E). Note hypertrophy and hyperplasia of the epithelial cells with regular and proportionate distension of all tubular lumina as well as enlargement of the glomeruli but no signs of nephrosclerosis in E (pure renotrophic effect). — F. G. H and I. Oil-immersion view of various fields from the kidney of a rat which received the hypophyseal preparation plus thyroxin. Note various types of mitotic divisions, budlike protrusion of cytoplasm reminiscent of apocrine secretion (F); a cell in mitotic division in the process of being discharged into the lumen (G), several binucleated cells and marked anisocytosis indicative of rapid proliferation. (After Selye et al.: Canad. Med. Assoc. J., 52, 571, 1945.)

Treatment with crude ANTERIOR-LOBE EXTRACTS restores the atrophic kidney of the hypophysectomized animal to normal and may even induce marked hypertrophy and hyperplasia of the kidney in intact animals. This action is not entirely due to thyrotrophin, since it is only diminished and not prevented by thyroidectomy. Nevertheless, the thyroid plays an important rôle in this "renotrophic effect," since simultaneous treatment with anterior-pituitary extract and thyroid hormone elicits a much more pronounced renal enlargement than can be obtained by either of these preparations alone. The renotrophic steroids (e.g., testosterone) likewise potentiate the kidney-stimulating effect of crude anterior-lobe extracts. In animals treated with renotrophic anterior-pituitary preparations, there is marked hyperplasia and hypertrophy of the epithelium in the proximal and distal convoluted tubules, as well as enlargement of the renal glomeruli. This effect is not to be confused with the nephrosclerotic action, which is presumably due to corticotrophin and will be discussed below. Unlike the latter, the renotrophic action is not markedly

influenced by the sodium content of the diet.

Renotrophic anterior-lobe extracts also increase the functional capacity of the kidney as judged by clearance tests and render the organ more resistant to the pressure atrophy normally produced by occlusion of the ureter. (See : p. 246.)

Impure anterior-pituitary extracts, however, may have an opposite, kidney-damaging effect characterized by *nephrosclerosis*. This action is particularly evident in animals sensitized by unilateral nephrectomy and given diets rich in sodium and protein. It is accompanied by hypertension and is apparently due to the same principle which elicits periarteritis nodosa and cardiac (Aschoff ?) nodules. The nephrosclerotic effect cannot be elicited after adrenalectomy and is apparently due to the corticotrophin content of the crude preparations. Simultaneous administration of thyroid hormone aggravates the nephrosclerotic action of pituitary extracts, and hence the thyrotrophin content of the crude preparations probably also plays a rôle (see : General-Adaptation-Syndrome). The renotrophic and nephrosclerotic actions of crude an-



Production of nephrosclerosis by anterior-pituitary extract. — A. Section through the kidney of a rat sensitized by sodium and unilateral nephrectomy and treated with lyophilized anterior-pituitary material during a period of four weeks. Note large, partly hyalinized, glomerulus with pericapsular fibrosis and thickening of afferent arteriole in the center of the field; there are also numerous dilated tubules with hyaline casts. — B. Section through the kidney of a similarly treated rat in which the development of pathologic renal changes was completely prevented by simultaneous ammonium-chloride administration which caused Na-depletion.

(After C. E. Hall and H. Selye: Rev. Can. de Biol., 4, 197, 1945.)



Renal infarct produced by vasopressin. Several belt-like anemic infarcts in the kidneys of a female adult rat which received 50 U.S.P. units of "surgical pituitrin" twice daily on two successive days. The distribution of the infarcts corresponds to the territories of the larger renal arterial branches.

terior-pituitary preparations do not necessarily run parallel, but it has not yet been possible to completely separate the two principles.

Large doses of vasopressor posterior-lobe extracts cause multiple anemic infarcts in the kidney, presumably due to their vasoconstrictor effect. For reasons which are not yet understood, pretreatment with folliculoids greatly increases

the incidence and severity of renal infarction following vasopressin administration.

Accessory Sex-Organs. — Almost all the actions of anterior-lobe extracts upon the accessory sex-organs are mediated by the gonads. These indirect actions have been discussed in detail in the chapters on the ovary and testis, hence they need not be reviewed here. However, a few accessory sex-organs are responsive to direct stimulation with pituitary hormones.

Thus, for instance, the MAMMARY GLANDS can be directly influenced by *anterior-lobe hormones*. Impure anterior-lobe extracts stimulate mammary growth even in gonadectomized (male or female) animals, due to a so-called "mammogenic action." This effect is greatly enhanced by simultaneous administration of various steroid hormones (folliculoids, luteoids, testoids), but it is manifestly not exclusively mediated through the gonad.

The effect of luteotrophin (prolactin) upon milk secretion is likewise independent of the gonads since it is not abolished by ovariectomy; indeed, even normal lactation is maintained (often actually increased) after spaying.

The galactagogic action of *posterior-pituitary extracts* is also direct; it depends merely upon the stimulation of the smooth muscle cells in the mammary gland and the resultant expression of accumulated milk. Unlike luteotrophin,



Effect of combined treatment with pituitary extract and Δ^5 -Pregnenolone upon the preputial glands of the rat. — A. Preputial glands of a control, not treated female [this as well as all other animals of this series (A-D) were hypophysectomized and ovariectomized in order to avoid complication of the results by endogenous ovarian or pituitary hormones]. — B. Slight enlargement of the preputial glands by Δ^5 -pregnenolone treatment. — C. Marked enlargement of preputial glands, due to combined treatment with pregnenolone and an impure mammogenic pituitary extract. — D. Moderate enlargement caused by treatment with pituitary extract alone, this illustrates peripheral synergism of hypophyseal and steroid hormones on a target organ.
(After H. Selye and E. Clarke: Rev. Canad. de Biol. 2, 319, 1943.)

posterior-pituitary extract does not alter the total amount of milk produced.

The stimulation of the PREPUTIAL GLANDS, especially in the rat, is likewise under the direct control of some pituitary hormone. As previously mentioned, anterior-lobe extracts cause an increase in the size of these glands in both sexes, irrespective of the presence or absence of the gonads. Certain steroids (e.g., Δ^5 -pregnenolone), which could come from the gonads, cause only mild preputial gland stimulation in themselves but they greatly enhance the corresponding effect of anterior-lobe extracts. As in the case of the mammary gland, there appears to be a peripheral synergism between the steroid and the anterior-lobe hormone. It has not yet been determined which, among the anterior-lobe principles, is responsible for this effect.

The contractions of the UTERUS are greatly stimulated by oxytocin. Small doses merely augment the tonus and increase the amplitude, while larger doses cause tetanic contractions, which may last several minutes. In certain animal species, especially the rabbit, folliculoids increase, while luteoids diminish the contractility of the uterine musculature and its response to oxytocin. It is noteworthy, however, that the human uterus responds to oxytocin at least as well during the luteal, as during the follicular phase of the cycle and its contractions are not inhibited by simultaneous progesterone administration. Even during pregnancy the human uterus contracts under the influence of oxytocin.

It has been claimed that increased secretion of oxytocin is responsible for the intense uterine contractions during normal delivery. In hypophysectomized animals, the process of delivery is often delayed and the contractions tend to be weak; yet parturition can occur after complete ablation of the hypophysis, so that the posterior-lobe is not indispensable for this process.

Clinically, oxytocin should only be used : (1) for the induction of labor at

term if this fails to occur spontaneously; (2) to control hemorrhage after delivery in the case of uterine atony; (3) to hasten the normal involution of the uterus during the puerperium. The administration of oxytocin is strictly contra-indicated during the first and second stages of labor, because the type of contractions elicited is too intense and sustained. If the hormone is given before suitable dilatation of the cervix and rupture of the membranes, it may cause severe laceration of the cervix, rupture of the uterus or excessive trauma to the infant. In the event of prolonged tetanic contraction of the uterus, the fetus may even die from asphyxia.

The addition of thymus extract to posterior-lobe extracts was recommended by some physicians, but the efficacy of this practice has not been proven.

As has been stated above, the PLUMAGE of certain birds exhibits a sexual dimorphism. This is apparently under the control of the hypophysis and due to the influence of certain anterior-lobe hormones, mainly the gonadotrophins and thyrotrophin.

Sexual Cycle. — Hypophysectomy abolishes, while administration of impure anterior-lobe extracts, or of purified gonadotrophins, deranges the sexual cycle both in animals and in man; this is due to the resulting ovarian changes (see: p. 374). Intermediate and posterior-lobe extracts exert no specific effect upon sexual cyclicity.

Pregnancy. — Hypophysectomy during the first half of gestation causes abortion in most animal species; during the second half of pregnancy, however, the pituitary may be removed in some animals (e.g., rat) without interfering with the subsequent progress of gestation, the growth of the embryo and the development of the mammary glands. Delivery is usually delayed (interference with oxytocin production ? delayed involution of the pregnancy corpora lutea ?) and sometimes the embryo dies in utero. In other instances, however, delivery of normal young has been observed.

Treatment with crude ANTERIOR-LOBE EXTRACTS or gonadotrophins, especially luteotrophin, tends to prolong the normal duration of gestation, presumably because of an increased and protracted progesterone secretion.

Lactation. — After delivery, milk secretion commences in a fully-developed mammary gland, even in the absence of the anterior-lobe, but these secretory phenomena soon cease and the breasts involute. Probably during the second half of gestation the placenta is sufficiently well-developed to take over most of the endocrine functions of the anterior-hypophysis. Through the production of luteotrophin the placenta maintains the structure and function of the large corpora lutea of pregnancy; these, and the placenta, produce adequate amounts of progesterone to maintain the developing ovum. After termination of the normal "life span" of the placenta, the latter becomes detach-

ed and transitory milk secretion ensues as a "withdrawal phenomenon." The maintenance of the mammary glands in the hypophysectomized, pregnant animal has been ascribed to placental "mammogenic hormones," and milk secretion at parturition to placental "prolactin."

Hypophysectomy during lactation causes immediate cessation of milk secretion, unless adequate prolactin therapy is immediately initiated; indeed even active prolactin preparations rarely suffice to maintain adequate lactation after hypophysectomy for more than a very brief period.

Metamorphosis. — In hypophysectomized tadpoles metamorphosis does not occur, unless either thyrotrophin or thyroid hormone is administered. Thyrotrophin (through its action on the thyroid) can even accelerate normal metamorphosis in intact amphibia.

HYPOPHYSEAL HORMONE CONTENT OF BODY FLUIDS AND TISSUES

Blood and Urine. — A good deal of work has been done concerning the GONADOTROPHIN content of blood and urine, because of their diagnostic value and the comparative ease with which such determinations can be performed.

In *newborn children*, the body fluids contain appreciable quantities of LH which is presumably of placental origin. A few days later, however, only traces of gonadotrophins can be demonstrated in the blood and urine until *puberty*, when the urinary gonadotrophin excretion increases in both sexes and becomes approximately the same as in adults.

In normal *adult women* the urine always contains at least traces of gonadotrophins, but appreciable quantities (2-25 I.U./L.) of both FSH and LH have only been demonstrated during the 10th-14th day of the cycle, that is at the time of ovulation. A second — less constant — peak in gonadotrophin ex-

cretion is claimed to occur just before or during menstruation. (See : p. 806.)

Normal *adult men* excrete only traces of gonadotrophin without any cyclic variations in the daily amount. Most investigators report between 5 and 25 M.U./L. of FSH, but only traces of LH, if any.

In women with *menstrual disorders* gonadotrophin elimination is often abnormal; for instance a midmenstrual peak is generally absent in patients with anovulatory cycles. In primary hypövarianism both blood and urine contain greatly increased quantities of gonadotrophin. Conversely, in secondary ovarian failure due to anterior-lobe deficiency, the gonadotrophin elimination is diminished.

Large quantities of gonadotrophin (mainly FSH) are also eliminated during the *menopause* and in women following surgical or X-ray *castration*. The absolute values obtained by various

investigators are not readily comparable, because of differences in the bioassay technics and the definitions of the units. It has been stated however, that in menopausal or castrate women, the urine contains anywhere between 100 and 500 M.U./24 hrs. or 25-75 I.U./24 hrs., which is several hundred per cent above the maximum of the normal urinary excretion. All these observations are in accordance with the assumption that in the absence of gonadal hormones, the pituitary produces an excess of gonadotrophins in a, usually futile, effort to compensate for the ovarian deficiency by stimulating the growth of gonadal tissue.

In pregnant women extraordinarily large quantities of gonadotrophins appear in the blood and urine almost immediately after the first missed period. Between the 5th-6th week an average of 16,000 M.U./L. are found in the blood, but some investigators reported values as high as 500,000 I.U./L. of blood on the 14th day of pregnancy. The peak of urinary excretion of gonadotrophins (during pregnancy almost exclusively LH) is also reached between the 20th and the 60th day after the first missed period. At the time of maximum excretion, the urine contains about 60,000 I.U./L. After the 67th day the hormone level falls sharply and reaches a level of about 5,000 I.U./L. on the 140th day. The urinary concentration of LH tends to be considerably higher in multiple pregnancies, apparently because of the presence of two or more placentæ. (See : p. 821.)

Pregnant animals of most species do not excrete any appreciable amounts of gonadotrophins during gestation. In the mare the blood gonadotrophin content (mainly FSH) rises rapidly between the 37th and 42nd day of gestation and reaches a peak of about 50,000 I.U./L. between the 42nd and 80th day; it almost disappears completely from the blood between the 130th and 180th day. The high concentration of FSH in pregnant-mare-serum makes the latter

a valuable source for the preparation of this hormone (usually referred to as "PMS gonadotrophin"). Curiously, almost none of this blood gonadotrophin is eliminated in the urine of the pregnant mare.

It is noteworthy that in *ectopic pregnancy* the Aschheim-Zondek test, or its modifications, often give negative results. This is probably due to the fact that systematic hormone studies are rarely performed on patients with ectopic pregnancies until the appearance of symptoms; by that time the placenta is usually detached so that the gonadotrophins are no longer transmitted to the mother. As long as the placenta is viable, there is no reason to believe that it would produce less gonadotrophin if it were ectopically located.

In the event of *fetal death*, the LH content of blood and urine frequently declines to very low levels. Since LH is produced by the chorion and not by the fetus, it is debatable whether death of the latter is the cause or the result of diminished LH production. It is possible that damage to the placenta caused by fetal death diminishes LH production by the chorionic cells, but conversely, a deficient LH production may adversely affect the fetus.

In *hyperemesis gravidarum*, as well as in pre-eclampsia and eclampsia, the LH content of the blood is often extremely high, while that of the urine may be normal, high or low.

Tumors of placental tissue (chorionepitheliomas, hydatidiform moles) and teratoids (e.g., in the ovary or testis), which may also contain chorionic elements, likewise tend to raise blood and urine gonadotrophin titers. This is of considerable diagnostic value in patients in whom pregnancy can be excluded. Even in pregnant women, the LH elimination is usually much higher if there is placental neoplasia, but here the difference is less striking and hence not always sufficient to formulate a diagnosis.

It is noteworthy that the dysgerminoma or "false seminoma" of the ovary or testis likewise tends to augment LH excretion; this, as well as certain histologic characteristics of these tumors (see : p. 449) intimate a close relationship to neoplastic placental cells. That women with *uterine carcinomas* frequently also excrete excessive amounts of FSH is most probably due not to the tumor itself, but to the fact that the majority of these patients are of menopausal age.

In cases of so-called *pseudopregnancy* with corpus luteum cysts, the urinary gonadotrophin excretion is likewise high, thus increasing the difficulty of differentiating between this condition and pregnancy.

Increased urinary gonadotrophin excretion also occurs in the *male climacteric, cryptorchidism, the "hypergonadotropic eunuchoidism without a Leydigism"* (Klinefelter syndrome) and less regularly in patients with *anterior-pituitary tumors, increased intra-cranial pressure, anatomic lesions in the hypothalamus, adrenal-cortical tumors and hyperthyroidism*.

Injection of PMS into monkeys or rats does not cause any elimination of gonadotrophin in the urine, although these species excrete injected LH prepared from human urine or blood. In the pregnant mare, the urine is likewise almost free of gonadotrophins in spite of the high blood concentration, hence it appears that the FSH of pregnant mare serum cannot pass through the renal barrier. Unlike LH, PMS is as effective in single as in divided doses. This may likewise be due to the non-excretability of the latter and partly also to the accompanying proteins, which delay its absorption from the site of injection. Experiments in the rabbit and gelding show that injected PMS is demonstrable in the blood for several days without being excreted in the urine. Its final disappearance from the blood is apparently due to destruction by the cells of the organism, but it has not yet been possible to determine the exact site at which this destruction occurs. The gonads (the specific target organs of this hormone) are not involved in this destruction, since disappearance from the blood proceeds as rapidly in gonadectomized as in intact animals. This is in accordance with the previously expressed view (see: General Endocrinology) that hormones are not "utilized" by their target organs while they exert their physiologic effects (p. 20, 21).

Much less is known about the concentration of other hypophyseal hormones in the blood and urine, mainly

because the pertinent test methods are more complicated and less accurate. It has been shown however, that CORTICOTROPHIN excretion is high in Cushing's disease, while THYROTROPHIN elimination rises in some patients with thyrotoxicosis in accordance with expectations. Normally, only negligible traces of corticotrophin, thyrotrophin, or any other anterior-lobe hormones are demonstrable in the urine of man.

Allegedly, the SOMATOTROPHIN content of the blood is demonstrably increased in acromegaly.

INTERMEDIN elimination is greatly augmented in pregnant women; this has been used as the basis of a pregnancy test which appears to be reliable (*Konsuloff*).

OXYTOCIN AND VASOPRESSIN activity has also been demonstrated in human urine, but only in traces and there is some doubt concerning the specificity of the test methods used. It has been claimed that the vasopressin concentration of the urine rises during *dehydration* and that this represents a physiologic, compensatory mechanism which helps to retain body-water.

Intracarotid injection of hypertonic (25%) NaCl in dogs causes excretion of an anti-diuretic substance in the urine. Simultaneously, there is oliguria due to increased tubular reabsorption. Filtration remains normal, as judged by creatinine clearance. Probably NaCl stimulates the hypothalamus-pituicyte system directly.

Hypophysis. — Comparatively few systematic studies have been made with the object of determining the NORMAL, absolute concentration of the various *anterior-pituitary hormones* in the pituitaries of different species. It has been shown, however, that the corticotrophin content of the pituitary diminishes in the following order : pig, man, sheep, horse, cattle; while the prolactin content decreases in the order : sheep, ox, man, pig, horse. It is also known that cattle pituitaries are comparatively rich in growth hormone, but poor in gonado-

trophin; while pig pituitaries are a rich source of gonadotrophic activity.

The pituitary of the rat contains unusually large quantities of thyrotrophin, while that of the guinea-pig is poor in this principle; conversely, the rat thyroid is very insensitive while that of the guinea-pig is especially responsive to stimulation by thyrotrophin.

It is noteworthy, that although the human pituitary has no distinct intermediate-lobe, it contains very large amounts of *intermedin* within the anterior-lobe.

Oxytocin and *vasopressin* are especially plentiful in the posterior-lobe, but traces have also been demonstrated in the stalk and tuber cinereum.

Perhaps too much emphasis has been placed upon changes in the hormone concentration of the pituitary as reliable indicators of hormone production. The concentration of a hormone in the pituitary can rise due to increased production or decreased discharge into the blood. Hence a change in the hormone content of the gland gives no evidence of the amount produced. Nevertheless, it is interesting to note that GONADECTOMY in either sex increases the gonadotrophin content of the hypophysis, simultaneously with the proliferation of the "castration cells" and increased urinary gonadotrophin elimination. Hence, in this case there is adequate evidence of a rise in gonadotrophin production as a result of gonadectomy. This view is further supported by the fact that folliculoids and testoids diminish the high gonadotrophin content of the pituitary and urine in castrates of either sex.

Similarly, THYROIDECTOMY increases, while thyroid hormone administration decreases the thyrotrophin content of

the anterior-lobe. It has been claimed that THIOUREA decreases the thyrotrophin content of the pituitary, but the evaluation of this observation must await additional data.

During the latter half of GESTATION, the prolactin content of the pituitary begins to rise quite considerably, but after delivery, with the onset of LACTATION, this increase suddenly becomes still more pronounced.

The stimulus of *nursing* influences the luteotrophin content of the pituitary, since in postpartum rabbits the initially high prolactin concentration of the hypophysis declines more rapidly if the young are removed than if they are permitted to nurse. Folliculoids tend to increase the prolactin content of the pituitary even when given in doses sufficient to inhibit milk secretion.

Placenta. — The placenta is a rich source of hypophysoid hormones. It contains and secretes LH, FSH, luteotrophin and perhaps even "mammogenic hormone." The human placenta elaborates especially large quantities of LH, while the mare's placenta produces predominantly FSH. The placental (chorionic) cells of most other animal species elaborate comparatively small quantities of FSH and LH, but there is adequate proof of a high luteotrophin production by the chorionic cells of the rat and rabbit placenta. Data concerning the production of other pituitary hormones by placental cells is less convincing.

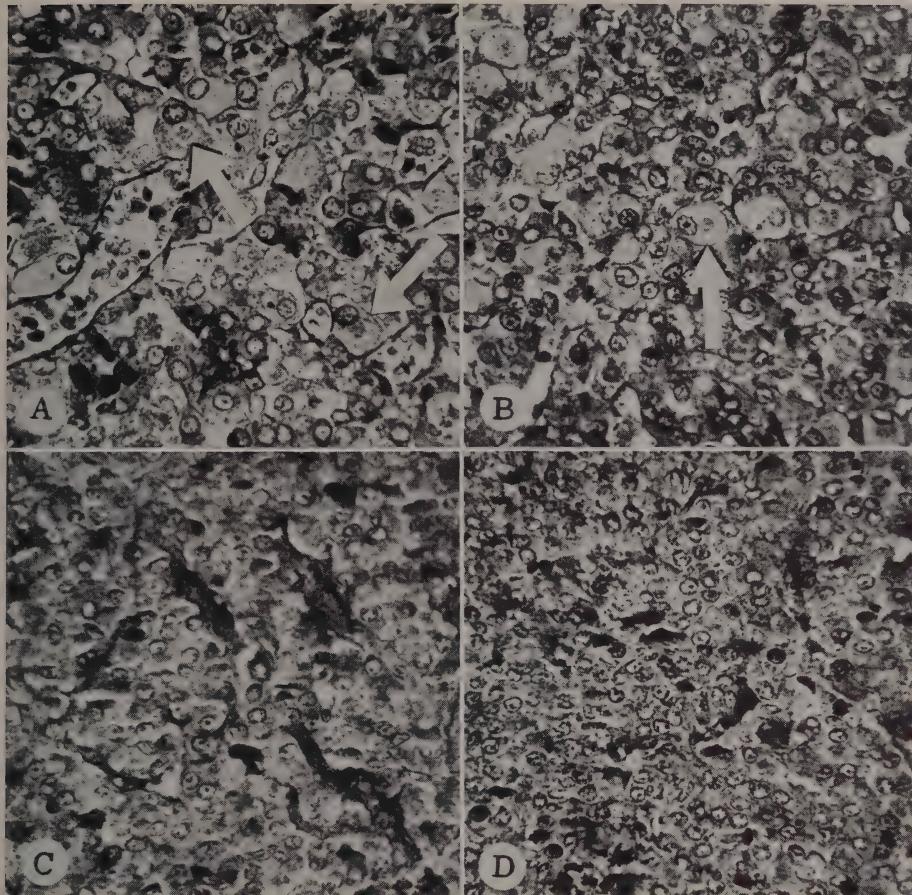
Plants. — It is interesting that gonadotrophic extracts have also been prepared from various plants (alfalfa-leaf meal, oak leaves, corn). These produce ovulation in the rabbit following intravenous injection, and some of them exhibit a gonadotrophic action even in the hypophysectomized male rat.

STIMULI INFLUENCING HYPOPHYSEAL STRUCTURE

Extrication of Endocrine Glands.

— ADRENALECTOMY causes no characteristic changes in the pituitary, although in the rat, some investigators report a decrease in eosinophils.

Following PARTIAL HYPOPHYSECTOMY, the remaining anterior-lobe tissue shows marked signs of compensatory hypertrophy. This is particularly obvious in the dog and rabbit, in which the



Effect of castration upon the hypophysis. — A. and B. Great enlargement of the pale basophil cells with beginning "signet-ring" formation (arrows). Note appearance of negative Golgi image beside the nucleus. The cytoplasm in the center of the Golgi reticulum is dark and homogeneous. Section taken 16 days after ovariectomy. In later stages, Golgi region is replaced by large vacuoles. — C. Hypophysis of spayed female rat, similar to that shown in Fig. A, but receiving 50 μ /day of α -estradiol. Note complete absence of "castration cells". — D. Hypophysis of rat similar to that shown in Fig. A, but treated with 5 mg./day of testosterone 10 days before autopsy. Note that testosterone also prevents the formation of "castration cells."

anatomically distinct pars tuberalis undergoes marked proliferation following ablation of the main pituitary body. In man, hypertrophy of the pars tuberalis has also been observed following destruction of the pituitary by disease. Regenerative phenomena in the intermediate and posterior-lobes are much less obvious.

CASTRATION, both in the male and female, produces essentially similar pituitary changes in animals of the same species. It is important to keep in mind, however, that the removal of the gonads causes qualitatively different changes in

various species. Thus, in the rat, gonadectomy (in either sex) produces a marked hypertrophy and hyperplasia of the basophils, which is quite obvious as soon as 14 days after the operation. At this time, enlarged basophils are especially numerous in the immediate vicinity of the middle-lobe. After three to four months, they are found throughout the anterior-lobe. These cells develop from normal basophils which first enlarge, then their Golgi apparatus becomes particularly prominent, so that the negative Golgi image is clearly visible in the shape of a light ring. Later,

a large vacuole is formed in the cytoplasm of the cell in the Golgi region, the subsequent enlargement of this vacuole pushes the nucleus to one side, giving the cell the characteristic "signet ring" appearance; the cytoplasmic border represents the ring itself and a large nucleus on one side is reminiscent of the signet.

Since among all laboratory animals, castration changes are most obvious in the rat, this species has been predominantly used for pertinent studies. Perhaps too far-reaching conclusions have hence been drawn, regarding the above-mentioned change in basophils and the increased gonadotrophic hormone production induced by castration. It is worth keeping in mind that in the majority of other animals (e.g., birds, cat, dog, pig, rabbit), as well as in man, gonadectomy causes proliferation of the eosinophils, sometimes accompanied by an actual decrease in basophils; yet in other animals (e.g., mouse) castration produces no conspicuous histologic change in the anterior-lobe. The only uniform effect of gonadectomy upon the pituitary of either sex, throughout the various animal species, is a more or less selective enlargement of the anterior-lobe, without significant influence upon either the intermediate or the posterior-lobe.

Administration of various steroid hormones to gonadectomized male or female rats inhibits the development of castration cells. The folliculoids, testoids, luteoids, corticoids and spermatogenic steroids are decreasingly less active in the order mentioned, but the degree of their activity is identical in the two sexes. From this, it has been concluded that the anti-castration-cell effect is subordinate to the folliculoid activity with which it runs parallel throughout the pharmacologic groups enumerated above. In this connection, it is worth reëmphasizing that the gonadotrophins (LH and FSH) unlike the gonadal hormones (folliculoids and luteoids in the female; testoids in the male) are not

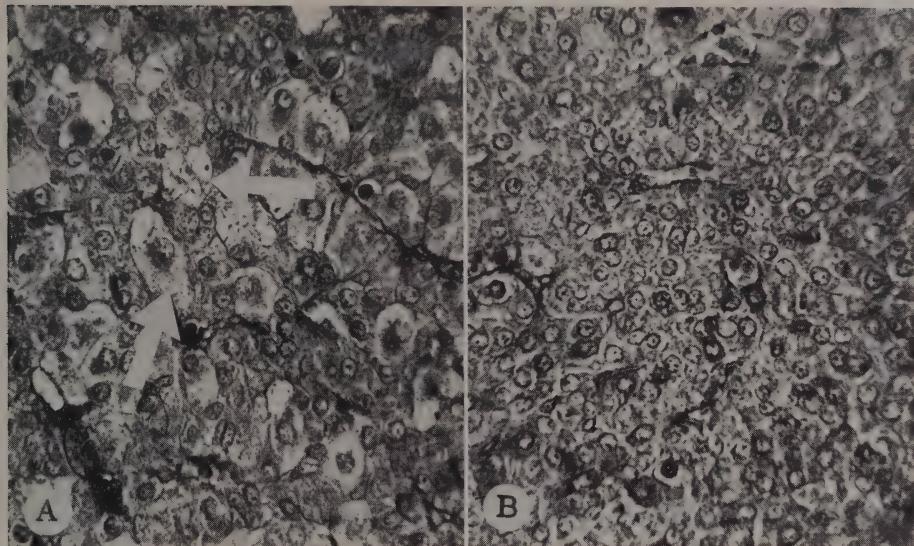
sex-specific. Correspondingly, the changes in anterior-lobe structure, accompanying increased gonadotrophin production, are the same in gonadectomized male and female animals and the inhibition of the castration changes can be accomplished by gonadal hormones of either sex, both in the castrate male and in the spayed female. *Ovarian or testicular transplants* likewise prevent the development of castration cells in either sex.

It has been claimed that *transection of the pituitary stalk* inhibits the formation of castration cells following subsequent gonadectomy in the rat, but administration of folliculoids succeeds in curing preëxistent castration changes, even in a pituitary whose stalk has been severed. It has been concluded that innervation of the anterior-lobe is essential for the formation of castration cells, but not for their cure by folliculoids.

Lactation inhibits the development of castration cells in spayed rats, perhaps due to the nervous stimulus of nursing.

PARATHYROIDECTOMY causes no conspicuous change in the pituitary of most species, although in the rat, some investigators claim to have found an increase in the number of eosinophils following this operation.

THYROIDECTOMY, whether or not accompanied by parathyroidectomy causes very pronounced and typical lesions in the anterior-lobe, especially in the rat. Among these, the most conspicuous is the formation of "signet-ring cells." These are similar to those produced by castration, but numerous small vacuoles appear after thyroidectomy instead of the single large one characteristic of the castration cell. Furthermore, in the thyroidectomized rat, the number of acidophils tends to decrease. Although the qualitative changes in the pituitary are different in the various animal species, it may be said that thyroidectomy rather uniformly results in a selective increase in the weight of the anterior-lobe, without noteworthy changes in the intermediate- or middle-lobes.



Effect of thyroidectomy upon the hypophysis. — **A.** Hypophysis of adult male rat 14 days after thyroidectomy. Note "multiple vacuolization" of the hypertrophic cells (arrows); this helps to distinguish them from the castration cells, in which only a single large vacuole develops in the Golgi region. — **B.** Hypophysis of a rat, similar to that shown in Fig. A, but receiving $350 \gamma/\text{day}$ of thyroxin. Note absence of "thyroidectomy cells".

In man, postoperative hypothyroidism likewise causes an increase in the pituitary weight, which is usually accompanied by the appearance of vacuolized basophils.

Administration of the various steroid hormones which are effective in curing the castration changes in the anterior-lobe of the rat do not influence the thyroidectomy cells, while conversely, *thyroid hormone* (thyroxin or desiccated thyroid) readily restores to normal the hypophysis of the thyroidectomized rat, but fails to influence the changes induced by castration.

Removal of OTHER ENDOCRINE GLANDS causes no constant, characteristic lesions in the pituitary.

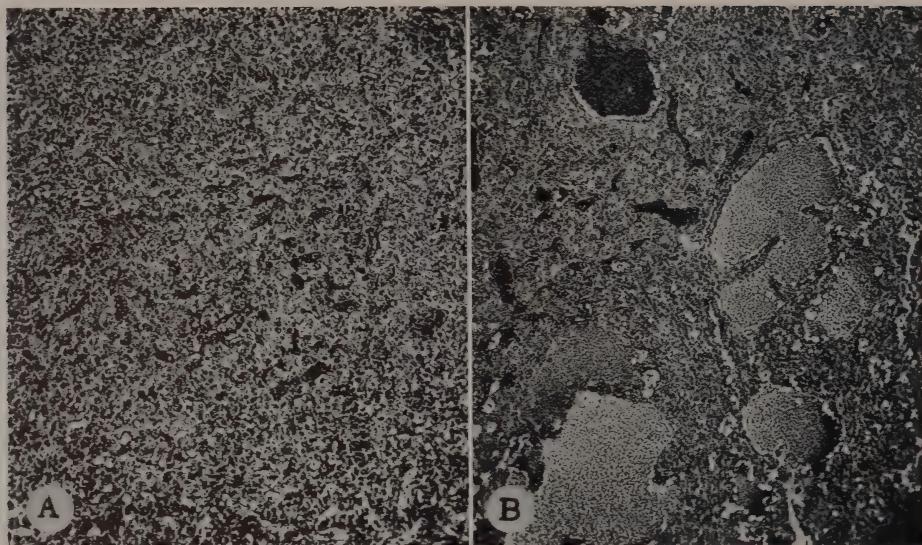
Hormones. — Reports concerning the influence of PITUITARY PREPARATIONS upon the anterior-lobe are somewhat conflicting, probably because, up to now, comparatively little work has been done with pure hormones. Chronic treatment with crude, cattle anterior-lobe extracts causes involution of the anterior-lobe in the rat; this is probably due to compensatory atrophy. Such ex-

tracts are poor in gonadotrophic potency.

Treatment, especially of immature females, with LH from pregnancy urine or placenta, causes an hypertrophy of the anterior-lobe, which runs approximately parallel with the resulting ovarian enlargement and fails to occur after spaying. Since folliculoids enlarge the anterior-lobe, it is probable that the gonadotrophins stimulate it, through the folliculoids produced by the animal's own ovary. This is all the more likely, since very chronic treatment with LH produces vacuolized anterior-lobe cells, similar to "castration cells," at a time when antihormone formation results in a secondary ovarian involution.

It is probable that anterior-lobe hormones can influence the pituitary in two ways: directly, causing compensatory atrophy of the anterior-lobe cells and indirectly, through the ovary, stimulating the anterior-lobe through the excess formation of folliculoids.

FOLLICULOIDS cause a very pronounced enlargement of the anterior-lobe in various animal species, especial-



Effect of folliculoids upon hypophysis. — A. Hypophysis of normal adult male rat (9 mg.). — B. Hypophysis of adult male rat treated with 100 γ /day of α -estradiol during 9 months. Note cystic cavernoma-like formation in the midst of chromophobe adenomatous anterior-lobe cells (54 mg.).

ly in the rat and mouse. This is accompanied by degranulation of both the acidophils and the basophils, as well as by development of cell types similar to those seen in pregnancy (see p. 260).

In the event of chronic folliculoid treatment, chromophobe adenomas, with large cavernous blood sinuses develop in the anterior-lobe of the mouse and rat. These tumors may become so large that eventually the animals lose their sight due to pressure upon the optic chiasm and excessive intracranial pressure is the final cause of death. Malignant changes have never been observed in such neoplasms.

TESTOIDS have no pronounced effect upon the pituitary when given by themselves, although they tend to counteract the hypophyseal enlargement otherwise produced by folliculoids.

THYROID HORMONE causes variable changes in the anterior-lobe, although most investigators agree that a proliferation of the basophils with hypertrophy of their Golgi apparatus is rather characteristic.

OTHER HORMONE PREPARATIONS have not been found to elicit any characteristic anterior-lobe changes.

Diseases. — In ADDISON'S DISEASE, the pituitary is usually small and the number of basophils diminished. These changes are especially conspicuous, when the adrenal insufficiency is secondary to a primary anterior-lobe failure (see: p. 263).

In juvenile DIABETES MELLITUS, the weight of the pituitary is diminished and the number of eosinophils decreased. These cells also show retrogressive changes, particularly atrophy. In rare instances the basophilic cells show hydropic degeneration. More often than in any other condition, foci of fetal cells, characterized by high cylindric shape, are seen in diabetes (E. J. Kraus).

In MYXEDEMA and HYPERTHYROIDISM, characteristic anterior-lobe lesions are not observed, although various investigators described minor changes which they consider to be typical. Usually, the anterior-lobe of patients with Graves' disease is small and the chromophils show signs of degeneration.

HYPERTENSION AND RENAL DISEASES are claimed to be frequently accompanied by the so-called "basophilic in-

vasion" of the posterior-lobe. In the anterior-lobe, the number of basophils is allegedly likewise increased. However, this process is common in normal individuals and hence difficult to evaluate. It is perhaps more significant that basophilic adenomas in the anterior-lobe are often accompanied by hypertension and secondary contracted kidney. The frequent occurrence of hypertension with nephrosclerosis in Cushing's disease, in which basophilic adenomas are found in the pituitary, is likewise noteworthy in this connection. In view of recent experimental work, showing that anterior-pituitary extracts can produce hypertension and nephrosclerosis (see experimental data on p. 248), these observations are rather significant. While it is true that not all cases of renal hypertension are accompanied by obvious lesions in the anterior-lobe, the bulk of the published data suggests some relationship between basophilism and renal hypertension.

ADIPOSITY, especially that of the constitutional type, is frequently accompanied by basophilia of the anterior-lobe and small chromophobe or basophil adenomas are unusually common in such patients. Perhaps the adiposity of Cushing's disease is also related to basophil proliferation.

In CHORIONEPITHELIOMAS, which produce excessive amounts of gonadotrophins, the pituitary shows "pregnancy changes."

Increased INTRACRANIAL PRESSURE, due to hydrocephalus, brain tumors, etc., is usually associated with hyperplasia of the anterior lobe. Only if the causative process destroys the stalk is there atrophy of both anterior and posterior lobes (*E. J. Kraus*).

It has been claimed that specific lesions occur in the anterior-lobe of patients suffering from DEMENTIA PRECOX, EPILEPSY, PROGRESSIVE PARALYSIS, ENCEPHALITIS EPIDEMICA and OTHER NERVOUS DISEASES, but these lesions are not

very characteristic. In ECLAMPSIA, basophilic invasion of the posterior-lobe, accompanied by proliferation of the basophils in the anterior-lobe, has also repeatedly been reported, although it is not very constant or conspicuous.

In patients bearing MALIGNANT TUMORS, degenerative changes have been seen in the anterior-lobe and in rats with transplantable carcinomas, large vacuolated cells are noted in the pars distalis. However, similar changes have also been produced by the injection of various protein extracts, and probably the decomposition of tissue in the necrotic tumor centers (rather than any specific humoral substance) is responsible for the pituitary lesions observed.

The hypophyseal lesions associated with various clinical forms of HYPO and HYPERPITUITARISM are discussed in the sections devoted to the latter; OTHER DISEASES rarely cause striking and specific changes in the hypophysis.

Diet. — Changes in diet may also lead to histologic lesions in the anterior-lobe, but it is often difficult to determine whether these are the direct result of the diet, or secondary consequences of the accompanying gonadal atrophy, adreno-cortical hypertrophy, etc.

AVITAMINOSIS-E induces castration changes in the pituitaries of male, but not of female rats, presumably because only in males do such diets cause gonad involution.

Nervous Stimuli. — In spite of the fact that the pituitary stalk carries numerous fibers to the pituitary, carefully performed stalk transections elicit no histologic change in the anterior-hypophysis. The positive findings of early investigators were probably due to incidental interference with the hypophyseal blood supply.

In animals in which copulation elicits pseudopregnancy (e.g., rabbit) the stimulus of mating decreases the eosinophil and basophil count. The chromophobes, on the other hand, become particularly abundant 5-6 days after mat-

ing. Bilateral extirpation of the superior cervical sympathetic ganglia in rabbits has also been claimed to decrease the eosinophil count of the anterior-lobe. Some investigators believe that the cervical sympathetic may be partly responsible for the hypophyseal changes elicited by mating especially since its electric stimulation (as that of the brain or spinal cord) can cause pseudopregnancy.

Age. — Changes in pituitary weight during the course of normal life, have been discussed under "Anatomy." Histologically, basophilic invasion of the posterior-lobe is alleged to be a characteristic manifestation of senility in man. It has also been claimed that the cystic portion, corresponding to the pars intermedia, increases in old people, although its colloid content remains the same. There is a decrease in the eosinophil, and an increase in the chromophobe count, while the basophils show no significant change (*Rasmussen*). Sometimes, lipid granules may accumulate in the anterior-lobe cells of very old people.

Sex. — In most laboratory animals, as well as in man, the anterior-lobe of females is larger than that of males. The weight of the other lobes is essentially independent of sex.

Estrus and Menstruation. — In several animal species, histologic changes characteristic of certain phases of the sexual cycle have been described; these do not lend themselves to generalizations, since they are qualitatively different in various species. In animals with a seasonal estrus, the anterior-lobe is usually largest at the height of heat.

Pregnancy. — The anterior-lobe is selectively enlarged during gestation in various species including man. Following repeated pregnancies, this growth becomes even more prominent.

In the rat, the eosinophils decrease and the chromophobes increase in number during gestation. Many large, light,

"pregnancy cells" arise from chromophobes or basophils. They contain very fine eosinophilic granules and resemble the cells which proliferate in the anterior-lobe after folliculoid hormone treatment.

In the human pituitary, no very characteristic histologic change is seen to accompany the anterior-lobe enlargement typical of gestation. It is questionable whether the disturbances of vision, often occurring in pregnant women, are due to compression of the optic chiasm by the enlarged hypophysis, or whether they result from functional disturbances.

Lactation. — In certain animals (e.g., guinea pig), almost all the eosinophils are degranulated postpartum, but in most species, the hypophyseal changes during lactation are not essentially different from those of pregnancy.

Season and Hibernation. — In animals with a seasonal estrus, and especially in hibernating animals (e.g., squirrel, wood-chuck, marmot), the weight of the anterior-lobe decreases considerably during the non-breeding or hibernating season and reaches a maximum at estrus. Concurrently, with the involution during the off-season, the chromophils lose their granules. There are no characteristic changes in the intermediate and posterior-lobes. It is probable that the above-mentioned seasonal variations are chiefly correlated with the sexual cycle rather than with hibernation in itself. However, a decreased production of corticotrophin, thyrotrophin and other metabolic anterior-lobe hormones during hibernation, is suggested by the decrease in metabolism and the atrophy of the adrenal cortex and thyroid.

Drugs. — Various drugs have been claimed to cause more or less characteristic lesions in the pituitary, but only a few of these appear to be specific.

ALLOXAN causes degenerative changes, ranging from hydropic degeneration to hyalinization necrosis and cyst for-



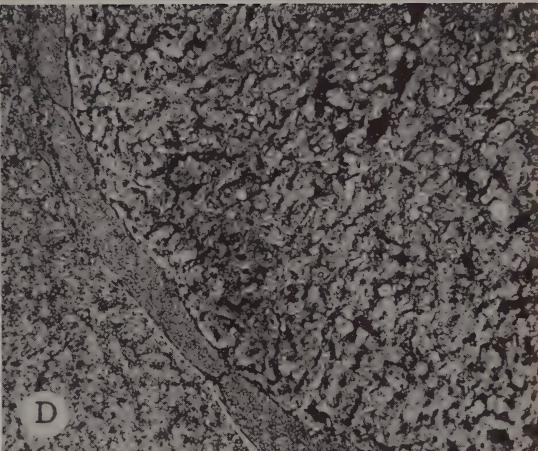
A



B



C



D



E

Effect of hypertonic NaCl on the pituitary. — A. Macroscopic view of the hypophysis of a normal control rat (as seen from above). — B. Macroscopic view of the hypophysis of NaCl treated rat (same magnification as Fig. A.). Note prominent and enlarged posterior-lobe and atrophic, thin intermediate-lobe. (The smaller size of the anterior-lobe is only apparent and due to fragmentation at the edges). — C. Section through anterior-, middle and posterior-lobe of a normal control rat. — D. Section through anterior-, middle and posterior-lobe of NaCl treated rat. Note atrophy of intermediate-lobe and infiltration of posterior-lobe with light edema fluid. The "honey-combed" appearance of the posterior-lobe is due to the presence of numerous vacuoles and light cells in mitosis. In many animals of this series (not in the one shown here) the pituitary cleft was distended with fluid. — E. High magnification of a cell in mitosis taken from the posterior-lobe of the rat shown in Fig. D.

(After H. Selye and C. E. Hall: Anat. Rec. 86, 579, 1943.)

mation in the anterior-lobe of various animals. The basophils appear to be particularly affected and there may be some correlation between this lesion and the concurrent degeneration of the Langerhans islets which results in diabetes.

THIOUREA and its derivatives, which interfere with thyroid hormone production, cause the same type of basophil degeneration in the anterior-lobe, as thyroideectomy. Accordingly, thyroid hormone treatment prevents the pituitary lesions produced by thiourea derivatives. In the case of complete inactivation of the thyroid by thiourea, about 2.25 γ of *dl*-thyroxin/100 gm. of body weight/day, is required to prevent these basophil changes in the rat. It was concluded that this represents the daily thyroid hormone requirement of the rat and that the pituitary change is indicative of an increased thyrotrophic hormone production, as part of the compensatory hypertrophy mechanism, elicited by thyroid deficiency.

Feeding or intravenous injection of hypertonic NaCl solutions causes swelling of the posterior-lobe, increased mitotic proliferation of pituicytes and fluid accumulation in the hypophyseal cleft in the rat. Presumably these changes are due to the increased demand for posterior-lobe hormones occa-

sioned by the resulting disturbance in water metabolism. (See : p. 261.)

Rays. — LIGHT increases the melanophore hormone content of the amphibian hypophysis and simultaneously, elicits absorption of the intermediate-lobe colloid into the capillaries, presumably a sign of increased secretion. In mammals, neither light nor ULTRA-VIOLET rays exert any noteworthy effect upon the structure of the pituitary, although it has been claimed that in the FERRET, exposure to ultra-violet irradiation causes the appearance of large cells in the anterior-lobe, concurrently with the induction of estrus.

X-RAYS may cause complete destruction of the anterior-lobe, but usually only in doses which damage the adjacent brain centers sufficiently to cause death. In embryonic chicks, X-ray treatment of the pituitary region may completely destroy the hypophyseal primordium, so that the gland fails to develop. This is usually accompanied, however, by other severe malformations of the cephalic end of the body.

In man, the pituitary is rather sensitive to X-ray treatment and temporary castration may result from irradiation of the hypophyseal region. The therapeutic value of X-ray treatment of the pituitary in hypophyseal diseases will be discussed in conjunction with the latter.

DISEASES OF THE HYPOPHYSIS

MALFORMATIONS

Among the malformations of the hypophysis, those which tend to give rise to hormonal disturbances are comparatively rare. Malformations of the cephalic end of the embryo, especially anencephaly may lead to abnormal development, and in rare instances to APLASIA of the hypophysis. In many cases of anencephaly, however, the pituitary is normal. HYPOPLASIA of the gland has repeatedly been described, even without anencephaly, in individ-

duals exhibiting signs of the adiposogenital syndrome, dwarfism or diabetes insipidus.

In rare instances, there is DYSTOPIA OF THE NEURO-HYPOPHYSIS, that is, separation of the posterior from the anterior-lobe so that the former becomes situated above the diaphragm and is connected with the latter by a thin stalk only. The condition resembles that normally existing in some animals (e.g., whale). Lesser degrees of pituitary malformation are occasional-

ly seen in mongolian idiocy, a syndrome regarded by some as a special type of hypopituitarism.

SIMPLE ATROPHY AND SCLEROSIS of the hypophysis may result from vascular disturbances or inflammatory lesions. Sometimes it is "idiopathic," that is, due to unexplained causes. It may occur as part of the "pluriglandular dystrophy" of Falta, a syndrome in which several endocrine glands undergo sclerosis and atrophy. Probably most of the allegedly pertinent cases are actually instances of hypopituitarism with secondary atrophy of those endocrines which are under pituitary control. However, in a few cases the endocrine glands exhibit sclerosis and chronic inflammatory lesions rather than the simple atrophy seen as a result of hypopituitarism. (See also : p. 867.)

VASCULAR LESIONS

Direct trauma may cause HEMORRHAGES AND NECROSIS of the hypophysis, sometimes conducive to acute hypopituitarism. THROMBOSIS, EMBOLISM and INFARCTS of the pituitary vessels themselves or of the sinus cavernosus can likewise result in acute hypopituitarism. Bacterial emboli are often the cause of pituitary infarcts. Such vascular lesions are especially common after complicated childbirth and in such cases tend to cause the postpartum type of Simmonds' disease. (See also : p. 267.)

DEGENERATIONS

"CLOUDY SWELLING" is difficult to recognize in the pituitary and it is doubtful whether it ever occurs. HYDROPIC DEGENERATION of the basophils has been described as a characteristic accompaniment of diabetes mellitus.

HYALINE DEGENERATION of the stroma may occur in the pituitary of old people as a sequel to tuberculosis, syphilis or other inflammatory lesions.

The term "COLLOID DEGENERATION" has often been used to describe the formation of colloid cysts within the anterior-lobe or at the border-line between



Amyloidosis of the hypophysis. Note homogeneous amyloid deposits which compress the anterior-lobe cell cords.

the anterior and posterior-lobe, a region in which some small colloid cysts are normally present. Allegedly this is particularly frequent in patients with intra-cranial growths. It is doubtful whether we should refer to this as a degenerative process.

AMYLOIDOSIS of the hypophysis is extremely rare and almost always occurs as part of generalized amyloidosis.

FATTY DEGENERATION of pituitary cells may occur in cases of acute yellow-liver atrophy, leukemia, phosphorus poisoning, septicemia, chronic nephritis, etc., that is to say, in diseases in which fatty degeneration of other organs is likewise common.

Among rare degenerative lesions, we might also mention the DEPOSITION OF CALCIUM, GLYCOGEN and PIGMENT GRANULES (especially hemosiderin).

INFLAMMATIONS

NON-SPECIFIC INFLAMMATORY LESIONS, usually described as "hypophysitis," may occur as a result of hematogenous infections, especially bacterial emboli, which sometimes cause abscess formation and extensive destruction of



Tuberculosis of the hypophysis. Caseous tuberculum in posterior-lobe. Note line of demarcation between homogeneous necrotic mass and remnant of healthy posterior-lobe tissue; in this region granuloma cells predominate. Anterior-lobe and pars tuberalis are normal.

pituitary tissue. Chronic inflammatory lesions with round-cell infiltrations may gradually lead to sclerosis of the hypophysis and hypopituitarism. In other instances, inflammations spread directly to the pituitary from the sinus cavernosus, the meninges or the sphenoid bone.

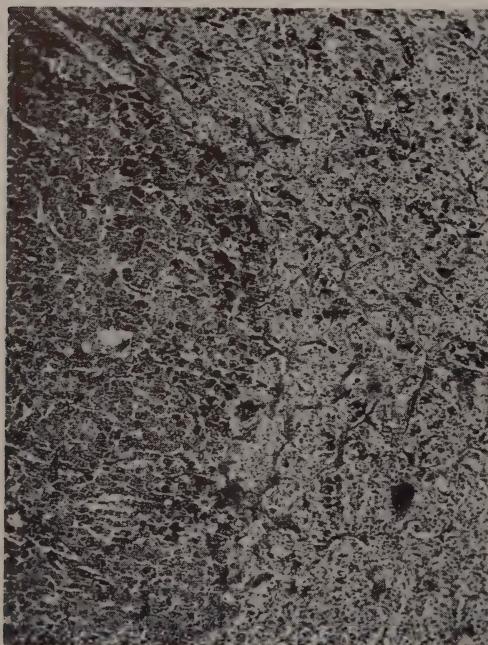
Among the specific inflammatory lesions, TUBERCULOSIS and SYPHILIS are

noteworthy since they can cause sufficient destruction to produce hypopituitarism.

TUMORS

The clinical aspects of pituitary tumors will be discussed in connection with the hormonal disturbances they produce. Here we shall consider them only from the morphologic view-point.

Adenomas. — Pituitary adenomas of small, often microscopic, size are comparatively common; they are found in about 10% of all human pituitaries. According to the prevalent cell type, we differentiate between: CHROMOPHOBIC, EOSINOPHILIC and BASOPHILIC adenomas. Only rarely does the same adenoma contain both eosinophils and basophils ("MIXED ADENOMAS"), or do separate eosinophilic and basophilic adenomas develop in the same pituitary. Some adenomas have no capsule and are consequently difficult to delimit from adjacent normal pituitary tissue. Hyperplastic nodules of this type form a



Eosinophilic adenoma of the hypophysis. Note dark eosinophilic cells forming adenoma which is clearly delimited from the remaining pituitary tissue (right).



Basophilic adenoma of the hypophysis. Note sharp delimitation of the irregular adenoma tissue from the normal anterior-lobe cell cords.

transition between simple hyperplasia and adenoma formation ("ADENOMATOUS HYPERPLASIA"). While eosinophilic adenomas are usually limited to the anterior-lobe, basophilic adenomas are sometimes found within the substance of the pars nervosa; here basophilic infiltration of the posterior-lobe is a frequent accompaniment.

A comparatively rare, special type of the chromophobe adenoma consists of tubularly arranged, high-cylindric chief cells, not unlike those of the embryonic pituitary. Sometimes these so-called "FETAL ADENOMAS" exhibit signs of malignancy.

The ADENOMA PSAMMOSUM is an anterior-pituitary tumor whose cells are poor in cytoplasm and surround hyalinized, often calcified, concretions. Occasionally, calcification may go so far as to cause an almost bone-like hardening of the gland. All types of pituitary adenomas are more frequent in advanced age and only 6% of them are seen in individuals younger than 20 years.

PRIMARY CARCINOMAS of the anterior-lobe usually consist of wider trabeculae and more irregular cells than those of normal pituitary tissue. Yet their differentiation from adenomas is not always possible, unless malignancy is demonstrated by infiltrative growth or metastases. Intermediate types have often been referred to as "*malignant adenomas*," which is a rather confusing designation.

Cysts of the pituitary are often lined by ciliated epithelium, such as that normally present in the "intermediate-lobe" region. If sufficiently large they may cause hypopituitarism due to compression of the anterior-lobe tissue.

The CRANIOPHARYNGIOMAS or "Erdheim tumors," are probably derived from the craniopharyngeal duct. They consist of cells, similar to those of the stratum spinosum of the epidermis, which form massive cell cords. Occasionally, they have a gelatinous, myxomatous stroma reminiscent of adaman-

tinomas. Pearl-like cornified masses are absent, but sometimes large cystic cavities (so-called pseudocysts) are formed in these tumors. Occasionally part of the tumor undergoes ossification or calcification; this may lead to confusion with teratomas, but the distinction is rather academic since the craniopharyngioma is derived from an embryonic vestige. Sometimes these growths contain a great deal of glia, bone, adenomatous tissue, pigment and cholesterol. The so-called "*cystic-papillomatous craniopharyngioma*" is a special type of this tumor; it contains large cysts, the interior of which is occupied by cauliflower-like outgrowths of stratified squamous epithelium which show a marked tendency to desquamate the superficial layers. Here again calcification and ossification of the stroma may occur.

MALIGNANT CRANIOPHARYNGIOMAS are actually basal-cell carcinomas of the craniopharyngeal duct. Unlike other carcinomas they are especially common in young people, while the benign craniopharyngiomas are more frequent in adults and older patients.

CHOLESTEATOMAS are occasionally found in the region of the infundibulum, but not in the pituitary itself. Unlike the craniopharyngiomas they show kerato-hyaline pearl formation. This has been considered a sign of their epidermal origin which distinguishes them from the craniopharyngiomas (arising from oral ectoderm, but not from the epidermis).

True TERATOMAS of the pituitary are supposedly rare since most of the allegedly pertinent cases of the old literature are now classed as craniopharyngiomas.

FIBROMAS, LIPOSAS, ANGIOMAS, and SARCOMAS of the hypophysis, as well as GANGLIONEUROMAS and GLIOMAS of the posterior-lobe are rare.

METASTATIC CARCINOMAS, and in very exceptional instances, SARCOMAS of the pituitary may also occur.

ANTERIOR-LOBE HYPOFUNCTION

DEFINITION

Anterior-lobe hypofunction is a condition in which the hormone production of the anterior-lobe is sufficiently disturbed to cause detectable manifestations of insufficiency. Among these, hypogenitalism, a decreased B.M.R. and muscular weakness are especially prominent. In young individuals this is accompanied by dwarfism; in adults by a tendency towards the development of cachexia.

CLASSIFICATION

The clinical types of anterior-lobe deficiency may be classified according to different viewpoints, such as the AGE OF ONSET, the intensity of the endocrine deficiency, the underlying PATHOLOGIC LESION in the anterior-lobe, etc. It is most customary however, to classify these syndromes according to the CLINICAL MANIFESTATIONS which they elicit. In this sense, we distinguish :

(1) *Late, general anterior-lobe deficiency* (Simmonds' disease), in which the condition develops after completion of normal growth. Since Dr. Simmonds' patients suffered from severe cachexia, some authors object to the use of the term Simmonds' disease when this typical manifestation is absent. Yet we shall employ it as synonymous with "late, general anterior-lobe deficiency" in order to avoid this cumbersome (though more correct) designation.

(2) *Early, general anterior-lobe deficiency* (Lorain-Levi Syndrome or pituitary dwarfism) in which the onset precedes the ossification of the junction cartilages. (See also : classification of infantilism and dwarfism, pp. 286, 287.)

(3) *Selective failure of certain anterior-lobe functions*, which is frequently due to the "shift in anterior-lobe-hormone production" elicited by a greatly increased requirement for one particular type of hypophyseal principle (e.g., decreased gonadotrophin and somatotrophin secretion at the expense of cor-

ticotrophin such as occurs during the general-adaptation-syndrome, or after folliculoid overdosage). — In *dystrophia adiposogenitalis* (*Fröhlich's syndrome*) and the *Laurence-Moon-Biedl Syndrome* there is selective failure of gonadal development and adiposity without any other manifest sign of hypopituitarism. These syndromes, and perhaps even the "metabolic craniopathy" (p. 287) probably result from deranged gonadotrophin production, presumably caused by lesions in the vegetative centers of the hypothalamus.

The word "*panhypopituitarism*" has often been used to designate complete failure of all anterior-lobe functions. Since it may give rise to confusion with simultaneous failure of both the anterior and the posterior-pituitary, we shall not use this designation.

PATHOLOGIC ANATOMY

Both SIMMOND'S DISEASE and PITUITARY DWARFISM may be caused by any local lesion which destroys most or all



Hypophysis in Simmonds' disease. Note almost complete replacement of hypophysis by scar tissue. Only a few anterior-lobe cell groups persist.
(Courtesy of Dr. W. Boyd.)

of the pars glandularis. Among such lesions are: tumors, hemorrhage, inflammatory processes and granulomas, hypoplasia, atrophy, etc. Among the tumors, the chromophobe adenomas and crano-pharyngiomas are the most common cause of hypopituitarism, because they themselves are not hormone-producing. However, even eosinophilic adenomas with acromegaly, or chromophobe adenomas with Cushing's disease may eventually result in secondary hypopituitarism, if the neoplastic tissue undergoes necrosis or extensive infarction as a result of vascular disturbances.

FRÖHLICH'S SYNDROME may be unaccompanied by any detectable lesion in the hypothalamico-pituitary region, but usually there are signs of an intrasellar, or suprasellar tumor and a chromophobe adenoma or Rathke-pouch cyst is found at operation.

We have no data concerning the causative anatomic lesion in the LAURENCE-MOON-BIEDL SYNDROME.

INCIDENCE

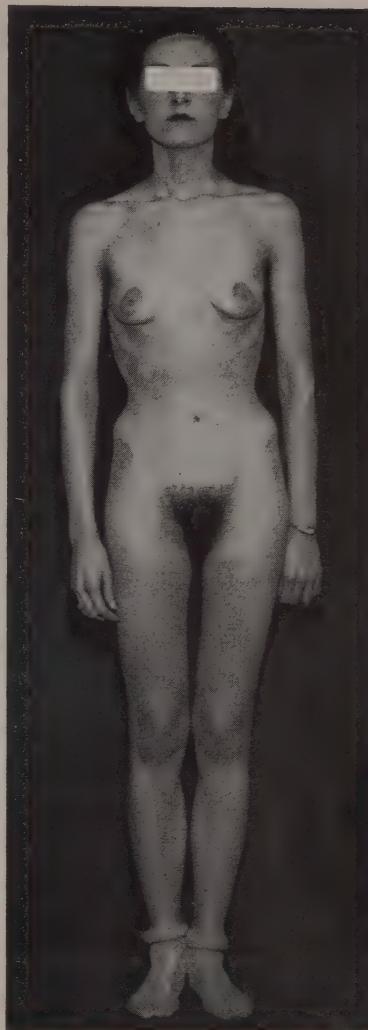
General anterior-lobe deficiency may occur at any AGE, but in children it is comparatively rarely accompanied by the profound cachexia, characteristic of late Simmonds' disease.

Mild adiposogenital dystrophy, as well as other types of selective gonadotrophin deficiency are more common in prepubertal than in older individuals, while the classic syndrome of severe adiposogenital dystrophy tends to develop in adults.

PREGNANCY definitely predisposes to Simmonds' disease, and the latter often appears immediately postpartum. In such cases the causative lesion is usually a pituitary infarct. (See : p. 263.)

Simmonds' disease is about 4 times more frequent in the female SEX, adiposogenital dystrophy is more common among boys, while the incidence of Laurence - Moon - Biedl syndrome is about equal in the two sexes.

HEREDITY also plays an important rôle in the development of various



Ovarian failure with malnutrition. 28-year-old woman with ovarian failure and malnutrition, following delivery 4 years earlier. Hypomenorrhea, vaginal smears markedly deficient, endometrium atrophic, FSH: 26-53 M.U./24 hrs., 17-KS: 6.8 mg./24 hrs., visual fields normal. (Postpartum Simmonds' syndrome?)
(Courtesy of Dr. E. P. McCullagh.)

hypopituitary conditions. Dwarfism, combined with obvious signs of infantilism, has repeatedly been seen in families in which other members also show definitely stunted growth. The hereditary occurrence of dwarfism in certain strains of mice (with aplasia of pituitary eosinophils) also indicates the importance of genetic factors. Since severe anterior-pituitary failure causes



Familial dwarfism. — A. Father of normal size (5'6", weight 175 lbs.). Three sons (from left to right) 25, 11 and 22 years old respectively, show signs of hypopituitarism, hypothyroidism and hypogonadism. Mother obese (weight 270 lbs.), but of normal height (5'6"), two sisters of normal stature. — B. Oldest of three brothers shown in A. Note immaturity of facies, genitalia and body appearance at 25 years of age. The sella and visual fields were normal. Epiphyseal maturity markedly delayed. Urinary gonadotrophins too low to measure, 17-KS: 0.1 mg./24 hrs., B.M.R.: -35%, blood cholesterol: 326 mg.%. — C. 5 years later. Note striking maturation in appearance of face and body due to treatment with thyroid, methyltestosterone and LH.

(Courtesy of Dr. E. P. McCullagh.)

sterility, direct transmission of the manifest disease is rarely possible.

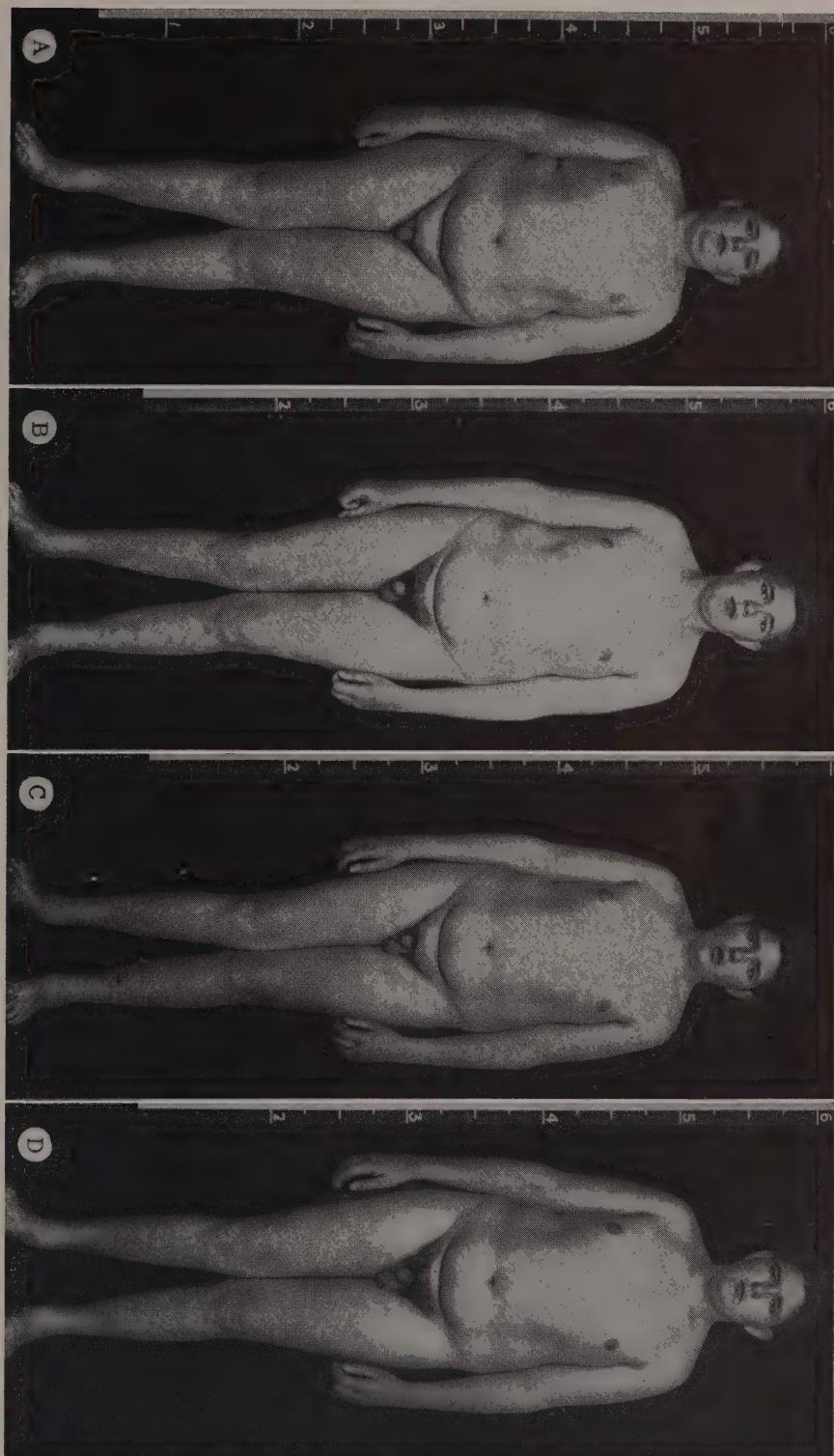
PATHOGENESIS

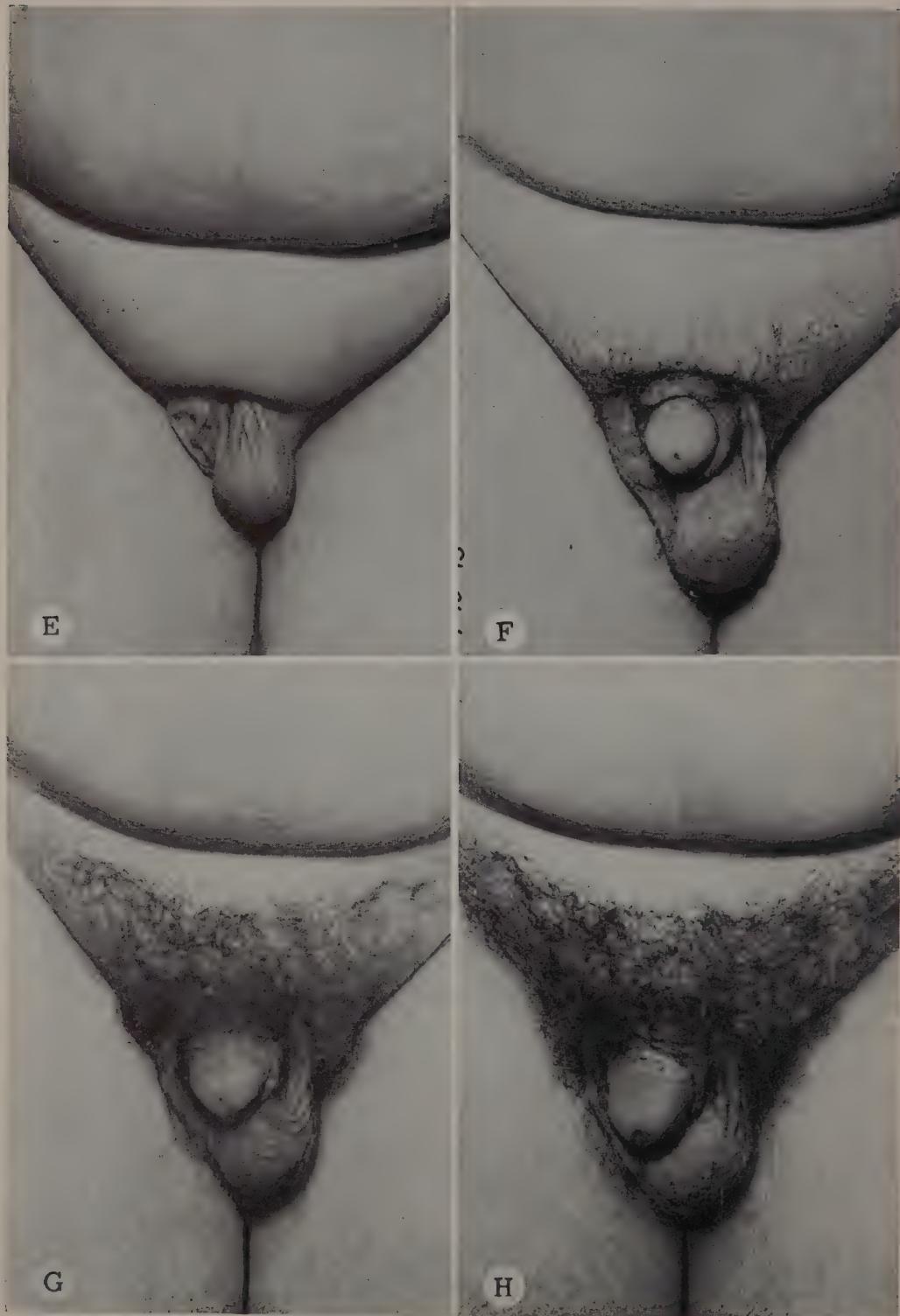
We have already discussed the ANATOMIC LESIONS found in the hypophyses of patients with anterior-lobe deficiency (see : pages 262 to 264). These destructive lesions interfere with the normal production of anterior-pituitary hormones and consequently cause derangements in metabolism as well as inhibition of growth, thyroid, adrenal-cortical and gonadal development.

It is especially important to remember, however, that many types of anterior-lobe deficiency are due to purely FUNCTIONAL CAUSES. Among these the repeatedly mentioned "shift in anterior-lobe-hormone production" is

especially important. It is apparently due to an adaptive readjustment of hypophyseal-hormone secretion during periods of stress. Under such conditions somatotrophin, gonadotrophin and prolactin production are diminished, presumably in order to permit maximal elaboration of the vitally needed corticotrophin. The severe cachexia and hypogenitalism of anorexia nervosa, often accompanied by a decrease in the B.M.R., probably also represents a special type of this protective adaptation phenomenon. Clinical evidence of pituitary failure is demonstrable in most cases of cachexia, regardless of its cause, presumably because the resulting decrease in metabolic and genital functions is advantageous under such conditions.

Obesity and testicular failure (adiposogenital dystrophy) without other signs of pituitary failure. — A. Age 14 years, sella turcica normal, visual fields normal, height 68", weight 209 lbs. Penis not visible, right testis small pea size, left testis pecan size. 17-KS: 2.2 mg./24 hrs. Urinary FSH, repeatedly over 105 MU/24 hrs. — B, C and D. Continued improvement on chorionic gonadotrophin (LH) 500 I.U. to 750 I.U. 3 times weekly. (B after 9 months, C after 13 months and D after 21 months of treatment). (Cont'd)





(See: p. 271 for legend.)

In adiposogenital dystrophy and in the Laurence-Moon-Biedl syndrome, the hypogenitalism is probably due to a more or less selective failure of gonadotrophin production caused by lesions in the "sexual centers" of the hypothalamus.

CLINICAL COURSE

State. — The most characteristic features of SIMMONDS' DISEASE are: loss of hair (particularly in the pubic and axillary regions), loss of teeth, trophic changes in the nails and skin (the latter is conductive to "geroderma" or "progeria" that is, the appearance of premature senility), atrophy of the genital organs (uterus, breasts, testes), amenorrhea, sterility, loss of libido, impotence, muscular weakness; a decrease in the pulse rate, blood pressure, body temperature, B.M.R. and fasting-blood sugar, mental apathy and greatly diminished sensations of hunger and thirst. Gradually profound cachexia may develop, but contrary to common opinion, this is a late manifestation of anterior-lobe insufficiency. The course of the disease is usually very slow and cases have been observed in which death

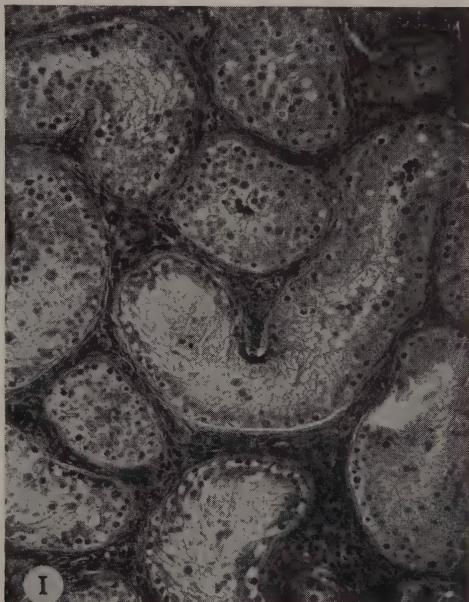
did not occur until 20 years, or more, after the onset of the first symptoms.

In PITUITARY DWARFISM body growth is inhibited due to a delay in the ossification of the junction cartilages. The various parts of the body retain their infantile proportions. The voice and body hair are child-like. Due to intense wrinkling of the skin, the facial expression of adult pituitary dwarfs is often suggestive of premature senility. The gonads and secondary sexual characteristics fail to develop, but cachexia is usually absent and the B.M.R. may remain essentially normal. Pituitary dwarfism is often associated with adiposity, diabetes insipidus or both these conditions, perhaps because of simultaneous lesions to the hypothalamic region. The mental development of these "midgets" is normal and frequently they are even unusually intelligent and industrious. This is remarkable in view of their secondary thyroid and adrenal insufficiency.

ADIPOSOGENITAL DYSTROPHY is actually a combination of adiposity and genital dystrophy. In certain cases these two cardinal manifestations may appear at different times, thus indicat-

— E. Close-up of sex organs in Fig. A. — F. Close-up of sex organs in Fig. B. — G. Close-up of sex organs in Fig. C. — H. Close-up of sex organs in Fig. D. — I. After 5 months' treatment with approximately 500 I.U. of LH three times a week, most tubules are fairly well-developed, although spermatogenesis is not yet detectable. The Leydig cells begin to proliferate.

(Courtesy of Dr. E. P. McCullagh.)

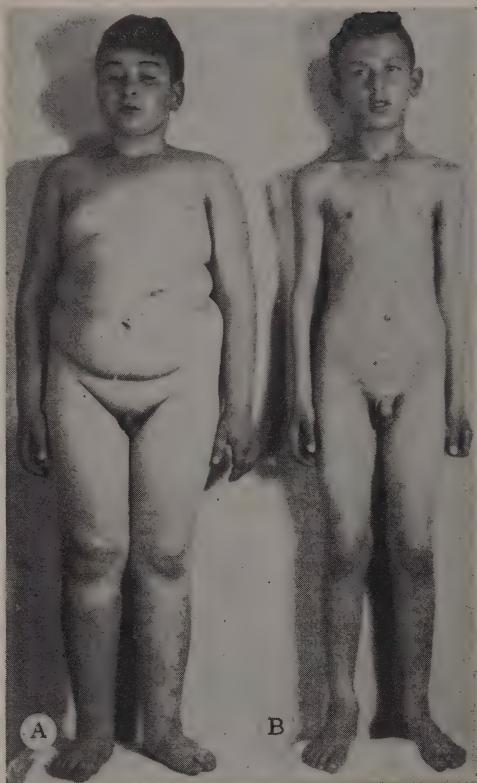




Obesity. Age 15 years, height 67½", weight 231 lbs. (ideal weight: 135 lbs.), sella normal by X-ray, visual fields normal, genital development entirely normal, urinary FSH normal. The patient is shown as an example of a type frequently mistaken for Fröhlich's syndrome or adiposogenital syndrome.

(Courtesy of Dr. E. P. McCullagh.)

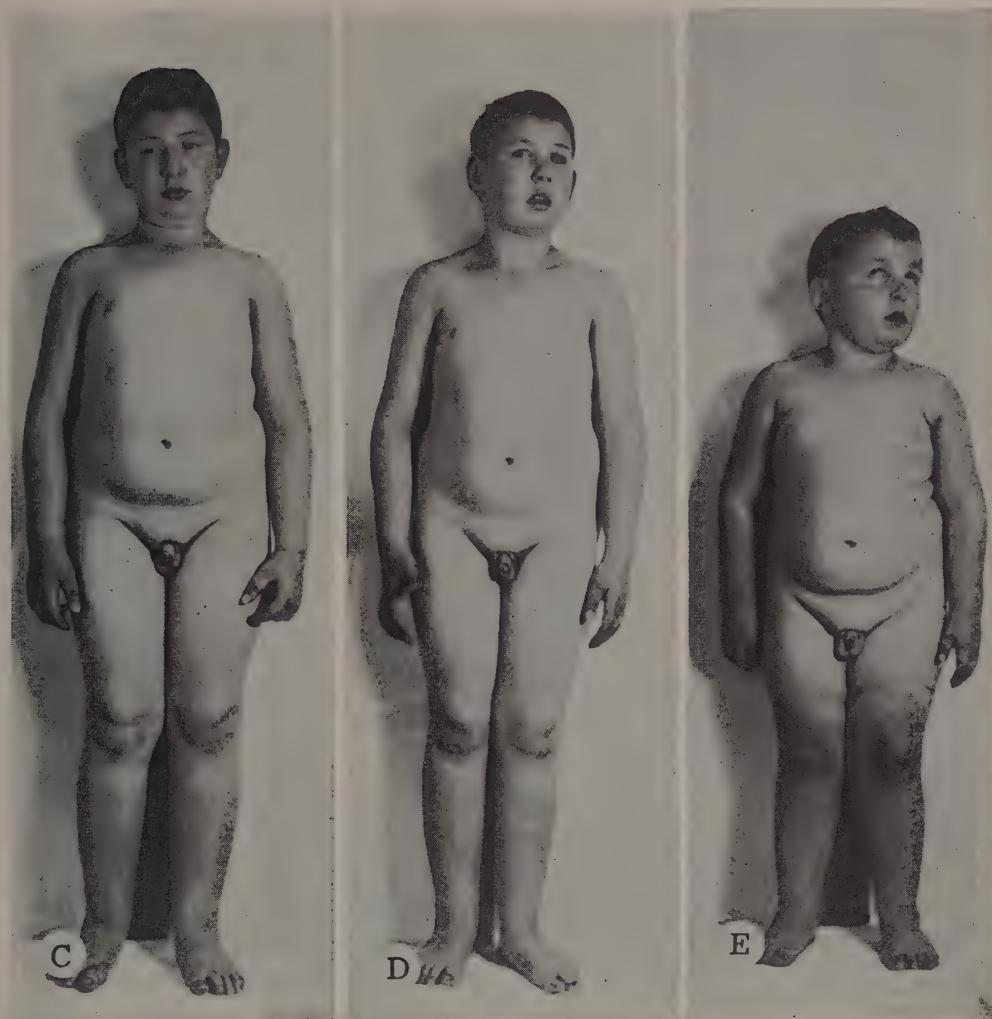
ing their relative independence of each other. The gonads and secondary sex characteristics are undeveloped, the skin is delicate and often unusually white. The patients may be mentally retarded but this is not always the case, although they are usually apathetic, lazy and sexually indifferent. Adiposogenital



(Cont'd on p. 273.)

dystrophy is sometimes associated with dwarfism and diabetes insipidus, presumably due to accompanying lesions of the anterior and posterior-lobe respectively, but in some cases growth is normal or even excessive.

The LAURENCE-MOON-BIEDL SYNDROME is a rare type of adiposogenital dystrophy, characterized by: adiposity, genital dystrophy, retardation of mental development, skull deformities, and congenital malformations such as atresia of the anus, polydactyly (formation of supernumerary fingers or toes) and retinitis pigmentosa. The syndrome is hereditary, often affecting several members of the same family. Only about 90 cases have so far been described and in the absence of adequate autopsy reports the underlying pathology is not known. It is probable that a congenital malformation of the hypothalamic region may be the cause of the adiposity



Familial occurrence of Laurence-Moon-Biedl syndrome. — A, B, C, D and E. Laurence-Moon-Biedl Syndrome in four of five brothers. The second boy (B) is entirely normal. The parents are first cousins. (Courtesy of Dr. E. P. McCullagh.)

and the genital dystrophy. There is no radiologic evidence of a pituitary lesion in these patients.

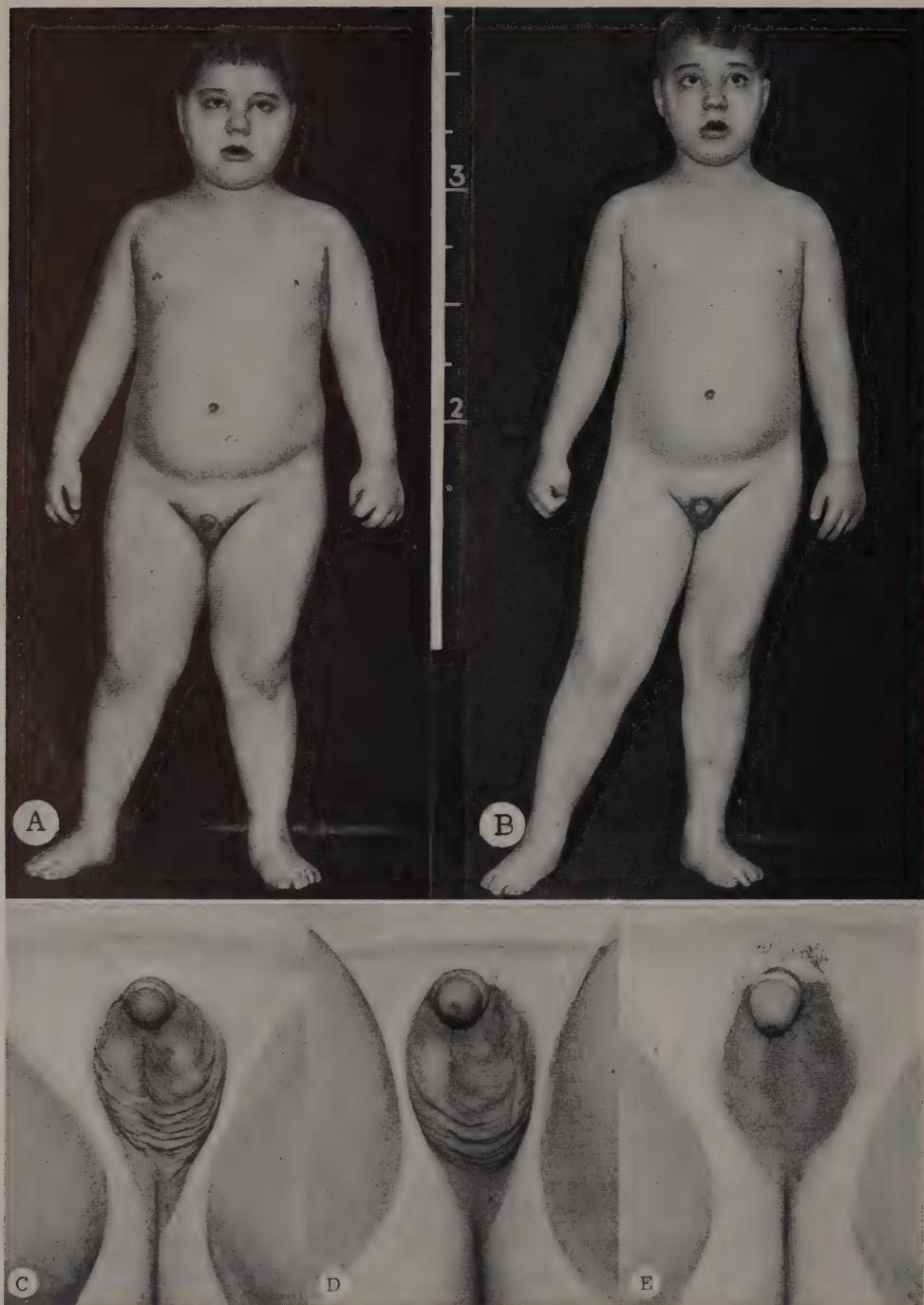
In the case of SELECTIVE FAILURE OF CERTAIN ANTERIOR-LOBE FUNCTIONS there may be severe cachexia or no change in the general appearance of the patient. The latter is frequently the case in menstrual disturbances of the "pituitary type" or sterility due to decreased gonadotrophin production.

Metabolism. — All types of anterior-lobe deficiency tend to cause some

(usually slight) decrease in the BODY TEMPERATURE.

The B.M.R. is always subnormal in Simmonds' disease and in many severe cases it was found to be even lower than in grave hypothyroidism. Figures as low as — 50% have been recorded.

In *pituitary dwarfism* the drop in B.M.R. is rarely severe, but the evaluation of pertinent data is difficult. It would not be rational to compare the B.M.R. of an adult pituitary dwarf with that of a child of equal size, nor



Laurence-Moon-Biedl Syndrome. — A, B, C, D and E. 5-year-old boy with obesity, mental retardation, polydactyly and retarded scrotal pigmentation. Penile growth produced by local methyl-testosterone ointment (10 mg. daily for 3 months). (Cont'd. on p. 275.)

(Courtesy of Dr. E. P. McCullagh.)



Note scars from removal of supernumerary fingers; patient shown on p. 274.

can one obtain standard normal values by extrapolation from healthy adult individuals taking weight or surface as a basis for comparison.

In *adiposogenital dystrophy* there is no constant diminution of the B.M.R.

Anterior-lobe deficiency also causes pronounced abnormalities of CARBOHYDRATE METABOLISM. A tendency towards pronounced fasting-hypoglycemia and a great sensitivity to insulin have been noted in Simmonds' disease and in pituitary dwarfism. In adiposogenital dystrophy this is not regularly present and indeed some patients with Fröhlich's syndrome exhibit a relative insensitivity to the hypoglycemic effects of insulin and fasting.

Glucose tolerance is usually significantly increased in Simmonds' disease and pituitary dwarfism, less constantly in adiposogenital dystrophy.

FAT METABOLISM is not specifically influenced by anterior-lobe insufficiency, although in Simmonds' disease loss of fat reserves is very characteristic of the late cachectic phase. Adiposogenital dystrophy causes considerable excess storage of fat; this is probably conditioned by an increased appetite combined with the apathy and laziness char-



Laurence-Moon-Biedl's Syndrome. Note characteristic polydactyly (six toes on each foot).
(Courtesy of Dr. A. Pinto Viégas.)



Hypothalamic obesity. 44-year-old man, height 77", weight 330 lbs., suffers from extreme drowsiness (patient fell asleep while this picture was taken), marked thirst (fluid intake about 5 L./day), complete impotence, sella normal to X-ray, glucose tolerance of diabetic type, urinary testoids normal, B.M.R.: +24% (?).

(Courtesy of Dr. E. P. McCullagh.)

acteristic of these patients. Pituitary dwarfs may have normal or excessive fat depots and rarely exhibit any severe cachexia. The blood cholesterol is usually normal.

The NITROGEN BALANCE becomes markedly negative in the late stages of Simmonds' cachexia, but the N.P.N. and blood protein concentration remain essentially normal in the various types of anterior-lobe failure.

The excretion of SALT is often delayed in Simmonds' disease, as judged by chloride tolerance tests. In patients with diabetes insipidus the subsequent development of Simmonds' disease — which may follow upon secondary in-

vovement of the anterior-lobe — cures the polyuria and polydipsia.

Growth and Bones. — In PITUITARY DWARFISM the growth of the skeleton, the appearance of ossification centers and the second dentition are greatly delayed. Union of the junction cartilages may not occur until adulthood so that some growth may be expected from somatotrophin therapy, even if it



Hypothalamic obesity. 16 years ago, patient suffered a head injury, followed by polydipsia, polyphagia, somnolence and increase in weight from 175 to 315 lbs. He had one 17-year-old son, but now has complete aspermia and impotence. His glucose tolerance is of the diabetic type, urinary FSH: 105-212 M.U./24 hrs., 17-KS: 5.2 mg./24 hrs., B.M.R.: —10%. There is no evidence of pituitary tumor.

(Courtesy of Dr. E. P. McCullagh.)

is instituted at an age when normal growth would no longer occur. (For relationship between pituitary and other types of dwarfism see : "Diagnosis," pp. 286, 287.)

In SIMMONDS' DISEASE there is usually some osteoporosis and a tendency to lose otherwise apparently normal teeth. The latter is followed by atrophy of the jaws.

ADIPOSOGENITAL DYSTROPHY may be accompanied by dwarfism, but sometimes growth in length is actually excessive. Quite frequently Fröhlich's syndrome is associated with flat feet, coxa vara and genu valgum, and patients with (pituitary?) adiposity tend to show sclerosis of the calvarium with hypoplasia of the paranasal sinuses. (See : "Metabolic craniopathy," p. 287.)

Blood. — Anemia is a constant finding in Simmonds' disease. The color index is usually less than one, and the hemoglobin averages 50%. In some instances, there is eosinophilia and lymphocytosis, but the latter are inconstant.

Cardiovascular System. — Hypoplasia of the cardiovascular system and a low blood pressure are characteristic of Simmonds' disease. In pituitary dwarfism, the blood pressure may be normal and diabetes insipidus, or Fröhlich's disease may even occur in combination with hypertension (influence upon vasomotor centers of hypothalamus?). In Simmonds', as in Addison's disease, exposure to stress fails to elicit the usual pressor reaction and generally tends to cause an inverse response. Thus, even slight muscular exercise may cause a significant drop in blood pressure. This is presumably due to secondary adrenal-cortical insufficiency, since it can be prevented by suitable corticoid therapy.

Lymphatic System. — The striking hyperplasia of the thymico-lymphatic apparatus so characteristic of Addison's disease, is rarely prominent in anterior-pituitary failure. "Lymphatism" is often



Pituitary dwarfism. 16-year-old boy with deficient sexual development and growth. Height 119 cm. (48"), bone age corresponding to 9th year, sella normal to X-rays, no 17-KS in urine. 100 mg. of testosterone propionate weekly, during six months, caused no increase in growth rate in this case.

(Courtesy of Dr. A. B. de Ulhôa Cintra.)



Dwarfism with hypogonadism. — A. Patient at 18 years of age before institution of therapy (height 49"). — B. Patient at 26 years of age. He received one year's treatment with a growth-hormone preparation, but then was left without therapy 7 years (height 53 1/4"). Methyl-testosterone therapy instituted in doses of 10 mg./day orally. — C. Patient at 29 years of age. Methyl-testosterone therapy was continued until 9 months before this picture was taken. After this, therapy was changed to testosterone propionate in doses of 25 mg. three times a week intramuscularly. Height now 56". Note also maturation of facial expression, genital and muscular development, approximating those of normal adult males.

(Courtesy of Dr. E. P. McCullagh.)



Pituitary dwarfism. On the left, a normal boy, age 15 years, weight 53 Kg. (103 lbs.), height 154 cm. (61 inches). On the right, 15-year-old boy, with anterior-pituitary deficiency. Weight: 19 Kg. (46 lbs), height 114 cm. (46 inches). Note also deficient development of the penis and lack of pubic hair. The measurements of this patient roughly correspond to those of a normal boy, age 5 1/2-6 years. (Courtesy of Dr. A. Pinto Viégas.)

conspicuous, however, with adiposogenital dystrophy.

Muscles. — The muscular system is usually atrophic, and muscular strength is considerably below normal in Simmonds' disease; to a lesser extent, this is also true of adiposogenital dystrophy and pituitary dwarfism.

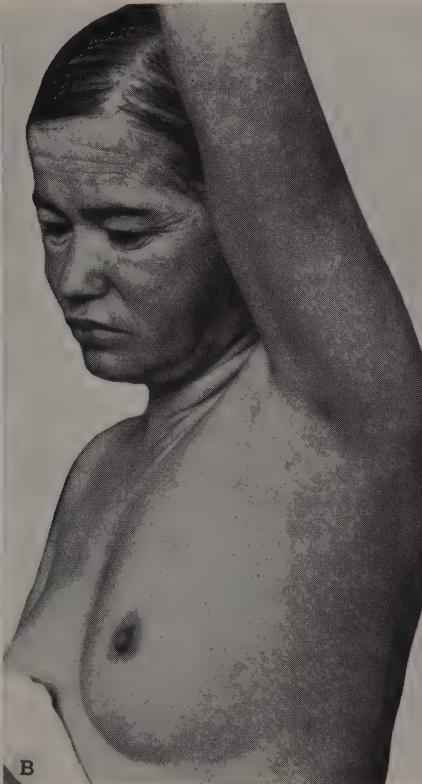
Nervous System and Sense Organs.

— In SIMMONDS' DISEASE, loss of libido, apathy and depression are common, while PITUITARY DWARFS are usually alert, intelligent people whose only manifestations of mental immaturity are those resulting directly from the deficient sexual development. The lack of libido and impotence may lead to serious psychologic disturbances, which are frequently aggravated by the tendency of other people to ridicule these dwarfs because of their small size and because of their sexual deficiency. In an effort

to compensate for the resulting inferiority complexes, pituitary dwarfs frequently marry, usually among themselves, although of course they are always sterile and almost invariably, sexually indifferent.

The indolence characteristic of ADIPOSOGENITAL DYSTROPHY and the often severe mental defects which accompany the LAURENCE-MOON-BIEDL SYNDROME have already been mentioned.

Visual disturbances are very characteristic of all hypopituitary conditions in which a disease process exerts pressure upon the optic chiasm. Usually there is bitemporal hemianopsia which sometimes develops from a central scotoma and often ends in complete blindness. Similar visual defects are seen in association with the tumors which give rise to hyperpituitarism. (See : p. 283.)

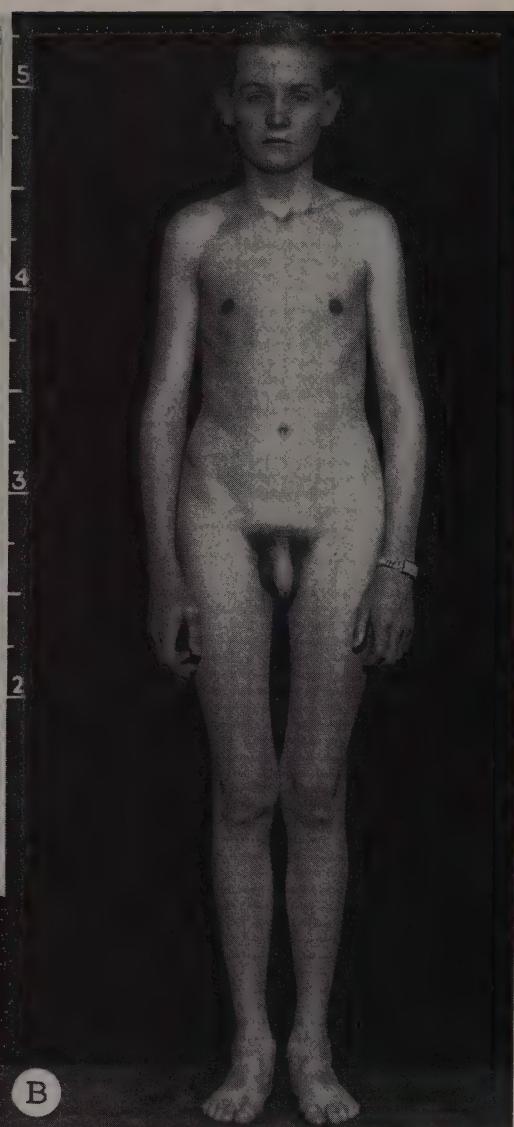


Simmonds' disease. — A. and B. 37-year-old woman who developed amenorrhea, loss of pubic and axillary hair, loss of weight and weakness immediately after her fourth delivery. She had a low B.M.R., low serum Na and Cl, high serum K, positive Robinson, Power and Kepler test, high blood cholesterol, urinary gonadotrophins and 17-KS close to 0.

(Courtesy of Dr. A. B. de Ulhôa Cintra.)



A



B



C



D

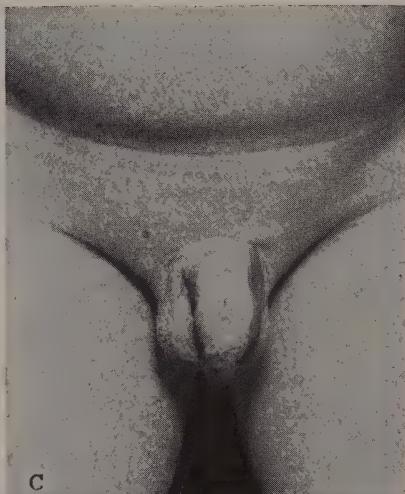
Severe anterior-pituitary deficiency with pituitary adenoma. — A. 16-year-old boy with emaciation, pallor, arterial hypertension (90/75), dry skin, mild polydipsia and polyuria, slight enlargement of the sella. B.M.R.: -38%. 17-KS: 0.7 mg./24 hrs., urinary FSH: 6 M.U./24 hrs. — B. Two months following removal of pituitary adenoma. X-Ray treatment to pituitary (2,000 r.u. to each temple), and 500 I.U. of LH three times weekly. Genital and somatic development greatly improved. — C. Testis biopsy before treatment. Note marked hyalinization of tubules and almost complete disappearance of tubular and interstitial cells. — D. Testis biopsy after therapy. Note proliferation of both tubular and Leydig cells; spermatogenesis not yet in progress.
(Courtesy of Drs. R. W. Schneider and E. P. McCullagh.)



A



B



C



D

Hypopituitarism. — A. — B. — C. 44-year-old patient with signs of gonadal and adrenal deficiency. Complaints were physical and mental fatigue. Note immature facies, distinct evidence of hypotestostroidism, present since before puberty. Blood pressure: 90/70, water excretion test (Robinson, Power and Kepler): 12.8, increased insulin sensitivity. Patient responded well to testosterone + desoxycorticosterone therapy. — D. Skull X-ray shows marked enlargement of sella with thinning of its walls and partial erosion of the clinoid processes. There is evidence of calcification, probably in the pituitary tumor or cyst wall.

(Courtesy of Dr. E. P. McCullagh.)

Digestive System. — Deficient secretion of gastric and pancreatic juice are rather characteristic of Simmonds' disease and subnormal development of all the abdominal organs forms part of the typical "splanchnomimicria" of anterior-lobe failure.

Skin. — The loss of hair, especially the loss of axillary and pubic hair is extremely characteristic of Simmonds' disease. Sometimes it is accompanied by loss of eyebrows. Histologically, the skin reveals atrophy of both hair follicles and sweat glands. Cutaneous pigmentation is comparatively rare in Simmonds' disease; when it occurs, differentiation from Addison's disease may become very difficult.

In adiposogenital dystrophy, absence of body hair is likewise characteristic, especially if the condition develops before puberty.

Accessory Sex Organs. — Atrophy or hypoplasia of the accessory sex organs is a constant characteristic of Simmonds' disease, pituitary dwarfism and adiposogenital dystrophy. In women there is complete amenorrhea, the uterus, vagina and breasts are atrophic and in the ovaries there is no follicle maturation, ovulation or corpus luteum formation. In men, there is atrophy of the testes and seminal vesicles, sometimes accompanied by complete absence of the prostate. Sterility in both sexes is a constant characteristic of all fully-developed cases of Simmonds' disease, pituitary dwarfism and adiposogenital dystrophy. (For relationships between pituitary and other types of genital infantilism see: "Diagnosis," below.)

Pregnancy and Lactation. — As previously stated, Simmonds' disease often occurs as a sequel to pregnancies which are followed by complicated deliveries. Since complete anterior-lobe failure is always conducive to sterility in both sexes, pregnancy and lactation do not occur in fully-developed cases of Simmonds' disease, pituitary dwarfism

or adiposogenital dystrophy. The alleged instances of fertility among (male or female) pituitary dwarfs are due to confusion with "primordial" dwarfism or with achondroplasia (see pp. 286, 287).

COMPLICATIONS

One of the most important complications of severe anterior-lobe deficiency is fatal hypoglycemia due to prolonged fasting or exposure to stress. Slight intercurrent diseases may elicit unduly serious complications or even death in these patients whose adaptive mechanism is severely deranged.

Other complications may be due to the causative pituitary lesion itself (e.g., visual disturbances, hemorrhages from pituitary tumors, increased intra-cranial pressure or invasion of the adjacent hypothalamic nuclei by hypophyseal growths).

DIAGNOSIS

Manifestations Characteristic of Pituitary Lesions in General. — The so-called "pituitary headache" is usually most intense between the temples, deep behind the eyes and is almost invariably due to the pressure of a pituitary growth.

Other characteristic local signs are the bitemporal hemianopsia and radiologic evidence of a lesion in the pituitary region (erosion or unusual smallness of the sella, calcification in a suprasellar tumor). Signs of marked intra-cranial pressure develop only in the presence of extensive pituitary growths.

Simmonds' Disease. — Fully-developed cases of Simmonds' disease are readily recognized on the basis of the above-mentioned characteristic manifestations. On the other hand, the diagnosis of incipient or very mild instances of anterior-pituitary failure may be extremely difficult and usually requires a great deal of clinical experience. In such instances, the peculiar waxy color of the skin, the sparse beard, genital and

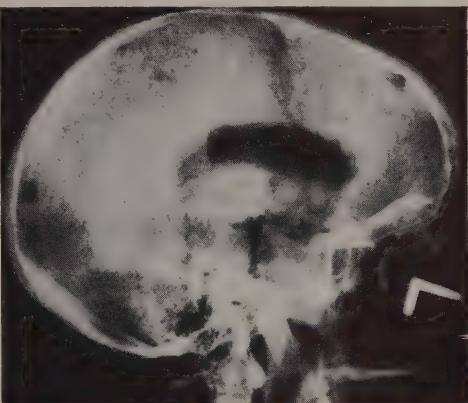
axillary hair development in conjunction with the deficient sexual development are of great diagnostic value. A very pronounced decrease in urinary 17-KS, folliculoids, corticoids and gonadotrophins is likewise typical of anterior-lobe failure.

The great insulin-sensitivity of patients with Simmonds' disease has also been recommended as a diagnostic measure. However, it is dangerous, since fatal hypoglycemia may ensue after very small doses, and it is not characteristic, since in Addison's disease, anorexia nervosa, hepatic disease and many other types of cachexia, insulin-sensitivity is likewise increased.

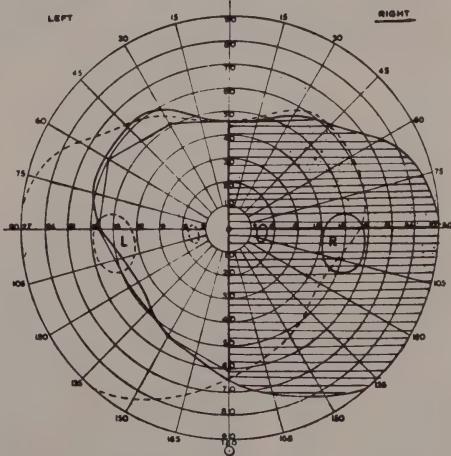
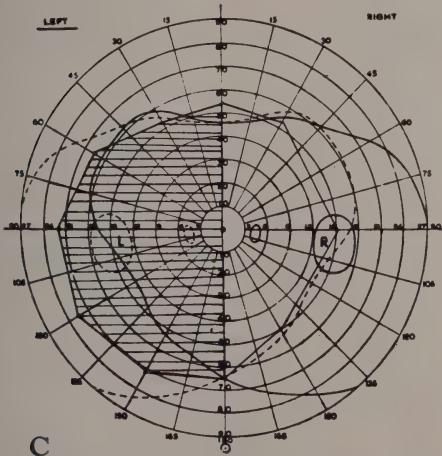
Differentiation from ANOREXIA NERVOSA is particularly difficult. In the latter condition the fundamental cause of the symptoms is a secondary hypopituitarism and hence the manifestations are essentially the same as in Simmonds' disease. However, in anorexia nervosa, psychogenic factors are the cause of the decreased food intake and the resultant cachexia. Correspondingly, the patients are amenable to adequate psychotherapy, in conjunction with an increased



A



B



Pneumo-ventriculogram in a patient with pituitary tumor. — A. Appearance of the cerebral ventricles after air insufflation in a normal individual. Note normal position of the (dark) lateral ventricles visualized by air insufflation; also normal appearance of sella. — B. 52-year-old woman with large chromophobe adenoma of the pituitary; there was bilateral hemianopsia with pronounced loss of vision and severe headaches. Note that the tumor caused obliteration of the interpeduncular cistern as well as great erosion of the sella with complete disappearance of the posterior clinoid processes. — C. Visual fields of patient shown in Fig. B. Vision was lost or impaired in shaded territory.

(Courtesy of Montreal Neurological Institute.)



Anorexia nervosa. — A. and B. 60-year-old woman with severe emaciation due to anorexia. Mental depression, loss of axillary and pubic hair, inexpressive face. 17-KS excretion normal. Marked improvement and gain in weight was accomplished with 10 mg. of methyl-testosterone/day. (Courtesy of Dr. E. B. del Castillo.)

dietary intake and thyroid administration to raise the B.M.R. Edema of the legs (unrelated to the level of the serum proteins) and a great delay in the excretion of orally-administered water are likewise characteristic. There is amenorrhea of the hypopituitary type that is unaccompanied by menopausal disturbances. The urinary excretion of 17-KS is low, but not as low as in Simmonds' disease where it is practically nil.

Confusion with MYXEDEMA may occur because of the pallor, intolerance to cold, decreased B.M.R. and apathy charac-

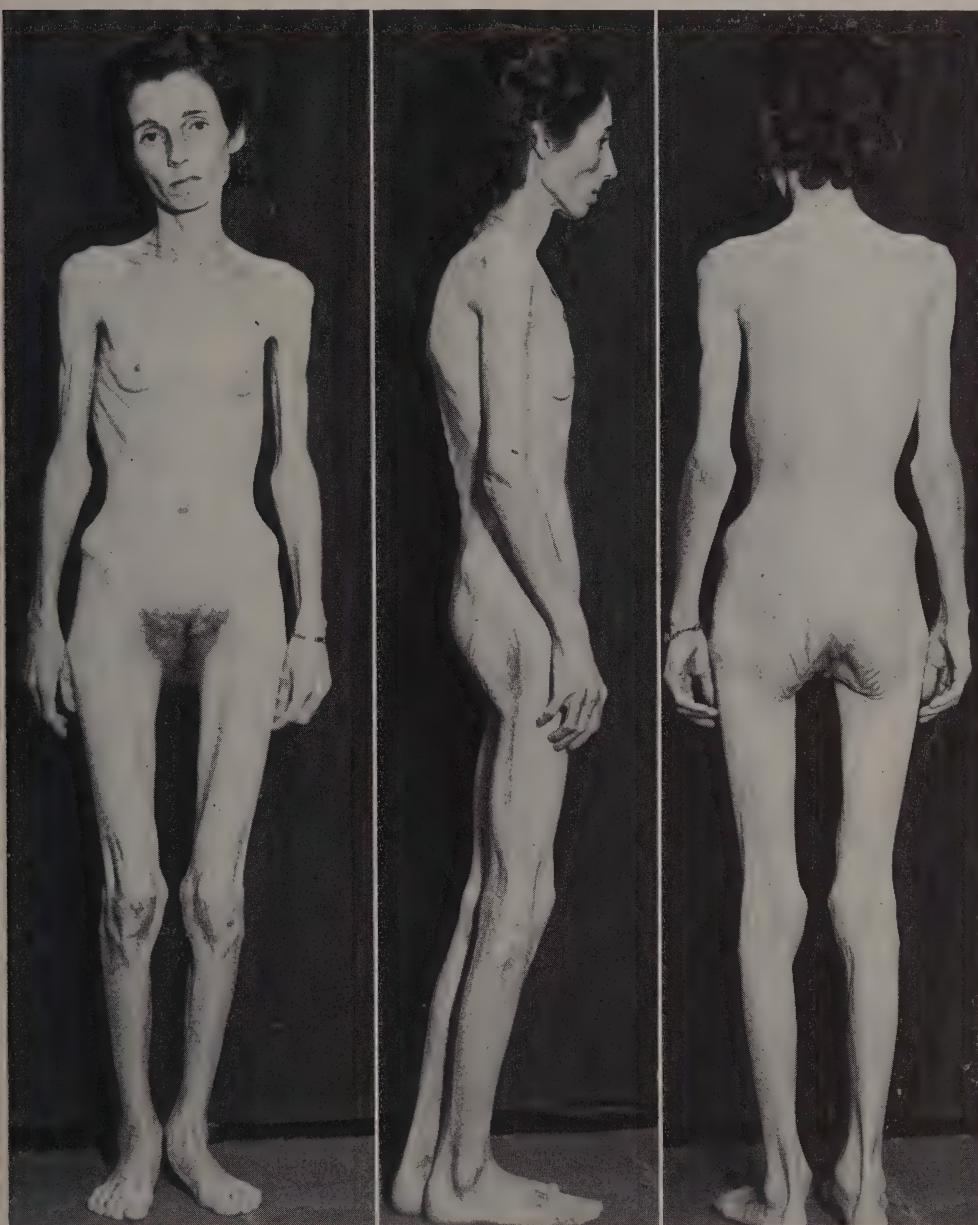
teristic of both these conditions. However, in hypothyroidism, the skin is coarse and dry and there is no great loss of beard, pubic or axillary hair. Hypercholesterolemia and myxedematosus infiltrations are absent in most cases of Simmonds' disease.

Differentiation from ADDISON'S DISEASE may be impossible in the absence of local signs in the pituitary or adrenal region. However, pigmentation of the skin is exceptional in Simmonds' disease and the blood-electrolyte changes, characteristic of cortical insufficiency if present, are much less pronounced.

Differentiation from PERNICIOUS ANEMIA is rarely difficult if this condition is kept in mind.

Pituitary Dwarfism. — At birth, patients with pituitary dwarfism are usually of normal size and appearance, but

later they show an inhibition in development which is inversely proportional to the age at which their anterior-lobe became deficient. The diagnosis is rarely difficult on the basis of the characteristic hormone-deficiency manifestations



Anorexia nervosa. Age 34 years, height 60", weight 59 lbs. X-ray of sella, chest, stomach, duodenum and colon normal; B.M.R.: -20%; Ewald meal and blood counts normal; water excretion tests normal; insulin tolerance low, glucose tolerance curves flat, as in hypopituitarism. Note great resemblance to Simmonds' disease.

(Courtesy of Dr. E. P. McCullagh.)

(see above) and the local signs in the pituitary region; the latter, however, are not always demonstrable.

From a differential diagnostic viewpoint, it is important to distinguish pituitary dwarfism from other types of growth inhibition and delayed psychic and genital maturation.

The term "INFANTILISM" has been used to designate conditions in which the child-like characteristics of body and psyche persist during adult life. It is well to remember however, that there is no disease which leads to the persistence of perfectly normal child-like somatic and psychic characteristics. Only some of the individual's features are truly child-like, hence perfect instances of "infantilism" do not exist.

In view of the very voluminous and confusing pertinent literature, the subject deserves some discussion. Certain authors placed special emphasis upon the retardation of genital development, but if this were taken as a sign of infantilism, adiposogenital dystrophy, eunuchoidism, eunuchism and even acromegaly (when accompanied by genital dystrophy) would have to be classified under this heading. Others take the stunting of somatic growth as the chief criterion, but in this sense all types of dwarfism (e.g., primordial dwarfism, renal rickets, and perhaps even achondroplasia would have to be classified under the same heading).

Even if we limit ourselves to those types of "infantilism" in which both the general somatic and sexual development are delayed, the following types must be recognized :

(1) *Pituitary infantilism* (Synonyms: nanosomia infantilis, Lorain-Levi syndrome), which has been described under a variety of names, depending upon the comparative conspicuousness of certain somatic or psychic manifestations. It is now generally accepted that all these types are due to a primary anterior-lobe failure.

(2) *Brissaud's infantilism* (Synonym : hypothyroid infantilism), which

is identical with hypothyroid cretinism. The mental and somatic development are retarded, but sexual maturation is less markedly impeded.

(3) *Herter's infantilism* (Synonym: intestinal infantilism) with retardation of somatic and sexual development due to some chronic gastrointestinal disease.

(4) *Paltauf's infantilism* is essentially identical with "status thymico-lymphaticus" accompanied by sexual retardation.

(5) *Pancreatic infantilism*, in which the retardation of somatic and sexual development are due to pancreatic disease.

(6) *Renal infantilism* or renal rickets with stunted somatic growth and sex development.

(7) *Other types of infantilism* associated with hereditary syphilis, malaria, tuberculosis, various types of intoxication (lead, mercury), malnutrition, etc. In all these cases, secondary pituitary failure is probably the causative agent. Just as anorexia nervosa may produce secondary anterior-lobe deficiency in adults, thus, non-specific stress can cause hypopituitarism with "infantilism" if it elicits a general-adaptation-syndrome during childhood.

For purposes of classification and differential diagnosis it suffices to distinguish the primary pituitary dwarfism (Lorain-Levi type) from secondary anterior-lobe failure due to a variety of stresses. The former can be differentiated from the latter by the absence of manifestations indicative of the specific diseases which tend to produce secondary anterior-lobe failure (e.g., hypothyroidism, infections, intoxications, and renal, cardiac or pancreatic disease).

All the above-mentioned types of "infantilism" are associated with growth inhibition, but there are other types of DWARFISM which differ from "infantilism" inasmuch as sexual development is not impeded. Among these we may mention :

(1) *Primordial dwarfism* (Synonyms : nanosomia essentialis, primary

dwarfism, pygmyism) which differs from pituitary dwarfism in that the individual is abnormally small even at birth (since the condition is apparently due to a congenital deficiency) while pituitary dwarfism develops later in life. The genital organs of primordial dwarfs are essentially normal, the body proportions of the adult type and the ossification of the function cartilages occurs at the normal age. Their psychic development is also normal even with respect to the sexual impulses and they are capable of normal reproduction, frequently transmitting the dwarfism to their children.

(2) *Achondroplasia* (Synonym : chondrodystrophia foetalis) is a congenital, sometimes hereditary, disease. It is presumably due to a disturbance in endochondral ossification — especially of the long bones and the base of the skull — caused by connective tissue invasion from the periosteum into the growth cartilages. These individuals are dwarfs only because their extremities are too short. Their trunk and head is of normal size, but due to the disturbance of bone growth at the base of the skull, the nose is flat and the lower jaw comparatively prominent. The hands are stubby and the fingers of equal length. The sex organs are normal and indeed, often precociously developed. The intellect is not affected.

The differentiation of these two types from pituitary dwarfism is simple, since unlike the latter they are not accompanied by genital dystrophy.

Adiposogenital Dystrophy. — It may be extremely difficult, or even impossible to differentiate adiposogenital dystrophy from EUNUCHOIDISM due to primary gonadal diseases, unless local signs call attention to the presence of a lesion in the pituitary or gonadal region. If the disease is due to some hypothalamic injury, an accompanying diabetes insipidus may help the diagnosis since other types of hypogonadism are not accompanied by polyuria.



Achondroplasia. Achondroplasia in a 16-year-old girl with amenorrhea. All epiphyseal centers about the elbow and hand are fused, but those at the wrist are not yet completely ossified. Unlike in pituitary dwarfism, the growth is disproportionate (the extremities being far too short for the trunk and head); sexual development is normal. (Courtesy of Dr. E. P. McCullagh.)

Confusion with the so-called METABOLIC CRANIOPATHY (Morgagni's or Stewart-Morel's syndrome) is likewise possible. This is a comparatively common, often familial, disease among middle-aged women. It is characterized by obesity, hypertension, secondary amenorrhea, hirsutism, neuropsychiatric disturbances and cancellous bone deposition on the lamina interna of the skull. It may be due to a hypothalamic disorder.

PROGNOSIS

The prognosis of SIMMONDS' DISEASE is extremely grave. However, patients may survive, in a condition of severe anterior-lobe failure for several years

and even decades if they are protected against all types of stress and if they can be persuaded to take adequate nourishment and therapy.

PITUITARY DWARFISM shows no tendency towards spontaneous improvement, but is compatible with a normal, and even a very long, active life. One such patient died at the age of 91 years. The fact that many pituitary dwarfs die early is merely due to their lack of resistance to such intercurrent stresses as infections, intoxications, etc.

ADIPOSOGENITAL DYSTROPHY due to anatomic lesions in the hypophyseohypothalamic region, and the LAURENCE-MOON-BIEDL'S SYNDROME likewise fail to regress spontaneously, but considerable improvement or even cures may be obtained in the former condition upon removal of the causative lesion (e.g., a craniopharyngioma). Flaccid, fat boys with "Fröhlich's Syndrome" often revert to normal at puberty without any treatment, but in such cases the causative derangement is apparently only functional.

THERAPY

Simmonds' Disease. — The logical therapy of Simmonds' disease would be substitution therapy with ANTERIOR-PITUITARY-HORMONES. Unfortunately, pure, active preparations of this type are not yet available in adequate amounts to make such treatment generally applicable. Treatment with TESTOIDS (to induce protein anabolism) or CORTICOIDS (to compensate for the secondary cortical insufficiency) has often been successful. THYROID therapy (to compensate for the secondary hypothyroidism) is dangerous, since hypopituitary patients are extremely sensitive to thyroid-hormone overdosage.

POSTERIOR-LOBE HYPOFUNCTION (DIABETES INSIPIDUS)

DEFINITION

In principle, posterior-lobe hypofunction would be a condition in which

Under present conditions, the most important therapeutic measures are: protection of the patient from any intercurrent disease or other types of stress (e.g., cold, emotional upset, infections), since these are likely to elicit a fatal hypopituitary crisis due to lack of adaptability.

Pituitary Dwarfism. — In pituitary dwarfism the therapy is essentially the same as in Simmonds' disease. ANTERIOR-LOBE EXTRACT or TESTOSTERONE administration has been recommended by some, not only to cause nitrogen retention and accelerate growth, but also to develop the sex organs. It is doubtful, however, whether the artificial induction of sexual maturity is desirable in these individuals. The awakening of a dormant libido may cause serious psychologic disturbances in dwarfs whose libido is rarely reciprocated.

Adiposogenital Dystrophy. — If the disease is due to a specific and anatomic lesion, such as a craniopharyngioma, SURGICAL removal of the tumor is necessary, especially if there is any danger of pressure upon the optic chiasm. Otherwise, treatment with pituitary GONADOTROPHINS or with TESTOSTERONE in males and with FOLLICULOIDS in females may be useful, especially if combined with reduction of food intake and with muscular exercise to cause loss of body weight. It is important to keep in mind however, that many adipose boys with genu valgum, flat feet and the "Fröhlich type of habitus" do not suffer from organic, hypothalamic or pituitary lesions, as their sex organs are actually normal, though hidden in the adipose tissue. In these cases, dietary restrictions and exercise alone may suffice to improve the condition; indeed at the time of puberty, such patients often become normal without any treatment.

the hormone production of the posterior-lobe is sufficiently disturbed to cause detectable manifestations of insufficiency.

In practice, only diabetes insipidus has been proven to result from such a derangement, although certain dystocias (difficult child-births) have been ascribed to uterine inertia due to a deficient oxytocin production.

CLASSIFICATION

As in the case of other endocrine diseases, diabetes insipidus may be classified according to various criteria; from a CLINICAL view-point it is most customary however, to distinguish :

(1) *Primary or idiopathic diabetes insipidus* in which the underlying cause cannot be determined.

(2) *Secondary or symptomatic diabetes insipidus* in which some organic disease of the brain and particularly of the hypothalamus (fracture of the base of the skull, basilar meningitis, pituitary or suprasellar tumors, chronic encephalitis) interfere with the hormone production of the posterior-lobe.

Some investigators like to classify diabetes insipidus according to the BLOOD CHLORIDE level into three groups :

(1) *Hyperchlloremic.*

(2) *Hypochlloremic.*

(3) *Normochlloremic.*

It is claimed that in the hyperchlloremic type, posterior-lobe extracts are especially effective in decreasing the polyuria and thirst while they increase the specific gravity of the urine. Furthermore, allegedly in such patients, salt-poor diets are beneficial while theophylline has little effect. Conversely, in the hypochlloremic and normochlloremic types, posterior-lobe extracts or salt-poor diets are often almost ineffective while theophylline causes marked hyperchlururia. However, this distinction is not generally recognized since some hypochlloremic cases respond well to posterior-lobe extract and the blood chloride level of the same patient may at times be below, and at other times above normal.

According to their SENSITIVITY TO POSTERIOR-PITUITARY EXTRACTS it has also been customary to distinguish :

- (1) *Vasopressin-sensitive cases.*
- (2) *Vasopressin-resistant cases.*

Anatomic studies suggest that usually, in the former, the tuberal nuclei are intact, while in the latter, they are destroyed by some local lesion.

PATHOLOGIC ANATOMY

Diabetes insipidus may result from any lesion which destroys (selectively or in combination) the posterior-lobe, the supra-optic nuclei of the hypothalamus or the stalk of the pituitary. Primary or secondary tumors (often craniopharyngiomas), inflammatory diseases (especially basilar meningitis, chronic encephalitis and syphilis), xanthomatosis, pellagra, trauma to the pituitary region, etc., may be the immediate cause of a secondary or symptomatic diabetes insipidus. Nevertheless, as previously mentioned in several carefully examined cases, no anatomic lesion was detectable either in the pituitary or the hypothalamus of otherwise typical instances of diabetes insipidus. These have been regarded as due to functional derangements and are referred to as the "primary" or "idiopathic" form.

INCIDENCE

Diabetes insipidus is a comparatively rare disease. Young people are most often affected and heredity appears to play an important rôle in some familial cases.

PATHOGENESIS

We have already discussed the ANATOMIC LESIONS which may elicit diabetes insipidus. To recapitulate, any lesion destroying the supra-optic nuclei, the tuber cinereum, the stalk or the posterior-lobe itself causes degeneration of all these structures, due to both retrograde degeneration of the neurons and peripheral degeneration of the nerve fibers together with the posterior-lobe which they supply.

The FUNCTIONAL MECHANISM through which posterior-lobe hormones (presumably vasopressin) act upon water metabolism has been discussed (see :

pp. 219 and 238) above. Since in the vast majority of the cases, posterior-lobe preparations exert a curative effect upon the polyuria of diabetes insipidus, it is reasonable to assume that the disease is due to a specific derangement in the production of such principles.

Suffice it to reiterate here that complete destruction of the anterior-lobe inhibits the development of diabetes insipidus irrespective of the presence or absence of lesions in the supra-optico-hypophyseal system; hence the anterior-lobe presumably produces some diuretic principle. Thirsting does not prevent the polyuria so that the latter must be regarded as primary and not merely a consequence of increased water intake.

CLINICAL COURSE

The most characteristic features of diabetes insipidus are: the excretion of large quantities of pale URINE with a "fixed" low specific gravity (that is, one which remains low even when fluid intake is restricted), decreased production of sweat and saliva and occasionally marked dryness of the skin, malaise, constipation and headaches.

The B.M.R. is usually normal, but there may be HYPOTHERMIA, due to interference with hypothalamic thermoregulation. There is no specific and characteristic derangement in carbohydrate, fat and protein metabolism, although the combination of diabetes insipidus with adiposogenital dystrophy (due to lesions in adjacent hypothalamic areas) is rather common.

As previously mentioned the blood CHLORIDE values may be normal, high or low. There is no strict correlation between the blood and urinary chloride concentration. It has been claimed that in the so-called "hyperchloremic-hypochloric diabetes insipidus" there is a specific disturbance in the chloride-concentrating power of the kidney, while in the "hypochloremic-hyperchloric cases" the renal chloride concentration is comparatively great. In the for-

mer type the urine volume is allegedly increased to help "wash out" chlorides, while in the latter, the excessive excretion of chlorides would carry the water with it.

The chloride concentration in the urine is usually markedly increased during the water retention induced by posterior-pituitary extract treatment, and this is especially evident in the hyperchloremic cases.

Diabetes insipidus is not conducive to any OTHER CHARACTERISTIC MANIFESTATIONS, except the difficulties of child-birth often (but not invariably) noted in women suffering from this disease. These dystocias are ascribed to derangements in oxytocin secretion.

The only important COMPLICATIONS of diabetes insipidus are those due to the local destructive effect of the causative lesion and those which may result from severe dehydration, if adequate amounts of water are not available to the patient.

DIAGNOSIS

In typical cases the diagnosis of diabetes insipidus is comparatively simple, although only LOCAL SIGNS can help to recognize the nature of the causative lesion (tumor, trauma, etc.).

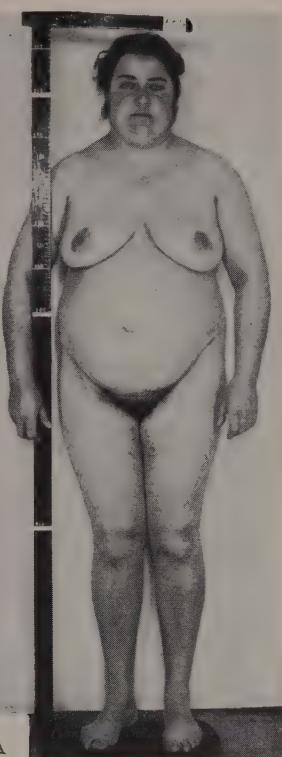
Differentiation from HYSTERIC POLYDIPSIA tends to cause the greatest difficulty. Some nervous individuals take enormous amounts of water as a result of psychic disturbances and this of course also causes marked polyuria. In this connection it is important to remember that in diabetes insipidus the specific gravity of the urine does not exceed 1.005-1.007, even if water is withheld for as long as the patient can endure it. In hysterical polydipsia, on the other hand, the specific gravity rises over 1.012 upon prolonged thirsting. The usually very marked anti-diuretic effect of vasopressin likewise helps to recognize true diabetes insipidus.

Differentiation from DIABETES MELLITUS causes no difficulty since in the latter the polyuria is always accompanied by glucose elimination in the urine.

RENAL INSUFFICIENCY with polyuria, is easily recognized by specific signs of kidney failure.

Diabetes insipidus with galactorrhea. — A. — B. and C. 33-year-old woman in whom obesity, pain in the bones, polydipsia, polyuria, headaches, diplopia, bitemporal hemianopsia, acne, hirsutism, insulin resistance and bilateral galactorrhea had developed 4 years ago. Urinary 17-KS: 42 mg./24 hrs., B.M.R.: normal. There is radiologically demonstrable enlargement of the sella. Pitressin decreases urine output. Following removal of an, apparently chromophobe, anterior-lobe adenoma, the vision became normal within 6 days. Presumably a case of diabetes insipidus with atypical Cushing's syndrome.

(Courtesy of Dr. E. B. del Castillo.)



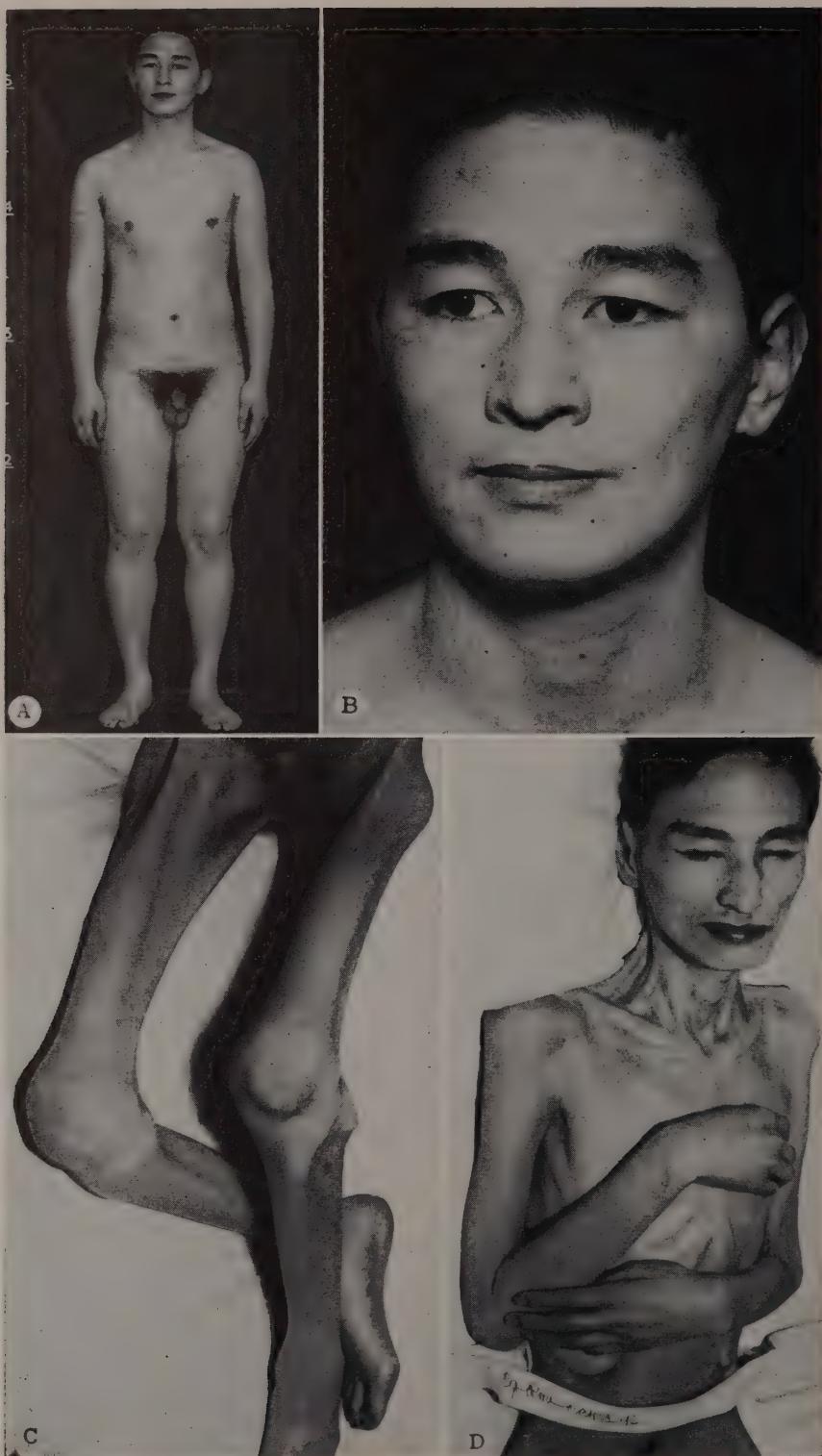
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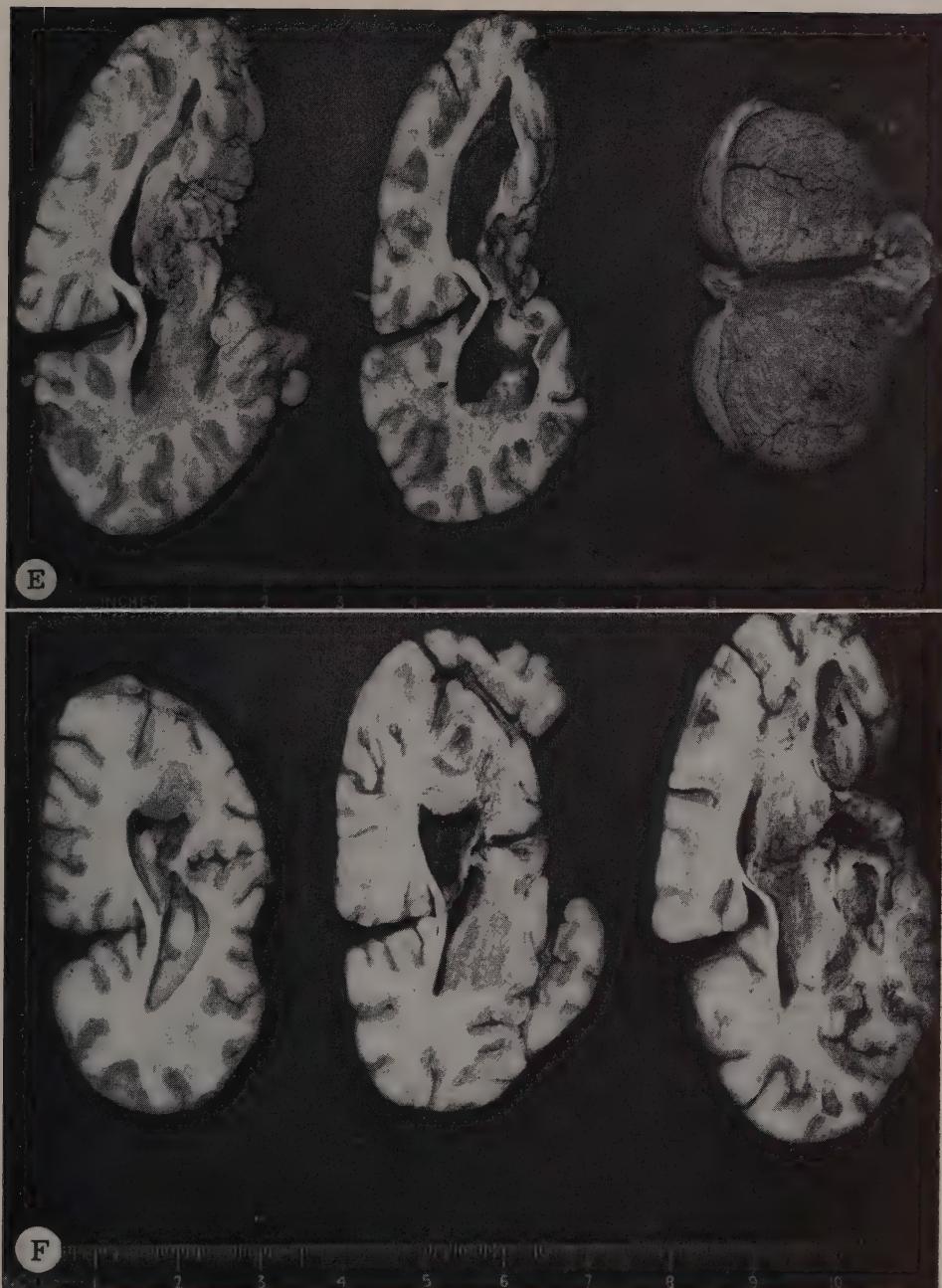
B



C



(Cont'd. on p. 293.)



Diabetes insipidus progressing to Simmond's disease. — A. and B. Man, age 25 years, who suffered from diabetes insipidus during 27 months. At first his nutrition was good. — C. and D. As the disease progressed, there was loss of libido and potency, drowsiness, anorexia, arterial hypotension, progressive weakness and loss of weight. B.M.R.: —46%, 17 KS: 1.1 mg./24 hours, urinary FSH: less than 6 M.U./24 hrs. The visual fields were normal and the sella showed no change upon X-ray examination. — E. and F. Tumor (pinealoma) invading the pituitary and hypothalamus. In this patient diabetes insipidus curiously persisted even after destruction of anterior-lobe. (Courtesy of Dr. E. P. McCullagh.)

PROGNOSIS

Spontaneous recovery from diabetes insipidus is most common if the derangement is caused by infectious diseases or trauma; it occurs especially frequently in children. Otherwise the derangement is usually permanent and tends to become worse unless the lesions spread to the pars distalis and secondary anterior-lobe deficiency inhibits the polyuria.

Temporary improvement during the period of posterior-pituitary extract treatment is the rule, except in the rare vasopressin-resistant cases.

THERAPY

The therapy should be directed against the causative anatomic lesion whenever possible; this means removal of tumors from the hypothalamo-pituitary region, anti-syphilitic therapy when the disease is caused by syphilis, etc. In all other instances purely symptomatic treatment with POSTERIOR-PITUITARY SOLUTION (0.5-10 U.S.P. units subcutaneously) generally alleviates the polyuria for a period of 6-18 hours. Most patients require about 1 cc. of the official preparation, given by hypodermic injection two to three times a day.

PITRESSIN TANNATE in oil may be used in doses of 3-5 pressor units injected intramuscularly every 36-48 hrs., since it is more slowly absorbed and hence longer-acting than the aqueous solution.

Following hypodermic or intra-muscular injection, the local vasoconstrictor effect of the hormone frequently leads to unpleasant complications, hence many physicians prefer to administer pitressin on cotton pledges soaked with the solution and APPLIED TO THE NASAL MUCOSA. Insufflation, by a spray (or atomizer) of posterior-lobe powder or aqueous pitressin preparations into the nasal cavity is likewise recommended. If dry posterior-lobe powder is to be used 50-65 mg. are placed on a piece of paper rolled in the form of a cylinder and placed into the nostril in order to aspirate it. This procedure generally has to be repeated two to four times daily. The powder may also be blown into the nose with an atomizer through a glass tube.

In the case of overdosage there may be : edema, convulsions, headache, abdominal cramps, diarrhea and sometimes even severe shock. All these symptoms are less likely to occur following nasal, than following subcutaneous or intra-muscular administration. If they occur, injection of a mercury diuretic is indicated as an antidote.

AMINOPYRINE (15 grains or 1 gm.) one to three times daily often helps to decrease the requirements for posterior-pituitary extract. Aminopyrine (and other drugs which act upon the hypothalamus) may also prove effective in certain cases which are resistant to the posterior-pituitary solution.

Furthermore, in order to decrease the reabsorptive work of the kidney it is advisable to LIMIT THE SALT INTAKE.

ANTERIOR-LOBE HYPERFUNCTION

DEFINITION

Anterior-lobe hyperfunction is a condition in which the hormone production of the anterior-lobe is sufficiently augmented to cause detectable manifestations of hyperpituitarism. Among these are the increase in the size and

function of those endocrines (adrenal cortex, thyroid, gonad) which are under anterior-lobe control. The hyperthyroidism and hypergonadism of pituitary origin have already been discussed in the chapters on the thyroid and gonads respectively, while increased

adrenal-cortical function has been considered in connection with the Cushing's syndrome of adrenal-cortical origin (see: p. 162). This disposition of the material was adopted because in pertinent cases it is often impossible to differentiate between disorders due to a primary, pituitary disease and those resulting from "idiopathic" hyperfunction of the thyroid, gonads or adrenals.

In the present section, chief emphasis will be placed upon HYPOPHYSEAL GIGANTISM, ACROMEGALY and CUSHING'S DISEASE since these syndromes are comparatively clear-cut clinical entities. In view of the frequent overlap in the symptomatology of the various clinical types of anterior-lobe hyperfunction it was considered advisable, however, to discuss this whole group conjointly in the present chapter.

CLASSIFICATION

The various types of anterior-lobe hyperfunction can be subdivided according to the age of onset, the intensity of the manifestations, the underlying pathologic lesions, etc., but for practical purposes it suffices to subdivide this whole group as follows:

- (1) Pituitary gigantism.
- (2) Acromegaly.
- (3) Cushing's disease.

(4) Mixed types in which manifestations of several among the above-mentioned groups are simultaneously present.

(5) Other types of anterior-lobe hyperfunction, mainly characterized by a more or less selective increase in the production of thyrotrophic, gonadotrophic, diabetogenic, mammogenic, lactogenic, etc., hormones. These latter diseases are not yet sufficiently understood to warrant detailed discussion here.

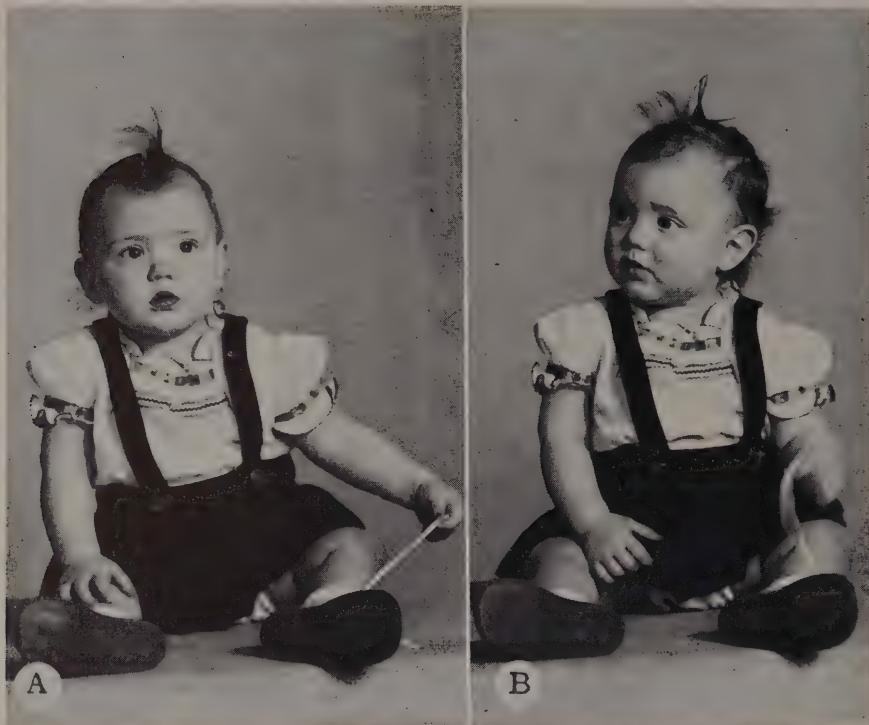
There are various types of GIGANTISM (e.g., familial, racial), but in the present chapter we shall only be concerned with that due to pituitary overfunction. The latter results from an excessive production of somatotrophin in a young individual whose epiphyseal junction cartilages have not yet ossified, so that the bones are still capable of growth in length.

ACROMEGALY is a type of gigantism which usually occurs in an individual whose growth in length has become impossible since the junction cartilages have already ossified when the increased somatotrophin production of the pituitary commenced. It therefore results mainly in appositional bone growth in width or at the ends of extremities, and in proliferation of soft tissues.

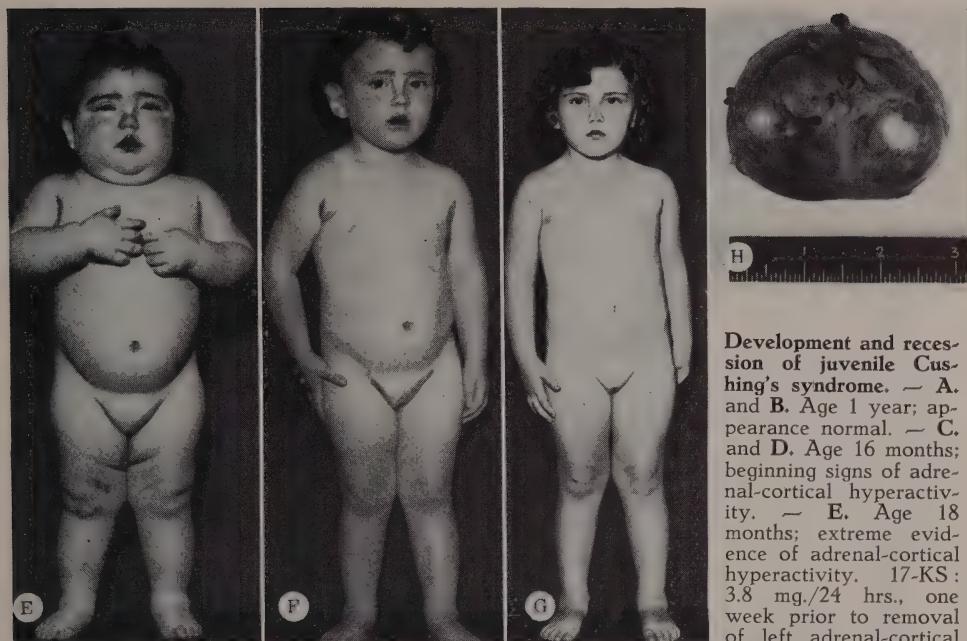


Acromegaly. — A. and B. 70-year-old Negro woman with a pituitary tumor. Note prominent lips and nose characteristic of this race but exaggerated by the disease.

(Courtesy of Dr. J. I. Lobo.)



(Cont'd on p. 297.)



Development and recession of juvenile Cushing's syndrome. — A. and B. Age 1 year; appearance normal. — C. and D. Age 16 months; beginning signs of adrenal-cortical hyperactivity. — E. Age 18 months; extreme evidence of adrenal-cortical hyperactivity. 17-KS: 3.8 mg./24 hrs., one week prior to removal of left adrenal-cortical adenoma. — F. Child 9

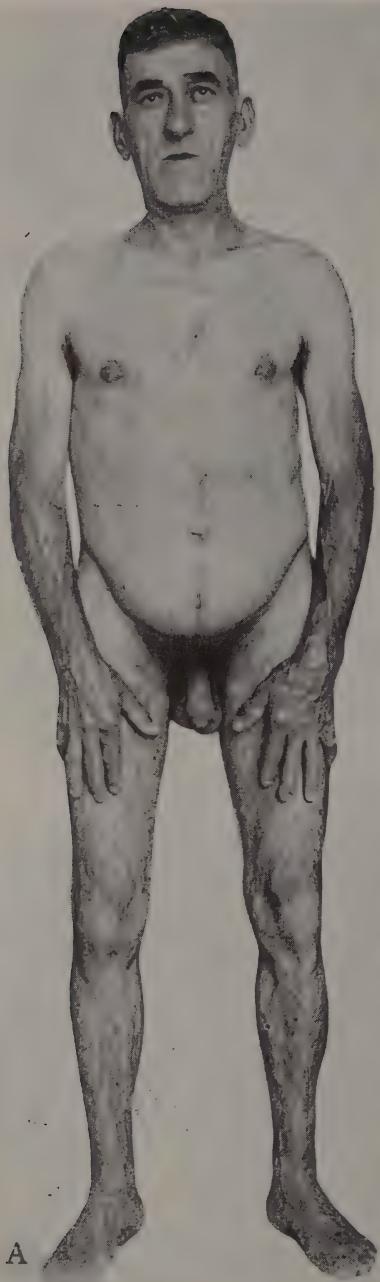
months postoperative; almost complete recession of the disease. — G. 2 years postoperatively; appearance normal. — H. Adrenal-cortical adenoma (140 gm.) removed; it contained hemorrhagic necroses and calcified deposits between neoplastic cortical cells.

(Courtesy of Drs. Robert W. Schneider and E. P. McCullagh.)

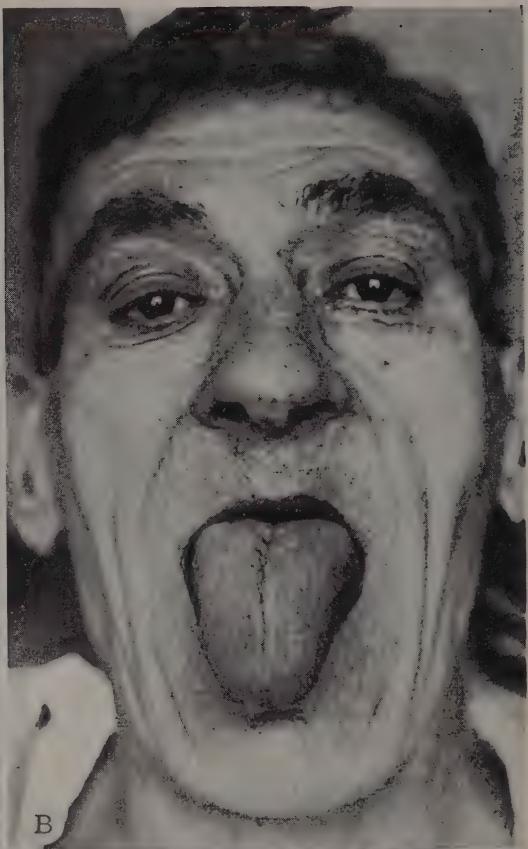


Clinical appearance suggestive of Cushing's syndrome (not verified by biopsy or autopsy). — A. and B. Woman age 36 years. Hirsutism, obesity (208 lbs.), blood pressure: 218/140, diabetic tendency in glucose tolerance curve, B.M.R.: +9%, Hb.: 110%, RBC: 5,400,000. Ruddy plethoric appearance of moon-shaped face. — C. Note plethora of face, obesity most marked on trunk, shoulders and neck.

(Courtesy of Drs. H. Lisser and C. K. Canelo.)



A



B



Acromegaly. — A. 53-year-old man who began to develop acromegalic features 20 years ago, and insulin-resistant diabetes 10 years ago. — B. Note large nose, tongue and chin. — C. X-ray shows enlargement of sella, large para-nasal sinuses and widening of the angle of the jaw.

(Courtesy of Dr. A. B. de Ulhôa Cintra.)

Sometimes we find mixed types of GIGANTISM WITH ACROMEGALY. Here the condition usually commences before the cessation of growth in length; this causes gigantism, but if increased somatotrophin production continues in later life, eventually acromegalic manifestations appear. Only rarely are there typical signs of acromegaly in children who are still capable of growth.

CUSHING'S DISEASE is a condition characterized by hypertension, glycosuria, osteoporosis, a peculiar type of facial and trunk obesity, purplish cutaneous striations and a florid complexion; in women there is hirsutism of the male type with amenorrhea and in men, impotence. The disease is due to basophilic or mixed-cell adenomas of the anterior-lobe and must not be confused with "Cushing's syndrome" in which the same clinical manifestations are due to adrenal tumors, lesions in the hypothalamus, thymus tumors or unknown causes.

MIXED ANTERIOR-LOBE HYPERFUNCTION in which manifestations of acromegaly and Cushing's syndrome appear

conjointly are not uncommon and this is one of the reasons why the various types of anterior-lobe hyperfunction are discussed here in the same chapter.

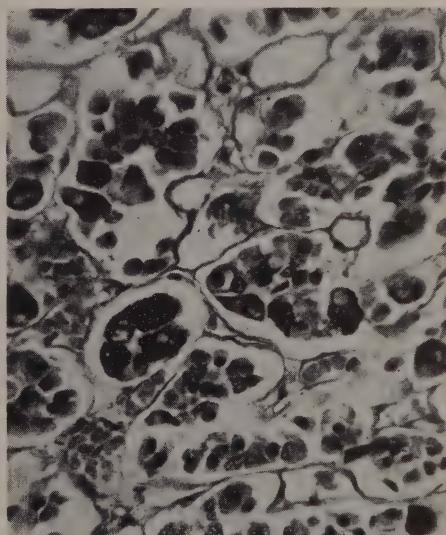
PATHOLOGIC ANATOMY

In PITUITARY GIGANTISM and ACROMEGALY the underlying lesion is an eosinophilic or mixed-cell adenoma (or carcinoma) of the anterior-lobe which may measure $1\frac{1}{2}$ to 2 inches in diameter (see also "Tumors," p. 264). In CUSHING'S DISEASE — as originally described — the pathogenic agent is a basophil adenoma of the anterior-lobe, but this is a comparatively rare cause of the Cushing syndrome. More frequently we find hyaline degeneration and vacuolization of the anterior-lobe basophils; in some cases the anterior-lobe shows no histologically-detectable abnormalities. It should be emphasized, however, that voluminous anterior-lobe tumors are rarely found in Cushing's disease; this accounts for the infrequency of radiologic signs of a hypophyseal neoplasm in patients suffering from this malady.



Hypophysis in acromegaly. Note uniformity of structure in this adenoma, which consists exclusively of eosinophils. (High magnification.)

(Courtesy of Dr. W. Boyd.)



Hyalinization of basophils in Cushing's syndrome. Note large, darkly staining basophils, whose cytoplasm is homogeneous ("hyalinized") and contains irregular, light vacuoles. Patient suffered from typical Cushing's syndrome.



Hyaline changes in pituitary basophils frequently associated with Cushing's syndrome. (1) Normal mature basophil cells. Note eccentric nucleus and light circle corresponding to "negative Golgi image"; the cytoplasm is filled with coarse basophil granules and there is one vacuole, but no homogeneous hyalin material. (2-8) basophil cells affected by increasing degrees of hyalinization. Some of the basophils are binucleated (4,8) and only in two of them (2,3) is the negative Golgi image detectable. Such lesions are not absolutely constant, but very characteristic of Cushing's syndrome. (After A. C. Crooke; J. of Path. and Bact. 41, 339, 1935).

INCIDENCE

Anterior-lobe hyperfunction is probably quite common but most pertinent cases manifest themselves as secondary hyperthyroidism, hypercorticoidism, hy-

pergonadism or hypophyseal diabetes mellitus.

True PITUITARY GIGANTISM is comparatively rare and of course occurs only in young, growing individuals.

ACROMEGALY is likewise rare; it is estimated to occur about once per 15,000 hospital admissions. About half of all cases appear during the third decade of life, but the fully-developed clinical syndrome may not be manifest before the fifth decade.

True CUSHING'S DISEASE is one of the least common endocrine diseases.

PATHOGENESIS

The causative pituitary lesions have been discussed above (see : Path. Anat. p. 299). Suffice it to say here that hypophyseal GIGANTISM is due to the growth-promoting effect of somatotrophin which accelerates new bone formation, especially at the junction cartilages. At the same time, there is a fairly proportionate increase in the size of the various organs and soft tissues.

In ACROMEGALY the increased somatotrophin production can no longer stimulate growth in length, since the disease occurs after the union of the junction cartilages. Nevertheless, the growth of those bones which are independent of junction cartilages (periosteal bone formation) and that of the soft tissues (e.g., internal organs, skin) continues; this gives rise to the typical acromegalic appearance.

CUSHING'S DISEASE is generally ascribed to an increased corticotrophin secretion which in turn augments the secondary corticoid hormone production. The fact that the adrenal cortices are often enlarged and that partial adrenalectomy tends to exert a curative effect is in accordance with this interpretation. Some investigators assumed, however, that at least the characteristic hyalinization of the basophil cells may be secondary rather than the cause of the disorder. In some instances, hypothalamic lesions appear to be the primary cause of the malady. It is possible that derangements in the hypothalamic region may increase corticotrophin production in the same manner in which tumors of the pineal region can appar-

ently increase gonadotrophin secretion and elicit precocious puberty.

CLINICAL COURSE

State. — In PITUITARY GIGANTISM there is generalized, symmetric overgrowth of the skeleton and soft tissues so that well-proportioned giant types result. These are usually physically strong, mentally alert, intelligent individuals whose libido and potentia are above normal. Occasionally however, gigantism is associated with genital hypoplasia and sometimes it is complicated by diabetes mellitus or hyperthyroidism.

In ACROMEGALY there is overgrowth of the short and flat bones, enlargement of the viscera, degenerative changes in the muscles, and often complication with hyperthyroidism or diabetes mellitus. The overgrowth affects mainly the ends (acra) of the body, namely the nose, chin, hands and feet; even the soft tissues of the "acra" (e.g., nose, lips, tongue) are enlarged.

Both in pituitary gigantism and in acromegaly, enlargement of the sella, due to pituitary tumor, is comparatively common and frequently accompanied by signs of intracranial pressure.

In CUSHING'S DISEASE the sella is rarely eroded, since large hypophyseal tumors are uncommon. However, the following manifestations are characteristic: obesity of face, neck and trunk, hypertension, diabetes, osteoporosis, cutaneous striations and "virilization." Occasionally the patient complains of backache, abdominal pain, pains in the eyes, choking and suffocating sensations, convulsions and even fainting spells. Usually the general resistance, especially to infections is greatly decreased in Cushing's disease.

Metabolism. — The BODY TEMPERATURE and the B.M.R. may be low, normal or high in the various types of anterior-lobe hyperfunction depending upon the functional activity of the pituitary. An increase in the B.M.R. is presumably the result of an excess

thyrotrophin production, while a decrease could result from a breakdown of pituitary tissue following compression or necrosis of hypophyseal tumors.

Disturbances in CARBOHYDRATE metabolism are characteristic both of acro-

megaly and of Cushing's disease, but are comparatively rare in gigantism. They can presumably be due to increased gluco-corticotrophin (and secondarily gluco-corticoid), diabetogenic principle and thyrotrophin production,



Acromegalic gigantism. — A. 27-year-old acromegalic giant (height approximately 7' 2½"). The two normal men standing at either side are 5'9" and 5'10½" high respectively. Note typical acromegalic facies, prognathism. The fingers are so wide that a U.S.A. 50 cent piece passes easily through his finger ring. Voice extremely deep and hoarse, musculature relatively weak. (Cont'd. on p. 303.)

since all of these hormones are conducive to hyperglycemia and even glycosuria.

Derangements in LIPID METABOLISM are especially noteworthy in Cushing's disease, which is characterized by a peculiar "buffalo type" of obesity, localized in the face ("moon face"), neck and trunk while the extremities remain comparatively thin. The blood lipid and blood cholesterol levels on the other hand show no constant deviation from the normal.

The anabolic effect of somatotrophin may result in marked NITROGEN retention in gigantism and acromegaly, but this is rarely the case in Cushing's disease. The N.P.N. rises only if secondary renal disease interferes with the excretion of the metabolic end-products of protein metabolism.

Disturbances in WATER AND SALT METABOLISM are likewise common in conjunction with the various types of anterior-pituitary hyperfunction. Polyuria is frequently observed, especially in acromegaly and Cushing's disease. It is sometimes due to accompanying nephro-

sclerosis and perhaps partly also to the diuretic effect of certain anterior-lobe hormones (e.g., thyrotrophin).

The blood PHOSPHATE level is often high (4-6 mg.%) in acromegaly, while Cushing's disease is sometimes conducive to the hypercorticoid type of electrolyte derangement, namely: a fall in blood potassium and chloride accompanied by alkalosis and a rise in blood sodium. These changes are presumably due to increased mineralo-corticotrophin production.

Growth and Bone Structure. — In PITUITARY GIGANTISM bone growth is excessive, but the skeleton remains proportionate. Many of these giants attain a size of 7 to $7\frac{1}{2}$ ft., and in a few cases a height of 8 ft. 9 to 10 inches has been claimed. Excessive growth may commence in early childhood, but usually it does not become evident before adolescence. If somatotrophin overproduction continues after ossification of the junction cartilages, acromegaly is superimposed upon the gigantism so that "acromegalic giants" result.

In ACROMEGALY usually one of the



— B. Skull showing widening of the angle of the jaw and enlargement of sella and para-nasal sinuses. — C. X-ray of the greatly enlarged hand of the patient in comparison with a normal hand.

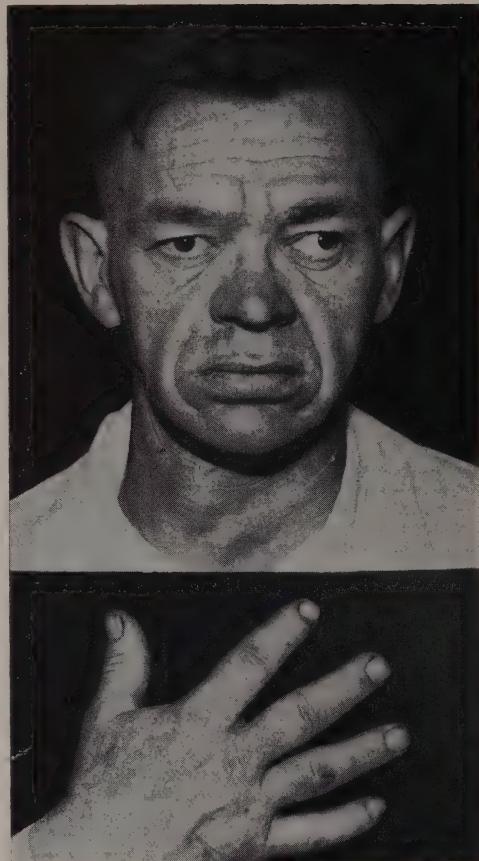
(Courtesy of Dr. E. P. McCullagh.)



first noticed signs is that the patient has to wear larger gloves and shoes than usual, because of the excessive growth of the hands and feet. The hands are not only large but especially broad ("spade hands") with thick, blunt "sausage fingers." The terminal phalanges may show "tufting." The supra-orbital ridges become very prominent and, if the disease develops comparatively early, the para-nasal (especially the frontal) sinuses are greatly enlarged. The growth of the mandible is especially pronounced. The angle of the jaw tends to straighten out while the chin turns upward; this often causes severe prognathism and a separation of the teeth, which "grow away from each other."

Acromegaly. Age 51 years, height $72\frac{1}{2}$ ", weight 220 lbs. Signs of acromegaly manifest during past ten years. Note enlargement of lips, soft tissues of face and hands; exophoria (outward rotation of eye-ball) due to weakness of internal rectus muscle. There is a large nodular goiter. Marked enlargement of sella turcica due to presence of pituitary tumor. Visual fields are normal, B.M.R.: +14%, urinary FSH: 26-53 M.U./24 hrs.

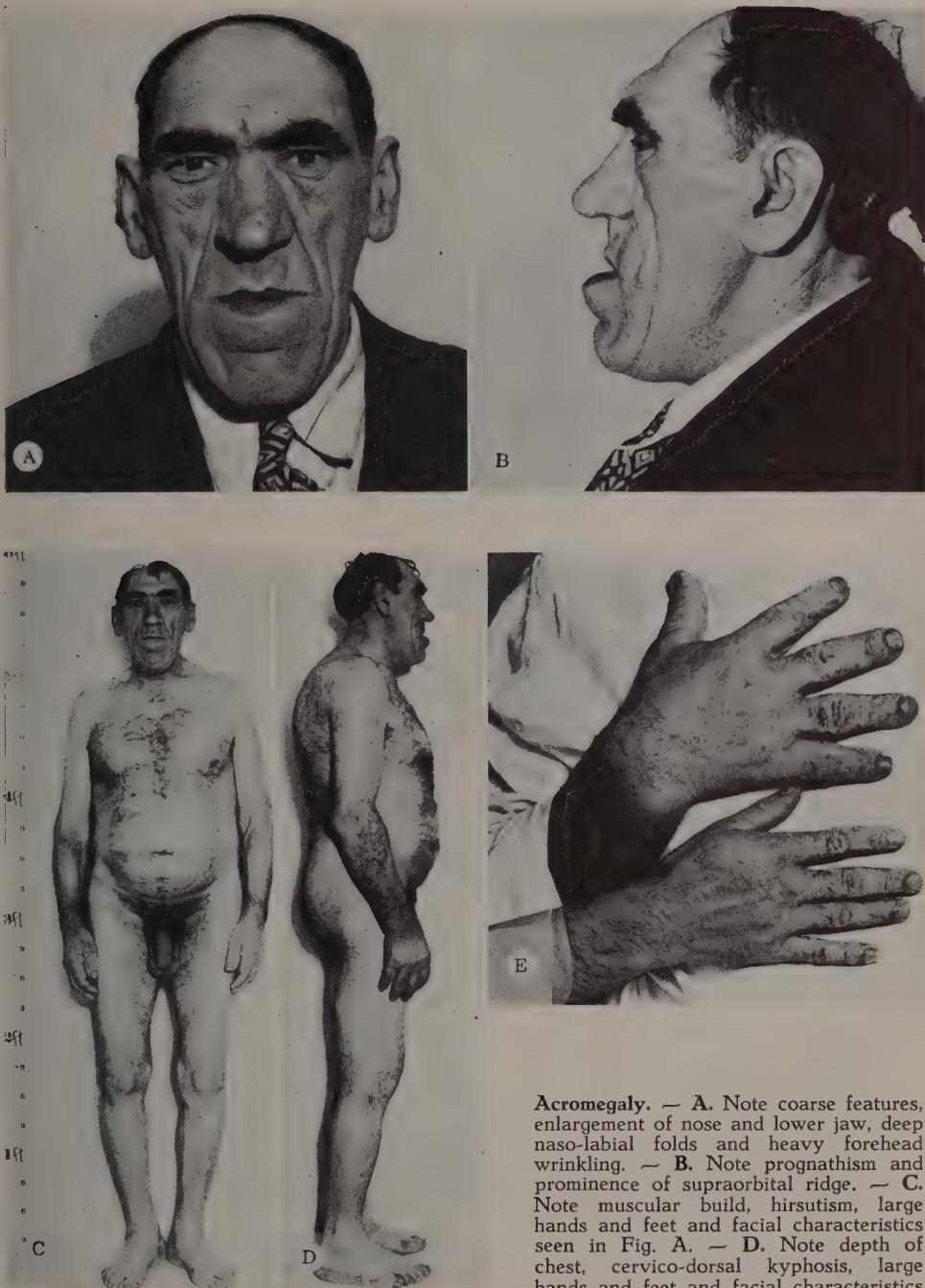
(Courtesy of Dr. E. P. McCullagh.)



Hands in acromegaly. Typical acromegalic hands. Note great increase in the development of the soft tissues.

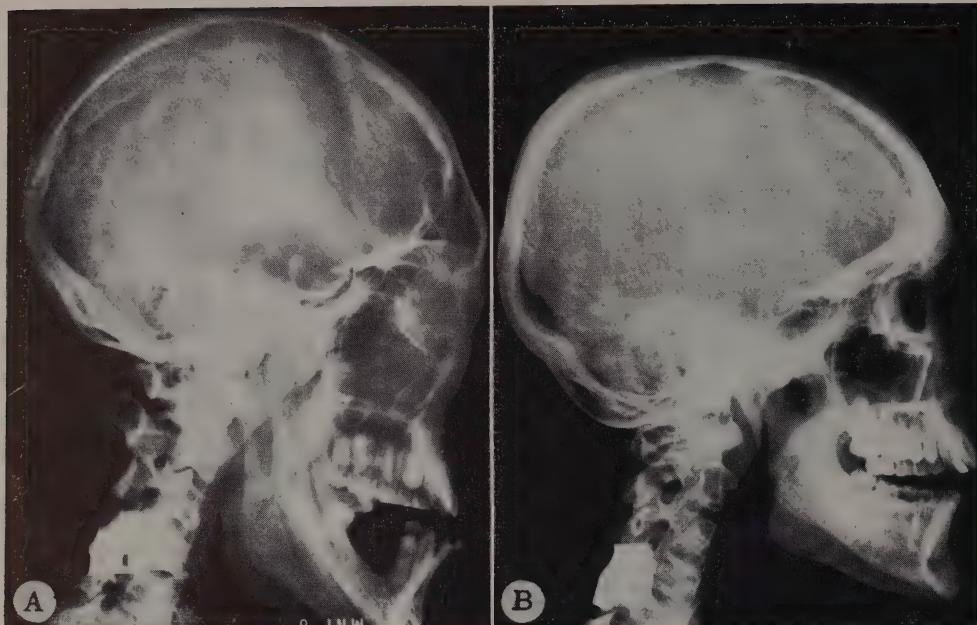
(Courtesy of
Dr. E. J. Kepler.)





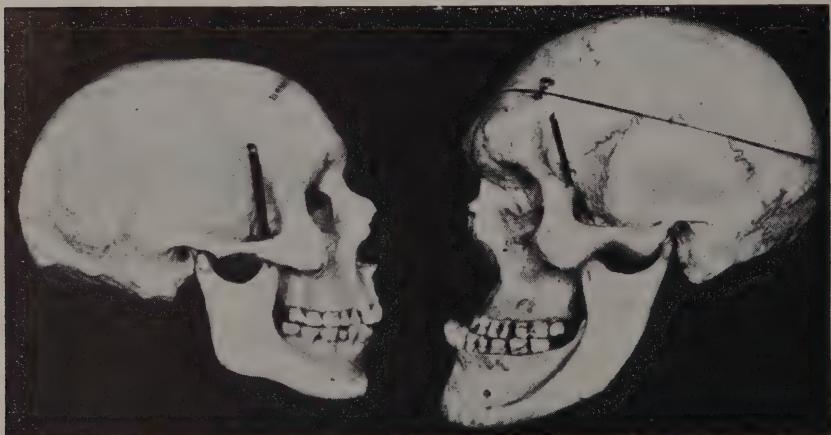
Acromegaly. — A. Note coarse features, enlargement of nose and lower jaw, deep naso-labial folds and heavy forehead wrinkling. — B. Note prognathism and prominence of supraorbital ridge. — C. Note muscular build, hirsutism, large hands and feet and facial characteristics seen in Fig. A. — D. Note depth of chest, cervico-dorsal kyphosis, large hands and feet and facial characteristics seen in Fig. B. — E. Note enlargement and sausage-like fingers (compared with normal male hand).

(Courtesy of Drs. H. Lisser and M. N. Goldberg.)



Acromegaly. — A. Man, age 28 years, with manifest signs of acromegaly. Note marked enlargement of sella, frontal and maxillary sinuses, marked opening of the jaw-angle with prognathism. — B. Man, age 22 years, normal skull for comparison.

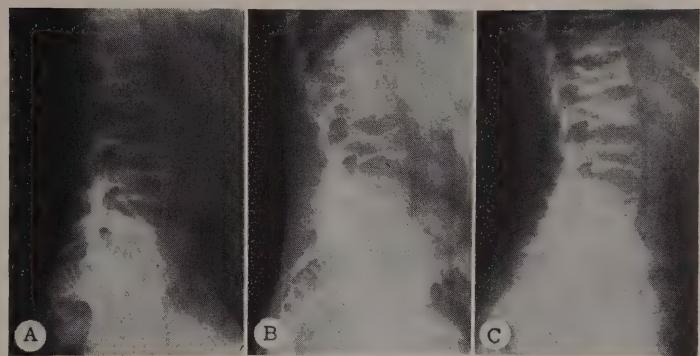
(Courtesy of Montreal Neurological Institute.)



Skull changes in acromegaly. Lateral aspect of a normal (left) and acromegalic cranium. Note prominent supraorbital ridges, opening of angle of mandible, prognathism, and separation of teeth. The maxillary spur had been removed in the trans-sphenoidal operation.

(After H. Cushing and L. M. Davidoff: Monographs of the Rockefeller Inst. for Med. Res. No. 22, 1927.)

Cushing's disease. — A. Man, age 28 years, suffering from typical Cushing's disease. Decreased glucose tolerance, hypertension, gibbus (hunch) formation, thin skin, "moon-face", markedly increased urinary corticoids. Patient had to wear brace, because of the great decalcification of the spinal column, clearly visible on this X-ray picture. The vertebrae are very transparent and separated by extremely wide intervertebral spaces. —



B. The patient was treated with irradiation of the pituitary, with calcium and vitamin-D supplements in the diet. This photograph was taken three years later, when, during this treatment, calcification of the vertebrae began. Cortical zone in vertebrae is denser, but several of them have partially collapsed; this led to a great reduction in the patient's height. — C. This radiogram was taken after one year's treatment with testosterone propionate (25 mg. twice weekly). The gibbus decreased, glucose tolerance and hypertension vanished. Calcification of the compressed vertebrae proceeds satisfactorily. However, in such cases, spontaneous remission cannot be excluded.

(Courtesy of Department of Radiology, Royal Victoria Hospital.)



Vertebral changes in acromegaly. Eleventh thoracic vertebra of an acromegalic (bottom) compared with the normal. Viewed from above (A) and from the side (B). Note numerous exostoses and osteoporosis in acromegalic.

(After H. Cushing and L. M. Davidoff: Monographs of the Rockefeller Inst. for Med. Res. No. 22, 1927.)



A



B



C



E



D

(Cont'd. on p. 309.)

Cushing's syndrome. Age 27 years. Outstanding characteristics: Obesity, chiefly of face and trunk, weakness, deep red "moon-face". Easy bruising, violaceous abdominal striae, arterial hypertension (216/148), gallop rhythm, headaches, retinal hemorrhages and exudates, decalcification of spine and pelvis. — X-ray therapy to pituitary (1200 r.u. to each temple) was applied without obvious benefit. Hence, about 4 months later, bilateral hemiadrenalectomy was performed. — A, B and C. General appearance before therapy. — D. and E. 7 months following bilateral hemiadrenalectomy. Complete symptomatic cure. Arterial hypertension remains, blood pressure 188/138. Decalcification of spine slightly less. — F. Appearance of patient three years after adrenal surgery. — G. X-ray five months preoperative. Note more marked demineralization of the spine, and of the pelvis. There appears to be a coincidental congenital deformity of the neural arch of the 5th lumbar vertebra. — H. Striking remineralization three years after operation. The partially destroyed head of the right femur with secondary hypertrophic changes is presumably the result of an unrecognized fracture due to the disease.

(Courtesy of Dr. E. P. McCullagh.)



F



G



H

In addition to the above changes, X-ray of the skull usually reveals an enlarged sella often with erosion of the posterior clinoid processes; the calva-

rium — as the other flat bones — becomes greatly thickened.

The bodies of the vertebrae increase in their antero-posterior dimensions, a

fact which is especially obvious in lateral X-rays of the thoraco-lumbar spine. Exostoses often result from periosteal over-growth. The degenerative changes in the vertebræ may lead to pronounced kyphosis.

In CUSHING'S DISEASE osteoporosis, especially of the spine, is frequently pronounced and may lead to a kyphosis similar to that seen in acromegaly. The shoulders tend to become round, the stature may actually grow shorter as a result of the osteoporosis and the consequent collapse of vertebrae. However, excessive growth of the bones, in length and width, or enlargement of the sella is likewise rare.

Blood. — In pituitary gigantism and acromegaly the blood count is rarely abnormal, but in Cushing's disease there often is polycythemia and an increase in blood volume which are responsible for the characteristic florid complexion of these patients.

Cardiovascular System. — In pituitary GIGANTISM the cardiovascular system remains essentially normal in proportion with the large size of the individual. In ACROMEGALY and CUSHING'S DISEASE there tends to be a pronounced enlargement of the heart ("cor bovinum") and of the blood vessels; these changes are probably secondary to the rise in blood pressure. Myocarditic scars are likewise frequent, especially in Cushing's disease. In this malady, apparently due to weakness of the peripheral vessels, there is also a great tendency to the formation of ecchymoses, even under the influence of very slight traumas.

Muscular System. — In pituitary gigantism the musculature is usually well-developed and physical strength is above average. In acromegaly and Cushing's disease on the other hand, the muscles show degenerative changes and there is great fatigability upon muscular exertion.

Nervous System and Sense Organs.

— All kinds of pituitary tumors which increase intracranial pressure tend to

cause HEADACHES, VISUAL DEFECTS, DIZZINESS and sometimes FAINTING SPELLS, irrespective of the type of their hormone production. Headache is estimated to occur in about 90% of all acromegatics but is much less common in Cushing's disease. The visual disturbances are sometimes due to choked discs but more frequently to encroachment of the tumor upon the optic chiasma with consequent bitemporal hemianopsia. The cause of this particular visual defect is that those fibers of the optic nerve tracts which supply the temporal part of the visual field are closest to the pituitary and hence first to be affected by an expanding tumor.

In gigantism and acromegaly an initial increase in LIBIDO, POTENCY AND GENERAL MENTAL CAPACITY is rather frequent. These individuals tend to be enterprising, energetic, ambitious persons until a final breakdown of the neoplastic tissue leads to secondary hypopituitarism with loss of libido and general apathy.

EXOPHTHALMOS is comparatively common in acromegaly, but rare with gigantism or Cushing's disease. It is frequently associated with an increase in the B.M.R. and presumably due to increased thyrotrophin production.

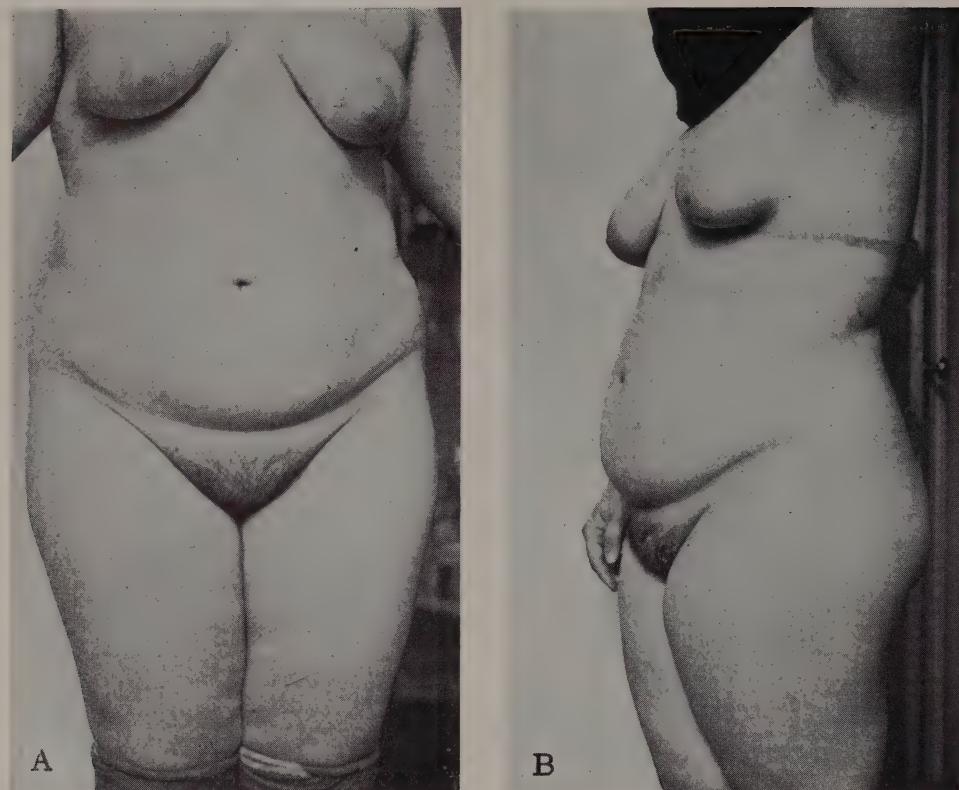
The extreme enlargement of the NOSE, TONGUE and LIPS so characteristic of acromegaly is not seen in typical gigantism or Cushing's disease.

Respiratory System. — A tendency towards deepening of the VOICE, due to excessive enlargement of the larynx, is noted in acromegaly but much less common in Cushing's disease.

Digestive System. — All the visceral organs are greatly enlarged in the various types of anterior-lobe hyperfunction. In pituitary gigantism this enlargement remains proportionate to the rest of the body, while in acromegaly the viscera (heart, liver, pancreas, intestines, kidney, etc.) are much greater than would correspond to the body size. In Cushing's disease the heart and kidneys are often enlarged and the latter



The skin in Cushing's syndrome. — A. Note the tendency of the skin to retain marks from clothing (due to skin edema?). — B. In this patient, hirsutism and keratosis pilaris are especially prominent. — C. Chest of a man with pituitary basophilism. Note prominence of acne, hirsutism and so-called "androgenic flush". The nipples are often pigmented, even in the male, and the areolae large. (Courtesy of Dr. E. J. Kepler.)



Cushing syndrome? — A. and B. 15-year-old girl with secondary amenorrhea, obesity and moderate hypertension. Note abdominal striae but no excessive pigmentation of the areolae and no hirsutism.

(Courtesy of Dr. J. I. Lobo.)

may show signs of nephrosclerosis, but general splanchnomegaly is rare.

Sclerosis or hydropic degeneration of the pancreatic islets is likewise noted in certain cases of acromegaly or Cushing's disease, especially in those accompanied by diabetes mellitus. These changes may well be due to excessive production of the so-called diabetogenic principle of the anterior-lobe.

Skin and Appendages. — In pituitary GIGANTISM the skin texture and hair distribution are essentially normal, while in ACROMEGALY there is pronounced increase in the thickness of the epidermis with hypertrophy of the skin papillæ and infiltration of the subcutaneous connective tissue. This causes great thickening and wrinkling of the skin, which is especially pronounced on the scalp, face and hands. It results in a coarsening of the features and adds

to the characteristic enlargement of the hands, nose, lips and chin. In early stages there often is marked hirsutism and apical baldness, but later there may be complete loss of genital and axillary hair, due to breakdown of the tumor.

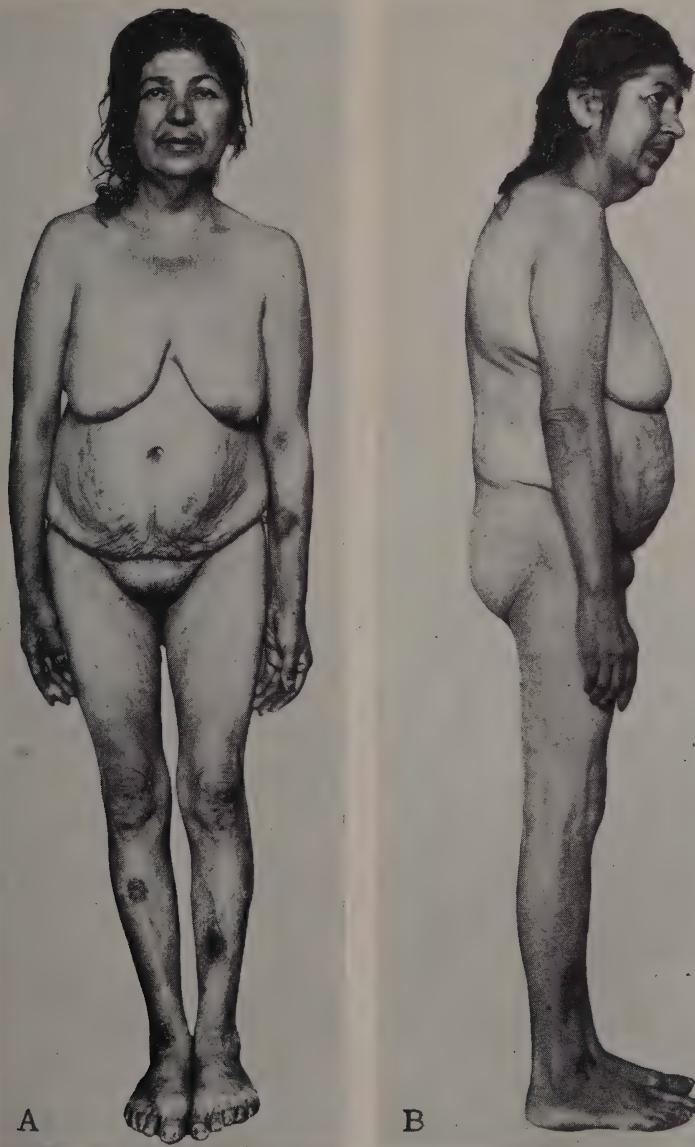
In CUSHING'S DISEASE the skin on the face becomes hyperemic with a reddish, sometimes bluish tinge which accounts for the florid appearance of these patients. There is hirsutism and shaving often becomes necessary in women. The body hair over the back, chest, arms, thighs and legs is likewise greatly increased and, due to some latent type of edema, the skin tends to retain the markings of tight clothing (e.g., brassieres, corsets, belts). Ecchymoses and purpuric spots appear readily on slight trauma to the skin of the abdomen, thighs, arms, back. Acne, keratosis pilaris and purplish, atrophic, cu-

taneous striations along the sides of the abdomen and thighs are especially characteristic of Cushing's disease.

Urinary System. — The kidney is enlarged in pituitary GIGANTISM, but only in proportion to the body surface. In ACROMEGALY and CUSHING'S DISEASE the dimensions of the kidney are disproportionately large and microdissection studies have shown that the entire nephron is increased in length and width. In late stages of Cushing's dis-

ease (less frequently in acromegaly) there often is marked nephrosclerosis; this is partly responsible for the accompanying hypertension. The great predisposition of patients with Cushing's disease to various types of nephritis, especially ascending pyelonephritis, is likewise noteworthy.

Sex Organs — In early stages of pituitary GIGANTISM and ACROMEGALY the sex organs may be stimulated by pituitary gonadotrophins; but in the



Cushing's syndrome. — A. and B. 38-year-old woman who developed some facial hair 2 years ago, amenorrhea 3 years ago and diabetes 18 years ago. Clitoris of normal size, voice feminine, osteoporosis, typical abdominal striae, vascular fragility (note subcutaneous effusions on forearm and legs), emaciation, skin and muscle atrophy, hypertension (blood pressure 170/110), insulin resistance, blood cholesterol: 132 mg.%, CO₂-combining power: 64 vol %, blood Cl: 80 m.Eq./L., blood Na: 134 m.Eq./L.

(Courtesy of
Dr. A. B. Ulhoa Cintra.)

final stages, if the causative neoplasm begins to break down, atrophy of the testes, ovaries and accessory sex organs, loss of pubic and axillary hair, impotence in the male and amenorrhea in the female are the rule.

In CUSHING'S DISEASE amenorrhea, loss of libido and impotence are usually noted from the onset. Curiously, in spite of other signs of virilization (hirsutism, acne, etc.) the clitoral enlargement, so characteristic of the adrenogenital syndrome is absent.

COMPLICATIONS

Most of the common complications of anterior-lobe hyperfunction are those due to the causative pituitary tumor itself. Among these are: visual disturbances, various neurologic manifestations due to invasion into, or compression of, the brain, hemorrhages into the tumor or liquefaction necrosis of the entire hypophysis with subsequent fatal hypopituitarism. All three types of anterior-lobe hyperfunction, but especially acromegaly and Cushing's disease, may be accompanied by marked, and often insulin-resistant diabetes mellitus or by hyperthyroidism.

Severe hypertensive disease with apoplexy and coronary complications are especially characteristic of Cushing's disease.

Acromegaly and particularly Cushing's disease decrease the patients resistance to various types of stress and intercurrent infectious diseases are a frequent cause of death among them. This is presumably due to a derangement of the normal, adaptive pituitary-adrenal mechanism.

DIAGNOSIS

Pituitary gigantism must be differentiated from simple PRIMORDIAL GIGANTISM. The latter is the counterpart of the so-called constitutional "primordial dwarfism" which has been discussed in the chapter on hypopituitarism. This is sometimes difficult since intermediate types between the two appear to be quite common. Even the constitutional

form in which several members of a family, or race, are above average in size, apparently acts through genetic factors whose influence is mediated through the anterior-lobe and its somatotrophin production. Some investigators even speak of a "hyperpituitary constitution." It is generally agreed however, that primordial gigantism should be differentiated from pituitary gigantism on the basis of a demonstrable hypophyseal tumor in the latter cases.

Acromegaly is often confused with PAGET'S DISEASE, chronic PULMONARY OSTEO-ARTHROPATHY or LEONTIASIS OSIUM, but the bone lesions characteristic of the latter conditions usually permit differential diagnosis. During PREGNANCY, changes suggestive of slight acromegaly are almost constantly demonstrable (thickening of the lips and nose, enlargement of the hands and feet, disturbances of vision, some splanchnomegaly) but the bony structures are usually not involved and after gestation the derangement disappears again.

This "forme fruste" of acromegaly has been ascribed to a temporary excess in anterior-lobe-hormone production during gestation.

CUSHING'S DISEASE may be difficult to differentiate from "Cushing's syndrome" (due to thymus neoplasms, adrenal tumors or hypothalamic injuries) especially since, unlike other types of anterior-lobe hyperfunction, Cushing's disease is rarely accompanied by manifest local signs of a tumor in the pituitary region. In doubtful cases it may be necessary to undertake surgical exploration of the adrenal and pelvic organs, the latter for possible accessory adrenals in the ovary or mesometrium, arrhenoblastomas and other virilizing neoplasms.

An increased elimination of corticoids and 17-KS is especially characteristic of Cushing's disease, but occasionally also tends to occur in acromegaly.

In the differential diagnosis of the three types of anterior-lobe hyperfunc-

Differential diagnosis of diseases due to anterior-lobe hyperfunction

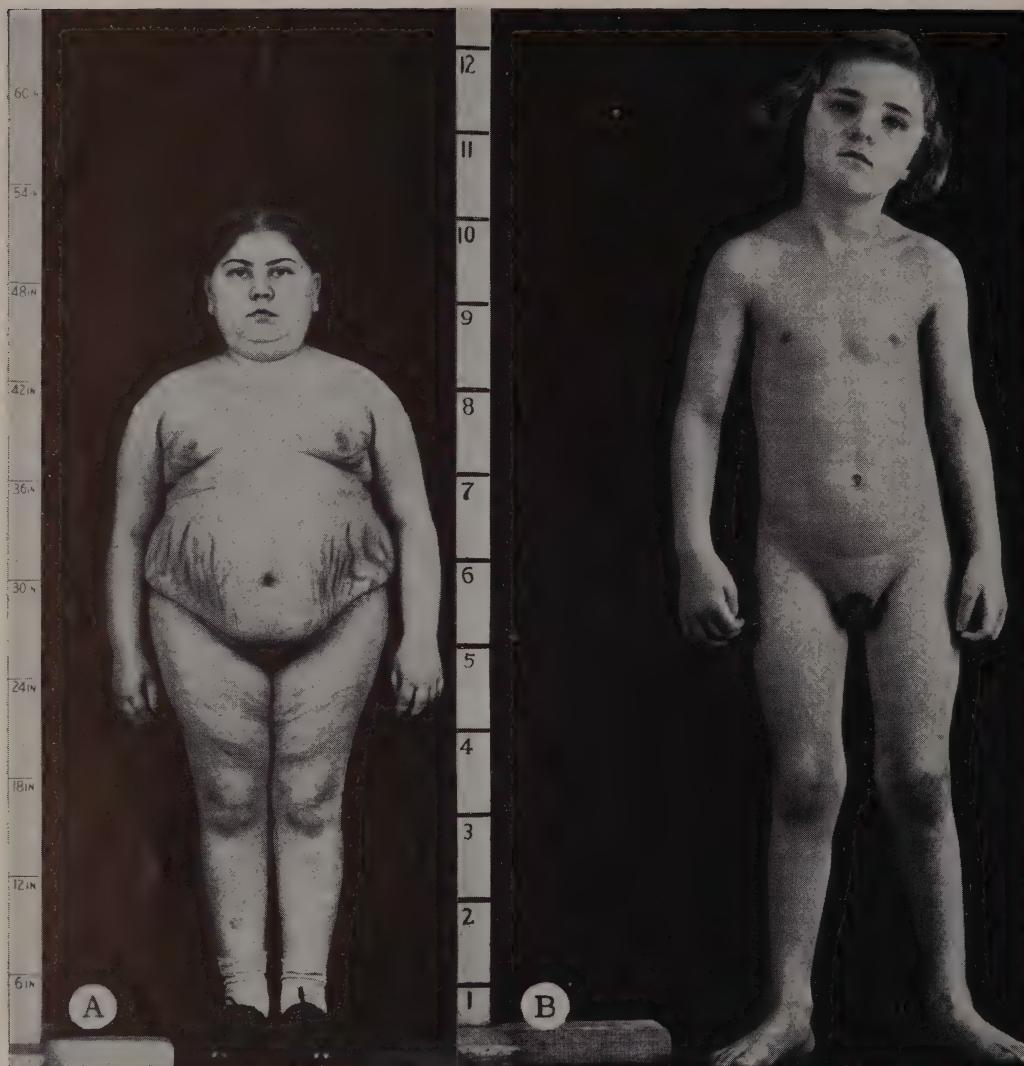
Signs and Symptoms		Diabetes		Hyperthyroidism and Exophthalmus		Hypertension		Nephrosclerosis		Skeletal changes		Splanchomegaly		Adiposity		Muscular strength		Sex organs		Skin		Age at onset		Visual disturbances		Other characteristics		Pituitary lesions	
Pituitary Gigantism	N	N	N	N	Proportionate gigantism	N	N	N or +	N or + later	Large hands and feet, turtling of terminal phalanges, broad bones, osteoporosis of spine with kyphosis, thickened flat bones especially calvarium, large para-nasal sinuses, prognathism, prominent supra-orbital ridges, exostoses.	++	N	First + later	Thickened with writhing; in late stages, often loss of pubic and axillary hair	Adults	++	Hyperphosphatemia	+	Growing children, rarely infants	N	Until late stages patient feels well	Eosinophilic or mixed adenoma; sella large	Other characteristics	Pituitary lesions					
Acromegaly	+	+	+	+	Osteoporosis, especially of spine; collapse of vertebrae; kyphosis	++	"Buffalo" obesity	—	—	Striae (amenorrhea, acne, ecchymoses)	Any age; usually adults	N	First + later	Thickened with writhing; in late stages, often loss of pubic and axillary hair	Adults	++	Hyperphosphatemia	+	Growing children, rarely infants	N	Until late stages patient feels well	Eosinophilic or mixed adenoma; sella large	Other characteristics	Pituitary lesions					
Cushing's Disease	+++	N	++	+++	Osteoporosis, especially of spine; collapse of vertebrae; kyphosis	++	"Buffalo" obesity	—	—	Striae (amenorrhea, acne, ecchymoses)	Any age; usually adults	N	Polycythemia, urinary calculi, high urinary 17-KS, hyalinization of cornea	Basophilic adenoma; sella small	Other characteristics	Pituitary lesions													

+ increase

— decrease

N no change (Normal)

In late stages of all three types of anterior-lobe hyperfunction, secondary break-down of the pituitary tumor may cause hypopituitarism.



Comparison of Cushing's Syndrome and adrenogenital syndrome. — A. Cushing's Syndrome. Patient at 11 years and 10 months, height 52". — B. Adrenogenital Syndrome. Patient at 5 years and 6 months, height 50". Note absence of adiposity and abdominal striae in this patient who, in spite of her age, was almost as tall as the girl shown in Fig. A. (Figs. A. and B. not to scale).

(After Albright et al.: The Harvey Lectures Series, 38, 123, 1942-43.)

tion the summary chart on p. 315, may be of value.

PROGNOSIS

Both pituitary GIGANTISM and ACROMEGALY tend to "burn out" spontaneously; this is often due to liquefaction necrosis of the causative neoplasm. Acromegaly very frequently runs a benign course lasting 30 to 50 years without ever resulting in secondary hypopituitarism. Only occasionally does

the causative adenoma become malignant and result in death due to infiltration and compression of the brain. In most cases the fatal outcome is due to intercurrent infection and final cachexia, or some complication such as diabetic coma, hyperthyroidism, congestive heart failure, etc.

CUSHING'S DISEASE may likewise take a very chronic course with repeated spontaneous remissions. Death is fre-

quently due to complicating, hypertensive cardiovascular disease, renal complications or intercurrent infections.

THERAPY

In pituitary GIGANTISM and ACROMEGALY most physicians consider X-ray treatment of the pituitary region to be the therapy of choice. The eosinophilic cells are claimed to be more sensitive to X-ray than the chromophobes and hence irradiation is more frequently successful in gigantism and acromegaly than in the treatment of chromophobe adenomas. Because of technical difficulties, operation is usually contra-indicated, unless the tumor is very large and begins to encroach upon the optic tracts, thus threatening blindness.

In CUSHING'S DISEASE, X-ray therapy of the pituitary region has also been used with considerable success. Bilateral partial resection of hyperplastic adren-

als or removal of complicating adrenal neoplasms is likewise often followed by temporary or even permanent remissions.

Treatment with folliculoids has been attempted in an effort to inhibit the hormone production of the hypophysis, but this is hardly justified in Cushing's disease, since folliculoids actually increase corticoid production (although they diminish somatotrophin secretion).

Several physicians recommend testosterone for the correction of the metabolic manifestations of Cushing's disease. In animal experiments, testoids tend to cause adrenal-cortical atrophy and to antagonize the nephrosclerosis-producing effect of anterior-pituitary extracts. The usefulness of this therapy in clinical medicine has not been fully verified, but it certainly causes symptomatic improvement.

POSTERIOR-LOBE HYPERFUNCTION

It has not yet been definitely established that excessive hormone production by the posterior-lobe can be the cause of a clinical syndrome. Nevertheless certain instances of DYSTOCIA, OLIGURIA, HYPERTENSION, DYSMENOR-

RHEA, HYPERGLYCEMIA and even the syndrome of eclampsia have at times been regarded as due to excessive production of oxytocin and vasopressin respectively. The evidence in support of these theories is still unconvincing.

INTERMEDIATE TYPES

As previously stated ACROMEGALY AND GIGANTISM may occur in combination, especially if hyperpituitarism commenced before, but continued after, ossification of the junction cartilages. Less frequently, symptoms characteristic of both ACROMEGALY AND CUSHING'S DISEASE manifest themselves in the same patient. It is undoubtedly true therefore that overdosage with anterior-lobe hormones does not necessarily produce clear-cut types of the three main forms of anterior-lobe hyperfunction, since intermediate types are common. Especially noteworthy among these are the forms in which otherwise typical acromegaly or Cushing's disease is associated with hyperthyroidism, hypergonadism, gynecomastia, persistent lacta-

OF HYPERPITUITARISM

tion, or diabetes mellitus, presumably due to excess production of thyrotrophin, gonadotrophins, "mammogenic principle," prolactin, or "diabetogenic principle" respectively.

The existence of a real "PANHYPERPITUITARISM" in which all parts of the pituitary would simultaneously become hyperfunctional has never been demonstrated, but it is not uncommon to observe hyperfunction of the anterior-lobe in combination with deficiency in posterior-lobe-hormone secretion. Thus, there may be acromegaly in combination with diabetes insipidus due to hyperfunction of an anterior-lobe tumor which compresses the posterior-lobe, and hence disturbs its function.

REFERENCES

- ASCHNER, B.: *Ueber die Funktion der Hypophyse*. Arch. f.d. ges. Physiol. **146**, 1 (1912).
- A monograph (147 pages, 50 figures, numerous early references) concerning the first systematic experimental studies on the physiology of the hypophysis. Mainly based on the author's personal observations on hypophsectomized dogs. (In German.)
- ATKINSON, F.-R.-B.: *Acromegaly*. John Bale, Sons & Danielsson, Ltd., London, 1932.
- A book (260 pages, 3 figures, more than 1200 references) in which the symptomatology of 1319 published cases of acromegaly is analytically tabularized according to the presence or absence of thyroid enlargement, kyphosis, albuminuria, glycosuria, etc.
- BERLINGER, W.: *Pathologie und pathologische Morphologie der Hypophyse des Menschen*. Handbuch der inneren Sekretion. Ed. by Max Hirsch, Curt Kabiszsch Publ., Leipzig 1, 910 (1932).
- This is one of the best pertinent reviews now available. (187 pages, 86 excellent illustrations, numerous references). (In German.)
- BRAUN MENENDEZ, EDUARDO: *Infuencia del diencefalo y de la hipofisis sobre la presion arterial*. Tall. Graf. Gasperini & Cia. Publ., Buenos Aires (1934).
- A thesis (75 pages, 14 illustrations, few references) concerning the rôle of the hypothalamo-hypophyseal system upon the blood pressure. (In Spanish.)
- CHAUVET, S.: *L'infantilisme hypophysaire précédé d'une introduction à l'étude des infantilismes et d'une classification des syndromes hypophysaires*. A. Maloine, Publ., Paris (1914).
- A book (333 pages, 68 illustrations, numerous references) concerning the various types of "infantilism" and their relation to pituitary dysfunction. (In French.)
- COLLIN, R.: *L'hypophyse*. G. Thomas, Publ., Nancy (1933).
- A volume (326 pages, 72 illustrations, few references) in which the author reproduces his numerous — mainly histophysiological — publications concerning the pituitary. (In French.)
- CUSHING, HARVEY: *The pituitary body and its disorders. Clinical states produced by disorders of the hypophysis cerebri*. J. B. Lippincott Co., Publ., Philadelphia (1912).
- A book (341 pages, 319 illustrations, 256 references) which undoubtedly represents one of the classics concerning the hypophysis, written by a prominent clinical investigator. Main emphasis is placed upon the clinical manifestations, diagnosis and surgical therapy of hypophyseal diseases. Highly recommended reading, for physicians and students.
- CUSHING, H. AND L.-M. DAVIDOFF: *The pathological findings in four autopsied cases of acromegaly with a discussion of their significance*. Mon. Rockefeller Inst. f.M. Res. No. 22 (1927).
- A monograph (131 pages, 104 figures, 94 references) in which the autopsy findings of four acromegalics are discussed in great detail.
- EVANS, H. M., K. MEYER AND M.-E. SIMPSON: *The growth and gonad-stimulating hormones of the anterior hypophysis*. Mem. Univ. of California **2**, 1 (1933).
- A monograph (446 pages, numerous illustrations and few references) mainly based on the authors' personal experience with the isolation and bioassay of somatotrophin and gonadotrophin. Highly recommended reading.
- FORSSMAN, H.: *On hereditary diabetes insipidus*. Håkan Ohlssons Boktryckeri, Tr. by C. Hannay-King, Lund (1945).
- A monograph (196 pages, numerous charts and tables, 170 references) in which the genetics of diabetes insipidus are surveyed with special emphasis upon the "sex-linked form".
- FRIEDGOOD, H.-B.: *Chapter XIV. Endocrine functions of the Hypophysis*. Oxford University Press, Publ., New York, (1945).
- A monograph (235 pages, 36 figures, numerous references) concerning the normal morphology, histophysiology and biochemistry of the pituitary.
- FUTCHER, PALMER H.: *Giants and dwarfs. A study of the anterior-lobe of the hypophysis*. Harvard University Press, Cambridge, Mass. (1933).
- A booklet (80 pages, no illustrations, 97 references) which gives a brief summary of growth disturbances due to derangements in hypophyseal function in man.
- GOULD, G. M. AND W. L. PYLE: *Anomalies and Curiosities of medicine*. Sydenham, Publ., New York (1937).
- A treatise (968 pages, numerous illustrations, 847 references) discussing various medical curiosities in a very amusing and instructive manner. A special extensive section is devoted to pituitary disorders.
- GUÉNOT, E., K. PONSE AND E. DOTTRENS: *Action physiologique et séparation des hormones auxogène, crinogène et thyroéostimulante de l'hypophyse*. Arch. d'anat., d'histol. et d'embryol. **20**, 15 (1935).

- A monograph (203 pages, 107 illustrations, 36 references) reviewing the biochemistry of anterior-lobe hormones. (In French.)
- HAMBURGER, C.: *Studies on gonadotrophic hormones from the hypophysis and chorionic tissue with special reference to their differences.* Acta path. et microbiol. Scandinav. Suppl. XVII. (1933).
- A doctor's thesis (184 pages, 33 illustrations, numerous references) mainly concerned with the technic and interpretation of gonadotrophin assays in urine and pituitary.
- HENDERSON, W.-R.: *The pituitary adenomata. A follow-up study of the surgical results in 338 cases (Dr. Harvey Cushing's series).* Brit. J. Surg. 26, 809 (1939).
- A very instructive monograph (110 pages, 68 illustrations, 28 references) describing the long-term results of Harvey Cushing's operations for pituitary tumors.
- HUG, E.-L.-J.-A.: *Hipoftisis y Crecimiento.* Imp. Frascoli y Bindi, Buenos Aires (1928).
- A doctor's thesis (147 pages, 11 illustrations, 194 references) concerning the rôle of the pituitary in somatic growth. (In Spanish.)
- KRAUS, E.-J.: *Die Hypophyse. Handbuch der speziellen Pathologischen Anatomie und Histologie.* Ed. by F. Henke und O. Lubarsch, Julius Springer Publ., Berlin 8, 810 (1926).
- A monograph (950 pages, 77 figures, numerous references) concerning the pathologic anatomy of the human hypophysis. An excellent guide to the relevant literature. (In German.)
- LI, C. H. and H. M. EVANS: *Chemistry of Anterior Pituitary Hormones.* In: *The Hormones.* Pincus G. and K. V. Thimann (Ed.). Academic Press Inc., Publ., New York (1948).
- An excellent up-to-date review (62 pages, few figures, numerous tables, 236 references) based on the authors' great personal experience in this field.
- LUCIEN, M., J. PARISOT, G. RICHARD: *Traité d'Endocrinologie — l'Hypophyse.* Gaston Doin & Cie, Ed. Paris, (1934).
- A treatise (686 pages, 96 illustrations, comparatively few references) concerned mainly with clinical, and to a lesser extent with morphologic and physiologic problems relating to the hypophysis. (In French.)
- MARK, L.-P.: *Acromegaly. A personal experience.* Baillière, Tindall & Cox. Publ., London (1912).
- A most instructive book (160 pages, 11 figures, no references) on acromegaly, by a physician who personally suffered from this disease. Main emphasis is placed upon the description of subjective symptoms.
- PAULESCO, N. C.: *Morphologie de l'hypophyse du Cerveau.* Imp. R. Oger, Paris (1907).
- A monograph (144 pages, no illustrations, few references) concerning the morphology and physiology of the pituitary. The book is of historic interest since it is written by one of the early masters in this field and reviews his personal experience with experimental hypophysectomy. (In French.)
- RANSON, S.-W. AND H.-W. MAGOUN: *The Hypothalamus.* Ergebni. d. Physiol. 41, 56 (1939).
- An excellent monograph (107 pages, 35 figures, numerous references) on the morphology and physiology of the hypothalamus. Highly recommended reading.
- RIDDLE, O. ET AL.: *Studies on carbohydrate and fat metabolism.* Carnegie Institution of Washington Publ., Washington D.C. (1947).
- A monograph (128 pages, numerous tables and charts, 203 references) in which the effects of the pituitary hormones upon carbohydrates in metabolism receive special attention.
- ROUSSY, G. AND M. MOSINGER: *Traité de neuro-endocrinologie.* Masson & Cie., Publ., Paris (1946).
- A treatise (1106 pages, 261 illustrations, numerous references) on the histophysiology and pathology of the hypophyseal-hypothalamic system and neuro-humoral correlations in general. (In French.)
- SWEZY, OLIVE: *Ovogenesis and its Relation to the Hypophysis.* The Science Press Printing Co., Lancaster, Pa. (1933).
- A monograph (87 pages, 17 illustrations, few references) concerning the rôle of the hypophysis upon ovogenesis.
- VAN DYKE, H. B.: *The physiology and pharmacology of the pituitary body.* University of Chicago Press, Chicago, Ill. Vol. I (1936), Vol. II (1939).
- A treatise (Vol. 1, 577 pages, 55 illustrations; Vol. 2, 402 pages, 28 illustrations, numerous references) mainly concerning the pharmacology of the pituitary but also discussing physiologic questions. A new edition is about to appear.

IV

THE OVARY

HISTORIC INTRODUCTION

Morphology. — The existence of the ovary, "the female testis" as it was called, has been known to ancient physicians since time immemorial, but ovary and testis were considered to be identical in structure. In 1673, the Dutch physician *Regnerus de Graaf* described small fluid-filled blisters which are visible on the ovarian surface during the fertile period of life. These "Graafian Follicles" were the first of the morphologic details observed in the ovary, merely because they are macroscopically visible. Subsequently, in 1827 the Russian anatomist *von Baer* saw the human ovum. In view of its large size, this structure is on the borderline of naked eye visibility, hence it could be detected with comparative ease, using the primitive microscopes of that era.

Removal and Transplantation of the Ovaries. — The first ovariectomies in women were performed by *H. Hauston* (1701), *MacDowell* (1809) and the famous Georgia surgeon *Robert Battley* (1872) who were severely criticized by their contemporaries for undertaking such a dangerous task. These operations were done to remove ovarian tumors, without realizing that the gland exerts an endocrine function.

In 1896, *Knauer* proved that ovarian TRANSPLANTS may re-awaken the extinguished sexual cycle of spayed animals and concluded that apart from the production of ova, the female gonads exert an important function in regulating estrus phenomena.

Folliculoids. — Subsequently, *Marshall and Jolly* (1906) and *Adler*

(1911) demonstrated the artificial production of estrus in spayed animals receiving aqueous OVARIAN EXTRACTS. Most of these early observations were not very clear-cut however, since ovaries store remarkably little hormone and hence the latter is difficult to demonstrate without using methods of purification and concentration.

In 1912, *Fellner* reported the surprising fact that PLACENTAL EXTRACTS have an effect upon ovariectomized rabbits similar to that of ovarian extracts, probably because the placenta contains ovarian hormones.

Work along these lines was seriously handicapped however by the absence of a convenient BIOASSAY METHOD for the estimation of folliculoid compounds. Prior to 1923, most investigators used the uterus of immature or spayed animals as an indicator of folliculoid activity and hence they had to kill the animals for each test. It was of great importance therefore, when, based on previous observations (*Stockard and Papanicolaou*, 1917; *Long and Evans*, 1920) of regular cyclic variations in the vaginal epithelium of rodents, *Allen and Doisy* (1923) described their well-known test for folliculoid substances. This technic is based on the cornification of the vaginal epithelium of spayed rats and mice produced by treatment with folliculoid compounds. It may be safely said that this simple and accurate test has been the basis for all further work on the purification of folliculoids and acted as one of the greatest stimuli for the rapid development which

has occurred in the field of steroid hormones during the following quarter of a century.

Within a short time *Doisy et al.* (1924) in the United States, and *Laqueur et al.* (1925) in Holland, not only demonstrated folliculoid activity in the fluid of Graafian follicles, but were able to prepare highly active concentrates of it.

The high concentration of folliculoid activity in human PREGNANCY URINE was discovered by *Aschheim and Zondek* (1927) in Germany. Subsequently *Haeussler* (1934) reported the surprising fact that STALLION'S URINE contains about 400 times as much folliculoid activity as that of women and *Zondek* (1935) found that stallion's testes are richer in folliculoid activity than any other tissue, containing about 500 times as much as human ovaries. Thus we learned that ovarian hormones are present in a variety of tissues and are by no means limited to the female sex.

Crystalline ESTRONE was first prepared in the United States by *Doisy et al.* (1929) (who described it under the name of "theelin") and shortly afterwards in Germany by *Butenandt* (1929). ESTRIOL was isolated from the urine of pregnant women by *Marrian* (1930) in England, and was originally referred to as "trihydroxyestrin" or "estrone hydrate."

From human placenta, *Collip* (1930) prepared a crude extract having folliculoid activity; he gave it the name "emmenin" in the belief that it represented a new hormone. *Brown* (1932) isolated a crystalline compound from human placenta, which at that time was believed to differ from estriol in being comparatively less active in spayed rats. This material was vaguely identified with "emmenin." *Butenandt and Browne* (1933) showed, however, that these crystals were pure estriol and that the originally observed differences in biologic potency were due to contamina-

tion with estrone of the early estriol specimens which had been used for comparison. Subsequently *Marrian* isolated estriol-sodium-glucuronide from placenta and it is now generally assumed that the folliculoid activity present in the original crude "emmenin" extracts was due to this substance.

ESTRADIOL, previously known as "dihydroxyestrin" was first obtained by reducing the ketone group of estrone (*Schwenk and Hildebrandt*, 1933). It probably represents the physiologic ovarian folliculoid as it is produced by the gonad and is the most active among the naturally-occurring substances of this kind.

From the urine of pregnant mares, several folliculoids were extracted by *Girard et al.* (1932-36) in France. They all proved to be chemically and physiologically very closely related to the previously known compounds of this type. In order to emphasize their equine origin, they were designated EQUILIN, EQUILENIN and HIPPULIN respectively.

Dodds et al. (1938) in England discovered that "stilbestrol," a stilbene derivative, possesses pronounced folliculoid properties and is highly active by mouth. This opened a new field by showing that compounds other than steroids may exert actions similar to those of steroid hormones.

LUTEIDS. — *Prenant* (1898) in France was the first to call attention to the GLANDULAR APPEARANCE OF THE CORPUS LUTEUM CELLS, a fact which led him to believe that this organ was an endocrine gland. In Germany, *Ludwig Fränkel* (1902) a few years later, showed that in the rabbit, NIDATION OF THE OVUM cannot occur if the corpora lutea are destroyed. This observation gave support to the endocrine theory of the corpus luteum cells. *Bouin*, the well-known histologist of Strasbourg (France) showed, a few years later (1906) that whenever a corpus luteum develops, the endometrium of the rab-

bit undergoes a PROGESTATIONAL TRANSFORMATION assuming the appearance of lace ("dentelle utérine"). Later, in Germany, *Fellner* (1913) and *Herrmann* (1915) were able to produce this same characteristic lace-like endometrium with corpus luteum extracts. A few years earlier in the United States, *Loeb* (1908) had discovered the importance of the corpus luteum in the formation of DECIDUOMAS in guinea-pigs. These are tumor-like structures resembling the maternal placenta, which occur following local trauma in the endometrium but only if corpora lutea are present in the ovary. All these observations helped to support the view that the corpus luteum is a distinct gland of internal secretion.

The important rôle played by the corpus luteum in the MAINTENANCE OF PREGNANCY was demonstrated by *Corner* (1928) who succeeded in maintaining gestation in the spayed rabbit by means of a corpus luteum extract.

Progress along these lines was very slow however, until 1929 when two American anatomists *Corner* and *Allen* described a simple bioassay technic based upon the ability of luteoids to cause progestational transformation in the rabbit.

Using this test and modifications of it, the work of chemists advanced rapidly. CRYSTALLINE CORPUS LUTEUM HORMONE preparations have been described in the United States by *Hisaw et al.* (1930); *Fevold et al.* (1932) and *Allen* (1932) and in Germany by *Fels and Slotta* (1931), but none of these authors gave an adequate description of the physical and chemical characteristics of their crystals. The ketonic properties of the hormone were first recognized by *Butenandt* (1934) and *Hartmann and Wettstein* (1933). The first pure PROGESTERONE crystals were prepared by *Butenandt* (1934) in Germany, and independently, by *Wintersteiner and Allen* (1934) in the United

States. Almost at the same time, *Slotta et al.* (1934) were able to establish the now generally accepted formula of progesterone. The partial SYNTHESIS OF PROGESTERONE (from stigmasterol) has been accomplished by *Butenandt et al.* and *Fernholz* (1934).

The most interesting aspect of this historic survey is the great part played by the above-mentioned two bioassay methods for folliculoid and luteoid activity respectively. These were described within the same decade and made possible an extraordinarily fruitful period of work by chemists who within a few years, isolated and clarified the structure of the most important ovarian hormones. This is all the more noteworthy since before the description of the above tests practically no progress had been made along these lines, in spite of centuries of persistent and industrious work.

It must be realized that a test object for a hormonal activity is of little use unless it is accurate and simple. Tests for ovarian hormone activity were known much before 1923, but since they were too cumbersome and inaccurate, they served merely to demonstrate the possibility of preparing active ovarian extracts. The above-mentioned superior technics had to be developed before chemists could readily check the biologic value of their preparations whenever a new fraction or compound was obtained. This simplicity of assay was instrumental in interesting chemists of distinction to pursue these problems.

PREGNANEDIOL was first prepared from human pregnancy urine by *Marian* (1929) in England. *Venning and Browne* (1937) in Canada, found that the compound is actually eliminated as pregnanediol sodium glucuronide — that is, a sodium salt of the glucuronide — since it is in this form that they were able to isolate this steroid

from human pregnancy urine. Subsequently, in 1938, these same investigators demonstrated that injected progesterone is eliminated in the urine as pregnanediol sodium glucuronide indicating that at least in man, this compound is an important end-product in the metabolism of progesterone. This observation opened the way for the many interesting studies concerning the metabolism of progesterone which were performed using pregnanediol elimination as an indicator.

Gonadotrophins. — The fact that the ovary is dependent upon certain trophic stimuli had been foreseen as early as 1914 by the Danish investigator, Knud Sand, who spoke of "X-SUBSTANCES" as indispensable for gonadal development and function. A. Lipschütz (1925) found that ovarian grafts grow better in spayed than in intact animals, hence he also concluded that extragonadal trophic influences are important regulators of ovarian growth. As previously stated in the chapter concerned with the history of pituitary research, this conception received ample confirmation later through the demonstration of the pituitary gonadotropic hormones.

Ovarian Diseases. — Female HYPOGONADISM as well as various types of MENSTRUAL ANOMALIES, were known to the physicians and laymen of antiquity.

Parallel with the previously mentioned experimental observations, it gradually became clear that the folliculoids secreted by the ovarian follicle are responsible for estrus in animals and post-menstrual changes in women, while the luteoids, coming from the corpus luteum, cause pregestational transformation of the endometrium and prepare the uterus for the nidation of an ovum. Menstruation was recognized as due to sudden withdrawal of these ovarian hormones at the end of the sexual cycle

(Fränkel, 1910; R. Meyer, 1913; Schröder, 1913; Novak, 1921; etc.).

A great deal can be learned from the spontaneous experiments which nature performs when one or the other ovarian structure is hypofunctional or selectively proliferates in an exaggerated manner. In the ovary, true neoplasms are not always readily distinguishable from other types of abnormal growths. They will be discussed here conjointly, grouped according to the type of clinical syndrome they provoke. In this manner we shall see how the ovarian diseases helped us to recognize the cell types which are responsible for the production of the different hormonal principles.

Complete primary APLASIA or AGENESIS of the ovaries was repeatedly reported in the early literature, but it was not until recently that Turner (1938) and Albright et al. (1942) clearly recognized that the clinical syndrome associated with this malformation is characterized by short stature, inhibition of sexual development, high urinary gonadotrophin titers and congenital anomalies, such as coarctation of the aorta, webbing of the neck, etc.

AMBISEXUALITY has raised a great deal of interest, ever since the earliest periods of recorded history. Hermaphroditos, the ambisexual "son" of Aphrodite and Hermes, was a beautiful, sacred and rather romantic figure in early Greek mythology. Some of the most distinguished Hebrew writers interpret the first chapter of Genesis as describing Adam as being of both sexes. Be this as it may, Theophrastos (372-287 B.C.) in his "Characters" describes ambisexual human beings to whom the name "hermaphrodite" is now assigned.

Most of the ambisexual people are PSEUDOHERMAPHRODITIC, that is, they only possess the gonads of one sex, but have both male and female accessory sex characteristics. Probably the first

instance of a TRUE HERMAPHRODITE, that is to say, a person having both testicular and ovarian tissue, was described by *Klotz* in 1879.

It is interesting to note that in view of recent observations, the whole conception of ambisexuality as an essentially abnormal process has to be revised. We know now that "female" sex hormones, such as folliculoids, are produced by the testis as well as by the ovary and that even certain hormones themselves (e.g., ethynodiol-diethylstilbestrol) are ambisexual since they stimulate both male and female accessory sex characteristics. It becomes increasingly more evident that a certain degree of ambisexuality is normal and that only the extremes of this condition are pathologic.

TUMORS OF THE OVARY have interested physicians since time immemorial, because of the enormous size which they may attain and because of their great morphologic and functional diversity. Here we shall mainly consider the history of those tumors, and related abnormal growths, which cause or are caused by endocrine disturbances.

The SIMPLE FOLLICLE CYST was first differentiated from other ovarian cysts by *Rudolph Virchow* (1848). It was not until the beginning of the 20th Century, however, that this distinction was supported by functional evidence of a resulting HYPERFOLLICULOIDISM, especially metropathia hemorrhagica (*Robert Meyer; Schröder*).

v. Rokitansky (1855) is usually credited with having described the earliest typical case of OVARIAN SMALL-CYSTIC DEGENERATION; however, his patient suffered from a hydatidiform mole and the ovarian lesion was only secondary. The first characteristic cases, associated with menstrual disturbances and other signs of hyperfolliculoidism, were reported towards the end of the 19th Century (*Bulius; Petit, etc.*).

A typical OVARIAN FOLLICULOMA was apparently described by *v. Rokitansky* in 1859 although he did not specifically classify the tumor *v. Kahlden* (1895) deserves credit for first distinguishing as a separate entity, an "adenoma of the Graafian follicle," containing folliculoid and cylindromatous areas which produced hyperfolliculoidism.

METROPATHIA HEMORRHAGICA was described by *Olshausen* (1875) under the name of "endometritis fungosa." It was subsequently shown by *Schröder* (1915) that this is not an inflammatory disease. He gave it the now generally accepted name "metropathia hemorrhagica."

Hewitt (1857) was the first to describe "Ovaries showing false corpora lutea with commencing cystic disease," but it was only much later (*Halban*, 1915; *Fraenkel*, 1922) that the corresponding pregnancy-like clinical syndrome was ascribed to the PERSISTENT CYSTIC CORPORA LUTEA.

v. Rokitansky (1859) was probably the first to describe a LIPID CELL TUMOR in the ovary. Later *Peham* (1899) attempted a detailed analysis of such a case both clinically and morphologically. He emphasized the resemblance of the growth to the zona glomerulosa of the adrenal cortex and called attention to the high glycogen content of its cells.

TUBULAR ADENOMAS of the ovary, frequently associated with virilism, have been known for a long time. *Dick* (1905) described them under the name of "adenoma tubulare, testiculare ovarii," *Robert Meyer* (1914-30) first referred to them as "adenoma tubulare ovarii," subsequently as "andreiblastomas," and still later, as "arrhenoblastomas." He deserves credit for having clarified many points in connection with their morphology and pathogenesis.

The "SEMINOMAS" of the testis were first described by *Chevassu* (1906). He recognized that, contrary to the opinion of earlier workers, these tumors

are not alveolar sarcomas, but represent a distinct type resembling certain ovarian neoplasms, which were subsequently described by *Masson* (1912) as "seminomas of the ovary," *Strong* (1919) spoke of them as "embryonal carcinomas"; the term "dysgerminoma" was proposed by *Meyer* (1930) because he claimed that the tumors arise from the germinal epithelium which loses its "germinal" properties as a result of some derangement occurring before its differentiation into ovarian or testicular tissue. The recent discovery of true seminomas of the testis, which proved to be essentially different from all ovarian tumors, clearly shows that the "dysgerminoma" is a false seminoma. (See : False seminomas.)

The STRUMA OVARI was usually misinterpreted by pathologists who found it very difficult to understand that true thyroid tissue could occur within the ovary. Thus tumors now known to belong to this group were diagnosed as "folliculoma malignum," a type of "endothelioma," or ovarian metastases of undetectable primary thyroid growth.

Thyroid tissue in the ovary had been described by several early investigators, but it was *Pick* (1920) who recognized that these growths are actually teratoids in which the thyroid tissue predominates to such an extent that

all other constituents are "crowded out" by it. This interpretation is further supported by the many intermediate types between ordinary embryomas with small nodules of thyroid tissue and tumors consisting exclusively of the latter.

The CHORIONEPITHELIOMA of the ovary was originally described in 1900 by *Kaufmann* but it was only as a result of considerable additional work that it was definitely recognized as a placental neoplasm.

Probably many cases of "vicarious menstruation" — known to physicians and laymen since the early ages — are the result of bleeding from an ectopic endometrium. The same is true of the "tarry" or "chocolate cysts" in the ovary and the pelvic peritoneum, frequently described early in the nineteenth century. v. *Rokitansky* (1860) was probably the earliest investigator to recognize internal ENDOMETRIOSIS as an independent form of disease. *Russell* (1899) was the first, however, to describe endometrial tissue in the ovary. *Sampson's* (1921) classic investigations convinced most workers that at least in some cases, if not in all, ectopic endometrium grows in the pelvis because it has been regurgitated there through the oviducts at the time of menstruation.

NORMAL MORPHOLOGY

ANATOMY

In man, the ovaries are paired, almond-shaped bodies situated on either side of the uterus, near the lateral wall of the pelvis. They are attached to the back of the broad ligament, behind and somewhat caudad from the uterine tubes. Their color is greyish-pink and their surface smooth or, especially in older women, puckered by numerous small scars. During the fertile period of life, small fluid-filled blisters are distinguishable on the ova-

rian surface; these are the follicles of de Graaf.

The two ovaries are approximately of equal size, measuring about 4 cm. in length, 2 cm. in width and 8 mm. in thickness. They weigh 2.0-3.5 gm. each. In its natural location, the ovary is so oriented that its upper pole is near the free end of the Fallopian tube, the lower pole or uterine extremity is connected with the origin of the tube from the uterus. The tubal pole lies near the external iliac vein, attached to the ova-

rian fimbria of the oviduct and a fold of peritoneum, the SUSPENSORY LIGAMENT of the ovary. The uterine pole is connected with the uterus near the origin of the oviduct by a rounded solid cord, the OVARIAN LIGAMENT. The latter is enclosed within the broad ligament and contains smooth muscle fibers. Between the two poles is the mesovarian border of the ovary, which is attached to the dorsal lamina of the broad ligament by a short fold, the MESOVARIUM. It is through this fold that the ovarian nerves, vessels and lymphatics pass.

The organ is enclosed in the OVARIAN FOSSA on the lateral wall of the pelvis. This pocket is delimited by the external iliac vessels above, the obliterated umbilical artery in front and the ureter behind.

Near the ovary are two vestigial structures :

(1) The EPOÖPHORON (organ of Rosenmüller; parovarium) which lies in the mesosalpinx between the ovary and the oviduct. It consists merely of a few short tubules which converge towards the ovary, while their opposite ends open into a rudimentary longitudinal duct (duct of Gartner).

(2) The PAROÖPHORON, which consists of irregular rudimentary tubules in the broad ligament between epoo-phoron and uterus. Usually these tubules are well-developed only in children.

HISTOLOGY

The ovary consists of a peripheral CORTEX which contains the various stages of maturing follicles, corpora lutea and their scars, and of a MEDULLA consisting of connective tissue stroma, blood vessels, lymphatics and nerves.

The Germinal Epithelium. — This is a simple or stratified lining of cuboidal or columnar cells separated from the underlying ovarian tissue by a basement membrane. Between these undifferentiated cells are found at irregular inter-

vals large round cells, the primordial sex cells. In the embryo, and perhaps to some very slight extent even during early postnatal life, this epithelium gives rise to ova which invade the underlying tissue. In adults it becomes gradually flattened and increasingly similar to the ordinary peritoneal mesothelium. Strictly speaking, the ovary possesses no capsule although the egg-free surface layer of dense stroma is often described as such under the name of TUNICA ALBUGINEA.

The Follicles. — Scattered throughout the ovarian cortex are about 500,000 FOLLICLES in various stages of maturation. Ordinarily only one of these reaches full maturity during each menstrual cycle and most of them undergo degeneration without ever having fully developed. Hence in the adult, the number of follicles gradually decreases and in senile women, very few, if any, persist.

The most immature stage in the process of maturation is represented by the PRIMARY FOLLICLES. The vast majority of the ova in the normal adult ovary are stored in such immature follicles, only a few maturing at any one time. The primary follicles, most numerous in the periphery of the gonad, are roundish bodies of about 45μ in diameter. They consist of a centrally-located large, roundish, egg cell or ovum surrounded by a single layer of flat or cuboidal so-called *follicular or granulosa cells*. The latter are so poor in cytoplasm that early histologists described them merely as granules; hence the term "granulosa layer."

The GROWING FOLLICLES enlarge chiefly through the proliferation of the follicular cells. The originally single granulosa-layer becomes stratified, while the ovum reaches a diameter of $60-80\mu$ and surrounds itself with a thick cell-membrane, the *zona pellucida*, so called because of its semi-transparent aspect.

In the granulosa, certain cells degenerate and droplets of fluid accumulate around them. The deeply basophilic, degenerating granulosa cells are referred to as *Call-Exner* bodies, while the fluid droplets represent the first stages in the secretion of the so-called *follicular fluid or liquor folliculi*. Due to the confluence of several of these droplets and the ever increasing amount of liquor folliculi, a cyst-like cavity, the *antrum folliculi*, is formed between the granulosa cells when the follicle reaches a diameter of about 0.2 mm. The fluid pushes the ovum with some surrounding granulosa cells to one side, so that the egg is enclosed in a small hillock, the *cumulus oöphorus* or *discus proligerus*, which protrudes into the follicular fluid.

During this growth, the connective tissue surrounding the granulosa cells differentiates into a capsule, the *theca folliculi*, separated from the granulosa by a homogeneous *basement membrane*. Within the theca, we distinguish : (1) an internal layer (*theca interna*) immediately surrounding the basement membrane and containing many capillary vessels, and (2) an outer layer (*theca externa*) consisting of spindle-shaped, concentrically arranged cells and connective tissue fibers. In many animal species the theca does not differentiate into these two layers and even in man the borderline, between the external and internal layers, is indistinct.

The further maturation of the follicles is chiefly due to the accumulation of increasing amounts of follicular fluid. The MATURE OR GRAAFIAN FOLLICLES measure about 10 mm. in diameter and are fluid-filled cysts, lined by a thin layer of granulosa cells; within this lining is the cumulus oöphorus with the ovum. At this stage, the follicles reach the ovarian surface and bulge outward into the peritoneal cavity preparatory to rupture. The immature follicles tend to migrate towards the sur-

face in the course of their maturation, partly owing to their increasing size and partly by actual outward movement, perhaps due to traction by some of the theca cells. On that pole of the follicle which lies closest to the ovarian surface, a few theca cells, the "*theca interna cone*" of Strassman, develop a definite tropism for the ovarian surface and actually infiltrate through the stroma in the direction of the tunica albuginea. It is assumed that the granulosa secondarily protrudes into the loose connective tissue of this cone, thus gradually reaching the surface.

In the fully-developed mature follicle, the *granulosa cells* have a diameter of about 10μ . They contain but little cytoplasm and a nucleus with 1-3 nucleoli. At any time many granulosa cells are found in the process of mitosis. The follicular cells of the cumulus oöphorus region assume a columnar, elongated and polyhedral shape, their longitudinal axes taking a radial position from the center of the ovum. This region consisting of 12-20 layers of granulosa cells is called the *corona radiata*. It is attached to the zona pellucida by radial fibrils. The *ovum* of the mature follicle has a diameter of 120μ ; it is the largest cell in the human body. Its eccentrically-located nucleus measures 25μ in diameter and has a thick membrane and a large nucleolus, the "*macula germinativa*." Its cytoplasm contains some yolk granules, but the mammalian ovum is much poorer in yolk than that of oviparous animals (e.g., birds).

Eventually the superficial, bulging membrane of the mature follicle becomes extremely thin and *ovulation* occurs upon its rupture. At this time the follicular fluid and the ovum, together with the surrounding corona radiata and discus proligerus, are ejected into the peritoneal cavity or into the contiguous ostium of the oviduct. It is believed that the granulosa cells of the cumulus oöphorus furnish nourish-

ment to the young ovum until implantation. In some animals, ovulation occurs only after mating; in others, including man, it takes place at regular intervals. (See : Estrus and Menstruation.)

Preparatory to fertilization, the ova have to undergo a process of MATURATION. In the adult mammalian ovary, the ova are primary ovocytes, corresponding to the primary spermatocytes of the male. After completion of their growth, these undergo two successive maturation divisions, the resulting four cells having only half the number of chromosomes of the original primary ovocytes (haploid number).

The *first maturation division* takes place immediately before or after ovulation and divides the nuclear chromatin evenly, but one of the daughter cells receives practically the entire cytoplasm and forms the secondary ovocyte (corresponding to the secondary spermatoocyte in the male), the other is a minute granule, the *first polar body*, which subsequently divides again and eventually degenerates. Immediately after elimination of the first polar body, the secondary ovocyte undergoes a *second maturation division*, the spindle of which remains in the metaphase until fertilization is completed. The chromatin is again equally divided, but (each daughter cell contains only half the number of chromosomes present in the mother cell) the cytoplasm remains with one daughter cell, that which forms the ovum (corresponding to the spermatid of the male); the other minute abortive cell is the *second polar body*. As a general rule the halving of the chromosomes, or "miosis," occurs during the first maturation division, although in some animals it takes place only when the second polar body is formed.

The number of chromosomes varies in the different species, but two of them are slightly different from the rest in their shape; these determine sex. In man, there are forty-six somatic and two sex chromosomes. In the human

ovum, both sex-chromosomes are similar (X-chromosomes), while in the spermia, they are different, only one resembling the female type (X and Y-chromosomes). At the time of fertilization, the final mature gonocyte contains only one of the sex-chromosomes, after the haploid division. In ova, this is necessarily an X-chromosome, while in spermia, it can be either X or Y. It is evident that following the union of the male and female gonocytes, the resulting cell will either have the X + X or the X + Y constitution, depending upon which paternal sex-chromosome was added to the maternal chromosome. Hence, the father can only blame himself for his offspring's sex.

Comparatively little is known about the *migration of the ovum* from the ovary into the uterus. It is assumed that unfertilized human ova remain viable only for about 24 hours after ovulation, so that migration into the tube, where fertilization normally occurs, must be rapid. Opinions differ concerning the relative rôle played by the musculature and the epithelial cilia of the tube in promoting the downward migration of the ovum.

FOLLICULAR ATRESIA is a degenerative phenomenon which affects a great many follicles before they mature sufficiently to produce a fertile ovum. It is estimated that in a normal human ovary there are about 20,000 atretic follicles at any time. It has been pointed out that even in a woman whose reproductive life lasts 35 years and who discharges an ovum every month throughout this period, only 420 follicles can mature to the point of ovulation. Since, on the other hand, all of the approximately 500,000 ova of infancy disappear during the course of life, it is obvious that the vast majority of the follicles degenerate before reaching maturity.

Follicular atresia commences with the degeneration of the granulosa cells and

the ovum. Their place can be occupied by invading connective tissue (*obliterative atresia*), so that a hyaline scar-like tissue is formed; or by liquid so that a cyst results (*cystic atresia*). The former type more frequently affects the small, the latter the large follicles. Sometimes parthenogenetic egg-divisions accompany the phenomenon of atresia. The basement membrane between theca and granulosa becomes thick and assumes a hyaline, glass-like aspect during atresia. This so-called "*glassy membrane*," tends to persist much longer than other constituents of the follicle. Some histologists like to reserve the term "*corpus nigricans*" for the blood-pigment-containing, and "*corpus albicans*" for the pigment-free, corpus luteum scars; the atretic follicle being designated as "*corpus candidans*." In common usage the adjectives *candidans* and *albicans* (both of which mean white) are employed interchangeably for the pigment-free scars of either corpora lutea or follicles. This is especially justified in the case of old scars in which degenerated ova are no longer visible so that it is not possible to differentiate between follicle and corpus luteum remnants.

In certain animal species, and even in man, we encounter POLYOVULAR FOLLICLES which contain several ova, or ANOVULAR FOLLICLES in which there are no ova. The latter can arise due to the formation of follicles without germinal cells or due to subsequent selective degeneration of the ova in follicles which were originally normal. Although the ovum is essential for the normal maturation of the follicle, the existence of anovular follicles shows that granulosa and theca can develop to some extent without the egg-cell.

The Corpus Luteum.—Immediately after the discharge of the ovum with its cumulus oöphorus, the remaining granulosa cells, which line the cavity of the follicle, undergo considerable hypertrophy. Both their nucleus and

cytoplasm are enlarged. There is also some hyperplasia, but judged by the scarcity of mitotic figures at this time, formation of new cells plays a secondary rôle in increasing the corpus luteum mass. The transformed granulosa layer is thrown into folds, since it becomes too large to fit smoothly into the follicular cavity. Hence, the cavity of the follicle becomes star-shaped when viewed in cross section. At the same time, the theca interna cells undergo a similar process of hypertrophy and transformation into corpus luteum cells. From the outside they invade the spaces between the granulosa-layer folds and bring blood vessels into the granulosa, which originally had no vascular supply. At a later stage, the differentiation between "granulosa corpus luteum cells" and "theca corpus luteum cells" or "paraluteal cells" may become impossible, although for a certain time, those derived from the theca are somewhat darker and poorer in lipids than those derived from the granulosa. By far the greater part of the mature corpus luteum is of granulosa-cell origin.

The typical CORPUS LUTEUM CELL differs from the granulosa cell in that it is larger, and has a well-developed, polyhedral cytoplasm which contains numerous lipid granules, cholesterol esters, granules of vitamins A and C as well as the lutein, a pigment which gives this structure its characteristic yellow color. The nucleus becomes vesicular and possesses a coarse chromatin network with one or two nucleoli. The corpus luteum cells strikingly resemble those of the adrenal cortex and also to some extent, the Leydig cells of the testis. This morphologic similarity is all the more noteworthy since these three types of cells produce chemically closely allied steroid hormones.

The CAVITY of the original follicle gradually becomes organized from the theca, which eventually penetrates across the granulosa to the central cavity. Here it forms a loose gelatinous

connective tissue which covers the inner surface of the cyst wall. It leaves some free space in the center which is filled with remains of liquor folliculi, serum and usually some decomposing erythrocytes, remnants of the minute hemorrhages which often accompany the process of ovulation.

The STROMA of the corpus luteum consists of a fine reticular network, in which sinusoids course radially from the periphery towards the center, between the corpus luteum cells.

Many histologists strictly differentiate between the various STAGES IN THE PROCESS OF CORPUS LUTEUM FORMATION. There is a stage of proliferation and hyperemia immediately following ovulation. As the name implies, this is characterized mainly by the proliferation and vascularization of the granulosa-cells accompanying the invasion of theca cells.

Secondly, there is the stage of full development, in the case of a regular 4-weeks-cycle, between the 17th and 26th day after the beginning of the last menstrual period. Macroscopically, this is characterized by the presence of a glassy, jelly-like coagulum in the central cavity, with a thin, red zone of incrotary cells surrounding it. At this time the folded glandular layer measures only about 250 to 350 μ in thickness. Histologically, the prevalent cellular element is the mature corpus luteum cell with a diameter of 25-40 μ and a cytoplasm containing very fine, dust-like lipid granules. In many animal species, the follicular rupture point (the "Blutpunkt" of the German investigators) remains visible on the surface of the corpus luteum in the form of a conically elevated point covered by a blood coagulum.

The third phase in the life of the corpus luteum is the stage of involution. It begins a few days before the menstrual bleeding and proceeds so rapidly

that within 14 days after menstruation, the diameter of the corpus luteum decreases to a few mm. and after six weeks, it is of microscopic size. At first the lipid granules, in the involuting corpus luteum cells, become coarse (storage instead of secretion?), but later they disappear completely and eventually connective tissue replaces the epithelium. Small hemoglobin-pigment-containing, hyalinized connective tissue scars persist for years after the involution of the corpus luteum. These correspond to the scars of atretic follicles. Thus in one young woman, who died 5 years after puberty, the number of corpora albicantia found upon histologic examination, corresponded to the number of her menstrual cycles.

The corpus luteum of pregnancy and that of pseudopregnancy (the latter develops only in certain animal species) are larger and persist for a longer time than those of menstruation. (See : Pregnancy, Lactation, Nervous Stimuli, on pp. 334, 381, 383.)

Stroma. — The CONNECTIVE TISSUE stroma of the human ovary consists of a network of reticular, elastic and collagenous fibers with fusiform cells resembling fibroblasts. True smooth muscle cells have also been described especially in the theca externa of the follicles in certain animal species (e.g., sow). The medulla is composed chiefly of loose connective tissue with elastic fibers and strands of smooth muscle cells accompanying the blood vessels.

Among the common fibroblasts are some, the so-called "INTERSTITIAL CELLS" of the ovary, which under ordinary conditions may not be distinguishable from fibroblasts. However, gonadotrophic hormones stimulate these cells selectively, so that they assume an epithelioid appearance and produce ovarian hormones (see below). In certain animals (e.g., rabbit) these cells are extraordinarily numerous.

They are probably derived from the theca of atretic follicles and in a certain sense are homologous to the interstitial cells of the testis. However, the sympatheticotropic cells (see below) are even more closely related to the testicular Leydig cells. The entire system of interstitial cells is sometimes referred to as the "interstitial gland" or "puberty gland" of the ovary.

In the hilus of the ovary or the mesovarium, certain nests of large, epithelioid cells are found in intimate contact with bundles of non-myelinated nerve fibers. They have been termed SYMPATHICOTROPIC CELLS (*Berger*); extraglandular, interstitial or Leydig cells of the ovary (*Kohn*); Berger cells or hilus cells. Their number and size are subject to great variations. They do not give typical chromaffin reactions and hence should not be considered as paraganglionic elements, but rather as the ovarian equivalents of the testicular Leydig cells. Sometimes their cytoplasm contains lipid and pigment granules as well as typical Reinke-crystalloids, similar to those found in Leydig cells. Their relationship to the latter is also indicated by the fact that their excessive development may lead to virilization.

True CHROMAFFIN CELLS, similar to those of paraganglia, are sometimes also found in the ovarian hilus, but these are essentially different from the "hilus cells."

WALTHARD'S RESTS are cystic or solid structures occurring in the cortex or hilus of the ovary, usually just beneath the lining epithelium. They probably arise from the celomic mesothelium, and may be the primordia from which certain ovarian tumors (Brenner tumors, pseudomucinous cystadenomas) develop. Under normal conditions, they do not seem to have any physiologic function.

Blood Vessels. — The ARTERIES of the ovary are 6-8 small branches orig-

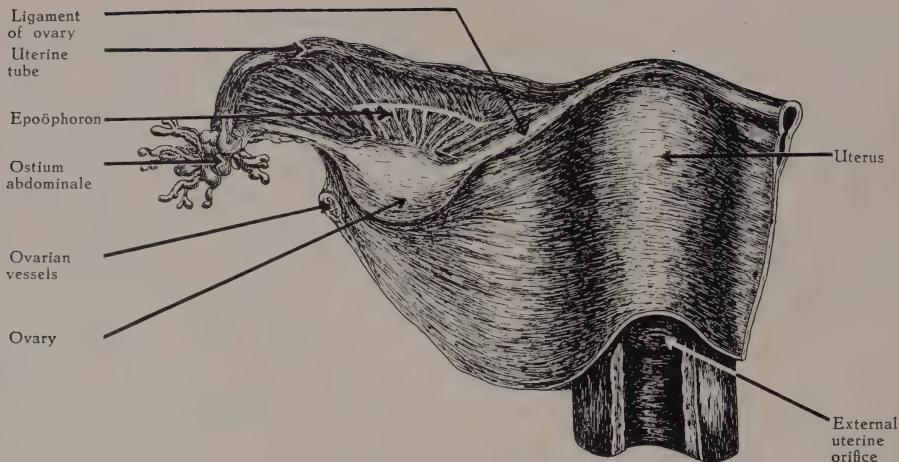
inating from an anastomosis of the uterine and ovarian arteries. They enter the gonad through the mesovarium, forming many convolutions. From the hilum, they proceed radially towards the periphery of the organ and split up into capillary networks around the follicles, penetrating the theca, but not the granulosa layer. The corpora lutea are richly supplied with blood vessels in all stages of their development.

The ovarian VEINS course from the perifollicular and stroma capillaries towards the hilus, where they form the cavernous-body-like ovarian plexus.

After puberty, a process of PHYSIOLOGIC SCLEROSIS takes place in the ovarian arteries, apparently due to the stress of constant changes in the nutritive requirements of the ovary during the menstrual cycles, pregnancies, etc. This has variously been referred to as "ovulation sclerosis," "menstrual sclerosis," "partial sclerosis" or "pregnancy sclerosis."

The LYMPHATICS are likewise very well developed in the ovary. Lymphatic capillaries are especially plentiful in the theca externa of the follicles and occur also in corpora lutea and albicanitia. The theca interna, granulosa and the albuginea are free of lymphatics.

Nerves. — The sympathetic nerves of the ovarian plexus originate from a ganglionic group which anastomoses with the celiac, renal and mesenteric ganglia. The nerves reach the hilum after a corkscrew-like course through the mesovarium. Then they proceed towards the periphery and distribute their branches to the vessels, stroma, theca and corpora lutea. The granulosa is free of nerves. Sensory fibers ending in Pacini-corpuscles have also been described in the stroma. The existence of a special ovarian "sympathetic ganglion" is doubtful; the ganglion-cell-like elements described by some investigators were probably hilus cells.



Posterior aspect of internal female sex organs



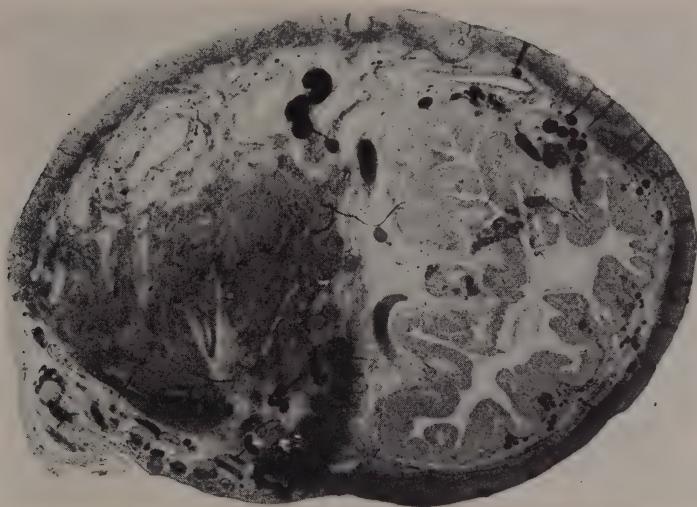
A



B

Ovary of newborn (Man). — A. Cortex of ovary under low magnification. Note irregularities of surface with invaginations of germinal epithelium. The large egg-cells are very prominent, but have not yet surrounded themselves with a stratified granulosa layer and no mature follicles are visible. — B. Same section under higher magnification. Note large vesicular nuclei in egg-cells and surrounding stroma. Differentiation of granulosa and theca not yet evident.

(Courtesy of Dr. W. Bonin.)



Corpus luteum of the menstrual cycle (Man). — Corpus luteum a few days prior to menstruation. Note that entire right part of the ovary is occupied by the highly corrugated corpus luteum. The hilum is at the left.

(Courtesy of Dr. W. Bonin.)



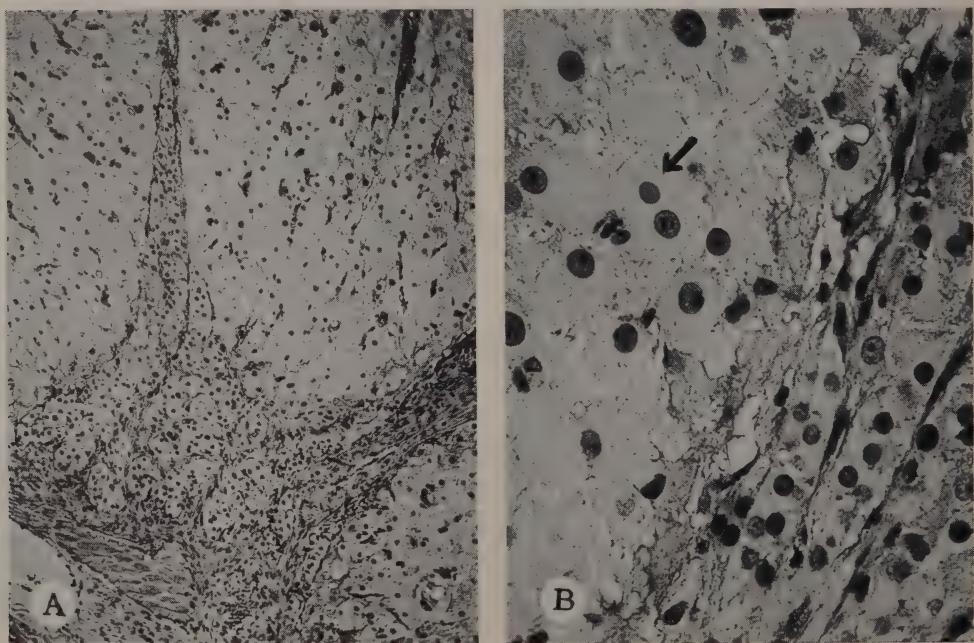
Corpus luteum of the menstrual cycle (Man). — A. Light, large, granulosa corpus-luteum-cells which form a convoluted layer. Surrounding them and between the convolutions, the stroma contains smaller, dark thecal corpus-luteum-cells. The central cavity is partly organized by loose connective tissue (very light). — B. Borderline between small (theca) and large (granulosa) corpus-luteum-cells. The latter contain no hyaline inclusions (such as are supposedly characteristic of gestation).

(Courtesy of Dr. W. Bonin.)



Corpus luteum of pregnancy (Man). — Corpus luteum during first month of pregnancy. Note corrugated surface of corpus luteum wall and large cavity filled with colloid. The remaining ovarian tissue is atrophic and contains only a few medium-sized follicles. The hilum is at the left. (Very low magnification.)

(Courtesy of Dr. W. Bonin.)



Corpus luteum of pregnancy (Man). — A. Note marked difference between large, granulosa corpus-luteum-cells and small, thecal corpus-luteum-cells. The latter fill out the triangular space between the convolutions of the former. — B. Higher magnification of the borderline between thecal and granulosa corpus-luteum-cells. Note dark homogeneous hyaline inclusion in one of the granulosa corpus-luteum-cells (arrow). These are allegedly characteristic of gestation.

(Courtesy of Dr. W. Bonin.)

COMPARATIVE MORPHOLOGY

Demonstrably endocrine cells have not been proven to exist in the ovaries of INVERTEBRATES. Large, so-called interstitial cells persist after discharge of the egg in the ovarian region of some species (e.g., hydra) but their function is not known.

In certain viviparous FISH the spent follicles bear some resemblance to the mammalian corpus luteum, but their possible endocrine function has not yet been adequately studied.

Morphologic similarities between certain cell-groups in AMPHIBIAN ovaries and endocrine elements are of doubtful physiologic significance. In some toads there is a small structure, *Bidder's organ*, in the vicinity of the ovary. Morphologically it resembles ovarian tissue and after ovariectomy it can substitute for the loss of ovarian incretions.

Among the REPTILES, certain viviparous Brazilian snakes (*crotalus terrificus*, *bothrops jararaca*) form corpora lutea whose physiologic function is clearly shown by the fact that their removal causes abortion, preventable by luteoids.

In most genera of BIRDS, ovulation occurs only from the left gonad, while the right ovary is rudimentary and contains testis-like elements. On the other hand in the hawk, falcon and many other birds of prey, both ovaries are normally developed and participate in egg production, although the right gonad may be somewhat smaller than the left. Certain cell-proliferations in spent follicles resemble corpora lutea, but are of questionable endocrinologic significance.

It must be kept in mind that the main object of the corpus luteum is to prepare the endometrium for the reception and maintenance of an embryo. Oviparous species (e.g., birds), which produce eggs containing large amounts of yolk, do not require such maternal trophic influences since nourishment is derived from the yolk-sac. There are

close functional analogies between the egg-yolk and the corpus luteum.

The ovaries of all MAMMALS are essentially similar; they have follicles, corpora lutea and varying amounts of interstitial-cell tissue. The number of corpora lutea formed at each cycle differs in the various species: in man, usually only one, rarely two or more, ova are liberated at each cycle; in the rat, the number of ovulating follicles is about 8; in one curious, small South African mammal "elephantulus myurus jamesoni" up to 60 or more. Elephantulus is also noteworthy for having corpora lutea of an unusual type since the granulosa is everted through the rupture point. This process occurs only as a rare anomaly in women and is referred to as "corpus luteum prolapse."

EMBRYOLOGY

In man, the primordium of the gonad can first be distinguished in a ridge called the UROGENITAL FOLD. The latter contains both the genital and the mesonephric (kidney) primordia. The germinal epithelium covering this region continually thickens and protrudes into the celomic cavity mediad to the gradually separating mesonephric fold. At the same time (as early as the 3.5 mm. stage), in the caudal end of the embryo, large cells appear which migrate through the dorsal mesentery into the genital fold. The nuclear structure of these so-called primitive or PRIMORDIAL GERM CELLS reveals that even those which are still outside the gonad are true sex cells. It has been thought that these are the source of all definitive germ cells, but most contemporary embryologists believe that the definite ova arise directly from the sex cells of the germinal epithelium and that the primordial germ cells degenerate.

In the six-week-old human embryo, sex can not yet be identified. The surface of the gonad is covered by a germinal epithelium, while most of its substance consists of an inner epithelial

mass derived from an ingrowth, the so-called "first proliferation," of germinal epithelium.

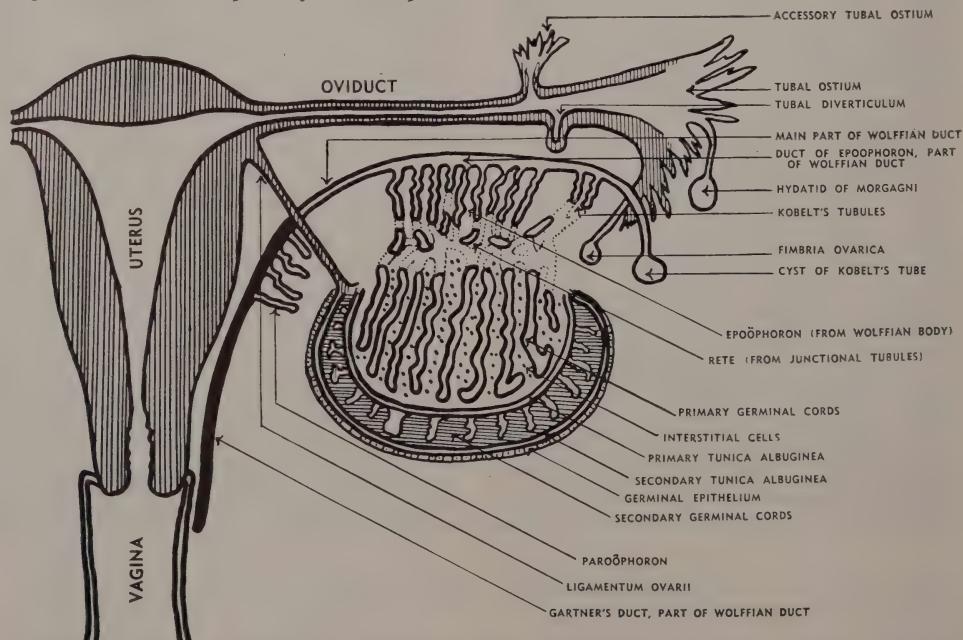
In the seven-week-old (13 mm.) embryo, the testes begin to be recognizable, because of the differentiation of cord-like structures, but the ovary has not yet acquired any distinguishing characteristics. It is not until the eleventh week that a dense cortex begins to separate from a loose medullary zone, while in the mesovarium the primitive rete appears. During the third month, stroma and blood vessels invade the ovary from the hilus and the connective tissue septules eventually reach the tunica albuginea. Simultaneously, beginning at the center, most of the cells of the inner epithelial mass are transformed into young ova. In fetuses of three to five months there is rapid ovarian growth, because a new cortical zone is formed, mainly as a result of a "second proliferation" of the germinal epithelium. While new ova are formed at the periphery of the gonad, the more centrally located germ cells degenerate. Subsequently, some prim-

itive sex-cells of the germinal epithelium (the future ova) surrounded by undifferentiated lining cells (the future granulosa) invade the cortex, thus producing PRIMORDIAL FOLLICLES. The theca cells are probably mainly of mesenchymal origin. It is possible, however, that derivatives of the celomic epithelium remain in the stroma and are transformed into theca cells.

The DESCENT OF THE OVARY from the kidney region into the pelvis is accomplished mainly by the cephalad elongation of the trunk in comparison with the fixed gonad. This produces a relative shift of the ovary caudad.

The RETE OVARII is a vestigial organ homologous to the testis. It may persist in the adult and can give rise to rete cysts and adenomas (see: Ovarian tumors).

The EPOÖPHORON is a rudiment developing from a cranial group of mesonephric tubules and corresponds to the epididymis; the PAROÖPHORON arises from the caudal group of these tubules and corresponds to the paradidymis of the male.



Schematic drawing illustrating the development of the ovary and para-ovarian structures
(After H. Selye: "Ovarian Tumors," Encyclopedia of Endocrinology, 1946.)

Table illustrating probable development of gonadal structures and tumors

which may arise from them in the female

(After H. Selye: "Ovarian Tumors," Encyclopedia of Endocrinology, 1946.)

Embryonic Structure	Mature Structure		Tumor to which it may give rise in the Female
	In Male	In Female	
Germinal epithelium	Seminiferous tubules	Ova and follicles	All types of <i>serous cysts, papillomas</i> and <i>carcinomas</i> ?
Primary germinal cords	Seminiferous tubules	Medullary cords	<i>Arrhenoblastomas, mixed teratoids</i> (by parthenogenesis)?
		Rete of ovary	<i>Rete adenomas, rete cysts, Brenner tumors</i> ?
	Leydig cells	Leydig cells	<i>Leydig (or hilus) cell tumors</i>
Secondary germinal cords	Seminiferous tubules	Ova and follicles	<i>Folliculomas, corpus luteum tumors, Mixed teratoids</i> (by parthenogenesis)? All types of <i>pseudomucinous cysts and carcinomas</i> (by parthenogenesis as "pure enteroid teratoids")? <i>Brenner tumors</i> (from Walthard's rests or parthenogenetically as abnormal "enteroid" teratoids)? <i>Struma ovarii, rhabdomyomas, chorionepitheliomas, false seminomas, etc.</i> , (by parthenogenesis as pure one-sidedly-developed teratoids)
	Testis stroma	Ovarian stroma	<i>Fibromas, sarcomas</i>
Pronephros?	Appendix of epididymis	Hydatid of Morgagni and Kobelt's tubules	<i>Cysts of infundibulum tubae</i> <i>Cysts of Kobelt's tubules</i> ?
Urogenital junction tubules	Connection between rete and epididymis	Absent	<i>Junctional cysts</i>
Wolfian body (cranial part)	Ductuli efferentes of epididymis	Epoophoron (Epi-oöphoron, par-ovarium organ of Rosenmüller)	<i>Cysts, adenomas and carcinomas of epoophoron-mesonephromas?</i> "Epithelioma Wolfien"?
Wolfian body (caudal part)	Paradidymis	Paroöphoron (Parepoöphoron)	<i>Cysts, adenomas and carcinomas of paroöphoron</i>
Wolfian duct (cranial part)	Duct of epididymis	Duct of epoöphoron	<i>Cyst of Kobelt's tubules</i> ?
Wolfian duct (caudal part) = Gartner's duct	Duct of epididymis	Gartner's duct remnants	<i>Gartner's duct adenomas</i>
Müllerian duct (cranial part)	Appendix testis	Oviduct with infundibulum (accessory tubal ostium, tubal diverticulum)	All types of <i>serous cyst papillomas and carcinomas</i> , ("salpingiomas") Ca of Fallopian tube, <i>para-ovarian cysts</i> due to hydrosalpinx of accessory tubal ostium or diverticulum
Müllerian duct (caudal part)	Utriculus prostaticus	Uterus and vagina	<i>Endometriosis</i> , (tumors of uterus and vagina not to be discussed here)
Adreno-cortical primordium	Accessory adrenal cortex in region of testis	Accessory adrenal cortex in region of ovary	<i>Hypernephroma ovarii</i>

THEORIES CONCERNING THE HISTOPHYSIOLOGY OF THE OVARY

In considering the histophysiology of the ovary, it must be kept in mind that we are dealing with an organ having both exocrine (production of ova) and endocrine (production of ovarian hormones) functions. Normally these two activities are coördinated, but under abnormal conditions ovulation is not necessarily accompanied by normal folliculoid and luteoid hormone formation and conversely, the cyclic production of these ovarian hormones may proceed even in gonads whose ova have been destroyed, or fail to be discharged at ovulation.

The Mechanism of Ovulation. — The mechanism of ovulation is still incompletely understood. It is probable, however, that under the influence of the continuously increasing intrafollicular pressure caused by the accumulating follicular fluid, the subcapsular wall of the follicle becomes so thin that it eventually ruptures. Just beneath the cumulus oophorus, the granulosa cells degenerate even prior to ovulation, so that the ovum with the corona radiata is then but loosely attached to the follicular wall. Hence the sudden decrease in intrafollicular pressure, occasioned by the rupture of the wall, readily detaches the ovum with its cumulus and flushes it out through the rupture point with the follicular fluid. The small hemorrhages, which sometimes accompany ovulation, may also be ascribed to the sudden decrease in intrafollicular pressure. It is possible that the smooth muscle cells of the ovarian stroma and theca may help to elicit follicular rupture, especially in those species in which such cells are plentiful.

The accumulation of follicular fluid and the final maturation of the follicles and ova are due to gonadotrophic hormones. Hence they are prevented by hypophysectomy and can be elicited at will by gonadotrophins, both in imma-

ture and hypophysectomized animals (in which follicle maturation would normally not occur).

The secretion of the follicular fluid is presumably a function of the granulosa cells, since it fails to occur in follicles whose granulosa has been destroyed (e.g., X-rays). On the other hand, the granulosa itself contains no blood vessels, so that the material for the formation of follicular fluid must reach it from the theca interna.

Migration of the Ovum. — After ovulation, the human ovum is discharged into the peritoneal cavity. However, laparotomy performed at that time reveals that the fimbriæ of the Fallopian tubes usually attach themselves closely to the ovarian surface when a follicle is about to rupture and hence the egg tends to enter directly into the oviduct. Under abnormal conditions however, the ovum may lose its way and implant itself anywhere in the pelvic peritoneum, thus giving rise to ectopic pregnancy. In certain animals (e.g., rat), the ovary is enveloped by a closed ovarian capsule which opens directly into the oviduct and does not communicate with the peritoneum. This obviates the possibility of peritoneal implantation.

In women in whom one ovary has been removed, ectopic pregnancy may occur in the tube of that side. Here we are evidently dealing with a **TRANSMIGRATION OF THE OVUM** from the contralateral ovary. Similarly, in animals having two separate uterine horns, removal of one ovary does not preclude the possibility of implantation in the ipsilateral uterus. Some investigators believe that both internal (transuterine) and external (transperitoneal) transmigration of the ovum are possible.

The Law of Constant Numbers in Ovulation. — With but minor variations, each species produces a certain, rather constant, number of ova at each ovulation. Hence the number of off-

spring during any one pregnancy is also fairly constant. This is referred to as "the law of constant numbers in ovulation" (*Lipschütz*). It is probably due to a species-specific, constant relationship between hypophyseal gonadotrophic hormone secretion and ovarian development. If one ovary is removed, the contralateral gonad tends to produce twice the normal number of ova at each ovulation, thus compensating for the loss. Presumably, a decreased ovarian hormone production (due to the unilateral ovariectomy) automatically occasions a compensatory increase in hypophyseal gonadotrophin secretion. In agreement with the conception that the number of maturing follicles depends upon pituitary stimuli, treatment with exogenous gonadotrophins increases the number of ovulating follicles beyond that expected on the basis of the law of constant numbers.

To illustrate this point, in a rat whose two ovaries would produce an average of 12 ova at each cycle, unilateral ovariectomy so influences the remaining gonad that it alone will produce approximately this number. On the other hand, treatment with hypophysoid gonadotrophins may lead to simultaneous ovulation from several dozens of maturing follicles.

Which cell produces which hormone? — FOLLICULOIDS are normally produced by the maturing follicles. This is shown by various observations. Only traces of folliculoid hormones are demonstrable in prepubertal or senile animals whose follicles do not mature. Injection of hypophysoid gonadotrophins greatly augments follicle maturation and correspondingly induces folliculoid hormone secretion. Hypophysectomy, which prevents the maturation of follicles, inhibits folliculoid hormone formation.

It is much more debatable whether the granulosa or the theca cells are

chiefly responsible for folliculoid hormone formation. In large ovaries (e.g., those of cattle) the parts of the follicle can be separated and direct bioassays indicate that follicular fluid, granulosa and theca tissue all contain very high concentrations of folliculoids. Destruction of all granulosa cells by X-rays (to which these cells are particularly sensitive) does not produce anestrus in mice if the theca cells are preserved. Treatment of hypophysectomized rats with pure LH fails to stimulate the granulosa, but causes marked proliferation of the theca accompanied by manifestations of estrus. These findings clearly indicate that the theca cells are capable of folliculoid hormone production. Ovarian tumors consisting of granulosa cells (folliculomas) or theca cells (thecomas) both cause manifestations of marked folliculoid hormone overdosage.

In view of these facts and of the close embryologic relationship between theca and granulosa, it is possible that both these parts of the follicle participate in the secretion of folliculoids, but since the granulosa is not vascularized at least the raw-materials for these hormones must come from the surrounding theca. It should be kept in mind furthermore that FSH causes granulosa proliferation without stimulating folliculoid secretion, while LH augments the production of folliculoids without affecting the granulosa. The bulk of evidence favors the view that the theca is the source of folliculoids. However, the corpus luteum, testis, adrenal cortex and placenta are also capable of folliculoid production, so that the elaboration of these hormones is certainly not the monopoly of any one cell type.

In the ovary, LUTEOIDS are apparently produced exclusively by the corpora lutea. Progestational changes occur in the endometrium only in the presence

of functional corpus luteum tissue. Extirpation of the corpora lutea causes a breakdown of the pregestational endometrium even if the remaining ovarian tissue is preserved. However, some evidence indicates that certain extra-ovarian tissues, especially the adrenal cortex and placenta, can also produce luteoids.

The maintenance and function of the corpus luteum depends upon stimulation by the *luteotrophic hormone* of the anterior-pituitary or placenta. Hypophysectomy causes a breakdown of a functional corpus luteum unless a fairly mature placenta is present in the organ-

ism or luteotrophic hormone is administered.

The *lipid granules* in the corpus luteum and theca cells are probably carriers of the (highly lipid-soluble) ovarian hormones.

There is no evidence that either the afferent or the efferent *nerves* of the ovary play any important rôle in the formation of ovarian hormones, since denervated ovaries or transplanted ovaries (whose nerves are of course severed) continue to form hormones under the influence of gonadotrophic hormones. (See also : Stimuli Influencing the Ovary, pages 381, 382.)

CHEMISTRY OF THE OVARIES

CHEMICAL COMPOSITION OF THE GLAND

The chemistry and biogenesis of the ovarian hormones will be discussed in subsequent chapters. Here, we shall merely consider the most important data concerning the general chemical composition of the gland.

It is difficult to make generalizations concerning the chemical composition of ovaries, because of the marked structural changes which they undergo between childhood and senility, as well as during each sexual cycle in the adult.

In general it may be said however, that the CARBOHYDRATE content of the ovaries is comparatively low, although traces of glycogen and lactic acid are demonstrable in them.

The LIPID content of the corpora lutea is especially high, but in the remaining parts of the ovary it is rather low. It appears that the phospholipid concentration rises in the corpora lutea until about the 10th day after ovulation, following which it begins to diminish. Conversely the cholesterol ester content — which indicates involution — is low until the 10th day and then rises suddenly. The corpus luteum also contains rather considerable amounts of neutral fats and fatty acids; the quant-

ity of these is comparatively low during the most active phases, but rises when involution begins. On the other hand, the scars of corpora lutea are almost completely devoid of lipids.

The bright yellow pigment so characteristic of the human corpus luteum is generally referred to as *lutein*. It belongs to the lipochromes or lipid pigments and is probably similar to the yellow pigment of egg yolk, but it has not yet been clearly characterized. It is presumably closely related to the vegetable pigments, carotene and xanthophyll, both of which have been demonstrated in corpus luteum tissue. A large percentage of the corpus luteum lipids occurs in the ovary in combination with protein as lipoprotein.

The major part of the dry material in ovarian tissue is PROTEIN, but so far the characteristics of the ovarian proteins have not been adequately studied.

Among the INORGANIC CONSTITUENTS, sodium, chloride and potassium are most important; but bromine, iodine, sulphur, iron and copper are also present.

Various ENZYMES occur in comparatively high concentrations in ovarian tissue. Lipases appear to play an impor-

tant rôle in the discharge of the lipid granules (e.g., from the corpus luteum and theca); these are presumably the solvents in which the ovarian steroids are stored. Phosphatases, especially adenosine triphosphatase (ATP-ase) — the adenosine triphosphate splitting enzyme important in energy-requiring reactions of tissues (e.g., muscles) — is found in comparatively high concentrations in corpora lutea. In general, the ATP-ase activity per unit weight is lower in functional than in non-functional corpora lutea. Among other enzymes, proteases, arginase, asparaginase, estrinase, oxidases and glycolytic enzymes appear to be most important.

Ovarian tissue is also comparatively rich in various VITAMINS, especially vitamin-C, but their function has not yet been determined.

Several OTHER METABOLITES (choline, glutathione, citric acid, avidin, various amines, purines, amino-acids and thymonucleic acid) have been claimed to play a more or less important part in the chemical composition of ovarian tissue, but their physiologic rôle is incompletely understood.

CHEMISTRY OF THE OVARIAN HORMONES

All the ovarian hormones known to date are steroids. Their fundamental chemical characteristics have been described in the section : "The Steroids."

GENERAL PHARMACOLOGY OF THE OVARIAN HORMONES

STANDARDIZATION

The direct gravimetric determination of folliculoids can only rarely be employed because of the minute quantities present in body fluids and tissues. The most commonly used methods are based upon chemical or biologic determinations of varying degrees of specificity and accuracy. These are usually performed on partially-purified extracts containing the desired fraction. Because of the differences in the solubilities of the various naturally-occurring folliculoids and their excretion products, it is possible to concentrate and purify the total folliculoid activity of body fluids and tissues, and to subdivide it into its constituent fractions (separation of ketonic folliculoids such as estrone, from the non-ketonic such as estradiol and estriol; differentiation between free and conjugated folliculoids, etc.). The conjugated folliculoids are usually biologically less active and more water-soluble than the free compounds, while the latter are more fat-soluble. By acid hydrolysis it is readily possible to transform the conjugated

into the free type. At an alkaline pH, however, even free folliculoids are quite soluble in water because their acid phenolic hydroxyl groups form water-soluble salts with alkali.

Analytic Methods for the Detection of Folliculoids. — A number of analytic methods have been devised for the quantitative estimation of folliculoids, but these are useful only when comparatively large amounts are present, as in the urine of pregnant women. The most commonly employed analytic methods are the following :

(1) The COLORIMETRIC TESTS are based upon the observation of *Wieland et al.* (1929) that impure folliculoid concentrates prepared from human pregnancy urine, give a yellow color when treated with concentrated sulfuric acid and chloroform (*Salkowski test*). A reddish color with green fluorescence (*Liebermann-Burchardt test*) is obtained when they are treated with acetic anhydride and sulfuric acid. Subsequently, *Kober* (1931) showed that the initial orange color, produced by estrone and sulfuric acid, changes to a

clear green-fluorescing red, when heated with water. In this manner the folliculoids can be differentiated from cholesterol, pregnanediol, bile acids and many other steroids, as these give a yellow color with sulfuric acid and this is decolorized by water. *Cuboni* (1934) used this test for the diagnosis of pregnancy in mares, which excrete large amounts of folliculoids during gestation. *Kober* (1931) also showed that the intensity of the fluorescence may be diminished and the intensity of the red color enhanced by using a mixture of phenol and sulfuric acid instead of the acid alone. This is due to the resulting formation of phenolsulfonic acid. Subsequently, he (*Kober*, 1936) introduced a second modification substituting β -naphtholsulfonic acid for the phenolsulfonic acid, the color being measured photoelectrically.

More recently, *Bachman and Pettit* (1941) devised a particularly accurate and sensitive modification of the *Kober* reaction. Their method permits the quantitative extraction of urinary folliculoids in a form pure enough to yield, with appropriate color-reagents, products closely resembling those obtained with pure crystalline hormones. The procedure also allows the complete separation of estriol from the sum of estrone and estradiol. The technic permits the determination of estriol when this substance is present in the urine in concentrations exceeding $1000\gamma/L$, and the estimation of the sum of estrone and estradiol when it exceeds $500\gamma/L$. Thus, it is practically applicable in the assay of human pregnancy urine after the fourth month of gestation.

Alkaline picric acid solutions have long been used for the determination of creatinine (*Jaffe*, 1886). It has been found that similar color-reactions are given by various compounds containing the group — CO-CH₂ —, such as that at C₁₇ in estrone. *Zimmermann* (1935) took advantage of this by devising a

colorimetric method, in which estrone solutions are treated with m-dinitrobenzene and KOH, whereupon a violet color results. The method is obviously not specific and it cannot be used for the detection of the non-ketonic folliculoids (estradiol and estriol) which do not contain the above grouping. However, under certain experimental conditions, the test can differentiate between ketonic testoids and ketonic folliculoids since, as previously mentioned, the latter are phenolic and can be removed from fat-solvents by extraction with alkali, leaving the neutral ketonic testoids behind.

(2) The characteristic ULTRA-VIOLET ABSORPTION spectrum of estrone (maximum absorption at about 2820 Å with a well-marked minimum at about 2500 Å in neutral or acid solution; and a maximum at 2950 Å with a corresponding shift of the minimum in alkaline solution) and of other folliculoids may also be used for analytic purposes, but only highly purified samples are suitable for spectrography and hence this is not yet practical for routine use.

Analytic Methods for the Detection of Luteoids. — There are no practical methods for the determination of progesterone in biologic material, but its chief excretion product, pregnanediol, can be detected in the urine (though not in the blood) by gravimetric methods. *Venning* (1937) developed such a method which permits the determination of pregnanediol-sodium-glucuronide. This is expressed in terms of free pregnanediol by calculation. *Bucher and Geschichter* (1940) described a technic which permits separate recovery and estimation of the free and combined forms from the same urine specimen. It is doubtful, however, whether free pregnanediol occurs naturally in the urine; most of it certainly arises due to spontaneous hydrolysis of the conjugate after voiding. Hence care must be taken either to prevent hydrolysis,

or to effect quantitative hydrolysis of the conjugate and determine the free compound as such.

Bioassay of Folliculoids. — The INTERNATIONAL UNIT (I.U.) of folliculoid activity is by definition that equivalent to the folliculoid potency of 0.1γ of pure estrone under the same bioassay conditions. It is therefore recommended to use estrone as a reference standard in such assays. The INTERNATIONAL BENZOATE UNIT (I.Bz.U.), which is equivalent to the activity of 0.1γ of estradiol-3-monobenzoate, has been recommended as a standard for the assay of slowly-acting folliculoids.

For the determination of folliculoids in body tissues and fluids, bioassays are usually more satisfactory than chemical methods of determination since they are more specific and can be performed with less completely purified material, minimizing losses during the process of purification.

In principle, BIOPSY SPECIMENS taken from patients (e.g., endometrium, vaginal smear) represent "internal bioassay technics" obviating the necessity for chemical purification of the folliculoids. The responses of the donors' tissues (e.g., follicular-phase endometrium, vaginal cornification) are themselves the indicators of hormone action. The great advantage of these procedures — as of all internal bioassays — is that they indicate the activity of hormones under the conditions existing *in vivo*. Their interpretation is less likely to be blurred by such factors as changes in the renal threshold for the hormones, variations in the degree of destruction and detoxification after the hormones have exerted their normal effects, transient fluctuations in hormone production and especially losses during extraction for bioassay. On the other hand, these methods are fundamentally qualitative and permit only an approximate estimation of the amounts of hormones present.

The technics devised for the bioassay of folliculoids in body fluids and tissues are usually based upon their extraction from the biologic material and the study of their effects upon sensitive test objects. The following are the most commonly used procedures:

(1) The ALLEN-DOISY TEST (1923) and its various modifications, is performed on immature or spayed mice or rats, whose vaginal epithelium is not cornified. Repeated subcutaneous injection of folliculoids into such test-animals causes disappearance of leukocytes and vaginal cornification within one or two days. This is readily detected by the histologic examination of stained (or even unstained) vaginal smears. Since spayed animals which have not been used for such tests for some time become insensitive, it is essential that prior to the test they be sensitized by a preliminary injection of a minimal effective dose of a folliculoid. The sensitivity of this assay may be increased by partial hepatectomy, since the liver inactivates much of the injected folliculoids. If the duration of action of a folliculoid is to be measured (e.g., prolongation of action by esterification) a single subcutaneous dose is administered and the length of the induced vaginal estrus is measured by taking vaginal smears bi-daily for several days.

(2) The METROTROPHIC TEST (Astwood, 1938) is performed on immature female rats (25-49 gm.) receiving a single subcutaneous injection of the folliculoid. The animals are killed 6 hours later and the increase in the weight of the uteri is taken as a criterion. The advantage of this test lies mainly in its rapidity.

(3) In EMMENS' S/L TEST (1940) two groups of spayed mice are given folliculoid by the subcutaneous and intravaginal route respectively. The criterion is the presence in the vaginal smears of cornified or nucleated epithelial cells in the absence of leukocytes.

The S/L ratio is that between the effective dose of Systemic and Local administration. The test is used for the differentiation of folliculoid precursors from active folliculoids. For the former, the ratio is in the neighborhood of unity, while for the latter it is much greater, since they act directly upon the vaginal epithelium without having to be absorbed and activated in the organism.

Several other indicators, such as the growth of the NIPPLES in male guinea pigs, the lengthening of the OVIPOSITOR in certain fish (bitterling), the feminization of the PLUMAGE in capons, the OPENING OF THE VAGINA in spayed adult guinea pigs, prepubertal mice or rats (in which the vagina is normally closed by a membrane), the MUCIFICATION OF THE VAGINAL EPITHELIUM of the rat (elicited by folliculoids in doses insufficient to cause cornification) and many other characteristic changes caused by folliculoids, have been used for bioassay purposes.

As previously stated, unlike the internal, the external bioassays must be preceded by at least partial chemical purification of the hormones; the hormone concentration in tissues and body fluids is usually not high enough to permit bioassay of the source material (blood, urine, etc.) in its original state. Several methods have therefore been developed for the quantitative EXTRACTION AND SUBSEQUENT BIOASSAY of folliculoids. Among these, the following are most useful :

(1) The BLOOD-FOLLICULOID TEST of *Frank and Goldberger* (1935) which is based upon the fact that in regularly menstruating women, the blood-folliculoid titer rises significantly a few days before the onset of bleeding; hence venous blood is taken shortly before the expected period. If the menses are irregular, blood specimens must be obtained weekly for more than a month, in order to detect possible cyclic variations in blood-folliculoid concentration.

50 cc. of blood are extracted, according to a certain procedure, and assayed by the Allen-Doisy test.

(2) The BLOOD-FOLLICULOID TEST OF *Fluhmann* (1934). 25-40 cc. of venous blood are taken, immediately centrifuged and 0.5 cc. of the clear serum is injected subcutaneously, 3 times daily, to spayed adult female mice on 3 consecutive days, each animal receiving a total of 4.5 cc. In this test, the serum itself can be used without extraction since the slightest degree of vaginal response, the mucification of the atrophic epithelium, is taken as positive. An approximately quantitative estimation is possible, even using only a single dose level, since various degrees of response between mucification and complete cornification can be distinguished with this technic.

(3) In the URINARY FOLLICULOID TEST of *Kurzrok and Ratner* (1932) the folliculoid activity of the urine is extracted with ethyl acetate in a continuous extractor. Following further purification, the extract is dissolved in oil for bioassay in the Allen-Doisy test.

(4) In the URINARY FOLLICULOID TEST of *Smith and Smith* (1935) the conjugated urinary folliculoids are first hydrolysed and thus transformed into the free, more active form; then the total urinary folliculoids are determined. Following concentration and purification, the active material is dissolved in oil and assayed on spayed female rats by the vaginal smear test. This method is superior to those carried out on non-hydrolyzed urine, since the proportion between free and total folliculoids varies and hence, bioassays cannot yield accurate data concerning the total folliculoid elimination unless there is total transformation into the biologically-active form.

Subsequently, *Smith and Smith* (1939) devised a modification for the extraction and separation of estrone, estriol and other folliculoids in the

urine. However, space does not permit the detailed discussion of this and the many additional methods recommended for the quantitative analysis of folliculoids in biologic material.

Bioassay of Luteoids. — The INTERNATIONAL UNIT of luteoid potency is defined as the activity corresponding to that of 1.0 mg. of progesterone under the same bioassay conditions.

Among the many bioassay technics used for the estimation of luteoids in solution, the following deserve special attention :

(1) **IN THE CORNER AND ALLEN TEST** (1929) adult female rabbits are spayed 18 hours after having been mated during estrus. The substance to be tested is then injected subcutaneously, daily for 5 days. The minimum amount necessary to cause complete progestational proliferation of the endometrium is taken as a unit. This is equivalent to 1.25 mg. of progesterone.

(2) **THE CLAUBERG TEST** (1930) is a modification of the former in which intact immature rabbits (pretreated with 8 daily injections of folliculoids) are used. The luteoid substance to be tested is subsequently given in 5, daily, subcutaneous injections and the total dose necessary to produce a "definite" progestational transformation of the endometrium is taken as the unit. This corresponds to 0.75 mg. of progesterone.

(3) **THE MCPHAIL TEST** (1934) is performed on immature female rabbits (750-950 gm.) treated with 150 I.U. of estrone over a period of 6 days, after which the unknown solution of the luteoid is administered in 5, daily, subcutaneous doses. In a clearly defined scale ranging from 0 to ++++ of progestational proliferation, an average reaction of ++ is taken as the unit. This corresponds to 0.25 mg. of progesterone.

(4) **THE PINCUS AND WERTHESSEN TEST** (1937) is carried out on rabbits

spayed 18 to 20 hours after mating. They then receive bi-daily injections of the luteoid to be assayed, on the 2nd, 3rd and 4th day after copulation. On the 5th day, the animals are killed and the fertilized ova (blastocysts) washed out of the uterus. The luteoid activity is estimated by determining the relationship between the degree of progestational development (measured with a planimeter) and the growth of the fertilized ovum. 0.38 mg. of progesterone can be detected with this technic. Various other modifications are based upon the ability of luteoids to maintain gestation in rabbits, which otherwise regularly abort following ovariectomy during gestation.

(5) **THE TEST OF FEVOLD ET AL.** (1930) is based on the ability of certain corpus luteum extracts to cause relaxation of the pelvic ligaments in guinea pigs. The test is supposed to be specific for a special hormone of the corpus luteum ("relaxin"). However, since folliculoids and various mixtures of folliculoids and progesterone induce relaxation of the pelvic ligaments, the specificity of the technic is doubtful.

(6) **THE SEXUAL RECEPTIVITY TEST** is based upon the fact that ovariecomized virgin guinea pigs, pretreated with folliculoids, exhibit the copulatory reflex (arching and straightening of the back and elevation of the pudenda) following the subsequent administration of luteoids, if the vulvar region is stimulated.

(7) **THE VAGINAL MUCIFICATION TEST**, performed in mice, guinea pigs or rats, is based upon the observation that immature or castrate females respond with mucification of the vaginal epithelium if luteoids are administered following sensitization with folliculoids. Since minute doses of folliculoids in themselves cause similar reactions, the test is not very specific.

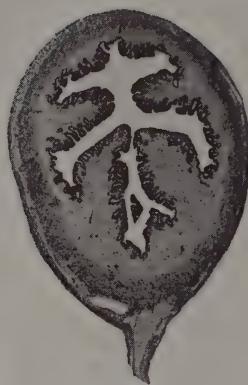
(8) **THE VARIOUS DECIDIUMA TESTS** are based upon the fact that in folli-



A



B



C



D



E



F

Biassay of luteoid activity in the McPhail test. — A. Cross-section through the uterus of an immature, untreated rabbit. B. Cross-section through the uterus of an immature rabbit, treated

culoid hormone pretreated animals (rats, guinea pigs) subsequent treatment with progesterone so sensitizes the endometrium that it responds to local trauma by the formation of a deciduoma (maternal placenta tumor).

(9) The inhibition by luteoids of the UTERINE CONTRACTIONS normally produced by oxytocic posterior-pituitary extracts in the rabbit, is not sufficiently specific to serve for bioassay purposes.

(10) The LOCAL APPLICATION OF LUTEIODS TO THE RABBIT ENDOMETRIUM following folliculoid pretreatment, is one of the most sensitive tests as it permits the detection of as little as 0.5-5.0 γ of progesterone. It is a valuable qualitative indicator of luteoid activity, but due to great individual variations in response, its applicability as a quantitative technic is rather limited.

(11) From the clinician's point of view, the most satisfactory bioassay is that based upon the induction of PROGESTATIONAL PROLIFERATION IN THE ENDOMETRIUM OF SPAYED OR POSTMENOPAUSAL WOMEN pretreated with folliculoids. This method gives direct evidence of luteoid activity in man, but for obvious reasons, it is not suitable for accurate quantitative assays on a statistically significant basis.

(12) The "INTERNAL BIOASSAY" of endogenous luteoid production is performed by evaluating endometrial biopsy specimens obtained by curettage.

PHARMACOLOGY OF OVARIAN HORMONE DERIVATIVES AND OF ARTIFICIAL OVARIAN HORMONES

Folliculoids and Artificial Folliculoids. — It is not within the scope of this book to discuss the special pharmacology of the numerous natural and artificial folliculoid compounds. It should be emphasized; however, that

unlike the derivatives of the other steroid hormone groups (testoids, luteoids, corticoids, renotrophic and spermato-genic steroids) the most potent folliculoids (like some of the most active anesthetic steroids) exhibit their chief activity to the exclusion of all other hormone actions. This is what we defined as a "simple" action. (See : The Steroids.) Even the various artificial folliculoids, many of which are chemically very different from the naturally occurring hormones, exhibit exclusively folliculoid potency. It is interesting, however, that most of the artificial folliculoids possess some anesthetic action as do the corresponding natural compounds. Indeed, some of the amine-substituted dihydrostilbestrol derivatives possess morphine-like analgesic effects, although they exhibit no obvious folliculoid potency.

Most of the artificial folliculoids are particularly active by mouth. This represents a great practical advantage over the natural hormones which are comparatively inactive when administered orally. It is noteworthy, however, that certain metabolites of the natural folliculoids such as estriol glucuronide (emmennin) which is present in human pregnancy urine, or estrone sulfate (premarin) from pregnant mare's urine are both orally active derivatives of naturally occurring hormones. The natural folliculoids have been discussed in the section : The Steroids. In the following table, we list merely the main characteristics of the most important artificial folliculoids. For the sake of simplicity, folliculoid activity is expressed in international units (I.U.) but the relevant data are only *very approximate* estimates since they are based on bioassays performed in different laboratories under not quite comparable conditions.

with a folliculoid (note estrus development of endometrium). — C, D, E and F, 1, 2, 3 and 4 plus pregestational response in estradiol-pretreated immature rabbits, given increasing doses of progesterone. Note the increasingly more pronounced "lacing" of the six longitudinal folds of the endometrium.

Principal artificial folliculoids and some related compounds

Name of Compound	Formula	Characteristics
ETHYNYL-ESTRADIOL [17(β)-ethynyl-Δ ^{1,3,5;10-} -estratriene-3,17(α)-diol] <i>Syn</i> : 17-ethynil-estradiol		I.U. = about 0.05γ (subcutaneously). Due to ethynyl side-chain, highly active when given by mouth.
DOISYNOLIC ACID [16 17-Δ ^{1,8,5;10-} -estratriene-3-ol-17-carboxy acid]		When assayed as the sodium salt I.U. = about 0.1γ (subcutaneously). More active than estrone when given by mouth. Note that this and especially the following compound possess great folliculoid activity in spite of the opening of ring D.
BISDEHYDRO-DOISYNOLIC ACID [16 17-Δ ^{1,3,5;10,6,8-} -estratetraene-3-ol-17-carboxy acid.]		When assayed as the sodium salt I.U. = about 0.01γ (subcutaneously), equally active when given by mouth. This is the most active folliculoid substance known to date.
DIETHYLSTILBESTROL [4:4'-dihydroxy- <i>a</i> : <i>β</i> -diethylstilbene] <i>Syn</i> : stilbestrol		I.U. = about 0.05γ (subcutaneously) almost equally active by mouth. In women stilbestrol by mouth is about as active as estrone by injection. Depending upon the position of the ethyl groups, the compound can exist in two isomeric forms. Both formulae given here represent the biologically more active isomer, but the lower formula is written in a manner emphasizing its resemblance to steroids.
HEXESTROL [3,4-di(p-hydroxyphenyl)-hexane] <i>Syn</i> : dihydrodiethylstilbestrol		I.U. = about 0.03γ (subcutaneously), almost equally active by mouth. In women, claimed to be slightly more active and less toxic than stilbestrol.
[1-(m-hydroxyphenyl)-3-(p-hydroxyphenyl)-hexane]		I.U. = more than 20 mg. Note that in spite of similarity to steroids the activity is greatly decreased in comparison with stilbestrol, due to the change in the aliphatic block between the phenolic rings.

Principal artificial folliculoids and some related compounds (Continued)

Name of compound	Formula	Characteristics
BENZESTROL [2,4-di(p-hydroxyphenyl)-3-ethyl-hexane] <i>Syn</i> : octofollin.		When most active isomer is tested I.U. = about 0.04γ (subcutaneously), almost equally active by mouth.
DODDS' COMPOUND M4 [β -hydroxy- α, β -diphenyl- <i>n</i> -ethylamine]		Not folliculoid. This and other diphenylethylamines are not only chemically closely related to dihydro-diethylstilbestrol, but also to morphine the formula of which is:
[1 methyl-2 (p-hydroxyphenyl)-6-hydroxy-3,4-dihydro-naphthalene]		Like morphine and the steroid hormones it possesses anesthetic and analgesic properties.
(5,6-cyclopenteno-1,2-benzanthracene)		Strongly carcinogenic benzanthracene derivative possessing only trace of folliculoid activity.
TRIPHENYLCHLORETHYLENE (α -phenyl- β -chlorostilbene)		I.U. = about 80γ (subcutaneously), equally active by mouth. Note that this, and the above mentioned compound, exhibit folliculoid activity, although they contain no oxygen.

Luteoids. — Only very few artificial luteoids have been prepared as yet. The most commonly used among them is ethynodiol. It is much less luteoid than progesterone, but has the advantage of oral activity. In high doses it is slightly virilizing.

More recently, ethynodiol has been used experimentally as an orally active, artificial luteoid. It is

somewhat less luteoid than ethynodiol, but considerably less virilizing.

The following table lists the main characteristics of the two most important artificial luteoids. For the sake of simplicity, luteoid activity is expressed in international units, but, as in the case of the previous table, the relevant data are only *very approximate*.

Principal artificial luteoids

Name of Compound	Formula	Characteristics
ETHYNYL-TESTOSTERONE [17(β)-ethynyl- Δ^4 -androstene-3-one-17(a)-ol] <i>Syn</i> : pregnenolone; pregneninol; anhydrohydroxyprogesterone.		I.U. = about 10 mg. (subcutaneously), almost equally active by mouth, due to the ethynyl side-chain. Possesses some testoid activity but this is very slight in women.
ETHYNYL-ANDROSTENEDIOL [17(β)-ethynyl- Δ^5 -androstene-3(β),17(a)-diol] <i>Syn</i> : ethynyl- Δ^5 -androstenediol-3,17.		I.U. = about 20 mg. (subcutaneously), almost equally active by mouth, due to ethynyl side-chain. Less testoid than ethynyl-testosterone.

MODE OF ADMINISTRATION AND CHIEF INDICATIONS

Folliculoids.— In administering folliculoids, it must be kept in mind that the response is subject to great individual variation. Hence the doses recommended are always approximate; they should be adjusted to the individual case by controlling the vaginal smears and other manifestations of folliculoid activity. If systemic effects are desired, oral or parenteral administration is most convenient, but if only one specific target organ is to be affected, topical application may prove preferable.

The cost of prolonged therapy with natural folliculoids is rather high and since the artificial folliculoids (especially those of the stilbene series) are equally effective, preference may be given to them if financial considerations are important. However, some patients suffer from nausea following oral treatment with stilbene derivatives. Since in most instances, folliculoid therapy takes the form of repeated short periods of treatment, the implantation of crystal pellets or the injection of crystal suspensions is rarely desirable.

INTRAMUSCULAR OR ORAL administration are recommended whenever systemic actions are desired. **SUBCU-**

TNEOUS administration of oily solutions is contraindicated because poor absorption leads to accumulations of oil. The average dosage for typical cases of *hypogonadism* with *amenorrhea*, or *suppression of lactation*, varies between 5000 and 50,000 I.U., that is, an activity equivalent to 0.5-5.0 mg. of estrone. This corresponds approximately to 2-10 mg. of stilbestrol and 0.2-3.0 mg. of hexestrol or benzestrol per os. Estrone-sodium-sulfate (premarin) is usually administered orally in tablets containing 1.25 mg. The latter compound is less likely to cause nausea than the stilbene derivatives. In most instances, these compounds are given in the above dosages two or three times weekly until the desired effect is obtained, but it is advisable to interrupt therapy every two or three weeks in order to prevent the development of metropathia hemorrhagica. If regular menstrual cycles are to be induced, progesterone should be given after interruption of the folliculoid treatment. Ethynyl-estradiol or estriol may also be administered per os in equivalent doses, since they have a comparatively high oral activity. In the therapy of *dysmenorrhea* and *menopausal disturbances*, the lowest dose levels usually suffice (e.g., 0.5-1.0 mg. of stilbestrol per day).

SUBLINGUAL (or buccal) administration has the advantage that the compound is directly absorbed into the circulation without having to pass through the intestinal tract. Thus, destruction by gastrointestinal enzymes is avoided and detoxification in the liver is minimized, since — unlike after oral administration — the compound does not pass first through the liver by way of the portal circulation. For this purpose, ethynodiol-diethylstilbestrol, or estradiol itself, is instilled into the sublingual space, in propylene glycol solution, in daily doses of about 0.2-0.5 mg. For routine clinical use this technic is rather impractical since it is inconvenient and — because of variations in absorption rate — it does not permit accurate dosage.

RECTAL administration of folliculoids is also effective in women, but comparatively inefficient and hence rarely used. About 15 times the subcutaneous dose of natural folliculoids is required when given in this manner.

VAGINAL administration is recommended especially in the therapy of *gonorrhœal vulvo-vaginitis* in children, or *senile vaginitis* and *kraurosis vulvae* in postmenopausal women. Vaginal suppositories (containing 0.02-0.4 mg. of estrone or estradiol or 0.1-0.5 mg. of stilbestrol) are recommended for this purpose. These comparatively small doses are effective, because in this case the folliculoids act directly on the target organ and are less subject to destruction in the body.

NASAL application, by a spray, of oily solutions (10,000-20,000 I.U. per cc.) of various folliculoids is advisable in cases of *atrophic rhinitis*, after irrigating the nose with an alkaline wash in order to remove the crusts.

Some gynecologists advocate the PERCUTANEOUS use of folliculoids in an ointment base. While there is no doubt that folliculoids can be absorbed through the intact skin, the rate of absorption varies greatly and can not be foretold. This mode of administration

may be applicable in cases of *breast hypoplasia*, *acne* and *hypertrichosis*, where local action is desired, but the cosmetic value of folliculoids in various creams and ointments has been greatly overrated. When systemic effects are desired, the percutaneous route of administration is not to be recommended.

Direct INTRA-UTERINE application of folliculoids undoubtedly causes a pronounced local effect in experimental animals, indicating that these hormones exert a direct action upon this target organ. In women, the intrauterine application of folliculoid-containing suppositories, or actual injections of folliculoids into the substance of the uterine muscle have been advocated by some, but this technic is still in the experimental stage.

INTRASPLENIC, INTRAPERITONEAL AND INTRAHEPATIC application of folliculoids have played an important rôle in animal experimentation, in proving that these hormones are detoxified during their passage through the liver. They are less effective, when given by any of these routes, than when administered subcutaneously or intramuscularly in equivalent doses. In clinical medicine, the above modes of application are, of course, never employed.

INTRAVENOUS injection of water-soluble folliculoids, would only be justified if very sudden and transitory effects were desirable. This is hardly ever the case in clinical medicine.

TRANS-PLACENTAL application of folliculoids is also effective, as shown by the fact that if such compounds are injected into pregnant animals, typical actions are elicited in the embryos. The comparatively excessive development of the breasts and other accessory sex organs in newborn children, is probably due to such trans-placental absorption of maternal folliculoids.

Direct application of folliculoids to certain areas of the COMB causes a local inhibition of its growth in capons simultaneously treated with testoids.

Local injection of folliculoids into the FERTILE FOLLICLE of the fowl induces typical folliculoid changes in the treated feathers only.

Introduction of folliculoids into BONES caused local stimulation of osteogenesis in the duck and pigeon; hence this action is presumably also direct.

Luteoids.— Only two compounds of this series are in common clinical use, namely progesterone, which is given parenterally by the INTRAMUSCULAR route, and ethynodiol-testosterone, which is given ORALLY. SUBCUTANEOUS administration is not recommended (oil accumulations!). Progesterone is almost certainly the natural luteoid compound produced by the corpus luteum. Ethynodiol-testosterone is a synthetic, artificial luteoid, prepared from progesterone or testosterone. It is estimated that progesterone parenterally is equivalent to about 5 times as much ethynodiol-testosterone per os.

For parenteral administration, progesterone is usually injected daily, or every 2nd-3rd day, in oily solution, in doses varying between 1 and 10 mg. In amenorrhea, it is given over a 7 day period, following 2 weeks of folliculoid hormone pretreatment; in dysmenorrhea and menorrhagia 3 to 6 days before the expected flow; in metrorrhagia — especially the type due to metropathia hemorrhagica — cyclic administration is recommended. 4 doses are given every other day; this is usually followed by withdrawal bleeding and 20 days after the onset of this bleeding, the same 8-day course is repeated. In this manner, artificial luteal phases, followed by apparently normal bleedings are produced, and eventually spontaneous cycles may reappear, especially in young women.

Progesterone has also been recommended in approximately the same doses in sterility due to inadequate pregestational proliferation which interferes with the nidation of the ovum. Here it is administered daily, beginning on the 14th day of the regular cycle or

whenever the occurrence of ovulation is suspected. Treatment is continued until the onset of flow (if treatment was ineffective) or until conception occurs and is verified by the Aschheim-Zondek test, or one of its modifications.

In threatened or habitual abortion, progesterone injections are recommended, daily or every second day, until the danger of miscarriage subsides. The efficacy of the treatment in these cases has not yet been definitely demonstrated, but many of the reported results are encouraging.

Parenteral progesterone administration has also been advocated in the therapy of *hyperemesis gravidarum* and premenstrual tension. In the former instance, it may be given continuously until hyperemesis subsides, while, in premenstrual tension, it is usually administered daily during the week preceding the expected date of the menses.

For all these indications, ethynodiol-testosterone may be alternatively given by mouth in the form of tablets containing 5-10 mg. The main contraindication for ethynodiol-testosterone is its virilizing effect, but in women the latter is very slight in comparison with the response of some animals. If necessary, the less virilizing but also orally active ethynodiol-androstanediol may be tried.

PERCUTANEOUS administration of progesterone or ethynodiol-testosterone to the shaved skin of folliculoid-pretreated immature or castrate rabbits, causes typical pregestational proliferation of the uterus. This shows the possibility of absorption through the skin. By this route the hormone is particularly active in alcoholic solutions.

SUBLINGUAL or BUCCAL administration of progesterone is effective in women if the compound is dissolved in propylene glycol.

INTRASPLENIC, INTRAHEPATIC or INTRAPERITONEAL administration of progesterone is less effective than if the same amount is given under similar con-

ditions by the subcutaneous or intramuscular route. This is presumably due to detoxification during the passage through the liver, and to the fact that from these sites progesterone is especially rapidly absorbed, hence it is eliminated before it could fully exert its luteoid properties.

INTRAVENOUS administration of luteoids is rarely advisable, since in this event detoxification and excretion are too rapid to permit optimum efficacy.

The direct application of progesterone to the ENDOMETRIUM causes a local pregestational proliferation, indicating that the compound acts directly upon the uterine lining. Advantage has been taken of this in the bioassay of the compound when only minute doses are available. There are no clinical indications, however, for any type of topical application of luteoids.

WITHDRAWAL EFFECTS AND PERMANENT CHANGES CAUSED BY TEMPORARY TREATMENT

After adequate folliculoid treatment, whether or not it is followed by luteoid administration, WITHDRAWAL BLEEDING occurs from the primate endometrium. In most of the other vertebrates however, the endometrium involutes without breakdown after hormone withdrawal.

It is also noteworthy that PERMANENT CHANGES MAY BE CAUSED BY TEMPO-

RARY TREATMENT with folliculoids. Thus, in very immature or embryonic male animals, excessive treatment with folliculoids may cause permanent damage in the testes and accessory sex organs.

OTHER PHARMACOLOGIC PROBLEMS

Other pharmacologic problems concerning the folliculoid and luteoid hormones, as well as their biogenesis and fate in the body will not be reviewed here, since they have been discussed in the section : The Steroids and on p. 372.

Suffice it to re-emphasize that both folliculoids and luteoids appear to be detoxified mainly in the liver; partly by the formation of conjugates, and partly by oxidation or reduction. Certain folliculoids (e.g., estriol) are more active in intact than in spayed animals; this suggests activation in the ovary.

Numerous steroids are comparatively more active upon systemic than upon direct local application to the responsive target organ; this also suggests transformation of *pro-folliculoids* into the fully active folliculoids in some (unidentified) internal organ.

"*Relaxin*" (see p. 345) has been extracted from corpora lutea and pregnancy blood. Unlike folliculoids and luteoids it causes relaxation of pelvic ligaments even after hysterectomy.

EXPERIMENTAL PHYSIOLOGY OF THE OVARIES

EXPLANTATION OF THE OVARIES

Various investigators succeeded in preparing viable TISSUE CULTURES of ovarian parenchyme in a variety of media. Not only the stroma, but even the epithelial elements, especially the granulosa, seem to be capable of proliferation in vitro. The ovocytes may divide and proliferate. Exposure of ova to changes in temperature, hypertonic or hypotonic solutions, as well as insemination even with foreign spermia, leads to precleavage activity in the egg nu-

cleus, extrusion of a polar body and formation of pronuclei (Pincus, 1940). As a rule, immature (especially embryonic) gonads grow better in vitro than adult ovarian tissue. Even human embryonic ovaries can grow in vitro to some extent.

ORGAN CULTURES of cat ovaries have been successful using the Carrel-Lindbergh technic. At the end of several days, the ova in their different stages of maturation were well-preserved and the corpus luteum tissue, as well as

other cellular elements, appeared normal and indistinguishable from control ovaries fixed immediately after their removal from the body.

It is especially noteworthy that frozen, human ovarian tissue can be preserved *in vitro* for several days in a viable condition. Tissue cultures prepared from such material grew successfully after preservation at -12°C (Zondek and Wolff, 1924). Because of its possible practical applicability, this discovery stimulated a good deal of research concerning the *optimum temperature* for the preservation of ovarian tissue *in vitro*. Most of the relevant work was performed on guinea pig ovaries, whose viability was checked by subsequent transplantation into spayed female or intact male guinea pigs. Successful grafts were readily detected by their "feminizing" effect (nipple growth, lactation) and by the histologic structure of the transplants. At the optimum temperature (just a few degrees above the freezing point) guinea pig ovaries may be preserved in a viable condition *in vitro* for more than two weeks (Lipschütz, 1927-1932).

The ovary never took when preserved at temperatures beneath 0°C .

Partial drying of the ovary, sufficient to cause a 35-57% loss of weight during preservation *in vitro*, does not interfere with its survival and taking, as shown by similar experiments on the guinea pig.

As regards the *metabolism of ovarian tissue* *in vitro*, it is noteworthy that a slight oxygen consumption continues for several days, even at temperatures below $+1^{\circ}\text{C}$. The metabolism of the rat ovary rises during estrus and falls during the diestrus period. Furthermore, the ovaries of immature rats have a lower metabolism *in vitro* than those of adults.

TRANSPLANTATION OF THE OVARIES

Several investigators claim to have performed successful heterotransplantations of ovarian tissue, at least among closely-related species. Thus, following transplantation of mouse

ovaries into the ovarian capsule of rats, the subsequently discharged mouse ova can allegedly be fertilized and give rise to normal gestations in the uterus of the host rat. This claim requires further confirmation. It is undoubtedly true, however, that fertilized ova may be removed from the uterus and successfully transplanted into the uterus or even the peritoneum of other animals of the same species.

As with other endocrine glands, autotransplantation of the ovaries has the greatest chance of survival, homotransplants are somewhat less likely to be successful, while heterotransplants take only in very exceptional instances (if ever) because of the incompatibility of the tissue proteins of different species.

In women, autotransplants of ovaries (removed because of tumors, inflammations or other diseases in their vicinity) have frequently been successful; even pregnancies have been reported following ovarian autotransplantations into the Fallopian tubes. Nevertheless the likelihood of a successful ovarian graft is much smaller in man than in most laboratory animals. Since we now possess highly effective hormone preparations to compensate for the loss of endocrine activity after castration, the operation is rarely justified, except for the preservation of fertility.

In the presence of adequate ovarian or testicular tissue, ovarian grafts do not tend to develop well. The results are better after unilateral and best after bilateral OVARIECTOMY or TESTIS EXTRIPATION. In the body of the female castrate, corpus luteum formation, while in that of the male castrate, follicle maturation prevails, presumably because the male pituitary produces predominantly FSH, that of the female LH.

Intrasplenic transplants of ovaries in spayed animals often develop into folliculomas or luteomas. This is probably due to the fact that the hypophysis is freed of ovarian control when the ovarian hormones are first forced to traverse the liver, in which they are detoxified. Excessive luteinization of such grafts is readily accomplished by folliculoids. Progesterone or desoxycorticosterone treatment prevents this luteinization (see p. 417).

In HYPOPHYSECTOMIZED animals, ovarian grafts rarely take unless exogenous gonadotrophins are administered. All

these observations are consonant with the view that in grafts — as in the normal condition — the development of ovarian tissue is dependent mainly upon hypophysoid trophic hormones, and comparatively independent of nervous stimuli, which are, of course, eliminated in the transplants.

PARABIOTIC union of a castrate bearing an ovarian transplant with a castrate male or female partner causes excessive stimulation of the grafted ovary, due to the increased gonadotrophic hormone production of the hypophysis in the gonadectomized twin. In this respect again, the graft reacts like the normal ovary in similar parabiotic pairs.

TECHNIC OF OVARIECTOMY

In FISH, the usual procedure is to make a midline incision, using clean (but not necessarily sterile) instruments. The ovaries may then be removed by blunt dissection or by thermocautery. The operation has a comparatively high mortality.

In AMPHIBIA and most REPTILES the procedure is approximately the same as in fish, but it must be kept in mind that in toads, Bidder's organ (see: Comparative Morphology) undergoes compensatory hypertrophy following ovariectomy and this may interfere with the development of typical castration changes.

In BIRDS, removal of the well-developed left ovary is comparatively simple. After plucking the feathers, an incision is made between two ribs in the ovarian region and a self-retaining retractor is inserted into the wound, so as to spread the ribs as far as possible. The air sacs, which tend to interfere, may either be pushed aside or transected. After this, all the ovarian tissue is removed, tearing the hilum between two forceps. Vessel ligatures are rarely necessary, except during the egg-laying period, when the vascularity of the organ is

excessive. Two stitches put through skin and muscle layers suffice to close the wound. In very young chicks, we found it useful to remove the ovarian tissue by means of suction applied through a fine glass cannula.

It is somewhat more difficult to remove the right ovary which is vestigial in most birds. However, if the left ovary had previously been removed, the right gonad becomes more conspicuous.

In most LABORATORY MAMMALS (cat, dog, rabbit, rat, mouse, etc.) ovariectomy is a very simple procedure. It may be performed from a single suprapubic incision, pulling the ovaries into the operative field by traction on the uterine horns and oviducts. Some prefer two separate costo-lumbar incisions. For most purposes (especially for bioassays in rodents) it is best to remove the ovary together with the ovarian pouch and part of the oviduct in order to ensure that no remnants are left behind.

In MAN, ovariectomy at any age is also simple as long as the gonad is not tumorous or adherent to its surroundings. In typical cases, the usual procedure is to open the abdomen through a suprapubic, midline incision, after the customary preparation for abdominal operations. The gonad is seized with Allis clamps and 2-3 ligatures are placed through the pedicle; after this the hilum is severed and the wound carefully peritonized, in order to prevent the formation of adhesions.

Cystic ovaries may have to be punctured before they can be delivered through the abdominal incision, but care must be taken not to spread the cyst fluid into the peritoneum, especially if there is suspicion of infection or malignant transformation of the cyst. In many cases (e.g., malignancy) the technic of choice is HYSTERO-OVARIECTOMY.

VAGINAL OVARIECTOMY is rarely performed, although certain cysts and abscesses may become so solidly adherent

to the vaginal wall that an extraperitoneal, trans-vaginal approach may be advantageous.

PARTIAL OVARIECTOMY may take the form of mere excision or ignipuncture of medium-sized surface cysts, complete decortication of the ovary, or removal of the major part of its substance by wedge excision.

Under modern surgical conditions, ovariectomy in itself is not a dangerous operation and the wound usually heals by first intention without complications.

Instead of surgical ovariectomy, X-RAY CASTRATION is sometimes the

method of choice, as in the treatment of certain inoperable ovarian tumors, the production of an artificial (permanent or temporary) menopause, etc.

It will be seen from the section devoted to the various ovarian diseases that temporary sterilization due to X-ray amenorrhea has been recommended in functional uterine hemorrhages, endometriosis, inflammations of the ovary and diseases in which pregnancy would be dangerous. The details of ovarian irradiation as practised today have been summarized in the following table (Schmitz, 1944):

**Roentgen Doses in r Units for the Production of Permanent and Temporary Amenorrhea
Measured at the Ovary**

(Factors of Production : 195 KV., 0.5 mm. Cu+1.0 mm. Al filter, 50 cm. F.S.D.)

AGE GROUP	Permanent amenorrhea				Temporary amenorrhea			
	Me	My-a	My-b	My-c	Me	My-a	My-b	My-c
A — 20 to 25 yr	321	324	373	462	276	277	316	400
B — 26 to 30 yr	308	311	358	444	263	266	302	382
C — 31 to 35 yr	289	298	342	424	250	254	289	362
D — 36 to 40 yr	282	284	327	398	239	242	277	342
E — 41 to 45 yr	269	271	310	379	226	230	261	324
F — 46 to 50 yr	254	256	295	360	213	218	248	307
G — 51 to 55 yr	241	243	279	341	200	207	236	290
H — 56 and over	226	230	264	321	189	195	224	272

Me = Hemorrhagic metropathy or myopathy

My-a = Myoma reaching to symphysis pubis

My-b = Myoma reaching midway between symphysis and umbilicus

My-c = Myoma extending above umbilicus

Among the dangerous complications of *X-ray castration* (due mainly to faulty technic or individual variations in X-ray sensitivity) are: damage to the skin, unintentional production of permanent amenorrhea, and the danger of causing mutations and deformities in the offspring following temporary castration.

It is presumed, though not definitely proven, that while large doses of X-rays

destroy, small doses stimulate ovarian function.

EFFECTS OF OVARIECTOMY AND TREATMENT WITH OVARIAN HORMONES

State — The main effects of OVARIECTOMY are the typical manifestations of hypogonadism, and so-called "menopausal disturbances" (see : Diseases of the Ovary). In general it may be said that the younger the individual

at the time of ovariectomy, the more striking are the resulting somatic and psychic changes. In ovariectomized prepubertal children, the entire development of the sex organs (breasts, uterus, pubic hair, etc.) is severely inhibited, but after puberty the involution caused by spaying is less striking, and in postmenopausal women ovariectomy often fails to exert any detectable effect. Usually, though not always, women gonadectomized during adult life tend to develop excessive adiposity; in many patients this is merely due to the psychosomatic effect of castration on appetite.

FOLLICULOIDS rarely produce any pronounced signs of acute overdosage and even excessive amounts are comparatively well tolerated. On the other hand, adrenal deficiency, hypopituitarism or extensive destruction of the liver sensitize to the toxic effects of folliculoids, apparently because the anterior-lobe and the adrenal cortex play an important part in adaptive defence reactions against folliculoids, while the liver is the chief site of their detoxification.

In premature infants, treatment with folliculoids is claimed to exert a beneficial effect upon development in general. It has also been claimed that resistance to certain infections and intoxications can be altered (increased or decreased, depending upon conditions) by ovarian hormones.

There is some evidence indicating that folliculoids stimulate plant growth.

LUTEOIDS are not known to exert any characteristic effects upon the general condition of animals or man. Even in the largest doses so far administered, they failed to cause toxic overdosage phenomena; they may, however, produce anesthesia.

Temperature. — Neither ovariectomy nor treatment with ovarian hormones induce significant changes in body temperature. The skin tempera-

ture may be slightly raised by the vaso-dilator effect of folliculoids and during menopausal flushes. It is very probable that the characteristic mild variations in body temperature during the menstrual cycle, are likewise conditioned by ovarian hormones.

Tissue Metabolism. — The metabolism of the accessory sex organs, which are normally stimulated by folliculoids, is directly increased by these hormones *in vitro* (e.g., uterus, vagina). The effect of folliculoids upon the metabolism of other tissues is not conspicuous outside the body.

Basal Metabolism. — Ovariectomy often causes a slight decrease in the B.M.R. while folliculoids tend to raise it.

Carbohydrate Metabolism. — In women ovariectomy or treatment with luteoids causes no prominent changes in carbohydrate metabolism. On the other hand, folliculoids may elicit severe diabetes in animals (e.g., rat), particularly after sensitization by partial pancreatectomy and forced feeding with a high carbohydrate diet. This diabetogenic effect is not entirely mediated by the adrenals since, to some extent, it is still demonstrable after adrenalectomy.

The accumulation of glycogen in the endometrium during the luteal phase is the direct effect of progesterone action. Conversely, the lining cells of the vaginal epithelium, which are free of glycogen in postmenopausal and castrate women, accumulate this carbohydrate under the influence of folliculoids.

Lipid Metabolism. — The ovary exerts no very prominent and characteristic effects upon lipid metabolism, although fat deposition is often increased in ovariectomized women. In birds, treatment with folliculoids causes very marked lipemia and cholesterolemia, presumably because the ovarian hormones play an important part in the

formation of the large lipid-containing egg-yolk.

Nitrogen Metabolism.—The metabolism of protein and other nitrogenous products is not characteristically influenced by ovariectomy or the administration of ovarian hormones.

Water and Salt Metabolism.—Ovariectomy causes no very typical change in water and salt metabolism. It has been claimed that folliculoids decrease the URINE OUTPUT, but this effect is rather inconstant. In high doses progesterone exerts a marked diuretic effect in the rat and under certain con-

ditions occasionally also in women with premenstrual edema.

The BLOOD CALCIUM concentration is markedly increased by folliculoids in birds (probably related to egg-shell formation). In mammals this effect is extremely slight (see also: Effect of Folliculoids on Bone Formation).

It has been claimed that both folliculoids and luteoids decrease Na and Cl excretion, but this has not been confirmed. The blood chloride concentration, however, increases slightly following folliculoid treatment in most animal species, as well as in man.



A



B



C



D

Effect of folliculoids upon the bones.—A. Section through the femur of a mouse given 250 γ of estradiol benzoate during 41 days. A large part of the distal marrow has been replaced by bone; the epiphyseal cartilage is thin due to reduction of both the proliferating and hypertrophic layers. — B. Ground section through the femur of a control (bottom) and folliculoid-treated mouse showing the extent of endosteal ossification. — C. and D. Long bones from an untreated (top) and folliculoid-treated pigeon showing marked endosteal bone proliferation. Similar bone deposition occurs normally during the egg-laying period.

(Courtesy of
Dr. W. U. Gardner.)

Growth and Bone Structure. — Prepubertal ovariectomy tends to increase somatic growth and to delay the ossification of junction cartilages. On the other hand, folliculoids (but not luteoids) inhibit growth in length and cause premature ossification of the epiphyses. In the case of chronic treatment with folliculoids, the proliferation of bone tissue may cause a severe reduction of the marrow spaces, conducive to anemia. This effect is particularly marked in birds, but also clearly demonstrable in mammals.

It is presumably a direct action since in ducks local bone proliferation was noted after injection of folliculoids into bones.

In addition to the above-mentioned actions on the entire skeleton, folliculoids also exert a specific effect upon the pubic bones. Simultaneously with the relaxation of the pelvic ligaments induced by these hormones, the adjacent parts of the pubic bones undergo absorption. Certain corpus luteum extracts (containing "relaxin") are especially active in this respect. The underlying process is probably essentially analogous to the physiologic preparation of the pelvis for delivery.

Blood. — In certain species, especially in the dog, chronic treatment with high doses of folliculoids causes severe, sometimes fatal, anemia. The pathogenesis of this change is not clearly understood. It cannot be entirely ascribed to obliteration of bone marrow cavities by proliferating bone tissue, since it is not necessarily accompanied by excessive osteosclerosis. In birds and some rodents where such obliteration is especially prominent, this in itself largely accounts for the resulting aplastic anemia.

BLOOD COAGULATION is slightly delayed following ovariectomy in most species, and the platelet count tends to decrease, but these actions are inconstant.

Cardiovascular System. — The **BLOOD PRESSURE** is likewise not significantly altered by ovariectomy or by ovarian hormone administration. The

claim that *folliculoids* increase the blood pressure in the rat has not been substantiated. Indeed, women with menopausal hypertension often show a decrease in blood pressure following folliculoid hormone therapy, although this effect is less marked than the beneficial action upon the flushes.

Folliculoids exert a vasodilator effect which is readily verified by direct inspection (e.g., in the rabbit ear), or by the resulting increase in organ volume (e.g., finger volume in women). The vessels of the nasal mucosa, and those of the accessory sex organs controlled by folliculoids, appear to be especially sensitive to this effect. The innervation of the blood vessels does not appear to play an important rôle, at least as regards the vasodilatation in accessory sex organs, since this effect persists in transplants of uterine tissue. It has been assumed that the action is due to a peripheral acetylcholine discharge caused by folliculoids. Encouraging results have also been reported after folliculoid therapy in: acrocyanosis, angina pectoris, diabetic gangrene and endarteritis obliterans; but in these conditions the value of this treatment is still controversial.

Luteoids do not appear to share with desoxycorticosterone the ability to raise the blood pressure. Under certain conditions they are even claimed to diminish the high blood pressure of animals with experimental renal hypertension.

The **HEART** action is not significantly influenced by ovarian hormones although in certain cases the allegedly typical electrocardiogram of hypoovarian women and the cardiac manifestations of hyperthyroidism have been claimed to respond favorably to folliculoids.

Lymphatic System. — Ovariectomy tends to enhance, while treatment with folliculoids markedly depresses the development of the thymus; the spleen,

bone marrow, lymph nodes and other lymphatic accumulations respond similarly though less markedly. Luteoids are almost inactive in this respect.

Muscles. — Ovariectomy diminishes, while folliculoids increase the spontaneous muscular activity of certain animals, especially the rat. This is reminiscent of the increased spontaneous muscular activity during estrus.

Nervous System. — Ovariectomy sometimes, but not always, causes disappearance of LIBIDO in women and this may be restored by folliculoids. In certain animals (e.g., rat), normal females exhibit both feminine (sexual receptivity) and masculine (clasping reflex, pelvic thrusts, etc.) sexual patterns, and ovariectomy abolishes only the former. Injection of folliculoids and luteoids in such cases restores the female sexual behavior without influencing the male pattern. Injection of folliculoids into fowl eggs during the period of incubation leads to the development of intersexual males, whose reactions may vary between perfect masculine behavior patterns to neutral (inactive) behavior. Even chicks, in which folliculoid treatment was started as late as the 15th day of life, come to resemble sexually-receptive hens, in that they squat for treading males after 3 weeks of treatment.

The normally very stable social position of hens in the "peck-order" (the right to peck others without being pecked in return) of a flock tends to fall after folliculoid treatment. The ovariectomized chimpanzee on the other hand (unless treated with folliculoids) is apparently considered "socially inferior" by intact females, as judged by her low priority rating in obtaining bananas available in a common cage. Many other observations indicate that folliculoids are a social asset among the primates.

In most species, treatment with folliculoids evokes the typical female mating behavior and tends to inhibit

the maternal instinct. In some animals (e.g., guinea pig) simultaneous or consecutive administration of progesterone greatly enhances this effect. In males, however, folliculoids decrease the normal sexual drive and may even induce homosexual female behavior.

The complex hormonal regulation of sexual drive is very incompletely understood as yet. Depending upon the species examined, and the dosages used, progesterone can even decrease and testosterone increase the female type of sexual behavior induced by folliculoids. Clinical experience indicates however, that in man, folliculoids tend to increase the normal libido in the female and to decrease it in the male.

When given in very large doses, progesterone causes ANESTHESIA in various experimental animals. Folliculoids are very much less active in this respect. (See : The Steroids.)

Certain analeptic drugs (e.g., metrazol) are capable of awakening rats from progesterone anesthesia and conversely, the convulsions induced by the analeptic drugs are counteracted by luteoids. The well-known lassitude and somnolence of pregnant women could be, perhaps partly, due to excessively produced progesterone or its derivatives.

Digestive System. — The epithelium of the ORAL CAVITY tends to become atrophic following ovariectomy, and this change is corrected by folliculoid administration. (See : Menopause.)

In certain species (especially the mouse) folliculoids cause atrophy of the tubular portions in the SALIVARY GLANDS.

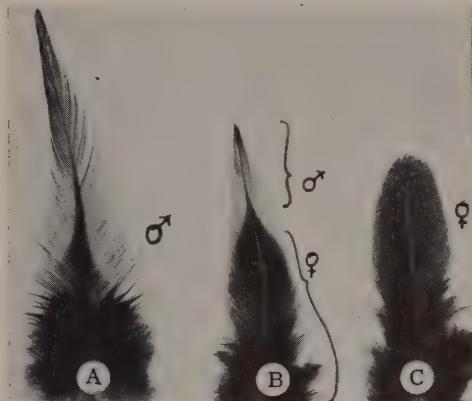
Neither ovariectomy nor treatment with ovarian hormones causes any consistently significant change in the morphologic structure, contractility and secretion of the INTESTINAL TRACT.

Degenerative changes in the LIVER, sometimes accompanied by necroses and jaundice, are produced by heavy overdosage with folliculoids in some ani-

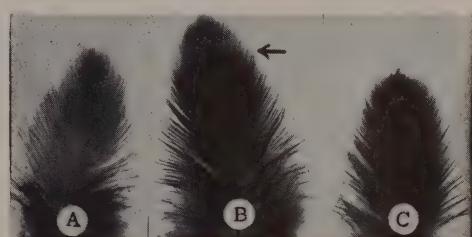
mals (e.g., certain strains of mice and rats) but hardly ever in man. This may be related to the hepatic detoxification of folliculoids. Luteoids exert no similar effect.

Skin. — Under the influence of folliculoids, the SEBACEOUS GLANDS of the skin involute. This may be responsible for the beneficial action of these hormones in acne. Curiously, in some women with secondary amenorrhea, acne may develop during folliculoid therapy if they previously suffered from this skin lesion at the time of menstruation. Folliculoids also tend to cause proliferation and increased keratinization of the dermal SURFACE EPITHELIUM. These effects, as well as a certain degree of local HYPEREMIA, can also be elicited by topical application of folliculoids to certain skin regions. In some animals (e.g., rat) there is severe loss of hair following chronic folliculoid hormone overdosage. In hirsute women, however, this depilatory action is rarely noticeable.

In certain strains of hairless mice GENERALIZED SKIN EDEMA ensues if folliculoids are given, while in most birds under the influence of such treatment, the PLUMAGE of male or gonadectomized animals becomes "hen-feathered."



Effect of ovary upon plumage. — A. Saddle hackle of spayed female silver dorking ("male type"). — B. Saddle hackle growing at time of ovarian regeneration. Lower part of female, upper of male type. — C. Saddle hackle growing after ovarian regeneration has occurred. The entire feather is of "female" type.



Effect of ovariectomy upon plumage. — A. Breast feather of normal brown leghorn pullet. The feather is brown. — B. Similar feather in which the part below the arrow grew after ovariectomy; this part is black. — C. Feather entirely grown after ovariectomy. The whole feather is black.



Effect of folliculoids on "sex skin". — A. Adult female Rhesus monkey treated with 200 mg. of diethylstilbestrol per os during one month. Note intense swelling of the skin not only in the perineal region but also along tail, back and thighs. This was accompanied by marked hyperemia. — B. Similar Rhesus monkey treated with increasing doses of 5-200 mg. of diethylstilbestrol/day and receiving a total of 38 gm. (!) during a period of one year. Note that under the influence of continued treatment with this folliculoid the swelling of the sex-skin region was not maintained and the skin on the back and thighs returned approximately to normal.

Even these huge amounts of folliculoid caused no neoplasia or anemia in this species.

During estrus the "SEX SKIN" in the genital region of certain primates (e.g., *maccaca mulatta*) becomes hyperemic, the derma undergoes gelatinous transformation and the epithelium thickens. Ovariectomy abolishes this cyclic skin-transformation, while folliculoids elicit it at any time, even in the spayed female or male. Following prolonged folliculoid treatment (weeks) this skin edema tends to become generalized involving most of the body surface, but after still longer treatment (months) it disappears in spite of continued hormone administration. Luteoids inhibit the sex-skin swelling elicited by normal estrus or by folliculoids.

The PIGMENTATION of the skin especially around the nipples is increased by folliculoids in women as well as in certain animals (e.g., guinea pig). This may have something to do with the "chloasma uterinum" characteristic of pregnancy and some ovarian dysfunctions.

It is questionable whether the induction of NUPTIAL-COLORING in the skin of lower vertebrates, especially fish (e.g., bitterling) is specifically due to folliculoids, since many other compounds have similar effects.

Urinary System. — Neither ovariectomy nor ovarian hormones exert important actions upon the KIDNEY, though folliculoids tend to cause atrophy of the renal tubules, sometimes accompanied by degenerative changes. Sometimes there are also extensive hematogenous (yellowish-green) pigment-deposits in the convoluted tubules. These presumably result from the erythrocyte destruction occasioned by marked hyperfolliculoidism. They are especially prominent, and accompanied by generalized icterus, in rats receiving thyroxine and folliculoids simultaneously. In certain species (e.g., mouse) there is also: distention of the ureters; hypertrophy of the walls of the ureters, bladder and urethra; and sometimes metaplasia of

the bladder epithelium or formation of urinary calculi.

Accessory Sex Organs. — We designate as "accessory sex organs," or sex characteristics, those structures other than the gonads which are characteristic of one sex. These may be subdivided into: (1) female, (2) male, (3) neutral and (4) bisexual sex characteristics.

In general, the female accessory sex characteristics are stimulated by ovarian hormones, and their physiologic development is inhibited by ovariectomy. The male accessory sex characteristics are dependent upon testoids and their normal development is impeded by orchidectomy. Neutral characteristics may occur in either sex, but their development is only inhibited by the hormones of the sex in which they are normally absent. Probably "neutral" sex characteristics are independent of the gonads and the hormones of the sex in which they are normally absent merely inhibit their development. Bisexual characteristics are identical manifestations of male and female sex hormones.

This somewhat complex problem may best be illustrated by the example of the FOWL, in which all four types of accessory sex characteristics are clearly distinguishable.

A typical *female* accessory sex organ of the fowl is the oviduct, which is well-developed in the sexually mature female, regresses after ovariectomy and is restored by folliculoid hormone administration.

Typical *male* sex characteristics are the comb, wattles, ear lobes, and the characteristic fighting instinct of the cock. They are well-developed in the sexually mature males, vanish following castration and are restored by testoids, but not by luteoids or folliculoids.

The spurs and the brilliant long feathers (especially the tail feathers) so typical of the male, are actually neu-

tral characteristics. They do not disappear after castration in the male and

appear following ovariectomy in hens. Testoids have no effect upon their development in intact or gonadectomized animals of either sex, but folliculoids specifically cause them to involute in intact or castrate males, as well as in spayed females, in which they are normally well developed. This explains why these characteristics, though essen-



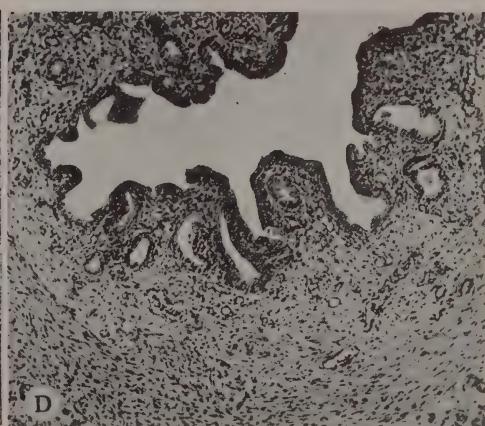
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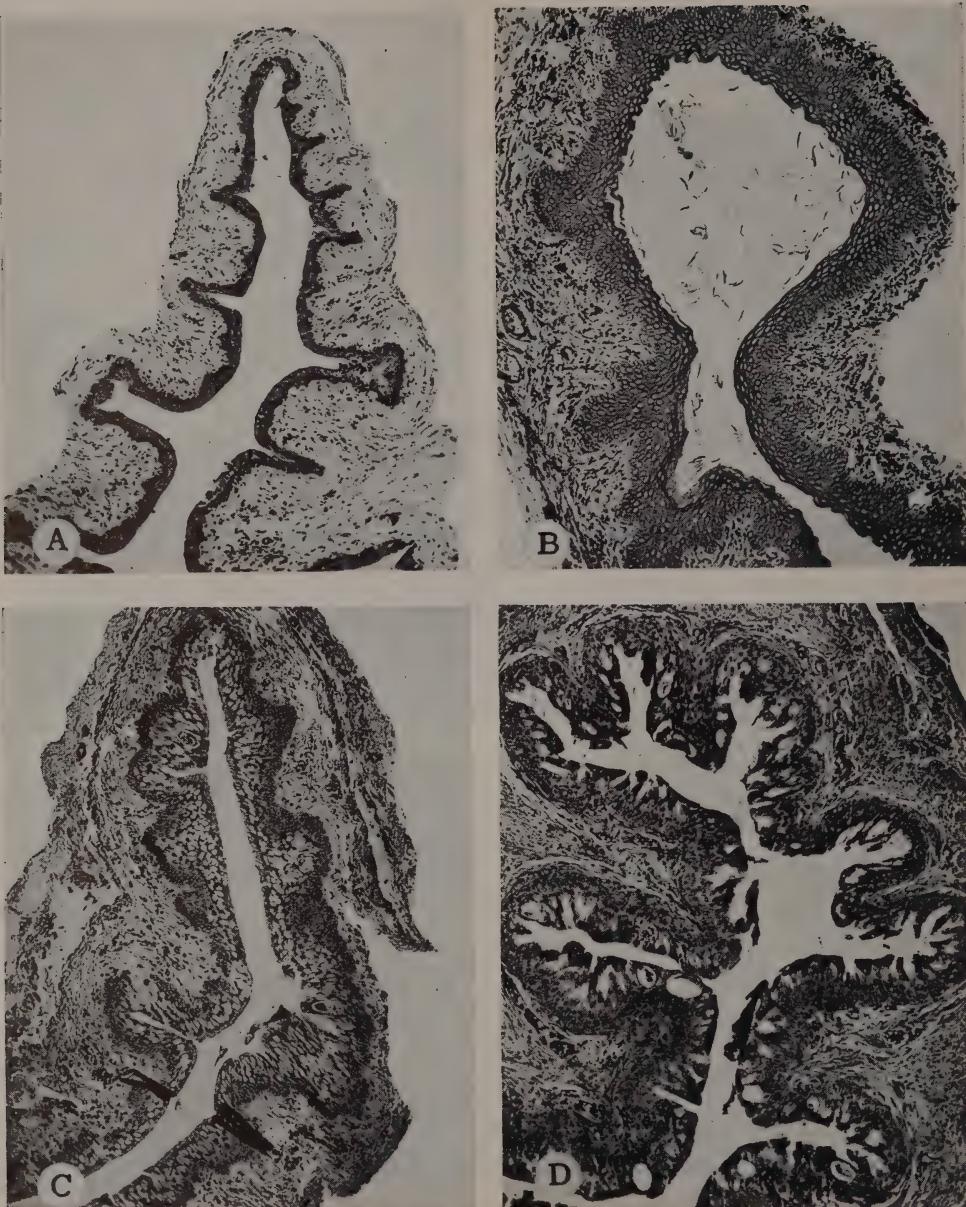


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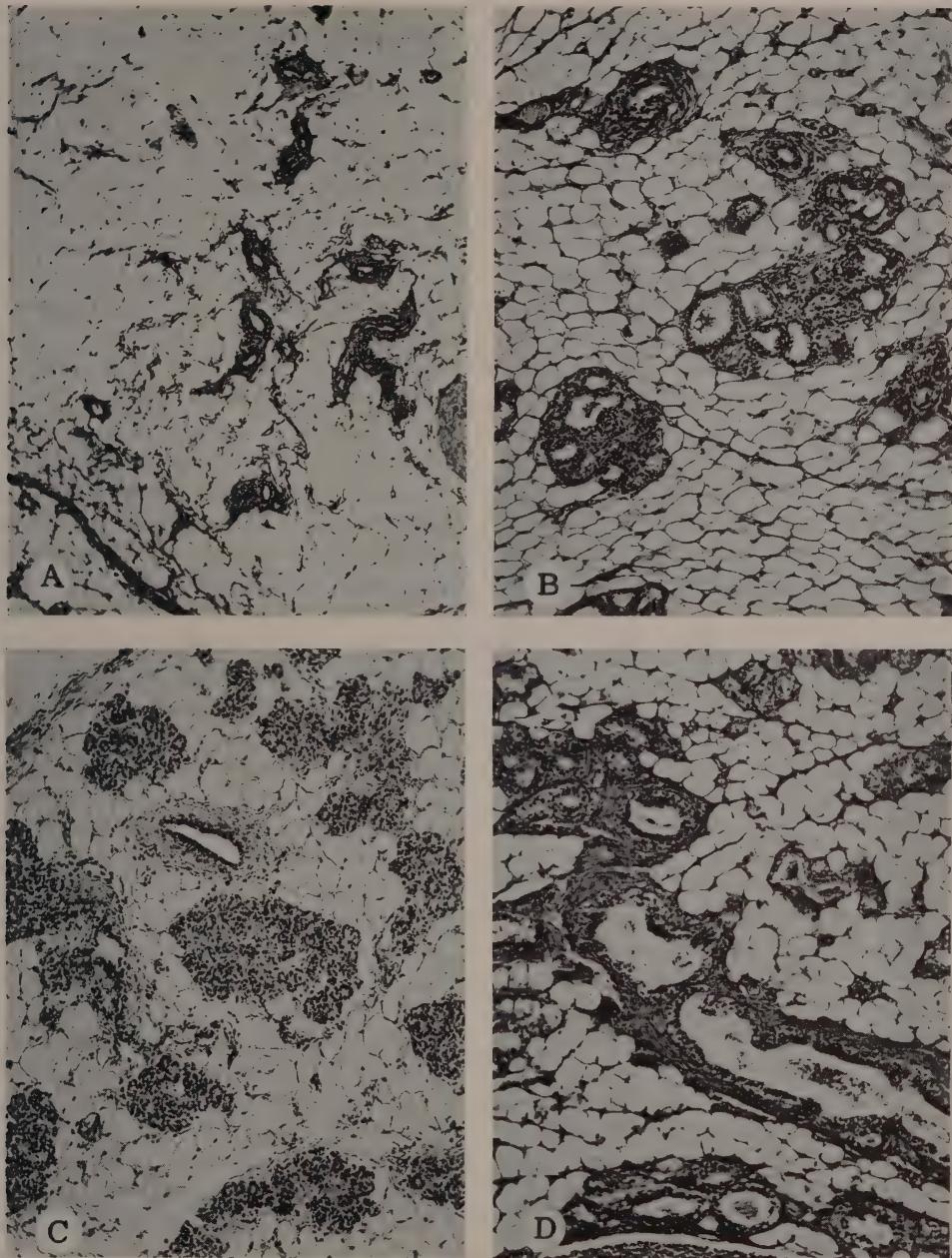


D

Effect of various hormones upon the uterus of the spayed female rat. — A. Cross-section through the uterus, of an adult ovariectomized control rat. Note pronounced atrophy of the epithelium, thin uterine wall and narrow lumen. (Same low magnification as figure B, C and D.). — B. Uterus of an adult spayed female rat in which estrus was induced by ten days' treatment with $5 \gamma/\text{day}$ of ethynodiol-estradiol. Note great enlargement of uterine lumen (so that only a small part of the circumference appears in the field). Hypertrophy of muscular layer and endometrial stroma (the latter is infiltrated with numerous eosinophils which appear as dark dots in this picture) and great enlargement of the endometrial cells, which assume a high-cylindric form. — C. Uterus of an adult spayed female rat in which progestational changes have been elicited by treatment during ten days with 15 mg./day of progesterone. Note hypertrophy of the muscle cells and pronounced "lacing" of the endometrium. Similar changes can be produced with much smaller amounts of progesterone if the animals are previously sensitized with folliculoids. The experiment shows, however, that such sensitization is not indispensable. — D. Uterus of an adult spayed female rat which received, during ten days, 5 mg./day of testosterone. Note that this testoid induced changes very similar to those obtainable by progesterone.



Effect of various hormones upon the vagina of the spayed female rat. — A. Vagina of a spayed female control rat. Note severe atrophy of the epithelium which consists of two layers of cells only. The basal layer is the matrix, the inner surface-lining exhibits a cuboidal cell type. (Same magnification as fig. B, C and D). — B. Vagina of a spayed adult rat in which estrus has been induced by treatment during ten days with 5γ /day of ethynodiol diacetate. Note stratification of the epithelium with cornification of the surface layer. Several cornified cells are shed off into the lumen. — C. Vagina of a spayed adult rat after 10 days' treatment with 15 mg./day of progesterone. Note stratification of the epithelium with pronounced mucification (white cells) of the inner layers. Similar changes can be produced with much smaller doses of progesterone in animals previously sensitized with folliculoids, but the experiment indicates that progesterone alone suffices to elicit this reaction, if the dose given is sufficiently large. — D. Vagina of a spayed adult female rat following 10 days' treatment with 5 mg./day of testosterone. Note that mucification is essentially similar to that produced by progesterone. The intra-epithelial cysts, which are particularly prominent in this field, may occur after progesterone treatment, although not as commonly as with testosterone.



Effect of various hormones upon the mammary gland of the spayed female rat. — A. Mammary gland of an adult spayed female control rat. Note that only a few atrophic ducts are discernible in the adipose tissue. (Same magnification as fig. B, C and D). — B. Mammary gland of an adult female rat, following 10 days treatment with 5 γ /day of ethynodiol diacetate. Note great proliferation of ducts whose lumen is enlarged and lined by a hyperplastic epithelium. — C. Mammary gland of an adult spayed female rat, following 10 days treatment with 15 mg./day of progesterone. Note proliferation of solid epithelial acinar tissue; ducts not particularly developed. Similar changes can be obtained with much smaller doses of progesterone in animals sensitized with estradiol. The present experiment merely proves that, if sufficiently high doses are given, progesterone alone suffices. — D. Mammary gland of an adult spayed female rat following 10 days treatment with 5 mg./day of testosterone. Note that the testoid also causes mammary development. Both ducts and acini are stimulated, though not as markedly as with folliculoids or luteoids.

tially neutral, are physiologically nonetheless characteristic of male birds.

The paucity of fat deposits is a *bisexual* characteristic since the adiposity of male or female castrates can be corrected by sex hormones of either sex.

The above statements hold for instance in most of the common varieties of fowl. It must be kept in mind however, that in many birds the development of the accessory sex organs follows a different pattern and their hormonal regulation, in each species, is not yet fully clarified. There are no clear-cut instances of neutral sex characteristics in man, although the elongation of the extremities in comparison with the trunk is a closely related phenomenon since it tends to occur after early gonadectomy in either sex, conversely, pubic and axillary hair growth is bisexual as it is stimulated by either type of sex-hormone.

It would hardly be profitable to discuss the hormonal regulation of the accessory sex organs in all the numerous species in which pertinent studies have been performed. Hence we shall limit our remarks to the most important laboratory MAMMALS and to man.

The *oviduct* involutes and its contractility diminishes following ovariectomy. Conversely, treatment with folliculoids stimulates its development in immature or castrate females. This effect is dependent upon an adequate supply of folic acid, especially in birds. Folliculoids also cause mitotic division and secretion of the lining epithelium and they enhance the motility of the oviduct musculature, thus promoting the passage of ova. Luteoids on the other hand, inhibit the motility of the oviduct in animals as well as in women.

The *uterus* involutes after ovariectomy. Folliculoids increase uterine size (myometrium and endometrium) and induce the so-called "follicular

phase or "estrous type" of endometrial change, while luteoids (especially if administered following pretreatment with folliculoids) cause progestational transformation, that is, the "luteal phase" type of mucosa. The histologic character of these changes is different in the various species but essentially, the folliculoids imitate the uterine changes of the preovulatory, and luteoids those of the postovulatory part of the sexual cycle.

In several animal species the motility of the uterus *in vitro* is increased by folliculoids and diminished by luteoids; in fact the latter can even counteract the oxytocic action of posterior-lobe hormones. However, in women the amplitude of the individual spontaneous uterine contractions and sensitivity to oxytocin actually increase under the influence of progesterone.

Chronic overdosage with folliculoids causes squamous metaplasia of the normally cylindric endometrial epithelium, especially in the rat. Traumatization of the endometrium elicits "endometrial moles" in folliculoid, and "deciduomas" in luteoid hormone, pretreated animals (see : Tumorigenesis, below).

The *vagina* responds essentially in the same manner as the uterus, inasmuch as it undergoes atrophy following ovariectomy, and estrous changes under the influence of folliculoids, while luteoids reproduce the vaginal changes normally seen during the luteal phase of the sexual cycle. (See : Sexual Cycle.)

The *clitoris* undergoes no great variations in its development, either following ovariectomy or under the influence of folliculoid and luteoid hormones. It appears to be a vestigial "male" organ, responsive mainly to testoids, which in the female are predominantly of adrenal-cortical origin.

The *preputial glands*, the ducts of which open near the clitoris, are parti-

cularly well-developed in rodents (e.g., mouse, rat) of either sex although they are normally larger in the male. They show but little atrophy following ovariectomy and are not significantly stimulated by folliculoids. Luteoids cause some increase in their size and secretion, but as in the case of the clitoris itself, these glands are mainly responsive to testoids. Crude anterior-lobe extracts enhance this effect of the testoids.

Bartholin's glands involute after ovariectomy and are selectively stimulated by folliculoids.

The mammary glands involute following ovariectomy and undergo proliferative changes (with little, if any, secretion) under the influence of folliculoid or luteoid hormones. This proliferation is most pronounced with mixtures of folliculoids and luteoids. Hypophysectomy almost completely prevents this effect and hence, it is probable that the mammary stimulation by ovarian hormones is predominantly mediated by the mammogenic principle of the anterior-lobe. Withdrawal of ovarian hormone treatment is usually followed by a transitory period of lactation.

In lactating animals and women large doses of folliculoids (but not luteoids) inhibit lactation. Advantage is taken of this in women when lactation is not desirable (stillbirth, abortion, mammary diseases, contagious diseases, etc.). Painful engorgement of the lactating, unemptied breast can be minimized in these patients by transitory treatment with high doses of folliculoids. Judged by experiments on the rat, combined treatment with folliculoids and luteoids exerts an even greater antilactation effect and folliculoids inhibit lactation more actively in intact (corpus luteum bearing) than in spayed rats. Very small doses of folliculoids actually stimulate milk secretion (e.g., goat).

All the *male accessory sex organs* undergo atrophy under the influence of folliculoids in intact animals and man. This is due to the resulting Leydig cell atrophy and not to a direct action on the peripheral target organs. In castrate males, whether the accessory sex organs are atrophic or well-developed due to testoid administration, folliculoids cause no inhibition. However, the folliculoids do exert a specific, direct stimulating effect upon certain male accessory sex organs. Thus either in intact or in castrate males (especially in rodents) they induce keratinization of the normally not-keratinized epithelium in the seminal vesicles and to a lesser extent, even in the prostate and the "utriculus prostaticus" (a "female" Müllerian vestige in the prostate). Simultaneously, there is marked proliferation of the fibrous and muscular tissue in the walls of the seminal vesicles (to a lesser extent in the prostate). The proliferation of these mesenchymal structures in the seminal vesicle walls has often been misinterpreted as a testoid effect, hence, it is well to keep in mind that not every increase in the weight of the seminal vesicles is necessarily indicative of the so-called "androgenic" stimulation.

Sexual Cycle. — The sexual cycle is interrupted by ovariectomy, folliculoids, or luteoids. Ovariectomy causes continuous anestrus in animals, and amenorrhea in women, because of the resulting sex-hormone deficiency. Extensive partial ovariectomies on the other hand, tend to cause continuous estrous changes due to the formation of persistent follicle cysts (see also page 376).

Folliculoids, if given in sufficiently high doses, interrupt the sexual cycle, because they cause ovarian atrophy and a continuously "follicular phase" endometrium (often metropathia hemorrhagica in women) or (at least in rodents) in high doses they prolong the life-

span and activity of the corpus luteum. In either case, the cyclic occurrence of a "withdrawal bleeding" is impeded.

High doses of luteoids maintain a pregestational endometrium beyond the normal span of the luteal phase and hence, they likewise prevent withdrawal bleeding at the end of each cycle. It must also be remembered that prolonged treatment with either folliculoid or luteoid hormones causes compensatory atrophy of the ovaries, a type of "functional castration." In senile or prepubertal individuals, brief treatment with folliculoids sometimes initiates a series of normal sexual cycles.

Pregnancy. — Contrary to common belief, it is extraordinarily difficult to produce abortion with folliculoids. In certain animals, exceedingly high doses have led to placental hemorrhages and abortion, but this is rather exceptional. Luteoids tend to prolong gestation, allegedly because they interfere with the uterine contractions which initiate delivery; however, as previously stated, progesterone increases the contractility of the human uterus *in situ* so that the prolongation of gestation must be due to a different mechanism.

Ovariectomy causes abortion in all animal species including man, if it is performed early enough, but the length of time during which the ovary is indispensable for the maintenance of pregnancy varies. In the rat, ovariectomy causes abortion at any time during pregnancy, while in women, after the third month of gestation, removal of the ovaries rarely interferes with the subsequent course of pregnancy and delivery. Apparently by that time the placenta has taken over the endocrine functions of the gonad so that the ovary becomes dispensable.

Lactation. — The influence of ovarian hormones upon lactation has been discussed above in connection with their other effects on the growth of

mammary tissue (see : Accessory Glands on page 367).

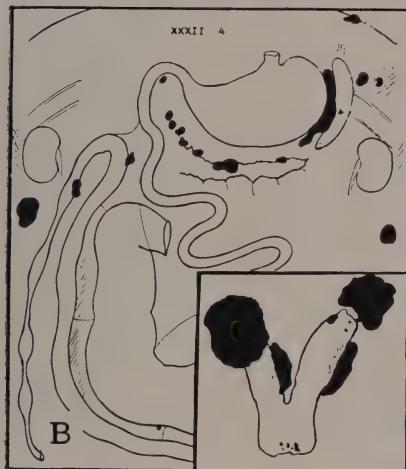
Fertility. — Not only ovariectomy, but even overdosage with folliculoids or luteoids may cause sterility due to disturbances in the cyclic transformations of the sexual organs and the compensatory ovarian atrophy.

Hibernation. — There is no reason to believe that ovarian hormones exert any important influence upon hibernation. The ovarian atrophy which occurs at that time is merely secondary.

Tumorigenesis. — The chemical resemblance between many of the "carcinogenic hydrocarbons" and the natural folliculoids has been emphasized in the section : The Steroids. There we also mentioned that many of these carcinogenic hydrocarbons exhibit marked folliculoid activity. We have reason to believe that the reverse is likewise true and that folliculoids can be carcinogenic.

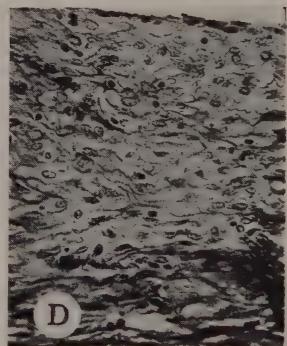
It has long been known to physicians that there are close relations between ovarian function and tumor formation. The great prevalence of carcinoma, especially in the accessory sex organs (uterus, mammary glands, etc.) during the climacteric age, and numerous experimental observations called attention to such interrelations.

In the UTERUS, cancer formation is especially frequent during the climacteric age when folliculoid hormone production, though diminished in quantity, is often more or less continuous because of the failure of corpus luteum formation. In women with folliculomas, in whom there is continuous excessive folliculoid production, uterine cancers are also common. In certain strains of mice (less readily in the rat), chronic treatment with folliculoids tends to produce endometrial cancers. It has not been clearly demonstrated that continuous folliculoid therapy can cause uterine carcinoma in women or in other primates (e.g., monkey), but "pre-



Experimental uterine fibroids. — A. Uterine and extrauterine fibroids in the abdominal cavity of the guinea pig, induced by six months' treatment with estradiol benzoate; 1, 2, 3 and 5, small tumors of the abdominal wall; 4 "apical" tumor of the mesosalpinx; 6, 7 and 8, tumors of the parametrium; 9, spheric mesenteric tumor; 10, enormous mesenteric tumor. — B. Diagram showing the typical localizations of abdominal fibroids induced by folliculoids in seventy-one days. Only 11 γ per day of α -estradiol absorbed from a subcutaneously implanted pellet. Parametric and "apical" tumors (inset). Tumors of the abdominal wall, diaphragm, pylorus, epiploon, mesocolon and spleen. — C. and D. Histologic structure of uterine fibroid induced in guinea-pig by three months' treatment with folliculoids. The tumor consists mostly of fibroblast-like, spindle-shaped cells and collagenous fibers.

(Courtesy of Drs. A. Lipschutz, R. Iglesias and L. Vargas.)



cancerous" lesions in the cervix uteri have been elicited with these hormones in *maccaca mulatta*. In the rat, chronic experimental hyperfolliculoidism causes keratinization of the entire endometrium.

Fibromyomas are also frequent in women suffering from hyperfolliculoidism. Since these growths tend to regress after ovariectomy, it is reasonable to assume that they are, at least partly, dependent upon stimulation by ovarian hormones. In the guinea pig, uterine and even extrauterine *fibromas* (usually without myomatous elements) can be produced by folliculoids. It is interesting that their development is inhibited by progesterone, testoids, and many other steroid hormones, which are commonly referred to as "anti-fibromatogenic." The relationship between these experimental fibromas, and the spontaneous fibromyomas of women has not yet been fully elucidated.

Whether *endometriosis* should be regarded as a neoplastic disease is debatable. It is noteworthy, however, that this invasive type of endometrial growth (which may even metastasize) is extremely prevalent among women with hyperfolliculoidism, never occurs before puberty and consistently regresses during the menopause or after castration. Experimental invasive endometriosis (with uterine glands penetrating the entire wall of the uterus and emerging under the peritoneum) has been produced by chronic folliculoid hormone administration in the guinea pig and rabbit.

Uterine polyps are likewise prevalent among women with hyperfolliculoidism; they also tend to regress after the menopause and ovariectomy. In rabbits and guinea pigs, chronic folliculoid treatment has been shown to produce experimental endometrial polyps. In this connection the frequent association of uterine polyps with *metropathia hemorrhagica* is also noteworthy, since the

latter is certainly due to hyperfolliculoidism.

The experimental *deciduoma* (or "placentoma") is a typical neoplastic growth elicited in the endometrium by local trauma. It can only be produced after pretreatment with luteoids, during the luteal phase of the cycle, or during pseudopregnancy, that is, in the presence of corpus luteum hormone. Here the rôle of both local trauma and ovarian hormones is manifest.



Experimental deciduoma. Numerous mitotic figures in a deciduoma which was produced in a spayed female rat, sensitized with estradiol and progesterone, prior to endometrial traumatization.

Endometrial moles are edematous, myxoma-like tumors of the endometrial stroma. They greatly resemble the so-called hydatidiform moles, which develop spontaneously from placental elements in women. The endometrial mole is elicited by uterine trauma in folliculoid hormone pretreated animals, especially the rat and rabbit. Like the deciduoma, the endometrial mole has a

limited life-span inasmuch as it regresses spontaneously in spite of continuous hormone treatment. It never metastasizes and is not invasive.



Experimentally-produced endometrial mole. Section through the uterus of a rat in which endometrial trauma, during the second half of gestation, produced a typical "endometrial mole." Note the epithelial lining cells and the hydropic stroma, which resembles that of the "hydatidiform mole" of the human placenta. Essentially similar tumors can be produced by trauma in folliculoid pretreated rats. The experiment indicates that during the second half of gestation there is apparently too much folliculoid hormone in the body to permit decidioma formation and under these conditions endometrial moles develop after trauma.

Many tumors of the MAMMARY GLANDS are indubitably under ovarian hormone control. In women with mammary cancers, ovariectomy has frequently been practiced with at least temporary success. Chronic treatment with folliculoids may cause a typical proliferation of mammary tissue, but it has not yet been shown that true mammary cancers can be provoked by such therapy in women. Indeed, high doses of folliculoids, especially stilbestrol, are

claimed to impede the development or cause regression of spontaneous breast cancers, particularly in elderly women; in younger women such treatment sometimes appears to cause the reverse effect.

In experimental animals, especially in certain particularly cancer-susceptible strains of mice (less readily in rats), chronic treatment with folliculoids produces mammary carcinomas or fibroadenomas. In the susceptible strains of mice, the breast cancers develop much more frequently in females than in males; they occur only some time after sexual maturity and their development is impeded by ovariectomy but enhanced by folliculoid treatment. The so-called "milk-factor" (probably a virus transmitted from the mother through the milk) predisposes the offspring to mammary cancer, and increases sensitivity to the tumorigenic



Uterine carcinoma following folliculoid treatment. Squamous carcinoma of cervix (or upper vagina) in a hybrid mouse that received 33.3 μ of estradiol benzoate weekly, for 73 weeks
(Courtesy of Dr. W. U. Gardner.)

effect of folliculoids. The development of experimental fibroadenomas of the mammary gland which can be produced by folliculoids in the rat, is inhibited by simultaneous progesterone treatment.

Whether *cystic-glandular hyperplasia* of the breast should be regarded as a neoplasm is doubtful, but it probably represents a precancerous condition. It is especially common in women with hyperfolliculoidism and can be experimentally produced by folliculoids in animals (especially the mouse and rat); the spontaneous form in women tends to regress after ovariectomy.

It is doubtful whether cancer of the VAGINA is related to ovarian hormones. *Krauosis vulvæ*, probably a precancerous lesion, is frequently ameliorated by folliculoid therapy.

In certain strains of mice, tumors (usually sarcomas) of the LYMPHATIC ORGANS, sometimes accompanied by LEUKEMIA — occur in a very high percentage of animals treated with folliculoids. In mice predisposed to the formation of spontaneous HEPATOMAS, the development of these tumors is likewise enhanced by folliculoids. It is somewhat more doubtful whether the TRANSPLANTABLE, SPONTANEOUS TUMORS of animals and those induced by CARCINOGENIC HYDROCARBONS are likewise influenced by ovarian hormones, although some investigators claim that their growth is also enhanced by folliculoids.

The effect of folliculoids upon tumorous growths in other endocrine glands

is discussed in the sections dealing with the adrenals, the hypophysis, and the testis, respectively. In order to complete this summary, we wish to mention, however, that chronic treatment with folliculoids can lead to the development of ADRENAL-CORTICAL ADENOMAS OR CARCINOMAS and LEYDIG CELL TUMORS (especially in predisposed strains of mice), as well as chromophobe ADENOMAS OF THE ANTERIOR-LOBE (especially in the mouse and rat).

CANCER OF THE PROSTATE appears to be, at least to some extent, under the influence of testoids produced by the Leydig cells. It is for this reason that castration often causes, at least a temporary, improvement (see : Testis) in patients suffering from prostatic cancer. Treatment with folliculoids tends to cause a similar improvement, probably because of a functional castration due to Leydig-cell atrophy.

IN CONCLUSION, we may say that the evidence mentioned above clearly indicates that folliculoids can produce neoplasms, especially in the accessory sex organs, the adrenals, the testis, and the pituitary. This tumorigenic effect is frequently inhibited by simultaneous treatment with luteoids or testoids. It is less clear whether folliculoids can produce tumors in organs other than the accessory sex organs and endocrines, except in hereditarily predisposed individuals. There is some indication that in certain specific instances, folliculoids may also have an antitumorigenic effect (prostatic cancer, mammary cancer?).

METABOLISM OF THE OVARIAN HORMONES (OVARIAN HORMONE CONTENT OF BODY FLUIDS AND TISSUES)

The main problems in connection with the metabolism and biogenesis of the ovarian hormones are discussed in the section : The Steroids. The variations in the ovarian hormone content of the blood and urine have been mentioned in connection with those diseases

in which they are of diagnostic importance. Additional pertinent facts will be found in the sections : Sexual Cycle and Pregnancy. Here, we shall merely give a brief outline of the main facts concerning the metabolism of the ovarian hormones.

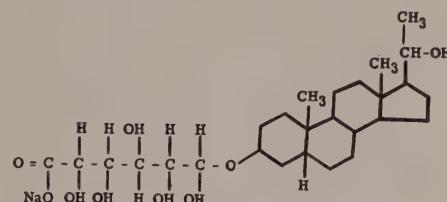
Apparently all steroid hormones, including the folliculoids and luteoids, are partly INACTIVATED in the body. Only comparatively small amounts are eliminated, through the urine or bile, in an active form. This inactivation is accomplished by : total oxidative degradation of the nucleus itself; by reduction (e.g., progesterone to pregnanediol); and by conjugation. All these processes appear to depend upon the activity of certain enzymes (e.g., "estrinase") present in various tissues, especially the liver.

Very little is known about the pathways through which complete OXIDATIVE DISINTEGRATION of the steroid nucleus occurs in the body. There is satisfactory evidence to show, however, that the side chain in some 17-ethyl-androstan (allo-pregnane) derivatives can be split off at the C₁₇ carbon atom by oxidative processes.

Much more is known about the CONJUGATION of steroids. The urinary steroids possess a hydroxyl group which can be conjugated by the formation of ethereal sulfates or glucuronides; these are usually excreted in the form of their sodium salts. Estriol glucuronide, estrone sulfate, pregnanediol glucuronide, and among the testoid derivatives, androsterone sulfate and trans-dehydro-androsterone sulfate (the latter prepared as a semicarbazone), have actually been isolated from urine. Unlike the free steroids, the conjugates are highly water-soluble and perhaps this facilitates their elimination through the urine. Conjugation lessens the biologic activity of the steroids when they are given parenterally, but increases it in the event of administration by the oral route.

In the glucuronides of estriol and pregnanediol the hydroxyl group of the steroid combines with the aldehyde group of glucuronic acid (glucosidic linkage). Since the reducing property of the latter depends on the aldehyde group, such conjugates do not reduce

alkaline copper solutions. However, following hydrolysis by boiling with dilute acids, free glucuronic acid is liberated from the conjugates and then its reducing properties (similar to those of glucose) become evident again. In pregnanediol glucuronide, the conjugation occurs at C₃, while in estriol, it takes place either at C₁₆ or C₁₇.



Pregnanediol Sodium Glucuronide

There is ample evidence to show that conjugation of steroids with either sulfuric or glucuronic acid occurs in the LIVER. Hepatectomy or liver damage by toxic substances (chloroform, carbon tetrachloride, etc.) prevents such conjugative processes.

Conversely there is evidence that certain folliculoid precursors or "pro-folliculoids" are activated in the body, judged by the fact that, for instance, estriol is more active in immature intact than in spayed rats. The ovary could be one such site of activation.

Occurrence of Folliculoids in Various Body Tissues and Fluids. — α -ESTRADIOL has been found to occur in various tissues such as : sow ovaries (0.014 mg./Kg.), human placenta (0.038 mg./Kg.) and horse testes (0.21 mg./Kg.). It is apparently not present in the adrenal cortex and occurs only in traces in the urine of normally cyclic or pregnant women and pregnant mares.

ESTRONE has been prepared from : sow ovaries (0.01 mg./Kg.), human placenta (0.035 mg./Kg.), horse testes (0.36 mg./Kg.), sow adrenal cortex (0.08 mg./Kg.), the urine of normally cyclic (traces) or pregnant (1 mg./L.) women and pregnant mares (100 mg./L.).

ESTRIOL has only been found in human pregnancy urine (9 mg./L.) and in human placenta (0.14 mg./Kg.), while none appears to be present in pregnant mare's urine, sow's ovary, and other tissues. Perhaps this compound is produced only by the human placenta.

A number of other folliculoid compounds, e.g., EQUILIN, EQUILENIN, and HIPPULIN, appear to occur only in the urine of horses, perhaps because of species-specific peculiarities of their intermediate metabolism.

The folliculoid hormone concentration in the corpus luteum is approximately the same as in ovarian follicles.

PROGESTERONE is not found in any part of the ovary except the corpus luteum, and is not eliminated as such in the urine. There is indirect evidence to show that the placenta can also pro-

duce luteoid compounds, perhaps progesterone itself.

The urinary excretion of PREGNANEDIOL SODIUM GLUCURONIDATE is only an approximate measure of ovarian progesterone formation, since the adrenal cortex can also produce progesterone and other steroids which are eliminated in the form of pregnanediol sodium glucuronide. Excretion of this compound commences one or two days after ovulation and reaches a maximum about one week prior to the onset of bleeding. It disappears from the urine almost completely two to three days before menstruation. It must also be remembered that progesterone can be excreted in the form of less completely reduced compounds (e.g., allo-pregnanolones) and perhaps, after oxidative degradation of the steroid ring structure, even in the form of smaller molecules. (See also pp. 80-84.)

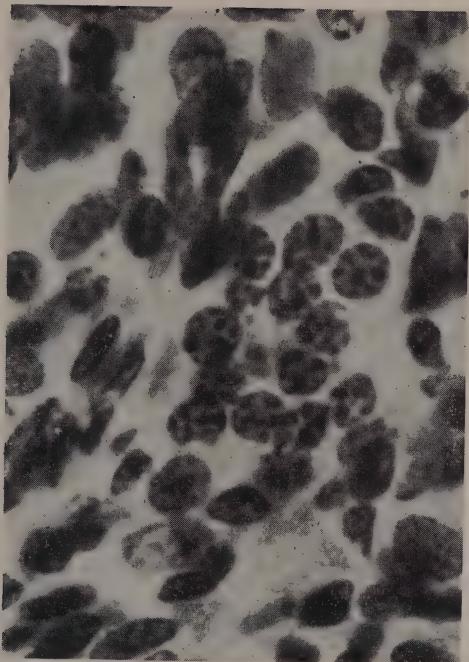
STIMULI INFLUENCING THE OVARY

Extrication of Endocrine Glands.

— HYPOPHYSECTOMY causes pronounced retrogressive changes in the ovaries of all laboratory animals. In adults, follicle maturation, ovulation and corpus luteum formation cease. At the same time, all pre-existent follicles which were fairly mature at the time of the operation, undergo atresia, while the very immature, especially the primary follicles persist indefinitely in spite of the hypophyseal deficiency. Indeed, even the formation of new follicles from the germinal epithelium is not abolished. The theca cells of the atretic follicles do not degenerate as completely as their granulosa cells and ova, but in several animal species (e.g., rat) they undergo a retrogressive process designated as "wheel-cell formation." During this transformation, the originally spindle-shaped theca cell becomes roundish and loses most of its cyto-

plasm, while the nucleus becomes similar to that of the plasma cells. The dense nuclear chromatin rearranges itself in such a manner as to leave radially oriented, clear, wheel-spoke-like spaces between the chromatin granules. In some animal species (e.g., rat) the pre-existent corpora lutea involute very slowly after hypophysectomy, while in others (e.g., mouse) their disappearance is rapid.

In the ovaries of prepubertally hypophysectomized animals, pituitary deficiency manifestations are also evident. Since advanced stages of follicle maturation or corpus luteum formation do not occur in any case at such an early age, only the wheel-cell formation is very obvious. However, this suffices to show that gonadotrophic stimulation is of importance for the development of ovarian structure even before the advent of puberty.



"Wheel cells" in the ovary of the hypophysectomized rat. Note peculiar arrangement of chromatin in rounded nuclei of theca cells (center of the field); the light spaces between the chromatin granules resemble spokes in a wheel.

Treatment with suitable combinations of *anterior-pituitary extracts* can restore the normal structure of the ovary in hypophysectomized animals. There are great species differences in the responsiveness to this substitution therapy; primates, including man, are least amenable to such treatment. As indicated earlier (see: Theories concerning the Hypophysoid Hormones), there is no agreement as yet concerning the number of chemically-distinct hypophysoid gonadotrophic principles. The chief factors probably responsible for the rather confused picture presented by the pertinent literature are: species differences in responsiveness, variations in the degree of purity, the dosage, or the relative proportions of the gonadotrophins administered and the timing of the injections. The following is a simplified summary of the

most important relevant facts (mainly gathered from experiments on rats):

(1) *FSH* causes follicle maturation without luteinization and without any evidence of either folliculoid or luteoid hormone secretion, as judged by the condition of the uterus and vagina.

(2) *LH* causes luteinization of the theca cells with formation of thecal corpora lutea. It also restores the wheel-cells to normal, subsequently transforming them into theca-lutein cells, and expedites the involution of pre-existent corpora lutea. In itself it does not cause luteinization of the granulosa. LH stimulates the pre-existent theca and corpus luteum cells to form folliculoids, but elicits no luteoid hormone secretion as judged by the condition of the accessory sex organs, which show estrus but no progestation.

In prepubertally hypophysectomized animals, LH produces no estrus unless precocious puberty has been elicited by gonadotrophins prior to the hypophysectomy.

(3) *FSH plus LH* causes follicle stimulation and luteinization, especially if the FSH treatment preceded LH administration. Under optimal conditions of dosage and timing, ovulation may also be obtained in the hypophysectomized animal by successive treatment with FSH and LH. It is not necessary therefore, to assume the existence of a special "ovulation hormone." Combined treatment with FSH and LH, causes folliculoid, but no luteoid hormone secretion by the ovary. It expedites the involution of pre-existent corpora lutea, even more than LH treatment alone. Excessive doses of FSH and LH, or combined treatment with both these substances may elicit the formation of many more follicles and corpora lutea in the ovaries of hypophysectomized animals than would occur, normally, in intact animals.

(4) *Luteotropic hormone (prolactin)* exerts no significant effect upon

the ovary of hypophysectomized animals, unless there are functional corpora lutea at the time of operation. In this event, it maintains the corpus luteum — both morphologically and functionally — in the absence of the pituitary. It causes luteoid hormone production by the pre-existent corpus luteum cells, as judged by the condition of the uterus (progestation) and vagina (mucification). Indeed, the amount of luteoid hormone thus produced suffices to permit the formation of decidiomas following local trauma to the endometrium.

(5) There is no reason to believe that pure *adrenotrophic*, *thyrotrophic*, *somatotrophic*, or any of the *intermediate* and *posterior-pituitary hormones*, exert any specific effect upon the ovary of the hypophysectomized animal.

(6) None of the steroid hormones appear to have a direct effect upon the ovary, except the folliculoids which may slightly delay the ovarian involution following hypophysectomy in certain species. When given in combination with gonadotrophic pituitary extracts, folliculoids enlarge the size of the developing follicles and corpora lutea, so that eventually, "pregnancy type" corpora lutea develop. This is presumably due to a peripheral synergism between the action of luteotrophin and of folliculoids upon the corpus luteum.

(7) Hypophysectomy during late pregnancy does not cause involution of the corpora lutea. Treatment of hypophysectomized, pregnant animals with LH prevents the formation of wheel cells in the theca, but results only in theca luteinization, not in follicle formation, as it would in intact animals. From this it was concluded that the placenta (though capable of maintaining the corpora lutea of gestation, thus substituting for the luteotrophic effect of the pituitary) does not possess the ability of the hypophysis to complement

the effect of injected LH by the secretion of FSH.

(8) Hypophysectomy during *pseudopregnancy*, elicited by lactation, copulation, or mechanical stimulation of the uterine cervix, causes involution of the corpus luteum of pseudopregnancy. In such an animal as the rabbit, in which copulation leads to ovulation within 10 to 12 hours, hypophysectomy prevents the formation of the corpora lutea, but only if it is performed during the first hour after coitus. Later removal of the pituitary is ineffective, apparently because sufficient gonadotrophin has been secreted during the first hour to insure ovulation and corpus luteum formation.

PARTIAL OVARIECTOMY causes a compensatory hypertrophy of the remaining ovarian tissue and, as long as the ovariectomy is not too extensive, the remnant tends to compensate for the deficiency according to the "law of constant numbers" (see : Theories concerning the Histophysiology of the Ovary). Hence, fertility and the number of offspring per litter are not necessarily diminished by partial ovariectomy. On the other hand, after very extensive partial resections, the small ovarian remnant tends to show cystic degeneration, due to excessive growth and persistence of non-ovulating mature follicles. This is accompanied by protracted estrus. The uterus then often enlarges, cystic glandular hyperplasia and adenomatous polyps may be produced, and proliferating glands may penetrate deeply into the myometrium. This condition has been ascribed to a derangement of the hypophyseal-ovarian relationship.

In birds (e.g., fowl) in which only the left ovary is fully developed and capable of egg production, removal of this gonad (especially if performed at an early age) may cause transformation of the rudimentary right ovary into an "ovario-testis" containing both male and female elements. Actual spermatogenesis may develop in these ovario-testes and their

testoid hormone production is demonstrated by the subsequent masculinization of the plumage, comb, wattles and sometimes even the growth of spurs. Probably most instances of spontaneous transformation of a laying hen into a rooster with male libido and all the male accessory characteristics, are due to destruction of the left ovary by disease and subsequent development of the rudimentary right ovario-testis.

If a spayed and an intact female rat are PARABIOTICALLY UNITED, the ovaries of the normal partner increase in size due to the development of many follicles and corpora lutea. Even if both the spayed and the intact partner are prepubertal, intense luteinization occurs, indicating that precocious, excessive gonadotrophin production by the castrate continues in spite of the ovarian enlargement in the intact parabiotic twin. Normally, the gonadotrophin-secretion inhibiting effect of ovarian hormones keeps the anterior-lobe activity down to a physiologic limit; however, in the case of parabiosis apparently only the gonadotropic, but not the ovarian hormones, pass freely across from one twin to the other. Thus, the gonadotropic hormones of both pituitaries would act on the ovaries of the intact partner, but the ovarian hormones of the latter could not effectively prevent the increased function of the spayed twin's hypophysis. It is also noteworthy that this increased ovarian stimulation may persist for more than a year without any evidence of antihormone formation against the endogenous gonadotrophins.

Parabiotic union of an intact female with a castrate male rat results in a similar ovarian response, but in this case usually follicle stimulation prevails over luteinization. Apparently the male castrate tends to produce more FSH than the spayed female.

Removal of the OTHER ENDOCRINE GLANDS (adrenals, thymus, thyroid, parathyroids) causes no noteworthy ovarian changes beyond those explici-

able on the basis of the non-specific damage occasioned by such interventions.

UTERUS EXTRIPATION tends to prolong the life span of corpora lutea, especially in the guinea pig. It is doubtful, however, whether the involution or cystic degeneration of the ovaries often seen in hysterectomized women, is due to any deficiency in hypothetic "uterine hormones." In most species including man, hysterectomy usually does not interfere with the ovarian cycle and it is quite probable that most, if not all, the ovarian lesions ascribed to uterine deficiency are actually due to incidental damage to the blood supply of the gonad. The responsiveness of the ovary to exogenous gonadotrophins is not significantly altered by hysterectomy.

SURGICAL INTERVENTIONS ON THE NOSE AND EYES may elicit structural changes in the ovaries which have been ascribed to specific derangements in the nervous regulation of gonadotrophin secretion. (See: Effect of Rays on pp. 384, 385.)

Hormones. — HYPOPHYSEAL EXTRACTS act somewhat differently in normal individuals from what has been said above about the response of hypophysectomized animals. This is due to the peculiar ability of the test-animal's own pituitary to complement the action of exogenous hormones by endogenous secretion of other, synergistic hormones. Without entering into details regarding species-specific responses, dosage, timing of the administration of certain principles, etc., the most important pertinent facts (mainly gathered from experiments on the rat) may be summarized as follows:

(1) *FSH* produces predominantly follicle stimulation, but this is rapidly followed by corpus luteum formation, presumably due to *LH* production by the animal's own pituitary.

(2) *LH* causes predominantly corpus luteum formation, but also some

follicle maturation, probably owing to the compensatory secretion of FSH by the animal's own pituitary.

(3) *FSH plus LH* causes a much more pronounced ovarian weight increase than either of these hormones alone, but the result is qualitatively similar to that produced by FSH or LH given singly. There is both follicle maturation and corpus luteum formation. While LH raises the ovarian weight only to a certain, limited extent irrespective of dosage, combined administration of FSH and LH is capable of producing a weight increase over a much greater range of dosages. Hence the most pronounced ovarian enlargement is obtainable by such combined treatment.

Highly purified preparations of FSH and LH from various sources (urine, blood, placenta, anterior-hypophysis) act essentially in the same manner.

In prepubertal, hibernating, or senile animals whose ovaries contain no mature follicles or corpora lutea, the effect of gonadotrophins is particularly striking. Yet in these, as in the normally cyclic adult animal, either FSH or LH results in the production of both follicles and corpora lutea, owing to complementary hormone production by the anterior-lobe.

The precipitous development of follicles and corpora lutea may result in abnormal types of luteinization; for instance, corpora lutea with enclosed ova due to luteinization before ovulation could occur (so-called "atretic corpora lutea"); partial luteinization of the granulosa in a follicle in which other portions of the granulosa remain uninfluenced (so-called "abortive luteinization"); hemorrhages into the antra of maturing follicles or corpora lutea (so-called "blood spots").

It is noteworthy that in birds (e.g., fowl, pigeon) LH, unlike FSH, causes no ovarian stimulation and may actually inhibit the development of the gonad.

This insensitivity to LH may be due to the fact that birds do not form corpora lutea.

The primate and particularly the human ovary is not as readily influenced by gonadotrophic preparations as that of most other animals. However, there is suggestive evidence that follicle maturation, ovulation and even corpus luteum formation may be obtained in women, if adequate doses are given at correct time intervals and especially if the injections are made intravenously, so that a high hormone concentration is obtained suddenly in the blood. — As judged by indirect evidence (pregnanediol excretion, endometrial histology), LH can cause premature luteinization of pre-existent follicles, or prolong the life-span of already formed corpora lutea in women. — Thus, depending upon the time of treatment it can either shorten or prolong the intermenstrual interval in patients with otherwise very regular cycles.

Chronic treatment with gonadotrophic preparations may cause complete involution of the gonads due to antihormone formation (see : Antihormones).

Local application of LH into the antrum of mature follicles results in the luteinization of these follicles only, while simple punctures without LH injections have no such effect. This suggests that the gonadotrophic hormones act directly upon the follicles.

(4) *Luteotropic hormone* tends to inhibit ovulation (especially in birds) and may result in an actual depression of ovarian weight. Indeed, it inhibits even the effectiveness of concurrently administered, exogenous gonadotrophins. However, it augments the life-span and luteoid hormone production of pre-existent corpora lutea.

(5) *Gonadotrophic hormones plus folliculoids*, result in the formation of excessively large, "pregnancy type" corpora lutea, which sometimes may

take the form of voluminous corpus luteum cysts.

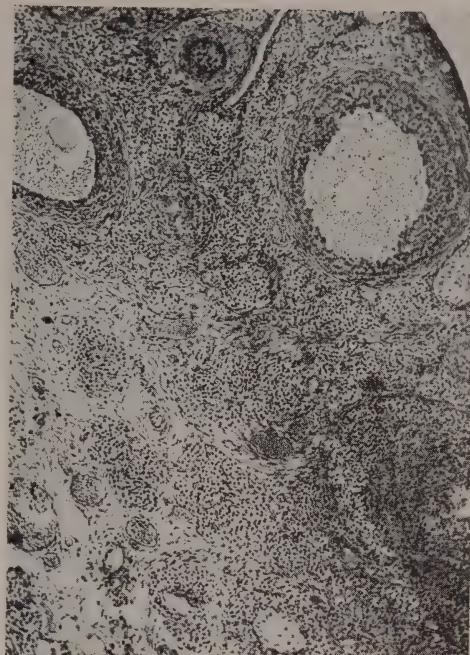
(6) *Other hypophyseal extracts*, including those of the middle and posterior-lobe, cause no specific ovarian lesions beyond those explicable on the basis of the non-specific damage produced by high doses.

FOLLICULOIDS, given in high concentrations over a short period of time, cause luteinization of pre-existent follicles and may even elicit precocious corpus luteum formation especially in the prepubertal rat. The resulting corpora lutea are unusually large, "pregnancy type," but do not persist very long, in spite of continued folliculoid administration. This effect is not obtained after hypophysectomy, unless anterior-pituitary extract is given with the folliculoids; presumably it is due to a peripheral synergism between folliculoids and luteotrophin.

Chronic treatment, even with small doses of folliculoids, causes pronounced ovarian atrophy. This is thought to be a compensatory atrophy due to inhibition of gonadotrophin formation in the pituitary.

TESTOIDS given in very high concentrations, may likewise elicit a transitory formation of "pregnancy type," large corpora lutea (especially in the rat). In general, testoids tend to produce more mature follicles rather than corpora lutea, especially in the mouse. Perhaps some types of cystic ovaries in women may be due to excessive testoid production by the adrenal cortex, or by virilizing tumors.

In embryonic and, less markedly, in immature postnatal ovaries, testoid treatment causes some "virilization of the ovary." It inhibits the development of the typically "female" cortex and stimulates the "male" medullary cords and rete tubules. Far-reaching transformations of this type have only been obtained in lower vertebrates (amphibia, birds) in which the testoids



Effect of estradiol on the rat ovary. Ovary of an adult rat which was treated with increasing doses (50γ gradually raised to 200γ) of estradiol, during a period of three months. The animal was killed after an additional period of two months, during which no treatment was given. Note degeneration of most follicles with necrosis of the granulosa and theca-nest formation. During the atrophy produced by estradiol the cells of these theca-nests are transformed into atrophic "wheel cells", but after the two month rest period intense thecal luteinization has occurred. The experiment demonstrates the long persistence of the ovarian atrophy elicited by chronic folliculoid overdosage.

were directly applied to the developing ovum. However, some degree of ovarian ambisexuality is also seen in mammals following testoid administration to the pregnant mother. This explains the case of "freemartin" cattle. Here a female and a male twin have a common placental blood supply and under the influence of testoids elaborated by the male, the gonad of the female partner assumes ambisexual characteristics. The sterile and ambisexual "tortoise-shell tomcat" probably owes its gonadal anomaly to similar placental conditions.

LUTEOIDS (e.g., progesterone) and CORTICOIDS (e.g., desoxycorticosterone) are rather inactive in producing large corpora lutea or follicle cysts. All steroids exhibit this action approximately in proportion to their folliculoid potency. (See: "The Steroids.")

Continued treatment with high doses of various OTHER STEROID HORMONES also results in ovarian atrophy. Here again, it appears likely that the effect is due to the folliculoid potency of these compounds and acts through inhibition of gonadotrophin secretion.

HORMONALLY INACTIVE STEROIDS (e.g., etiocholanolones, pregnanediol) cause no ovarian change.

Among the OTHER HORMONES, none exert a specific effect upon the ovary, although when given in toxic doses they all tend to inhibit follicle maturation and to produce ovarian atrophy as part of the general-adaptation-syndrome to their non-specific damaging effect.

Diseases. — As may be expected, the various forms of HYPOPITUITARISM (e.g., pituitary dwarfism, Simmonds' disease, adiposogenital syndrome) cause involution or lack of development of the ovary. Even the common ovarian involution with amenorrhea, produced by various non-specific damaging agents during the general-adaptation-syndrome, is probably of hypophyseal origin. It presumably results from a diminished gonadotrophin secretion at times when there is a more urgent, vital need for an increased adrenotrophin production, the hypophysis being unable to elaborate both types of hormones in adequate amounts.

HYPERPITUITARISM (e.g., acromegaly, Cushing's disease, etc.) causes rather variable ovarian changes. Gonadotrophin production may be normal, increased, or diminished, while the pituitary produces an excess of other anterior-lobe hormones.

HYDATIDIFORM MOLES and CHORIONEPITHELIOMAS are frequently accompanied by the formation of numerous, often cystic, large corpora lutea, and a great increase in the size of the ovaries due to the excessive production of gonadotrophins by the tumor tissue.

In the ADRENOGENITAL SYNDROME, the ovaries are usually undeveloped, in spite of the striking precocious uterine bleeding and sex-organ differentiation.

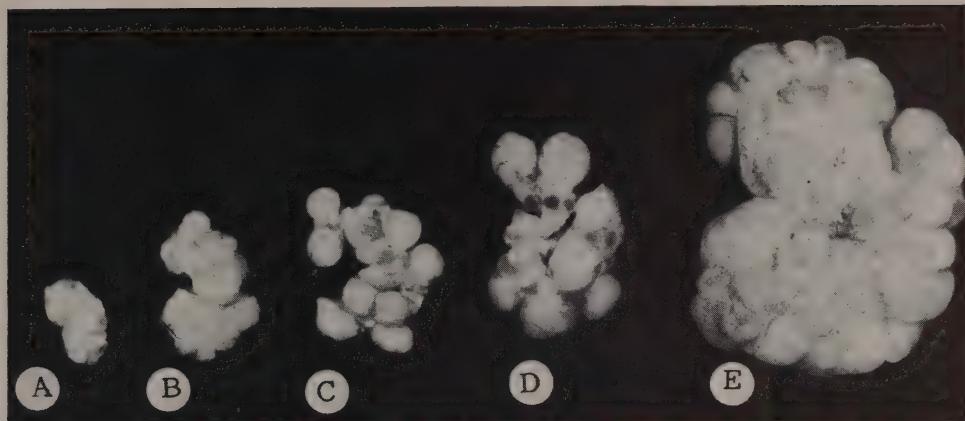
In TRUE PRECOCIOUS PUBERTY, there is a balanced and otherwise normal, but premature gonadotrophin formation by the pituitary; correspondingly the ovaries are similar to those of normal adult women. Children who are the bearers of such ovaries may become fertile at an unusually early age. In one case (described by *Escomel*) a girl of 5 years and 8 months gave birth to a healthy living infant.

BRAIN LESIONS, especially tumors of the pineal region or hydrocephalus, may produce precocious pseudopuberty with premature uterine bleeding. The ovaries rarely reveal any sign of precocious maturation, although sometimes they show "small cystic degeneration" (see: Diseases of the Ovaries). The mechanism of this type of pseudo-puberty is still unknown.

In women with FIBROMYOMAS of the uterus, persistent follicles or unusually large corpora lutea are common. Some investigators believe that hyperfolliculoidism is an important factor in fibromyoma formation.

OTHER DISEASES are not known to act upon the ovaries directly. However, in most women with serious, chronic diseases, follicle maturation and corpus luteum formation are disturbed, and the cycles become irregular, or chronic amenorrhea sets in as a result of the general-adaptation-syndrome elicited by the accompanying non-specific damage.

The ovarian lesions which are typical of the various clinical forms of HYPO- or



Appearance of rat ovary under various conditions. — A. Ovary (4 mg.) of untreated 2-week-old rat. Note absence of mature follicles and corpora lutea. — B. Ovary of adult untreated rat (16 mg.). Note several normal sized corpora lutea. — C. Ovary on 14th day of lactation (26 mg.). Note one set of fully developed corpora lutea, which are hardly much larger than those of the cycle. — D. Ovary of pregnant rat (about 14th day of gestation) (47 mg.). Note that number of corpora lutea is not increased and corresponds to that formed during average cycle. Size of individual corpora is greatly augmented however; several, very small follicles (greyish area) are also visible. — E. Enormous development of corpora lutea ("mulberry ovary") of a lactating rat which received 600 IU./day of PMS for 10 days during lactation (308 mg.). Size of the individual corpora lutea is not very different from those seen in normal lactation (smaller than those of pregnancy).

HYPEROVARIANISM are discussed in the sections devoted to the latter.

Diet. — The ovary is especially sensitive to qualitative or quantitative changes in food intake. Prolonged FASTING or UNDERNUTRITION causes atrophy of the gonads in various animal species. Only in certain fish is the development of the ovary remarkably independent of food intake. Thus, for instance the salmon swims upstream and takes no food during the breeding season, although it performs very active muscular exercise. The animal loses weight, while its ovaries enlarge so much that instead of the initial 0.4%, they constitute 27% of the total body weight. In women, prolonged malnutrition leads to fibrosis and atrophy of the ovaries. Mature follicles are absent and the interstitial cells become atrophic.

On the other hand, OVERFEEDING and various QUALITATIVELY INADEQUATE DIETS (vitamin, amico-acid, protein, salt

deficiencies, etc.) likewise interfere with the sexual cycle by causing ovarian atrophy. Here again, the increased production of corticotrophin and perhaps also of other pituitary hormones, during the resulting general-adaptation-syndrome, probably occurs at the expense of a diminished gonadotrophin production during the period of strain.

Certain diets, however, may exert a specific influence upon the ovaries. Thus, rats fed predominantly on LENTILS — or some of the other leguminous seeds — develop ovarian atrophy with persistent diestrus, perhaps due to the presence in such plants of a specific ovario-toxic substance (phaseolin).

Contrary to the marked degenerative changes observed in male gonads, the ovaries of VITAMIN-E DEFICIENT animals are of normal appearance. The sterility of such animals is merely due to embryo resorption during pregnancy.

Nervous Stimuli. — COPULATION OR STIMULATION OF THE UTERINE CER-

VIX causes pseudopregnancy in a variety of animal species (e.g., rat, rabbit, cat); this is accompanied by the formation of a persistent, functional corpus luteum. Here a single nervous stimulus suffices to produce endocrine changes which maintain the corpus luteum for a period of several weeks. The length of pseudopregnancy varies in the different animal species.

In the human female, follicle rupture is usually spontaneous, but some investigators believe that occasionally, ovulation may be provoked by sexual intercourse. In very exceptional cases, the corpus luteum may persist for several months, so that pseudopregnancy results. It has also been claimed that excessive sexual intercourse or masturbation may cause cystic degeneration of the ovaries or the formation of persistent follicles. In such instances, it is difficult to determine however, whether hyperfolliculoidism due to persistent, cystic follicles was not the cause, rather than the result, of the excessive sexual activity.

An interesting note illustrating the action of psychic stimuli upon the ovary has been published by LeConte, in 1884. He relates the case of two nannies who were struck by lightning. In one of them, a young woman, menstruation ceased completely after the accident, while in the other, a woman over 70 years of age who had not menstruated for 20 years, regular cycles reappeared every month for more than a year and her breasts enlarged considerably following the shock. Some gynecologists believe that while pleasant sensations elicited by sexual intercourse may provoke ovulation, disgust or fear of cohabitation can inhibit it. This may explain the sterility of certain marriages in which both partners prove to be fertile in subsequent marriages to other mates. Prolonged amenorrhea is frequent in women who are afraid of pregnancy, but the underlying ovarian

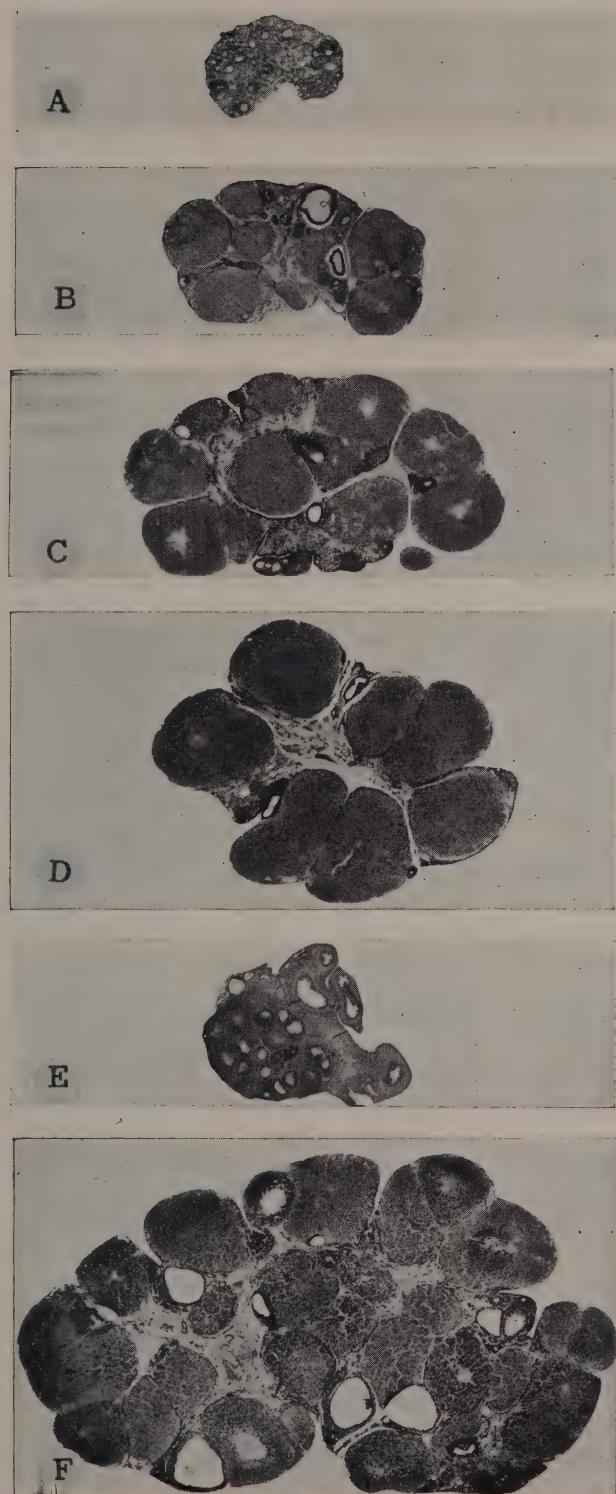
changes have not yet been studied in such cases.

The NERVOUS STIMULATION OF THE NIPPLES by the offspring is responsible for the persistence of the so-called corpora lutea of lactation. That these corpora lutea are not maintained owing to milk production has been shown by experiments on the rat in which the milk ducts (galactophores) were transected, but the young were allowed to continue nursing. Although milk could obviously not be secreted, the nervous stimulus in itself sufficed to maintain the corpora lutea in a functional condition. Unlike the stimulus of copulation, that of nursing must be continuous in order to maintain the corpora lutea of pseudopregnancy. (See : Pseudopregnancy.)

COMPLETE DENERVATION OF THE OVARIES does not interfere with the production of follicles, corpora lutea of the cycle, or even corpora lutea of pregnancy.

TRANSECTION OF THE PITUITARY STALK before or immediately after copulation prevents ovulation in the rabbit. On the other hand, LOCAL ANESTHESIA OF THE VAGINA AND VULVA or even complete ablation of the upper portion of the vagina and uterus do not prevent postcoital ovulation in rabbits. It was concluded that local nervous stimuli in the genital region are not essential for the copulatory reflex. Apparently, psychic stimulation alone suffices to elicit nervous impulses which pass down to the pituitary across the hypophyseal stalk. Such emotional stimuli must also be invoked to explain the pseudopregnancy produced in rabbits by merely showing them a male kept in an adjacent, but separate, cage.

Age. — Follicle maturation and corpus luteum formation commence at puberty. Usually the first few cycles are rather irregular and in some animals, as well as in women, they may be anovular.



Appearance of the rat ovary under various experimental conditions. — A. Ovary of a normal 3-week-old rat. Note immature aspect of the ovary with no large follicles and no corpora lutea. (Same low magnification as fig. B-F.) — B. Ovary of a normal adult rat. Note several large corpora lutea and one almost mature follicle with distinctly visible cumulus oöphorus and ova. — C. Ovary of a normal rat on the 16th day of lactation. No mature follicles but well-developed, medium sized, corpora lutea of lactation. — D. Ovary of a rat on 16th day of lactation, following treatment with 1 mg./day of estradiol commencing immediately after delivery. Note greatly enlarged corpora lutea (compare with fig. C.), but no formation of new corpora lutea. Milk secretion was arrested under the influence of this folliculoid therapy. — E. Ovary of an adult rat treated with increasing doses of estradiol (beginning with 50 γ /day and raised to 200 γ /day) during a period of three months. The picture was taken two months after discontinuation of this prolonged folliculoid treatment, but ovarian atrophy was still severe. Note that general aspect of the ovary resembles that of an immature rat (compare with fig. A). Part of the oviduct is also shown in this picture to demonstrate marked fibrosis and thickening of its wall. — While brief treatment with massive doses of folliculoids causes enlargement of preexistent corpora lutea (fig. D.) prolonged administration of even smaller doses causes severe ovarian atrophy, which persists after discontinuation of therapy. — F. Ovary of an adult rat following treatment with increasing doses of LH (150 I.U./day gradually raised to 450 I.U./day). Note typical appearance of "mulberry ovary" with numerous, but not particularly large corpora lutea.

Compare these histologic pictures with the macroscopic specimens reproduced on page 381.

The ovarian weight in man is about 0.3 gm. at birth. At this time several fairly mature follicles may be distinguishable, presumably due to exposure of the embryonic gonads to the large quantities of maternal gonadotrophins. The ovarian weight at 6-10 years is 2 gm. and at puberty, about 7 gm. Even after puberty, it tends to increase to about 10-12 gm. by the 21st-30th year, but subsequently it decreases to about 4 gm. in senile women. About three to four years after the menopause, no follicles are distinguishable, although the germinal epithelium remains intact. Sclerotic changes in the ovarian vessels are especially conspicuous in the aged.

Constitution, Race and Heredity. — Constitutional and racial factors play a rôle in ovarian development in animals and man. The claim that puberty tends to occur earlier in southern than in northern races is not clearly supported by statistics, but unusually early or late puberty is frequently seen as a familial characteristic. In uniovular twins, the menstrual cycles of both sisters often run synchronously.

In the dwarf mouse, ovulation does not occur under normal conditions, and consequently there are no corpora lutea. Certain breeds of sheep possess the ability to reproduce consistently at higher rates than others, and twinning in sheep is possibly also inheritable.

Sex. — The effect of SEXUAL INTERCOURSE upon the development of the ovary has been discussed above under "Nervous Stimuli"; the changes corresponding to the phases of the SEXUAL CYCLE will be considered in the chapter on Estrus and Menstruation.

Pregnancy and Lactation. — The ovarian changes characteristic of pregnancy and lactation are discussed in the sections specifically devoted to these conditions.

Season. — Seasonal variations in ovarian development are especially im-

portant in amphibia, reptiles, birds, and hibernating mammals in which the ovaries undergo atrophy during the resting season. For instance in the bat, ovulation occurs in the spring, while during hibernation the gonad is inactive and small.

Some non-hibernating mammals (e.g., dog, sheep) likewise produce mature follicles only during certain seasons of the year.

In the human female, seasonal variations have no marked effect upon the ovarian cycle.

Rays. — X-RAYS are especially damaging to the granulosa cells, which can be selectively destroyed by them. In the mouse, suitable X-ray treatment may lead to the complete disappearance of all follicles and corpora lutea, while the estrus cycles continue (*Parkes*). This shows that folliculoids can be produced in the absence of granulosa cells or corpora lutea. If immature mice are exposed to a full sterilizing dose of X-rays, irradiation is followed by complete degeneration of all granulosa cells and ovocytes. The germinal epithelium proliferates and forms epithelial cords. In the adult animal, the entire ovary may then be composed of the cords of this first proliferation, which tend to transform themselves into luteal tissue. In many cases, a second proliferation of small, spherical, slightly elongated cords occurs from the germinal epithelium. These resemble the so-called spermatic cords described in the ovaries of inbred rabbits and free-martin cattle. Even the ovaries of mice X-rayed in utero, show similar changes.

In certain strains of mice, X-ray treatment of the ovaries induces a very high incidence of folliculoid-producing (permanent estrus) transplantable granulosa-cell tumors (*Furth and Butterworth*).

Corpora lutea are much more resistant to X-rays than are follicles, as shown by experiments on lactating mice.

In man, treatment of the ovary with moderate doses of X-rays is frequently used as a therapeutic measure to produce temporary amenorrhea. Minute doses of X-rays are said to stimulate the ovaries and occasionally, to reinitiate menstrual cycles in women suffering from amenorrhea. Heavy X-ray treatment on the other hand, causes permanent ovarian involution and cessation of menstrual cycles with sterility.

In certain animals, LIGHT RAYS play a very important rôle in the regulation of ovarian activity. In the brook trout, spawning may be induced prematurely by exposure to light. In the salamander, ovulation fails to occur as long as the animals are kept in the dark. In the duck, pheasant, sparrow, etc., egg-laying may be induced by exposure to light both in the immature and (during

the off-season) in the mature female (*Benoit, Roan, Bissonnette*). In most mammals however, light rays exert no marked influence upon ovarian development, except perhaps in the ferret and a few other species with seasonal estrus.

It is most probable that light stimuli reach the pituitary through the stalk.

Other Stimuli. — CLIMATE, CAPTIVITY, TEMPERATURE VARIATIONS, EXCESSIVE MUSCULAR WORK, HEMORRHAGE, BURNS, TRAUMA and other non-specific agents capable of eliciting a general-adaptation-syndrome all tend to cause irregularities in ovarian function, usually accompanied by ovarian atrophy. None of these factors have been proven to exert a specific effect, that is, one unaccounted for by the disturbance in hormone production due to the resulting general-adaptation-syndrome.

OVARIAN DISEASES IN GENERAL

DEFINITION

In this chapter we shall briefly consider conjointly all those diseases which result from changes in the ovaries. In contradistinction to other glands, it is impractical in the case of the female gonad to strictly distinguish between hypo- and hyperfunctional diseases. Hyperfunction of one part of the ovary (e.g., a follicle cyst causing hyperfolliculoidism) may be associated with hypofunction in other respects (in the above example for instance, lack of corpus luteum formation). Hence we shall use this chapter on Ovarian Diseases in General, to outline briefly the changes produced in the various organs by hypo- or hyperfunction of the ovary, or one of its parts. Here we shall also discuss general problems (e.g., classification, pathogenesis, complications, diagnosis, prognosis and therapy) which are applicable to several ovarian diseases. The reader will be referred to other chapters for a more

complete description of specific syndromes.

CLASSIFICATION

It is customary to classify the ovarian syndromes according to their most prominent clinical manifestations. Thus amenorrhea, menorrhagia, or dysmenorrhea are often considered as distinct clinical entities irrespective of their etiology. This may be adequate from a purely clinical viewpoint but in this book we are using the following classification :

I. OVARIAN DISEASES IN GENERAL :

Definition.

Classification.

Pathogenesis.

Clinical Course.

State and Metabolism.

Growth and Bone Structure.

Blood.

Cardiovascular System.

Nervous System (dysmenorrhea, intermenstrual pain, ovarialgia, sense organ lesions).

Digestive System.

Skin.

Urinary System.

Accessory Genital Organs (lesions in the oviduct, amenorrhea, oligomenorrhea, hypomenorrhea, retention of menses, polymenorrhea, menorrhagia, midmenstrual bleeding, metrorrhagia, changes in the vagina, changes in the mammary glands).

Sterility.

Complications.

Diagnosis.

Therapy.

II. SPECIAL DISEASES OF THE OVARY :

Malformations and Anomalies (inc. ambisexuality).

Hernia and Prolapse.

Torsion.

Retrogressive Changes.

Vascular Disturbances.

Lymphangiectases.

Inflammations.

Tumors.

III. DISEASES OF THE SEXUAL CYCLE :

Diseases of the Pubertal Period (puberty hemorrhages, precocious and delayed puberty).

Diseases of the Menopause.

Derangements of the Correlation Between Ovulation and Menstruation or Estrus (Amenstrual Ovulation, Anovular Menstruation).

Vicarious Menstruation.

IV. DISEASES OF PREGNANCY :

Group IV will be considered in the chapter "Pregnancy" of the section "Correlations."

PATHOGENESIS

In general, ovarian syndromes are due to one of the following three causes:

(1) PRIMARY LESIONS OF THE OVARY. For instance, aplasia, destruction by local diseases, tumors causing hypo- or hyperactivity, can be the cause of ovarian malfunction.

(2) PRIMARY LESIONS OF THE PITUITARY or disturbances in the normal interaction between pituitary and ovary. Thus, destruction of the pituitary in Simmonds' disease causes atrophy of the ovaries because of the resulting lack of gonadotrophins; hyperplasia or tumor formation in the anterior-lobe may result in a variety of ovarian lesions, depending upon the resulting increase or decrease in gonadotrophin production; disturbances in the normal interaction between pituitary and ovary may cause anomalies of the sexual cycle.

(3) CHANGES IN THE INTERNAL OR EXTERNAL ENVIRONMENT. As part of the general-adaptation-syndrome which is elicited by most of the spontaneous diseases (including malfunctions of other endocrine glands), exposure to stress, malnutrition or even emotional upset, the gonadotrophin elaboration by the hypophysis may be so altered as to cause ovarian changes with derangements of gonadal hormone production.

CLINICAL COURSE

State and Metabolism. — In spite of the manifold systemic effects of ovarian hormones, the diseases of the ovary are generally not accompanied by any characteristic and uniform disturbances in the general condition of the patient. The so-called "FACIES OVARIANA," a drawn-out, tired facial expression, is not especially typical of ovarian diseases. It probably results from the continued effect of pain, repeated loss of blood, etc., which so frequently accompany ovarian diseases.

Slight water retention, with a corresponding gain in weight, occurs normally prior to or at menstruation in many women. When this is exaggerated we speak of "*premenstrual*" or "*menstrual edema*." The etiology of the condition is still unknown, but it is probably of endocrine origin. Treatment with a salt-diuretic, such as ammonium chloride is often very useful.

Growth and Bone Structure. — Although chronic treatment with folliculoids may cause increased bone deposition with osteosclerosis in experimental animals and perhaps even in women, spontaneous diseases of the ovary are rarely accompanied by typical skeletal lesions. **OSTEOMALACIA** — a disease characterized by deficient calcification of osteoid tissue — had previously been considered to result from ovarian hyperactivity and it was claimed that many patients improved following castration. It is now generally recognized, however, that osteomalacia is merely the adult counterpart of rickets, and as such, is due to a nutritional deficiency. Yet the disease is much more frequent in women than in men and hence the possibility of some ovarian influence cannot be excluded completely. The strain on calcium metabolism occasioned by repeated pregnancies and lactations is probably of importance.

Blood. — Various types of ANEMIA associated with ovarian diseases are most frequently due to the chronic loss of blood from the uterus. "CHLOROSIS VIRGINUM," a special type of anemia which occurred almost exclusively in pre-adolescent girls, had been regarded as due to ovarian insufficiency. This etiology has not been proven however, and the disease, prevalent during the 19th Century, seems to have disappeared almost completely. It was probably due to inadequate hygienic conditions, undernutrition and pubertal hemorrhages rather than to any specific ovarian malfunction.

HEMOPHILIA is an hereditary disease in which the coagulation-time of the blood is considerably prolonged. This disease is transmitted only by women, but is manifest only in men. It is claimed that the women of hemophiliac families are particularly fertile and predominantly produce male children who do not transmit the disease. It is very doubtful that ovarian hormones play

any direct rôle in this condition and contrary to the expectations of some authors, ovarian hormones exert no significant beneficial effect.

Cardiovascular System. — Ovarian diseases cause no characteristic cardiovascular symptoms, except those due to pressure by very large ovarian tumors, which may result in cardiac decompensation, edema in the lower extremities, etc. Congenital malformations of the ovary are often accompanied by malformations of the cardiovascular system.

The characteristic menopausal hypertension and flushes are discussed in the chapter : Diseases of the Menopause.

Nervous System. — Nervous disturbances are very common accompaniments of ovarian disease. EMOTIONAL INSTABILITY and even severe HYSTERIA may appear in women who suffer from chronic ovarian or uterine disease, due to a sense of sexual inferiority derived from the abnormal bleeding, sterility, etc. Even the name of hysteria is derived from "hystera," the Greek word for uterus, since ancient physicians sought the cause of the disease in that organ. FRIGIDITY and lack of libido are also frequent, but usually due to psychologic causes rather than to a diminished folliculoid hormone production; however, folliculoids, luteoids and testoids likewise play a rôle in conditioning the female libido.

The most important nervous manifestations of ovarian disease are those associated with pain. Among these we distinguish the following :

DYSMENORRHEA, which merely means painful menstruation. The pain in primary dysmenorrhea (see below) is usually of a colicky, laborlike nature in contrast to the mostly dull pain of secondary dysmenorrhea. Indeed, the pain may be so severe that nausea with vomiting ensue and the patient becomes bedridden for several days every month. It may begin one or two days before the onset of the flow, persisting

until the first or second day of the period; often it commences simultaneously with menstruation, and sometimes it continues throughout the period.

Most authors distinguish primary (essential, functional or intrinsic) dysmenorrhea due to functional factors from secondary dysmenorrhea, in which the pain is explicable by the accompanying morphologic lesions (inflammations, neoplasms, endometriosis, uterine displacement, lesions in the bladder, the ureter or in the sacro-iliac joints, etc.). In this chapter we shall limit our discussion to functional dysmenorrhea. Its etiology is still unknown, but the following factors have been considered:

(1) *The hormonal control of uterine motility.* The uterus undergoes contractions and relaxations throughout the cycle, and it is known that both folliculoid and luteoid hormones influence this contractility. It has not been possible to determine exactly what hormonal disturbance if any, is responsible for the painful contractions. The fact that dysmenorrhea may be beneficially influenced by various hormone preparations, and that pain is often limited to a certain phase of the cycle, speaks in favor of the conception that hormonal stimuli play an important rôle. Many gynecologists believe that functional dysmenorrhea never occurs in the absence of ovulation.

(2) *The development of the uterus.* In most patients dysmenorrhea is associated with hypoplasia of the uterus frequently accompanied by infantilism or displacement. Since contractions of anoxic muscles tend to be painful, it has been assumed that inadequacies in the blood supply to the myometrium may elicit pain by causing temporary anoxemia of the muscles during contractions.

(3) *Nervous factors.* Dysmenorrhea is particularly common in high-strung, nervous women, although the phleg-

matic type is by no means immune; frequently nervous excitability is the result, not the cause, of constant dysmenorrhea. Some investigators ascribe a prominent rôle to changes in the nerve fibers and ganglia of the uterus.

(4) *Passage of large fragments through the uterine canal.* The internal os is extremely sensitive to dilatation and the passage of large tissue fragments and blood clots through a resistant uterine canal may cause painful cramps during menstruation. The so-called "membranous" dysmenorrhea is due to the passage of large, undissolved fragments of uterine lining and represents the most clear-cut case of this type. Anteflexion of the uterus, with a kink in the cervix region, predisposes to the so-called "obstructive dysmenorrhea." It is doubtful however, whether similar mechanical causes are operative in the usual cases of functional dysmenorrhea.

(5) *Gastrointestinal disorders.* In many cases there is evidence suggesting that the cramps are of gastrointestinal origin; there is constipation during the premenstrual period, the pain being accompanied by a desire to defecate and the passage of hard stools, from which laxatives bring relief.

In any event, it is quite improbable that all types of menstrual pain could be ascribed to a uniform etiology. Dysmenorrhea is a symptom, not a disease. Even the distinction between primary and secondary dysmenorrhea is somewhat artificial, since as soon as the etiology of a certain type is recognized, we shift it from the first to the second category.

INTERMENSTRUAL PAIN (MITTELSCHMERZ) is a pain of varying intensity, which occurs approximately at the mid-interval between two menses. It usually lasts from a few hours to one or two days and is sometimes, but not invariably, accompanied by bleeding. The condition has variously been ascribed

to : rapid growth of, or bleeding from, the ovulating follicle (this has sometimes been demonstrated by biopsy); a temporary drop in folliculoid hormone formation at the middle of the cycle; a hormonal disturbance in the contractility of the uterus or the oviduct, or to adhesions between the ovary and other peritoneal organs which could be stretched and torn at the time of follicle rupture. It is very probable that several, if not all, of these factors, can be conducive to intermenstrual pain.

OVARALGIA (from the Greek : *algos* = pain) merely means pain in the ovary. It is frequently due to small-cystic degeneration, endometriosis of the ovary (see corresponding chapters), or to inflammatory lesions. Certain types of intermenstrual pain are transient instances of ovaralgia. The condition has no uniform etiology.

MENSTRUAL HEADACHE (or **MIGRAINE**) and **EPILEPSY** have been interpreted as of endocrine origin because of their relationship to the cycle, but too little is known about their etiology to warrant a detailed discussion here. Premenstrual treatment with luteoids or folliculoids is often beneficial. They may be due to vasomotor disturbances or transient slight edema of the brain, temporary enlargement of the hypophysis, etc.

"**PREMENSTRUAL TENSION**" is a condition of extreme nervous irritability, sometimes with emotional outbursts, compulsive ideas, etc., which occur before the onset of bleeding. It has been ascribed to abnormal sodium retention and subsequent brain edema due to a hormonal derangement. As in the closely allied premenstrual edema, ammonium chloride often brings relief.

Sense organs. — Blurring of **VISION**, **ENGORGEMENT OF THE NASAL MUCOSA**, often accompanied by **PISTAXIS**, may occur as accompaniments of ovarian diseases or of certain phases in the sexual cycle. The ocular manifesta-

tions have been ascribed to pressure by a temporarily-enlarged pituitary, the nasal lesions to the specific stimulating effect of folliculoids upon the mucosa of the nasal conchae.

Digestive System. — Gastrointestinal symptoms, especially loss of appetite, belching, abdominal pain and constipation are common in patients with a variety of ovarian diseases. Irritation of the pelvic peritoneum may result in nausea and vomiting; the latter disturbances, if they occur at the time of menstruation, are regarded by some gynecologists as a special type of dysmenorrhea. (See also : p. 388.)

By contiguity, appendicitis may lead to oöphoritis and vice versa. These possibilities should be kept in mind, in connection with the sometimes difficult problem of a differential diagnosis between appendicitis and ovarian lesions.

Skin. — Ovarian diseases undoubtedly exert an important action upon the skin. The so-called "**CHLOASMA UTERINUM**" (light brown, irregular patches on the skin, especially of the face) occur frequently in pregnancy and in connection with diseases of the ovaries, perhaps because of the well-known effect of folliculoids upon pigmentation.

SCLERODERMA is sometimes associated with ovarian disease, as are various other skin lesions, especially those of the **ALLERGIC** type.

Urinary System. — The urinary system is rarely involved in ovarian disease, except by mere contiguity (spreading of inflammatory or neoplastic processes, endometriosis, etc., from the ovarian surface to the bladder), or direct compression of the bladder or ureters by ovarian neoplasms.

Accessory Sex Organs. — Ovarian diseases may affect the **FALLOPIAN TUBES** either by direct contact, or through the intermediary of the ovarian hormones which influence both the morphologic development and the con-

tractility of the oviducts. However, changes in the tubes rarely play an important part in the general syndrome.

One of the most frequent accompaniments of ovarian disease is a derangement in the menstrual flow. We distinguish the following main types:

(1) AMENORRHEA, that is, absence of menstruation. In *primary amenorrhea*, menstruation has never occurred. Since in cases of delayed puberty the first bleeding may not appear until the 18th year of age, it is incorrect to speak of primary amenorrhea in patients under that age. This condition may occur as a result of an endocrine derangement (aplasia, hypoplasia, degeneration of the ovaries or pituitary, etc.), or it may be merely due to anomalies of the uterus itself.

In *secondary amenorrhea*, a previously established menstrual cycle ceases before the normal age (45-50 years) of the menopause. Surgical or X-ray castration, persistent corpora lutea, destruction of the pituitary or the ovaries by disease, systemic intoxications, infections, nutritional deficiencies, or even purely emotional disturbances may cause secondary amenorrhea; indeed, any chronic exposure to stress can abolish the menstrual cycle, due to the decrease in gonadotrophin secretion during the ensuing general-adaptation-syndrome.

(2) OLIGOMENORRHEA is a condition in which menstrual bleeding occurs rarely and usually at irregular intervals. There is no clear distinction between temporary amenorrhea and oligomenorrhea. Rarity of menstrual bleeding appears to be due to a disturbance in the normal interrelationships between the pituitary and the ovary, but the exact mechanism of its pathogenesis is unknown, and probably diverse disturbances may lead to this symptom. Oligomenorrhea is not incompatible with good health and fertility.

(3) HYPOMENORRHEA is a condition in which the menstrual bleeding is quantitatively deficient, although the length of the cycle, the progestational transformation of the endometrium, and even fertility may remain normal. It is common in hypothyroidism and in various types of ovarian insufficiency, especially those of the pre-climacteric age. Sometimes partial destruction of the endometrium by disease or partial hysterectomy causes hypomenorrhea. Quite frequently the condition is associated with oligomenorrhea.

(4) RETENTION OF MENSES (OR CRYPTOMENORRHEA) is a condition in which menstruation proceeds normally, but the flow does not make its way to the exterior because of an obstruction in the cervical canal, or an imperforate hymen. It should not be confused with amenorrhea. Retention of the menses is not an endocrine disease, but merely due to a malformation of the genital passages. It often leads to the accumulation of menstrual blood and subsequent cystic dilatation of the uterus or vagina, and predisposes to the backflow of endometrial particles through the oviducts into the peritoneum; thus, it may give rise to internal endometriosis.

(5) POLYmenorrhea is a condition in which the menstrual intervals are shortened, but the cycle is otherwise normal. In the majority of the cases, it is probably due to premature ovulation or early involution of the corpus luteum; correspondingly either the follicular or the luteal phase is shortened. Various types of pelvic inflammations, uterine fibromyomas, hypo- or hyperthyroidism and even purely emotional factors, such as fear of pregnancy, have been claimed to cause polymenorrhea.

(6) MENORRHAGIA (OR HYPERMENORRHEA) is a condition in which menstruation is more abundant and of longer duration than normal. It is often, though not always, accompanied

by polymenorrhea. Its main danger lies in the constant loss of considerable quantities of blood. It occurs in connection with hypothyroidism or hyperthyroidism; organic lesions in the pelvis (adenomyomas, salpingitis, subinvolution of the uterus, cardiac decompensation), or as a result of various blood diseases.

(7) METRORRHAGIA is a condition in which uterine bleeding is frequent and irregular. It is often caused by bleeding uterine neoplasms (polyps, cancers, fibromyomas, etc.), retained placental remnants, or endometrial inflammations. Its most common endocrine cause is hyperfolliculoidism, due to persistent cystic follicles which lead to metropathia hemorrhagica (see: Metropathia on pp. 426, 427). Like menorrhagia, it may also lead to severe anemia, due to the constant loss of blood.

(8) INTERMENSTRUAL (OR MIDMENSTRUAL) BLEEDING is a condition in which, usually slight, uterine bleeding (sometimes only a few spots of a brownish discharge) occurs between otherwise regular menstrual periods. It is frequently associated with intermenstrual pain. These scanty flows are often regularly interpolated between the normal periods and occur at ovulation time. In the German literature, the term "kleine Regel" (little period) has been applied to this condition. It corresponds to the uterine hemorrhage which occurs at estrus in some animals (e.g., dog), and probably results from small hemorrhages in an excessively stimulated follicular-phase-type of endometrium. It is not necessarily associated with any severe derangement in sexual functions and is compatible with normal fertility.

CHANGES IN THE VAGINA are comparatively unimportant accompaniments of ovarian disease. With the use of the vaginal smear method it is possible to estimate approximately the ovarian hormone production. In amenorrhea

due to ovarian failure, nucleated cells predominate, while, in hyperfolliculoidism most of the cells in the vaginal smears are cornified. Ovarian failure predisposes the vaginal epithelium to infections, since it reduces its resistance against micro-organisms. Sometimes deficient cornification renders the vagina hypersensitive and causes dyspareunia.

CHANGES IN THE MAMMARY GLANDS are common consequences of ovarian disease. *Underdevelopment of the breasts* is often the result of ovarian deficiency, but it may occur in women whose cycles and fertility are normal. In these cases it is perhaps due to a relative ovarian hormone insensitivity of the pituitary (decreased mammogenic hormone production) or of the mammary tissue itself.

Hypertrophy of the breasts often occurs at puberty (pubertal hypertrophy) and may persist throughout life. The breasts may become so large as to hang below the waist line. This may be due to excessive production of ovarian hormones or to an exaggerated hormone-sensitivity of the pituitary or breast tissue.

Chronic cystic mastitis (cyclomastopathy) is a condition which appears in two main forms: in one there is simple cystic dilatation of the galactophores (cystic form); in the other proliferation of the duct epithelium predominates (Schimmelbusch's disease, Reclus' disease, adenosis). In these latter cases, differentiation from malignancy may become difficult.

Cystic mastitis is especially common between 30 and 45 years of age, but it may occur in younger women. Usually it disappears after the menopause, presumably due to diminished ovarian hormone secretion, or even before that time without obvious reasons. It has a tendency to undergo periodic exacerbations and remissions. The breasts are often enlarged and localized cysts and

nodules are readily detectable by palpation. The etiology of this condition is not known. There may be an increased ovarian or hypophyseal hormone production, or an excessive hormone sensitivity of the breast.

Sterility and Infertility. — Sterility is often defined as the inability to conceive, while, in infertility, pregnancy can be initiated, but is terminated by abortion. It is customary to distinguish *primary* sterility in which conception has never occurred, from the *secondary* type, in which conceptions have taken place at an earlier time.

Here, we shall only consider the female factor in sterility and infertility. In order to permit insemination, it is manifestly necessary that the ovaries produce normal ova and discharge them from the ovarian surface into the tubal orifice, through which they must migrate without meeting an obstruction. Similarly, the spermatozoa must find favorable conditions in the vagina, the cervical canal and the oviduct; that is, the passages through which they have to ascend, in order to fertilize the ovum in the tube. Finally the endometrium must be progestational to permit implantation of the ovum, and the developing embryo must remain under favorable humoral conditions; thus, for example, during the first stages of development it is greatly dependent upon an adequate maternal progesterone supply.

VARIOUS TYPES OF STRESS may interfere with normal ovulation and menstruation (infectious diseases, alcoholism, dietary insufficiencies, emotional factors), and the same is true of ENDOCRINE DISTURBANCES, especially hypopituitarism, hypo- or hyperthyroidism, etc. Sterility can also be occasioned by COITAL DIFFICULTIES, such as dyspareunia (pain during sexual intercourse). This may be caused by psychologic reasons, trichomonas infection, senile vaginitis, inflammatory le-

sions in the pelvis, endometriosis, malformations, or occlusion of the genital passages due to developmental anomalies, inflammatory lesions, etc.

COMPLICATIONS

Among the complications of ovarian diseases, the most important is CHRONIC SECONDARY ANEMIA, due to continuous loss of blood through exaggerated menstrual bleeding. Occasionally, the rupture of large, cystic follicles into the peritoneum may lead to severe PERITONEAL HEMORRHAGES; rupture of an ovarian abscess to PERITONITIS, etc., but in general, ovarian diseases do not tend to cause important complications.

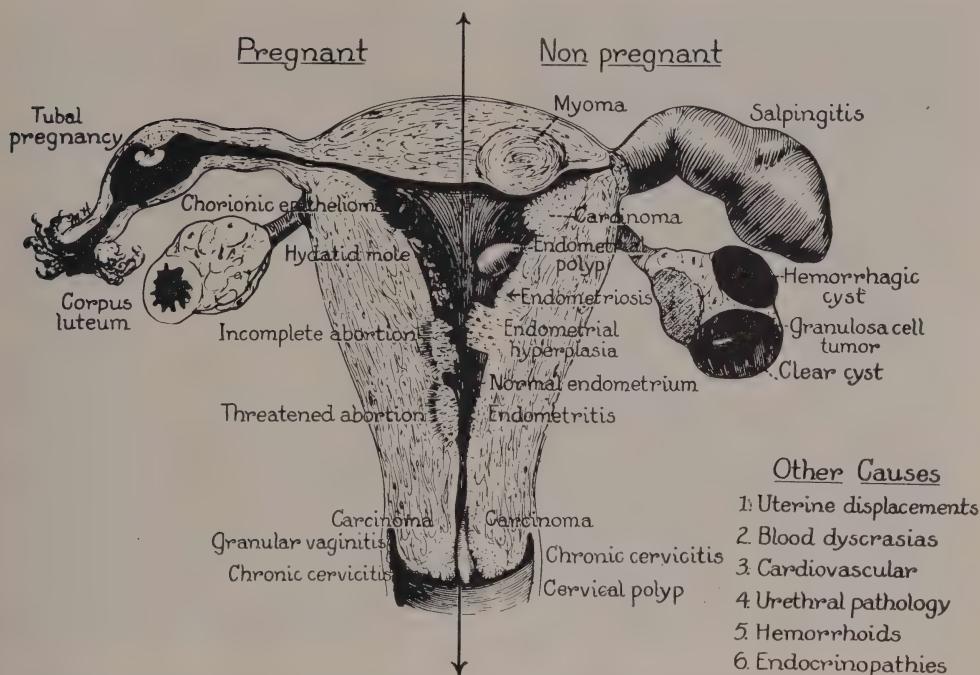
DIAGNOSIS

In the diagnosis of ovarian disease, it is important to inquire into the FAMILY HISTORY, since many diseases tend to be transmitted from the parents to the offspring.

The PAST HISTORY OF THE PATIENT furnishes valuable data concerning the onset of menstrual bleeding (menarche), the duration of the flow, the length of the intervals between menses, the amount of menstrual blood, the character of the discharge (with special reference to the presence of clots, membranes and bright red blood which are abnormal), intermenstrual bleeding and pain, dysmenorrhea, and the date of the last menstrual period. The marital history of the patient, the type of vaginal discharge (if any), urological symptoms (pain, increased frequency, hematuria, etc.), and the development of the PRESENT ILLNESS must also be recorded.

SYSTEMIC PHYSICAL EXAMINATION should be directed towards the detection of infectious, nutritional, endocrine, and other general diseases which may have an effect upon ovarian function.

GYNECOLOGIC EXAMINATION will help to reveal the presence of inflam-



Schematic drawing summarizing chief causes of uterine bleeding.

(After E. Henriksen: Am. J. of Obst. and Gynec. 41, 179, 1941.)



Sterility due to malformation of the uterus.
X-ray following injection of the uterus with X-ray opaque fluid (utero-salpingo-graphy). Note uterus unicornis. Obstruction of the abdominal ostium prevents the passage of the injection fluid.

(Courtesy of Dr. A. Pinto Viégas.)

Sterility due to blockage of the oviducts.
Note that X-ray opaque fluid completely fills the uterine cavity, which appears to be normal; the distal parts of both tubes are obstructed so that free passage of spermia and ova is impossible.

(Courtesy of Dr. A. Pinto Viégas.)

matory or neoplastic lesions in the pelvis; congenital anomalies in the development of the sexual organs, etc.

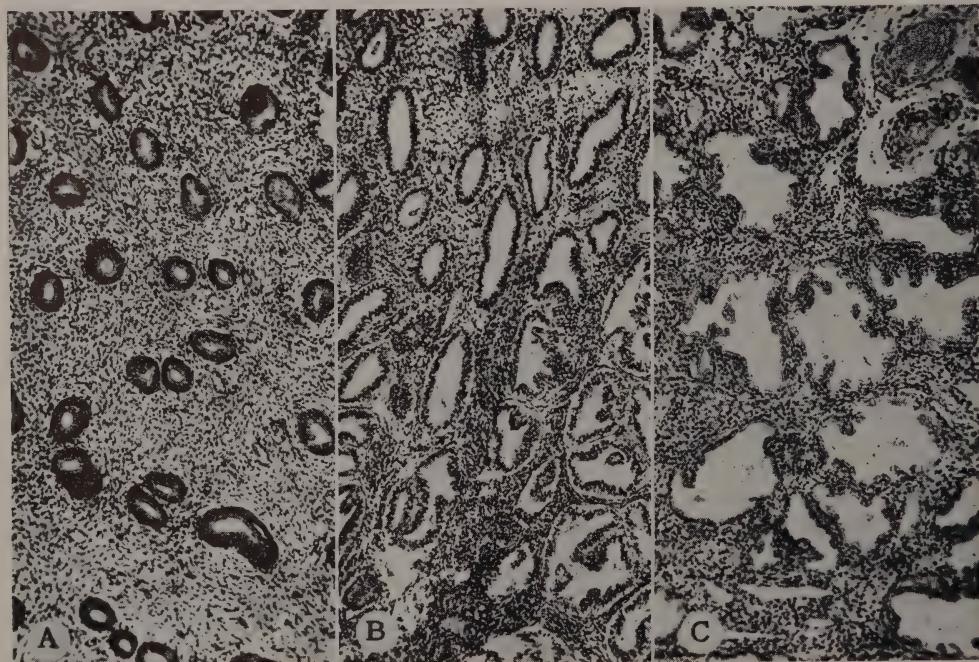
X-RAY EXAMINATION often helps to detect tumors in the ovarian, suprarenal or hypophyseal region. The patency of the oviduct can be examined by two commonly used technics. In the *tubal insufflation method* (Rubin, 1920) carbon dioxide is passed through the uterus into the tubes and the peritoneal cavity, the resulting pneumo-peritoneum can be demonstrated by X-ray examination or by subjective symptoms (shoulder pain, etc.). Furthermore, the pressure in the insufflation apparatus rises only slightly if the tubes are patent, but reaches very high levels if they are closed. The passage of air can also be detected, by its characteristic sound, if a stethoscope is applied to the lower abdomen. *Hysterosalpingography*, after injection of iodized oil

and other X-ray opaque substances, may give further details concerning the exact position and permeability of the tubes.

Examination of **ENDOMETRIAL BIOPSIES** and the **VAGINAL SMEARS** is of the greatest value in detecting ovarian diseases, since the histologic appearance of the uterine and vaginal mucosae exhibit characteristic changes under the influence of folliculoid and luteoid hormones respectively.

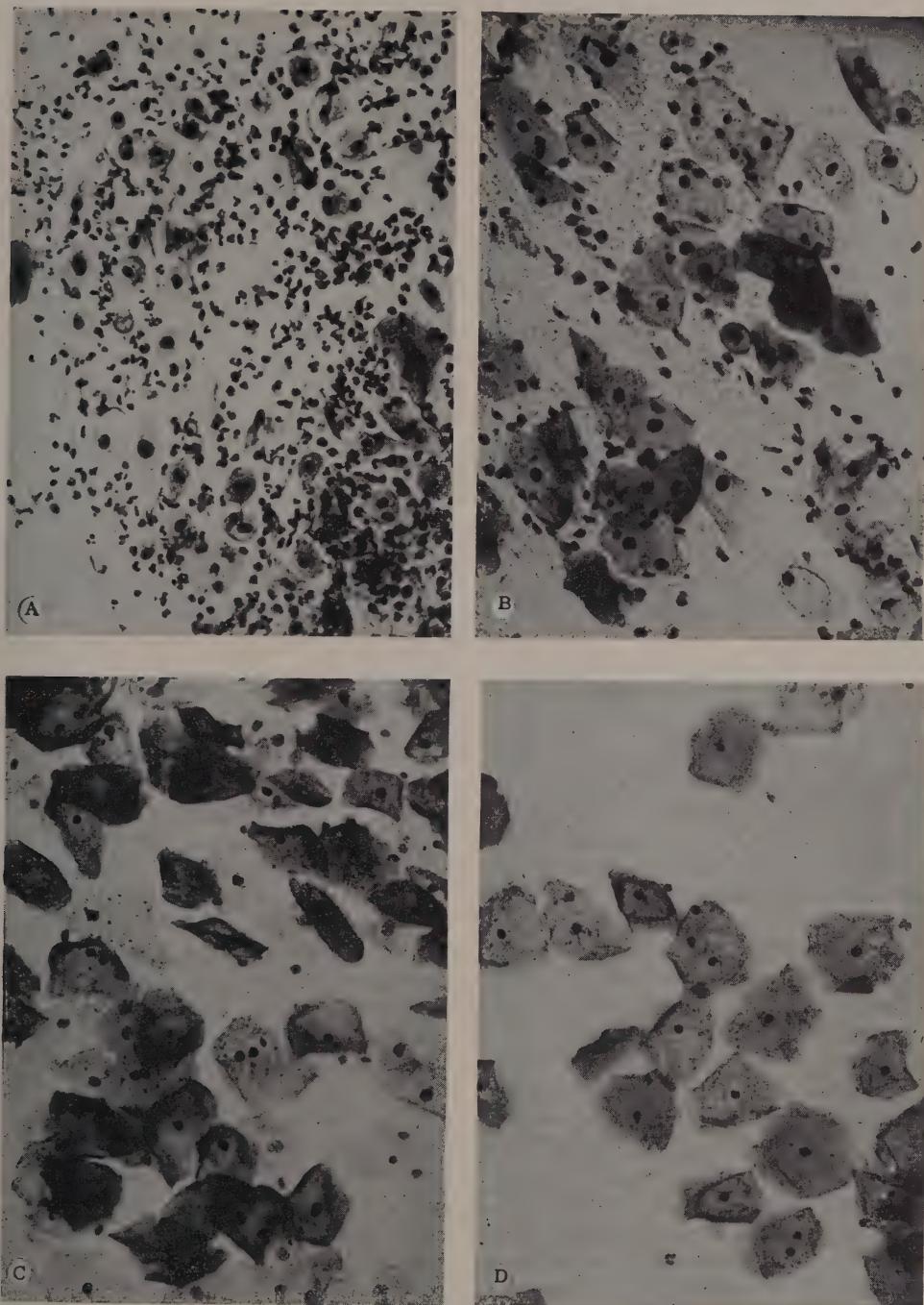
BIOCHEMICAL STUDIES, such as the B.M.R., blood and urine sugar and hemoglobin determinations are particularly valuable if changes in thyroid function, diabetes, anemia and other systemic diseases are suspected.

Among the **HORMONE DETERMINATIONS**, the urinary excretion of folliculoids, pregnanediol (as an indication of progesterone production), 17-KS and gonadotrophins are especially



Endometrial biopsies during the menstrual cycle. — A. Follicular phase. Specimen taken on 12th day of a 27-day-cycle. Note high cylindric cells of the glands with many mitotic figures (latter not clearly visible at this magnification). — B. Early luteal phase; 18th day of 27-day-cycle (2nd day of pregnanediol excretion). The nuclei of the glandular epithelia migrate towards the lumen, while the base of the cell contains light vacuoles. — C. Late luteal phase; 23rd day of a 25-day-cycle. Papilloma-like excrescences in convoluted glandular tubules.

(Courtesy of Drs. G.-S. Henry and J.-S.-L. Browne.)



Vaginal smears illustrating effect of folliculoids in primary amenorrhea. — A. Atrophic vaginal smear (primary amenorrhea) characterized by deep cells and leukocytosis. — B. Early changes induced by folliculoids. — C. More advanced changes induced by folliculoids. — D. Full follicular or estrous smear, indicative of full replacement therapy with folliculoids; characterized by leukopenia, thin mucus and the predominance of cornified cells with small pyknotic nuclei.

(Courtesy of Dr. E. Shorr.)

instructive. In all these determinations, it is advisable to obtain excretion curves extending over a whole or even several cycles, since single determinations are of comparatively little value.

THERAPY

In connection with the HORMONAL THERAPY of ovarian diseases, the folliculoid, luteoid, gonadotrophic, thyroid and perhaps also the testoid preparations are useful. The pertinent pharmacologic problems (dosage, route of application, sensitization by combined treatment with several hormones, etc.) have been reviewed in connection with the pharmacology of the individual hormones. For the therapy of well-characterized specific ovarian diseases, the reader is referred to the chapters dealing with these in particular (e.g., Metropathia Hemorrhagica, Ovarian Tumors). We shall limit ourselves, here, to the discussion of a few general principles, applicable to the therapy of a variety of ovarian diseases.

Hypogonadism. — In the treatment of hypogonadism and amenorrhea it is usually best to imitate the normal, sexual cycle by the successive administration of folliculoids during 18 days, and luteoids during the subsequent 8 day period. If, at the end of this time, hormone treatment is interrupted for a few days, true menstrual bleeding occurs from a progestationally prepared endometrium. This therapy also helps to develop the accessory sex organs (breasts, uterus, vagina) and to improve the otherwise subnormal libido.

A simpler and apparently equally effective procedure is the "2 day treatment" (*Zondek*), in which bleeding ensues 4 days following simultaneous intragluteal injection of 2.5 mg. of estradiol benzoate and 12.5 mg. of progesterone on two consecutive days. Of course, it will be realized that in the absence of ovulation, treatment with

ovarian hormones can not restore fertility. Yet, in certain instances where hypogonadism is not due to complete destruction of the ovaries, such treatment re-initiates spontaneous ovarian and menstrual cycles.

Some gynecologists limit treatment to 8-14 daily doses of a folliculoid compound (e.g., 0.5-1 mg. of stilbestrol, or 30 mg. of estrone sulfate per os daily), or to luteoids alone (50 mg. of progesterone, divided in 5 injections of 10 mg. each on 5 successive days), a procedure which is usually followed by uterine bleeding 60 hours after the last injection in cases of secondary amenorrhea. On theoretic grounds, the sequential administration of folliculoids and luteoids appears preferable, since it imitates natural conditions, but satisfactory results are sometimes obtained with folliculoids or luteoids administered singly. If the amenorrhea is primary, and the endometrium entirely unprepared by endogenous folliculoids, pretreatment for about 5 days with a folliculoid, in the usual doses is essential.

It should be kept in mind that as far as we know, absence of menstrual bleeding is not harmful in itself. Its artificial induction in an amenorrheic woman is chiefly of psychologic value unless ovulation is produced. Hence the importance of producing a progestational endometrium is somewhat dubious.

In order to stimulate the ovary itself, GONADOTROPHIC PREPARATIONS must be administered. A variety of hypophyseal, urinary, and pregnant mare serum gonadotrophins, as well as combinations of the above, are commercially available. Some gynecologists recommend the use of gonadotrophins in combination with folliculoids, for the treatment of hypogonadism and amenorrhea. Unfortunately, the commercial preparations of gonadotrophins are still impure, and unlike in experimental animals, it is rarely possible to produce

ovulation and thus to restore fertility with the gonadotrophins now on the market. We are as yet unable to lay down definite rules concerning the choice of the preparations and the clinician usually has to determine by trial and error which patient responds better to ovarian and which to gonadotrophic substances. If the ovary is completely destroyed, it is of course useless to administer gonadotrophins.

In view of the frequent occurrence of hypogonadism in the presence of, sometimes latent, hypothyroidism, the administration of small doses of DESICCATED THYROID ($\frac{1}{2}$ to $\frac{3}{4}$ grains daily) is often advantageous. Indeed, it is claimed that such therapy may help even if there are no signs of hypothyroidism.

PROSTIGMINE has been recommended in the treatment of retarded menstrual bleeding. It apparently produces pelvic hyperemia through its action upon the parasympathetic nervous system. It should be recalled that the folliculoids have been claimed to produce hyperemia through the release of acetylcholine, whose destruction is impeded by prostigmine. The applicability of this therapy is rather limited.

It is important to remember that the general condition of the patient plays a prominent rôle in the causation of many types of amenorrhea. Hence, a properly balanced DIET is recommended, particularly the control of excessive obesity, and in the event of anemia, IRON preparations. Light X-RAY treatment of the ovary or hypophysis have often been claimed to be beneficial in certain types of amenorrhea because of their alleged gonad-stimulating effect.

The THERAPY OF OLIGO- AND HYPO-MENORRHEA is based essentially on the same principles as that of other types of hypogonadism.

Hyperfolliculoidism. — For therapy see : Hyperfolliculoidism on page 431.

Hyperluteoidism. — For therapy see : Corpus Luteum Cysts and Hyperluteoidism on page 436.

Dysmenorrhea. — The therapy of dysmenorrhea is still almost entirely empirical. In some cases, SURGICAL intervention is indicated if the cause is obstruction of the genital passages, endometriosis or malposition of the uterus. In certain instances, dilatation of the cervix with curettage proves beneficial. In very resistant cases, sympathectomy or hysterectomy is recommended, but the latter must, of course, be performed only on patients refractory to all other types of therapy.

When excessive emotional reactions to the normal menses are the cause of dysmenorrhea, PSYCHOTHERAPY often gives excellent results.

Recently, HORMONE THERAPY has become increasingly more popular and favorable results have been reported after treatment with large doses of folliculoids (which prevent ovulation), or small doses of testoids (which in addition to inhibiting ovulation, cause involution of the uterus). Gonadotrophic preparations and progesterone have also been recommended, the latter because it was believed to inhibit tonic uterine contractions.

For the treatment of the pain itself, various SEDATIVES (salicylates, bromides and, in severe cases, morphine) may have to be prescribed.

Hypermenorrhea (or Menorrhagia). — In women with menorrhagias testoid treatment tends to diminish uterine bleeding because of the resulting atrophy of the endometrium. In some instances, gonadotrophins or progesterone prove effective in diminishing the flow, although the mechanism of their action is not yet quite clear. Thyroid treatment is indicated in hypothyroid women. In young women, temporary, in older, premenopausal women permanent X-ray or radium castration or even ovariectomy is recommended.

since it eliminates ovarian hormone production; but this should only be used as a last resource. In young women in whom the bleeding is very severe and irresponsive to all other therapeutic measures, hysterectomy without ovariectomy may be preferable.

Intermenstrual Bleeding. — Unless it is severe, this condition does not necessarily require any therapy. Curettage in itself may bring about a cure. In some cases, testoid or folliculoid hormones prove effective when given prior to the expected intermenstrual bleeding.

Chronic Cystic Mastitis. — This condition is very resistant to any form of therapy. Treatment with folliculoids and testoids has been recommended on the basis of rather doubtful hypothetic considerations. Both folliculoids and testoids stimulate mammary growth, and hence they are not likely to be beneficial unless their suppressing effect upon the endogenous hormone production of the ovary overcompensates for their direct breast-stimulating action. On the other hand, folliculoid therapy is often highly effective in the treatment of underdeveloped breasts.

Vaginitis. — Gonorrhreal vulvovaginitis in children and senile vaginitis in old women, respond very well to folliculoid therapy, and in such instances topical application in the form of vaginal suppositories is recommended.

Sterility and Infertility. — Many cases of sterility are due to vaginitis, cervicitis, pelvic inflammation, stenosis of the cervix, displacement of the uterus, fibromyomas, systemic diseases such as anemia, diabetes, etc., or malnutrition. In these instances, THERAPY SHOULD BE DIRECTED AGAINST THE SPECIFIC CAUSATIVE FACTOR. It must also be kept in mind that some women are constitutionally less fertile than others

and that the fertility of the male partner must be ascertained before the blame is placed upon the woman. It is likewise well to explain to the patient that INTERCOURSE DURING THE MIDMENSTRUAL PERIOD (at the time of ovulation), is most likely to bring about pregnancy, after several days of abstinence. The optimal time for conception can be determined by vaginal smears or basal body-temperature measurements. (See: "Estrus and Menstruation.")

ARTIFICIAL INSEMINATION with semen from a man of proven fertility is undoubtedly effective in cases in which the male partner is at fault. The procedure has been much criticized, however, both on moral and legal grounds.

As regards the endocrine therapy of sterility, THYROID preparations have often been claimed to be helpful, even in cases without any definite evidence of hypothyroidism. It is somewhat difficult to understand, however, why this measure is effective.

On theoretic grounds, treatment with GONADOTROPHINS would appear to be most logical, since these so readily produce ovulation in laboratory animals. As we have stated elsewhere, however, the gonadotrophins now available rarely, if ever, cause ovulation in primates. Correspondingly, results with various FSH and LH preparations or combinations of the two have not been very satisfactory in women.

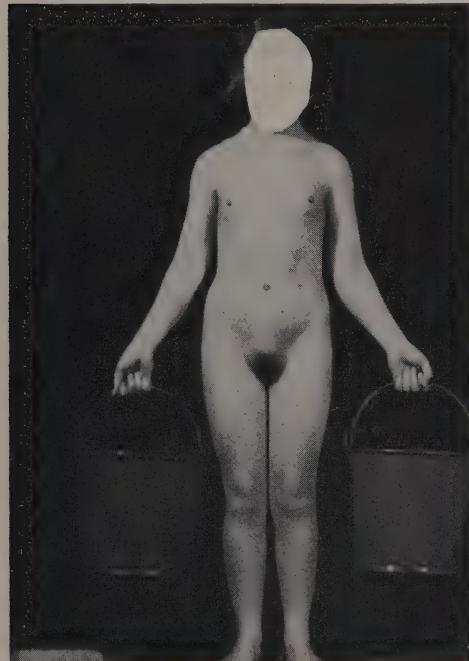
Treatment with FOLLICULOID or LUTEOID preparations is advisable only if there is clinical evidence (biopsy, bioassay) of a deficiency in the endogenous production of these hormones. In such cases, therapy should be adjusted to compensate for the functional deficiency, folliculoids being given during the first, luteoids during the second half of the cycle.

SPECIAL DISEASES OF THE OVARY

MALFORMATIONS AND ANOMALIES

Aplasia (or Agenesis). — Very few cases have been reported in which both ovaries were absent at autopsy in women in whom there had been no reason to suspect a secondary destruction of the gonads during embryonic or postembryonic life. True ovarian agenesis is usually accompanied by manifestations variously described as TURNER'S SYNDROME or ALBRIGHT'S SYNDROME. This is characterized by great shortness of stature ("ovarian dwarfism"); infantile development of the mammary glands, uterus and vagina; diffuse osteoporosis; increase in the urinary elimination of gonadotrophins; diminution, but not complete absence, of axillary and pubic hair; and various congenital anomalies (e.g., coarctation

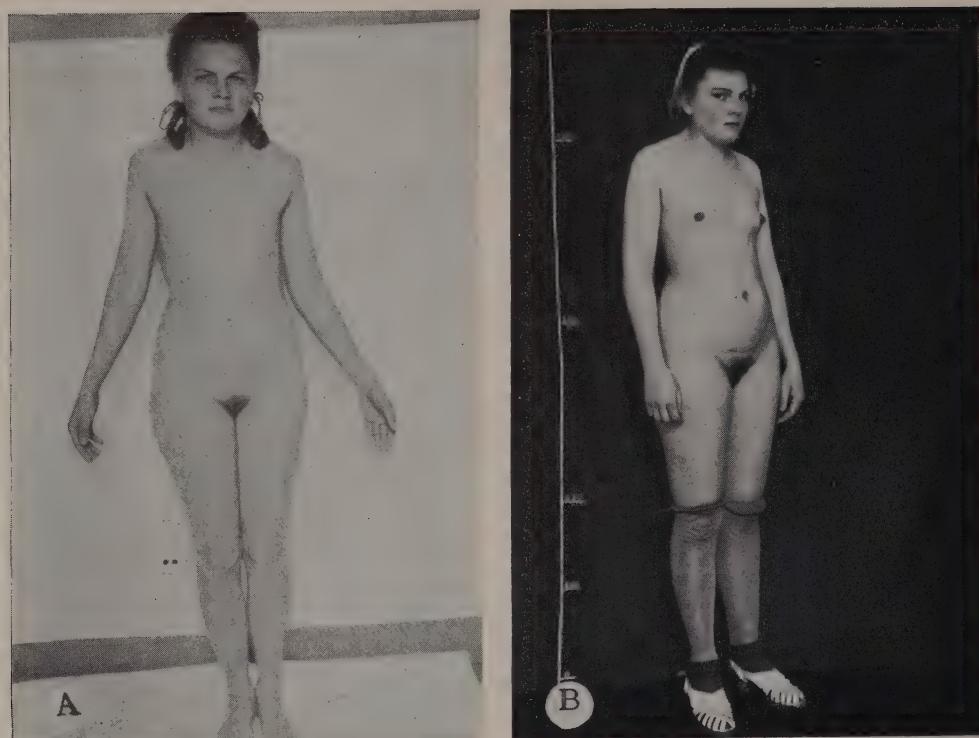
of the aorta, cubitus valgus, scoliosis, absence of upper lateral incisor teeth and webbing of the neck).



Turner's syndrome (Ovarian agenesis)
Age 14 years. Webbing of neck is hardly noticeable because of plastic operation. Note increased carrying angle of arms, lack of breast development and moderate amount of pubic hair.
(After F. Albright et al.: Am. J. Med. Sc. 204, 625, 1942.)

Primary Hypoovarianism. Age 26 years, primary amenorrhea, excessively long arms and legs, lack of breast and uterine development (uterine canal 4 cm. in length), endometrium immature with no signs of progestational proliferation, vaginal smears of castrate type, skeletal maturity less than corresponding to 21 years of age. Note immaturity of facial expression, sella normal as judged by X-rays, visual fields show no abnormality.

(Courtesy of Dr. E. P. McCullagh.)



Ovarian agenesis. — A. Girl with primary amenorrhea, absence of breast development and shortness of the neck. Cubitus valgus not very marked. — B. Same patient after 4 months of intensive folliculoid therapy. Note increase in pubic hair and breast development, with prominent pigmented nipples. (Courtesy of Dr. E.-B. del Castillo.)

The frequent accompanying mental retardation may be due to congenital defects in brain formation.

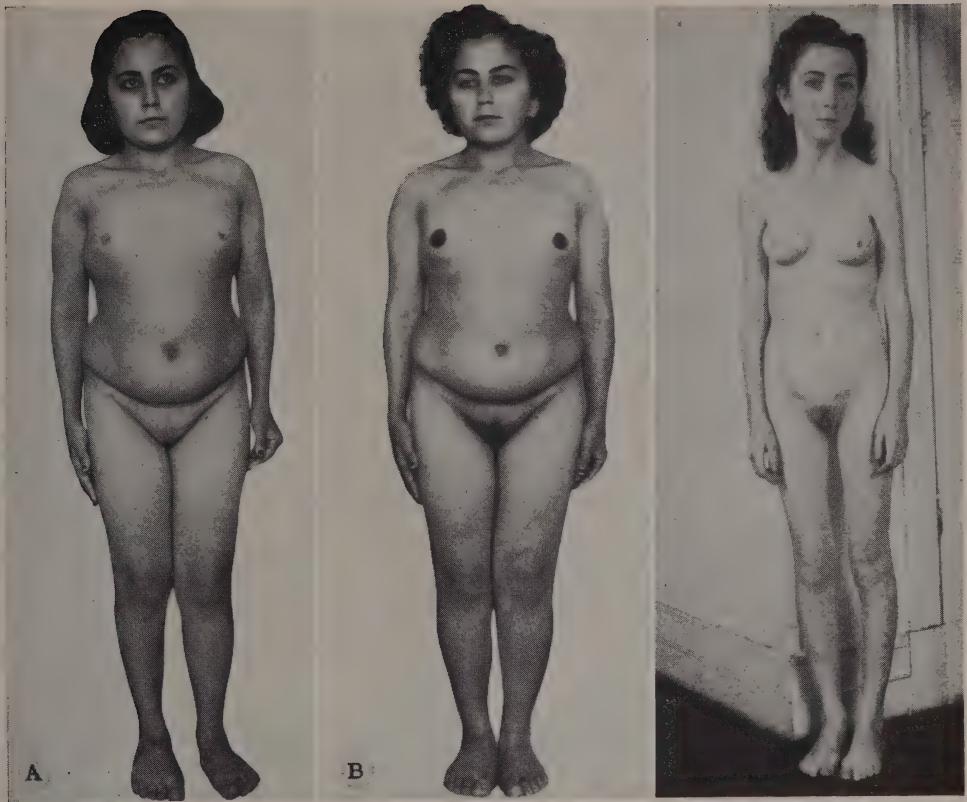
It is often difficult to differentiate these women from pituitary dwarfs, but unlike the latter, the ovarian dwarf exhibits an only slightly retarded bone age, a normal insulin tolerance curve, and an increased urinary FSH excretion. Furthermore, the ovarian dwarf responds to folliculoid therapy with marked axillary and pubic hair development while the pituitary dwarf does not. In some women, ovarian agenesis is accompanied by signs of virilism, especially hirsutism.

Hypoplasia of the Ovaries. — Primary hypoplasia of the ovaries is presumably a very rare condition. In allegedly pertinent cases, it is almost impossible to determine whether we are dealing with a true malformation of the

gonad, with the result of pituitary malfunction, or with originally normal ovaries which underwent secondary involution under the influence of local damage.

Clinically, this condition does not differ significantly from other types of severe hypoovarianism which, having commenced at a very early age is accompanied by primary underdevelopment of the accessory sex organs.

Accessory Ovaries. — Women with more than two ovaries are exceptional. Sometimes parts of an otherwise normal ovary are partially separated from the main gland and if this malformation goes further, certain portions of the gonad may become completely separated. These are usually referred to as SUPERNUMERARY or FALSE ACCESSORY ovaries. In exceptional instances, the additional ovary is fully equivalent —

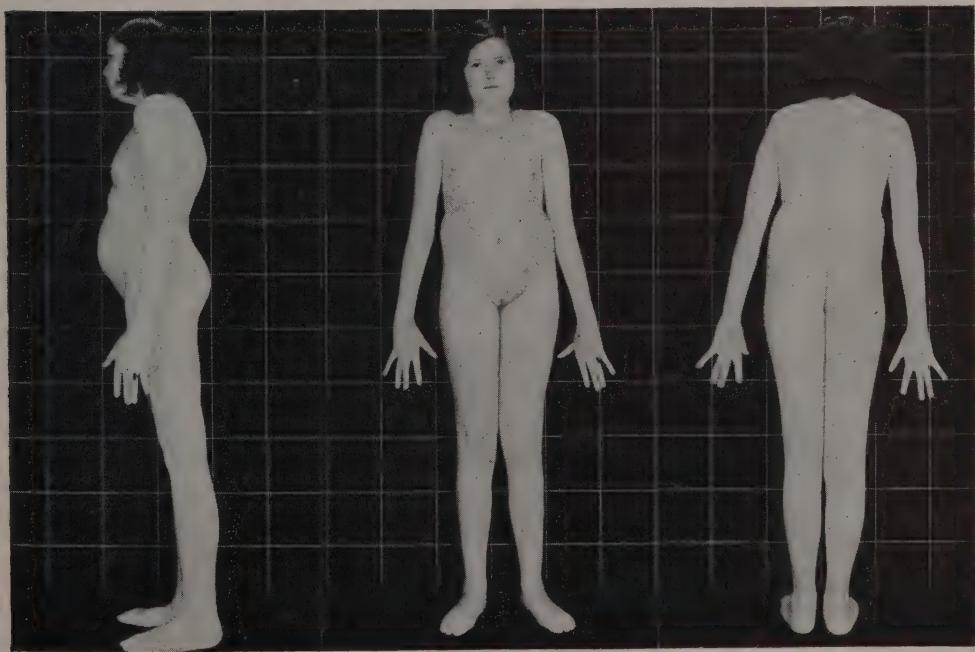


Ovarian dwarfism. — A. 17-year-old girl, with retarded growth and sex development. Several congenital anomalies (cubitus valgus, flat feet, nystagmus and short neck); osteoporosis; urinary gonadotrophins high, daily 17-KS low (2.8 mg.) — B. After folliculoids development of pubic hair and breasts, pigmentation of the areola.

(Courtesy of Dr. A.-B. de Ulhôa Cintra.)

Hypoovarianism. 17-year-old girl with primary amenorrhea and scarce pubic hair. Note that breast can be normal in severe hypogonadism.

(Courtesy of Dr. J.-I. Lobo.)



Primary hypoovarianism. 19-year-old girl with absence of pubic hair and breast development. Note disproportionately long extremities and fingers, open junction cartilage, lack of pigmentation in areola and short neck.

(Courtesy of Dr. E.-K. Shelton.)

in size and function — to the normal gonads, indeed it may be connected with a third oviduct. In these cases, we speak of TRUE ACCESSORY OVARIES OR THIRD OVARIES. A surprisingly large number of these rare third ovaries become the site of tumor formation (mostly teratoids). This may be interpreted as indicating that congenital ovarian malformations result in a predisposition to tumor formation. It must also be kept in mind that the torsion of tumor-bearing ovaries with subsequent self-amputation and reimplantation may be confused with tumor-formation in ectopic ovaries.

Hyperplasia and Hypertrophy. — It is doubtful whether true hyperplasia of the ovaries ever occurs as a primary malformation. Probably most, if not all cases described by such terms as "the giant ovary" and "the large ovary" are actually due to chronic proliferative inflammatory processes, or to increased gonadotrophin production, rather than to a true congenital malformation. As

with several other endocrine glands, it is impossible to distinguish between hyperplasia and hypertrophy due to causes inherent in the ovary itself, and corresponding changes resulting from increased pituitary trophic hormone production.

Primary Ectopia of the Ovary. — The malpositions of the ovary may be subdivided into two groups: (1) *primary ovarian ectopia* in which the ovary is abnormally situated, due to a disturbance in its embryonic development (usually incomplete descent during fetal life); (2) *secondary ovarian ectopia* comprising acquired malpositions, due to displacement of the gonads by growths, hernias of the ovary, etc. The distinction between these two groups is not always clear; for instance, inguinal hernias of the ovary are possible only if closure of the inguinal canal fails to occur during fetal life. In such instances, it is difficult to prove whether the malposition of the ovary itself is primary or secondary. Furthermore,



Pseudohermaphroditism. — A, B and C. Patient with pseudohermaphroditism, whose sex has not been verified by biopsy. Note unusually large clitoris or penis (3 cm. in length), female pubic hair distribution. The testes were not palpable, the urethral orifice was about 2 cm. below the penis-like organ. Rectal palpation revealed neither prostate nor testes or female accessory sex organs. The 9-year-old individual was mentally retarded and possessed a small palpable goiter.

(Courtesy of Dr. A. Pinto Viégas.)

ovarian torsion may lead to spontaneous amputation of the gonad, with secondary reimplantation in other parts of the abdomen. [See also : Hernia and Prolapse (Secondary Ectopia) of the Ovary on p. 406.]

Ambisexuality (Hermaphroditism and Pseudohermaphroditism). — We use the generic term "AMBISEXUALITY" to designate the condition of individuals possessing both male and female characteristics. As outlined elsewhere in this book, even the normal human individual is somewhat ambisexual. Both folliculoid and testoid compounds are produced by men and women and even certain purified steroids (e.g., testosterone) exhibit both folliculoid and testoid activity. For the purpose of the present discussion however, the expression "ambisexuality" will be used as a generic term, designating individuals in whom the simultaneous presence of the characteristics of both sexes is exaggerated. Hence it includes not only cases in which the disturbance is due to a congenital malformation (hermaphroditism), but also tumors or hyperplasia of endocrine glands developing during postnatal life (for a more detailed discussion of these, see : "Adrenogenital Syndrome," "Arrhenoblastomas," "Cushing's Disease," "Testoid Hyperthecosis," "Lipid Cell Tumors of the Ovary," "Diseases of the Testis," etc.).

The term "TRUE HERMAPHRODITISM" (HERMAPHRODITISMUS VERUS) is used to designate the condition of individuals possessing both male and female gonadal elements. Even this condition is normal, at least in some lower animals. Thus, many invertebrates are physiologically hermaphrodites inasmuch as every individual produces male and female gametes either at different times or synchronously. Some species are even self-fertilizing. Among vertebrates however, there are only few examples of true permanent hermaphro-

ditism under normal conditions (see below).

In the early literature, there was a great tendency to develop complex systems for the CLASSIFICATION OF THE VARIOUS TYPES OF AMBISEXUALITY. It is amusing to note, for instance, that at a time when the medical literature knew only of 7 or 8 proven cases of true hermaphroditism, Klebs (1876) devised a system of classification and terminology in which 16 categories of true hermaphroditism were distinguished, merely because they were theoretically possible. Such systems played a great rôle in the medical literature, but they are hardly justified, since neither the intensity nor the extent (number of organs affected) of the hermaphroditic changes fall into natural, well-defined categories.

Young (1937) proposed the following simplified classification of true hermaphroditism as being closely in agreement with general usage :

(1) *Lateral hermaphroditismus verus* (An ovary on one side and a testicle on the other).

(2) *Unilateral hermaphroditismus verus* (Both ovary and testis on one side and one of the two on the other).

(3) *Bilateral hermaphroditismus verus* (An ovary and a testis on both sides).

In man, cases of true hermaphroditism are extremely rare. A careful study of the relevant literature revealed only 38 cases which in my opinion may be accepted as belonging to this category. In one family, benign testicular tubular adenomas were observed in two hermaphroditic sisters (Novak, 1943). There were several additional "intersexual" individuals in the same family and the malformation was considered to be hereditary. The gonads were described as "ovotestes."

The testes of true hermaphrodites do not tend to descend into the scrotum but remain in the abdomen or the inguinal canal. Since spermatogenesis is inhibited even in otherwise normal

ectopic testes (see : Cryptorchidism), it is in accordance with expectations that ovo-testes usually contain under-

developed tubules, similar to those of ovarian tubular adenomas. This illustrates the close relationship between



True hermaphrodite. — A. Close-up of genitals showing phallus in position of clitoris surrounded by prepuce representing labia minora. Labia majora showing corrugations reminiscent of scrotum. — B. Close-up of genitals revealing phallus with urethra occupying position of clitoris. Beneath this is vaginal orifice. — C. Gross specimen of pelvic organs showing uterus, tube and bilateral ovo-testis. — D. Microscopic section showing primordial ovarian follicles and seminiferous tubules in the same organ. — E. Microscopic section showing presence of early-prostate-like tissue and primordial ovarian follicles in the ovo-testis of the opposite side.

(Courtesy of Dr. R.-C. Grauer.)

true hermaphroditism and virilization due to ovarian tumors.

In some instances however, both gonads consisted of comparatively normal ovarian and testicular tissue, while in others separate testes and ovaries were noted. In one individual only one of the gonads was examined by biopsy and proven to be an ovary, yet the patient had frequent intercourse with women and since living spermatozoa could be detected in the spermatic fluid, it is evident that the remaining gonad must have been a testis.

The term "PSEUDOHERMAPHRODITISM" (PSEUDOHERMAPHRODITISMUS) is used to designate the condition of individuals possessing either ovaries or testes, but both male and female secondary sex organs.

The classification of pseudohermaphroditism according to Klebs (1876), although extremely complex, is still so generally in use that we must mention it here. It distinguishes :

(1) *Pseudohermaphroditismus masculinus externus*. (The sex glands and inner secondary sex organs are female, while the external genitalia are male).

(2) *Pseudohermaphroditismus femininus externus*. (The sex glands and

inner secondary sex organs are male, but the external genitalia are female).

(3) *Pseudohermaphroditismus masculinus internus*. (The sex glands are male, but oviducts, uterus and vagina are also developed).

(4) *Pseudohermaphroditismus femininus internus*. (The gonads are female, but both male and female sexual passages are developed).

(5) *Pseudohermaphroditismus completus masculinus*. (The sex glands are male, but all secondary sex organs are female).

(6) *Pseudohermaphroditismus completus femininus*. (The sex glands are female, but all secondary sex organs are male).

In itself, the term pseudohermaphroditism is incomplete, unless modified by two of the following five adjectives, namely : masculine or feminine and internal, external or complete. The first two designate the gonad, the last three the manner in which the genital structures differ from the sex of the gonads. By the use of these adjectives, all possible combinations can be designated fairly accurately (Creevy). These considerations led to the following modification of Klebs' classification :

Pseudohermaphroditism

Designation	Gonads	Internal Sex Organs	External Sex Organs
Feminine External	Ovaries	Tubes and uterus present either normal or hypoplastic	To some extent, resemble those of the male
Feminine Internal	Ovaries	Tubes and uterus present; Vestigial male structures present	Appear normal
Feminine Complete	Ovaries	Tubes and uterus absent or vestigial. Vestigial male structures present.	May appear to be completely masculine. Well developed phallus with a urethra. Labia closed to form a scrotum, etc.
Masculine External	Testes	Normal and masculine	Hypospadias, cleft scrotum, etc.
Masculine Internal	Testes	Tubes, uterus present	Appear normal
Masculine Complete	Testes	Masculine organs absent or vestigial; tubes, uterus present	Almost entirely feminine

Most external and complete pseudohermaphrodites are reared as the wrong sex and when they marry, usually marry a member of the same sex.

Pseudohermaphroditism is much more common in females than in males and statistics indicate that about one out of every thousand women is pseudohermaphroditic. In many of these, the histologic structure of the ovary is approximately normal, while in others, excessive proliferation of the "male elements" in the hilum region (medullary cords, ovarian Leydig cells) or of heavily luteinized and sometimes cystic theca-cell nests (testoid hyperthecosis) account for the virilization.

The secondary sex organs of female pseudohermaphrodites show varying degrees of masculinization, such as development of a rudimentary prostate, hirsutism, a male type of skeleton, voice, libido and fat distribution. In male pseudohermaphrodites, cryptorchidism, hypospadias with hypoplasia of the penis and feminization of voice, libido and fat distribution are most striking.

HOMOSEXUALITY may be regarded as a psychic type of pseudohermaphroditism, but since this condition is not necessarily associated with any definite change in ovarian structure, it will not be discussed here.

The term "INTERSEX" has been used by Goldschmidt (1931) to designate animals which originally developed as pure genetic males or females, but then changed in the direction of the opposite sex. In such individuals there is an intermediate period in which characteristics of both sexes are manifest.

The term "GYNANDROMORPH" is often used to designate animals in which both the gonad and the secondary sex organs are male on one side and female on the other. This term may also be employed however for more complex types of ambisexuality, such as true hermaphroditism "due to a mosaic of genetically male and female cells" (Witschi, 1939).

Since neither intersexuality nor gynandromorphism have been definitely proven to exist in mammals, we shall not discuss them here.

Rare Malformations and Anomalies of the Ovaries. — The terms "ovarium lobatum," "gyratum," "succenturiatum," "bipartitum" and "disjunctum" are often applied to gonads with deep furrows in which part of the parenchyme is more or less completely separated from the rest. Some of these anomalies are obviously congenital, while others may be acquired during postnatal life.

"ECTROPION" OR PROLAPSE OF THE CORPUS LUTEUM (very rare) results from an eversion of the granulosa through the rupture point of the follicle at the time of ovulation. Thus, the resulting corpus luteum undergoes a prolapse which places the originally innermost layers of the granulosa to the outside. It will be recalled that in certain animals (e.g., elephantulus), this eversion occurs normally.

SPONTANEOUS AMPUTATION of the ovary is seen almost only in tumor-bearing gonads. The amputated ovary may reimplant itself in an ectopic position or remain floating in the peritoneum and become necrotic or even calcified.

The CORPUS LUTEUM LIBERUM (very rare) is a yellow body, which has become entirely separated from the ovary (presumably due to trauma or pressure from pelvic tumors). It may remain a free body in the peritoneum or reimplant itself in an ectopic position.

HERNIA AND PROLAPSE (SECONDARY ECTOPIA) OF THE OVARY

The most frequent types of secondary ovarian ectopia are the hernia and especially one of its varieties, the prolapse. The HERNIA may be defined as a transposition of the ovary from the abdominal cavity into a pouch covered by peritoneum. Because of its comparative frequency and its peculiar anatomic

characteristics, ovarian PROLAPSE deserves special attention. It is a transposition of the ovary into the posterior cul-de-sac where — depending upon the degree of prolapse — the ovary may or may not actually protrude into the vaginal lumen within a special peritoneal sac. Hence, a prolapse may merely represent a slight intraperitoneal transposition or a true vaginal hernia of the ovary.

The different sites of ovarian hernias are, in decreasing order of frequency : inguinal, femoral, sciatic, obturator, perineal, vaginal, umbilical, lumbar, broad ligament and abdominal scar.

From the endocrinologic point of view, herniation of the ovary is important because it is frequently accompanied by cystic degeneration or tumor formation in the gonad. It must be kept in mind that inguinal hernias can only occur in the case of a delayed occlusion of the inguinal canal and an exaggerated descent of the gonad. Both of these changes are "male" characteristics (typical of testis development), and hence, inguinal hernias may be regarded in a sense as due to partial pseudohermaphroditism. They are not uncommonly accompanied by other signs of virilism and in many instances of true hermaphroditism, the ovo-testes remain permanently in an inguinal location.

Frequently, the herniated ovary continues to function normally although it may cause discomfort and local pain at the time of ovulation and menstruation. In the case of vaginal prolapse, it tends to produce severe dyspareunia.

The most serious COMPLICATIONS of ovarian hernias are due to strangulation and torsion of the pedicle.

The THERAPY is replacement of the ovary into its normal position, if possible. Often however, it is preferable to perform an ovariotomy, especially if the gonad is cystic or inflamed.

TORSION OF THE OVARY

Because of the comparatively great mobility of the ovaries, they are rather subject to torsion at the pedicle. This may lead to severe nutritional disturbances in the gonads and even necrosis or self-amputation. Torsion occurs most frequently in tumor-bearing ovaries which have elongated, narrow pedicles. Pregnancy is an important predisposing factor. Perhaps the development of a large corpus luteum and the gradual enlargement of the uterus help to displace the ovaries during gestation.

CLINICALLY, the predominant symptoms are those of sudden intense pain in the lower abdomen with accelerated pulse and often severe shock. The most frequently made diagnostic errors are due to confusion of ovarian torsion with acute appendicitis, or ectopic pregnancy.

The TREATMENT of choice is the surgical removal of the twisted ovaries.

RETROGRESSIVE CHANGES

The retrogressive changes of the ovary are of no great endocrinologic importance. Among them, it suffices merely to mention COLLOID OR HYALINE DEGENERATION and AMYLOIDOSIS, which are very rare.

SIMPLE ATROPHY is almost invariably due to deficient gonadotrophin formation, while CIRRHOSIS is most commonly a result of chronic inflammation or cystic degeneration.

NECROSIS is almost always due to torsion, tumors, self-amputation and other diseases conducive to nutritional disturbances in the ovary.

CALCIFICATION and even OSSIFICATION may occur in the ovary during the healing of inflammatory or degenerative lesions. These are often detectable by X-ray examination.

VASCULAR DISTURBANCES

Ovarian HEMORRHAGES are of great clinical importance because of their comparative frequency and the serious clinical manifestations which they may elicit. Usually, they are due to excessive bleeding from a ruptured follicle, a corpus luteum or an ovarian tumor. The incidence of ovarian hemorrhage is estimated to be about 1% among all gynecologic and surgical laparotomies. They occur more frequently during the menarche, and usually at the time of ovulation. Corpus luteum hemorrhages are often seen during pregnancy and the increased pelvic hyperemia and intra-abdominal pressure of gestation also predispose to hemorrhage from ovarian tumors.

The main *clinical* characteristics in the typical case are midmenstrual pain, slight elevation of the temperature, some polymorphonuclear leukocytosis and — in the case of severe hemorrhages into the peritoneum with considerable loss of blood — a weak pulse with signs of severe shock.

In many instances, no therapy is necessary, since the bleeding stops spontaneously. If shock is present, blood transfusion and external application of heat are advisable after the operation. The simplest surgical procedure is to strip the hematoma cavity of its lining and approximate its walls with a fine catgut suture. If the ovary is severely damaged, complete resection may be necessary.

Because of the large number and comparatively great width of the veins in the ovarian hilum region, they are rather subject to VARICOSITY (varicocele), especially if circulation is impeded as in retrodisplacement or subinvolution of the uterus. Usually varicocele causes no endocrine derangement and in most cases it can be cured by ligature and removal of the varicose veins.

INFLAMMATIONS

Inflammations of the ovaries are rarely of endocrinologic significance, except in differential diagnosis. With regard to their COURSE, we distinguish acute and chronic oöphoritis; with regard to the LOCALIZATION of the inflammatory reaction, it is customary to delimit diffuse, interstitial oöphoritis from abscess formation in the cavities of follicles, corpora lutea or ovarian cysts.

Concerning the PORTAL OF INFECTION, it is known that microbial invasion of the ovary may be *hematogenous*, through the lymphatics, or by ascension, through the Fallopian tubes. Most frequently, the infection spreads by direct contiguity and leads to simultaneous development of oöphoritis in combination with salpingitis. Sometimes, oöphoritis is due to infection by contiguity from an inflamed appendix. Gonorrhreal infection tends to spread through the tubes, while streptococcus infection shows a predilection for propagation through the parametria, probably because it occurs most frequently postpartum or after abortion when the uterus is traumatized.

As regards the ETIOLOGIC microorganism, the *gonococcus*, *streptococcus* and *tubercle bacillus* are of the greatest importance, though *syphilis*, *staphylococcus* and many other microbial infections may also lead to ovarian inflammation.

The microorganism of mumps is believed to possess a special affinity for ovarian tissue, thus causing oöphoritis, comparable to the more common mumps-orchitis. Oöphoritis is a more serious consequence of mumps than orchitis, since it leads to peritoneal irritation and hence may cause severe systemic manifestations.

Oöphoritis is a frequent cause of sterility, mainly because it leads to adhesions preventing the migration of ova,

but only rarely does it seriously interfere with the endocrine activity of the ovaries.

Pregnancy and the puerperium appear to predispose to ovarian inflammation. It has frequently been stated that various types of strain (e.g., physical work, cold baths, dancing, alcohol and sexual excesses) may cause oöphor-

itis, especially if the patient is exposed to these stimuli during the menstrual period. This is mere superstition, since we have no proof that such agents could by themselves cause inflammation of the ovary. It is possible however, that they so decrease general resistance as to facilitate infection and subsequent inflammation.

DISEASES OF THE SEXUAL CYCLE

Definition. — For anomalies in the sexual rhythm (oligomenorrhea, polymenorrhea); the abnormalities in the quantity of the menstrual flow (hypomenorrhea, retention of menses, menorrhagia); as well as the phenomenon of midmenstrual bleeding, see : pages 385-398 in the previous chapter on Ovarian Diseases in General. Here we shall limit ourselves to the diseases of puberty, the menopause, the derangements of the normal correlation between ovulation and menstruation (in animals, estrus) and vicarious menstruation.

Diseases of the Pubertal Period. — Irregularities of the sexual cycle often occur during the first months or years after the menarche. The most common among these are the so-called PUBERTY HEMORRHAGES. They are usually due to metropathia hemorrhagica, with excessive and prolonged bleeding, at irregular intervals, from a purely follicular type of endometrium. In most instances, the bleeding is probably not preceded by normal ovulation, but it is incorrect to refer to any uterine hemorrhage as "anovular," merely because progestational proliferation of the endometrium has not occurred. It is quite conceivable that the follicle may rupture and the ovum be discharged, without there being subsequent luteinization of the granulosa with the formation of sufficient corpus luteum hormone to cause progestational uterine changes. In any event, the hemorrhages are due to a disturbance in the normal sequential pro-

duction of folliculoids and luteoids. The endometrium tends to exhibit the characteristics of mild metropathia hemorrhagica, but occasionally signs of progestational proliferation are detectable in endometrial scrapings.

The condition is presumably not merely due to excess folliculoid hormone production in itself, since the bleeding usually ceases under the influence of treatment with folliculoids. The hemorrhages are more probably the result of irregular fluctuations in folliculoid production and the absence of cyclic progesterone secretion. The intervals between flows may be shortened, prolonged or completely irregular.

If the loss of blood is severe, hemostasis may have to be secured by packing of the vagina or curettage. Administration of pitressin tannate in oil or ergot has also been found highly effective in some cases. In general, daily administration of 6 mg. of stilbestrol, 7.5 mg. of estrone sulfate, or of other folliculoids in equivalent doses, stop the bleeding within a few days. This treatment can be continued for 2 or 3 weeks, after which the discontinuation of therapy is followed by the usual withdrawal-bleeding. In some cases the resulting hemorrhage is of alarming severity, but often several cycles of folliculoid therapy eventually restore normal periods. Some gynecologists recommend cyclic progesterone administration. Gonadotrophin therapy is not as consistently successful.

It is well to realize that infrequent bleeding from a follicular-phase endometrium, or even occasional excessive loss of blood during adolescence, is no cause for grave concern; in most instances, these anomalies are readily amenable to the above-mentioned therapeutic measures and frequently they disappear spontaneously. More drastic interventions, such as X-ray treatment or partial resection of the ovaries, are to be avoided.

When puberty occurs before the 9th year of age, we speak of PRECOCIOUS PUBERTY, although this strict delimitation is perhaps somewhat arbitrary. It must be borne in mind that precocious menstruation or precocious development of accessory sex characteristics is not synonymous with precocious puberty. True precocious puberty is a rather rare condition in which normal, potentially fertile, ovarian cycles commence at an abnormally early age. This is accompanied by precocious development of the accessory sex organs and the soma in general.

The term "*precocious pseudopuberty*" is used to designate diseases in which external manifestations of sexual maturity occur while the ovaries remain immature. Folliculomas, adrenal-cortical hyperfunction, pineal and mid-brain lesions may cause precocious pseudopuberty as described in other chapters of this book. In these, the uterine bleeding is usually irregular and accompanied by an excessive or heterosexual development of the accessory sex organs.

True precocious puberty may lead to the commencement of regular, cyclic menstruation even in girls below two years of age, who are healthy and normal except for the precocious assumption of the mature female form (breast growth, development of pubic and axillary hair, precocious ossification of the cartilaginous skeleton, widening of the pelvis, etc.).

Although even precocious pseudopuberty (e.g., that due to granulosa cell tumors) may cause periodic menstrual bleeding from a purely follicular-phase endometrium, premature fertility occurs only with true precocious puberty. In such instances, pregnancy may ensue at an extraordinarily early age and several cases have been described in which girls of 5-12 years became pregnant, and subsequently delivered, normal children.

The primary cause of true precocious puberty is not known, but undoubtedly there must be a premature initiation of normal, cyclic gonadotrophin production, in order to explain the formation of follicles, the discharge of ova and the subsequent corpus luteum formation.

The so-called "overdevelopment-obesity syndrome" (early sexual maturity, obesity, advanced bone age, etc.) may also belong to this group.



Precocious puberty. 4-year-old girl in whom breast development began at three years and soon afterwards uterine bleeding commenced and reappeared regularly in 27-day cycles. There is pubic hair development and great precocity of ossification. Urinary folliculoid elimination was high. The child became bashful and self-conscious. Exploratory laparotomy revealed a mature ovarian follicle but no tumor. Apparently this is a case of true precocious puberty.

(Courtesy of Dr. J. I. Lobo.)



Precocious puberty. Age 8 years. Encephalitis followed by epilepsy at age of 5. Breasts, axillary and pubic hair well developed, sella normal as judged by X-rays, visual fields normal, bone-age about 12 years, urinary FSH 26-53 M.U. /24 hrs.

(Courtesy of Dr. E.-P. McCullagh.)

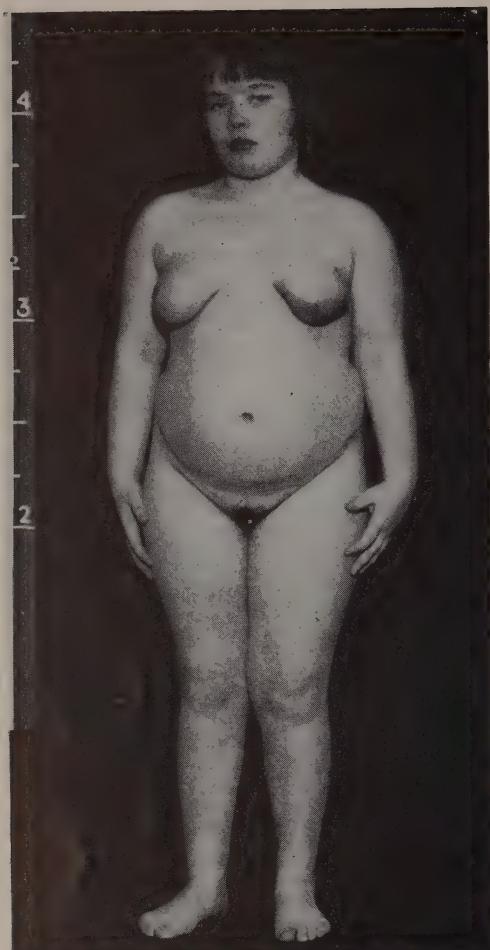
In very young girls with precocious sexual development, exploratory laparotomy is usually justified, in order to determine the cause of the condition. If the case is definitely identified as true precocious puberty, no radical therapy is necessary, but the child must be guarded against excessive self-consciousness and the possibility of sexual violations which are extraordinarily common in such girls, since their body is much more mature than their mind.

It is rather generally, though somewhat arbitrarily, agreed that if the menarche is postponed until after the 17th year of age, we may speak of **DELAYED PUBERTY**. It must be kept in mind, however, that the absence of external signs of menstruation may be due merely to malformations of the genital passages; in that case, other signs of sexual maturation proceed normally and physical examination usually reveals the cause of retention of the menses.



Precocious puberty. Age 15½ years, development of breasts, axillary and pubic hair, vulva, vaginal smears and uterus of adult type. Comparative shortness of limbs, reminiscent of achondroplasia. X-rays reveal no abnormality of sella. Urogram gives no indication of adrenal tumor. Bone-age about 21 years or more.

(Courtesy of Dr. E.-P. McCullagh.)



Precocious puberty due to cerebral lesion. 9-year-old girl with posterior cranium bifidum, hydrocephalus, precocious puberty and diabetes mellitus. Menses appeared at the age of 8 years, breasts began to develop at 7 years. Mental development greatly retarded, ataxia, B.M.R. + 18%, bone age corresponding to approximately 12 years, ventriculogram shows marked enlargement of both lateral ventricles and third ventricle, sella is normal to X-ray.

FSH: less than 13 M.U./24 hrs.

(Courtesy of Drs. R.-W. Schneider and E.-P. McCullagh.)

As previously mentioned, various climatic and dietary factors can retard sexual maturation; a delay in the onset of puberty may merely be one manifestation of the general-adaptation-syndrome to damaging agents. It is also noteworthy that obese individuals usually experience their menarche later than normal. Other common causes of

delayed puberty are hypopituitarism, hypothyroidism or primary under-development of the ovaries themselves. In the latter case, we are usually dealing with primary amenorrhea, that is, normal puberty fails to occur at any time.

A slight delay in the onset of menstruation requires no therapy, while the treatment of considerably delayed puberty is identical with that of primary amenorrhea (see : Ovarian Diseases in General).

Diseases of the Menopause. — The term MENOPAUSE (from the Greek : *menos* = month and *paeuin* = to cease) is used to designate the cessation of menstrual cycles. Some investigators prefer the term "climacteric" (from the Greek : *climakter* = the rung of the ladder) which is defined as the entire period between regular menstruation and its complete cessation. The word "climacteric" is also applicable to the corresponding period in men when sexual functions gradually cease. The average normal menstrual life lasts 33 years and the menopause occurs in women between 45 and 50 years of age. In men, the climacteric is less well-defined and hence more difficult to delimit, but usually it occurs approximately at the same age as in women.

PREMATURE MENOPAUSE may result from surgical removal or destruction of the ovaries by X-rays, radium or local disease. Various debilitating diseases may cause cessation of menses through the repeatedly mentioned decrease in gonadotrophin production, resulting from the general-adaptation-syndrome. Curiously, in the latter cases there are rarely any climacteric disturbances. Hysterectomy brings about a cessation of menstruation but is not necessarily accompanied by immediate signs of ovarian deficiency. In some instances, premature menopause sets in without any obvious cause, often in women whose menarche occurred at an unusually early age, though sometimes there is early menarche with late menopause.

and vice versa. In general, one does not speak of premature menopause unless the menses cease before the 35th year of age.

DELAYED MENOPAUSE is a condition in which normal menstrual cycles continue past the 55th year of age. This is an anomaly, but hardly a disease.

MENOPAUSAL DISTURBANCES are on the border-line of the normal. Certain derangements are manifest in every woman during this critical period of the "change of life," but their exaggeration may lead to severe pathologic conditions.

The *ovary* undergoes involution after the menopause, but there is no strict parallelism between the cessation of menstrual bleeding and that of ovulation. During the climacteric period monthly bleeding may continue, although ovulation has ceased (anovular menstruation). In other instances, ovulation appears to occur in the absence of menstruation (amenstrual ovulation). These derangements are identical with those described (p. 414) under the heading of "Derangements in the Correlation between Ovulation and Menstruation." Progestational transformation of the endometrium, and even pregnancy (a definite proof of ovulation) may ensue after the cessation of normal menses. The discrepancies between ovulation and menstruation are probably due to the fact that during the climacteric, ovarian hormone production may be sufficient to cause uterine bleeding, even though ovulations do not occur and, vice versa, ovulation can continue when the production of ovarian hormones has declined to a level incompatible with normal menses.

Extensive senile involution of the ovary does not occur until several years after the menopause.

The *uterine changes* of the menopausal period are particularly conspicuous. Sometimes menstruation ceases abruptly, while at other times, several periods are missed, but menstruation

recurs at irregular intervals. Frequently, the last periods of flow occur from a follicular-phase endometrium, because folliculoid hormone production continues after progesterone formation has ceased for some time. Eventually, however, both the endometrium and the musculature of the uterus involute as markedly as after complete castration.

The *vagina* atrophies, its lumen shrinks, the rugæ of the walls disappear, vaginal fluid is scanty and the mucosa becomes dry and glistening. Microscopically, the vaginal mucosa is reduced to 4-5 layers of atrophic, nucleated cells containing no glycogen. These changes predispose the mucosa to infection (senile vaginitis) and leukoplakia.

The *urethra* may be involved in the vaginal atrophy and senile urethritis, incontinence or abnormal frequency of micturition may result.

The *oral mucosa* sometimes undergoes an atrophy similar to that of the vagina. This predisposes to ulcerative stomatitis.

The *breasts* become flaccid and pendulous, often due to resorption of fat, while the nipples decrease in size and lose their erectile power. At the same time, there is a tendency towards fibrosis of the breasts, with the formation of minute cysts due to dilatation of the milk ducts.

Severe *psychologic changes* (psychoses, involitional melancholia) are uncommon, but many women suffer from depression, headaches, digestive symptoms and numerous other subjective manifestations, some of which are undoubtedly caused by a sense of inferiority, resulting from the cessation of sexual functions. Libido is largely preserved for several years and normal orgasm may be experienced after cessation of the menses.

The most important manifestations of the menopause are the *vasomotor symptoms*, such as hot flushes involving the neck, head and upper part of the thorax, or less frequently, the entire

body. These are often accompanied by profuse sweating, which tends to follow immediately upon the flushes. The pathogenesis of the vasomotor phenomena is not yet clearly understood, but their immediate cause is certainly a deficiency in ovarian hormones, since similar manifestations can be evoked even at an earlier age by ovariectomy.

Signs of *virilization*, the so-called "menopausal virilism" (deepening of the voice, growth of facial hair, apical baldness, virilization of body contours and facial expression) are rather common among women of the climacteric age. They are probably due to a diminution of ovarian folliculoid formation and perhaps also to increased testoid production by the adrenal cortex.

Climacteric *arthritis* is likewise very prevalent. It usually begins as a chronic synovitis, but later, becomes indistinguishable from hypertrophic arthritis. Its pathogenesis is not known.

The most characteristic *hormonal derangements* during the natural or artificially-produced menopause are: (1) a great decrease in the urinary elimination of *folliculoids*, although traces of such hormones are detectable even several years after cessation of the menses; (2) a greatly increased urinary elimination of *gonadotrophins* (10-50 times the normal amount) with a measurable increase in the gonadotrophin content of the blood; (3) *Pregnandiol* elimination (probably of adrenal origin) may continue, in traces, several years after the menopause; (4) the 17-KS and biologically-active *testoids* in the urine have been claimed to show a temporary increase, but this requires confirmation.

The comparatively common syndromes of mild *menopausal Cushing's syndrome* and *menopausal hyperthyroidism* indicate that the severe endocrine derangements of this critical period of life may involve a variety of

glands of internal secretion in addition to the gonads themselves.

Most women need no *therapy* other than reassurance. In severe cases, folliculoid treatment is highly effective, especially in combatting the vasomotor disturbances and the profuse sweating. Control by vaginal smear is helpful in the evaluation of this treatment. The danger of producing tumors with folliculoids has probably been somewhat overrated. However, it is well to interrupt the treatment from time to time, not only in order to decrease the probability of producing neoplasms, but also to prevent the formation of a cystic hyperplastic endometrium.

Essentially the same therapy is used whether the disturbances are due to spontaneous or artificially-induced (ovariectomy or X-ray castration) menopause.

Derangements in the Correlation between Ovulation and Menstruation or Estrus. — Certain diseases of the sexual cycle are due to a derangement of the normal correlations between production of ova and ovarian hormone secretion. We speak of **AMENSTRUAL OVULATION** when the ovum is discharged, but uterine bleeding fails to occur, or arises from a follicular-phase endometrium. Sexual intercourse may lead to pregnancy, even in women who have been amenorrheic for a long time. This could be explained on the basis of amenstral ovulation; there might be cases with sufficient progesterone production to permit implantation, but with a derangement in the normal cyclic withdrawal-bleeding. However, this etiology is difficult to prove; it is also possible that the act of sexual intercourse in itself has caused ovulation, although previously, in the absence of coitus, neither menstruation nor ovulation had occurred.

Corresponding derangements in animals lead to "anestrous ovulation,"

whose occurrence is much better established by simultaneous histologic study of vaginal and ovarian changes.

ANOVULAR MENSTRUATION is a condition in which no ova are discharged from the ovaries, but true menstrual bleeding occurs from a progestational endometrium. In these cases, apparently the follicles mature and are transformed into active corpora lutea, but the ova remain enclosed in the follicular cavity due to some interference with their discharge ("atretic corpora lutea"). Such ova subsequently degenerate and cannot be fertilized. Some cases of ovarian pregnancy within corpora lutea indicate, however, that in exceptional instances, not-discharged ova can give rise to gestation.

It is incorrect to speak of "anovulatory cycles" when there is neither ovulation nor corpus luteum formation, since then there can be no menstrual cycle but only repeated uterine hemorrhages from a "follicular" endometrium.

Vicarious Menstruation. — We speak of vicarious menstruation when

cyclic hemorrhages occur either simultaneously with, or instead of menstruation, from organs other than the uterus. The nasal, conjunctival and oral mucosæ; the intestines, ears and the mammary glands are sites of predilection for such hemorrhages. Here probably abnormal vasomotor phenomena are conducive to rupture of blood vessels. The alleged liberation of acetylcholine under the influence of folliculoids may have something to do with such a derangement.

The most common type of vicarious menstruation, however, is undoubtedly due to external endometriosis. The transplantation of endometrium onto the ovarian surface or other parts of the peritoneum, its invasion into the umbilicus and sometimes even the urinary passages, readily explains many instances of vicarious menstruation from such sites. The correctness of this interpretation has repeatedly been confirmed by the histologic demonstration of typical endometrial tissue at the site of the bleeding. (See : pp. 467-475.)

OVARIAN TUMORS IN GENERAL

DEFINITION

It is evident from our "definitions" of other endocrine gland tumors that a clear distinction between hyperplasia and tumor formation is always somewhat artificial. This is particularly true of the ovarian neoplasms. It has been said that "a tumor is an autonomous new growth of tissue" (*Ewing*); or that "a tumor proper is a mass of cells, tissues, or organs resembling those normally present, but arranged atypically. It grows at the expense of the organism, without, at the same time, subserving any useful function" (*Adami*).

There are several types of ovarian growths which are not true neoplasms in the usual sense of the word. Yet they represent tissue proliferations, which are comparatively autonomous and sub-

serve no useful purpose. In the present state of our ignorance, the concept of tumorous growth — as that of any other of life's mysteries and even the concept of life itself — is embraced by experience and not by rational delimitation. Hence we shall not enter into a theoretic discussion of this problem, but will include in the present section a number of ovarian growths which have so many characteristics in common that they are best studied conjointly, for mere didactic reasons.

Thus, we shall deal here with growths which are not considered to be true neoplasms (e.g., follicle-retention cysts, corpus luteum cysts), merely because their clinical behavior and their ability to produce hormones make it expedient to review them together with corres-

ponding true ovarian neoplasms. Endometriosis of the ovary is also included in the present section, partly because in its morphology and clinical course it resembles the more orthodox ovarian growths, and partly because we are not yet convinced of its non-neoplastic nature. The extra-ovarian form of endometriosis is reviewed conjointly with the ovarian, in order to avoid duplication of data.

The salient characteristics of hyperfolliculoidism (such as is produced by follicle-retention cysts and folliculomas) and hyperluteoidism (such as is produced by corpus luteum cysts and corpus luteum tumors), will be considered here in detail conjointly with the causative ovarian growths, hence only cursory reference was made to them in connection with other ovarian diseases.

CLASSIFICATION

"Ovarian tumors present a wider scope of structure, greater individual variation and a more complex embryologic and histogenetic basis than those of any other organ, and for these reasons, they have escaped satisfactory classification" (*Ewing*).

The criteria upon which classifications have been based are MORPHOLOGIC (e.g., separation of cystic from solid, epithelial from mesenchymal tumors); EMBRYOLOGIC or HISTOGENETIC (e.g., separation of growths originating from the follicles, interstitial cells, ectopic tissue inclusions) and CLINICAL (e.g., separation of the benign from the malignant, or of hormone-producing from endocrinologically "silent" growths).

Unfortunately, no matter how we formulate our subdivisions, there are always so many intermediate groups and overlappings between different groups, that in most instances a tumor appears to fit into several groups with almost equal justification.

In this book, we shall classify the ovarian tumors according to a system which appears to be most satisfactory from the endocrinologist's point of view:

it is partly based on the morphologic structure, but mainly on the hormone-producing ability of the neoplasms.

PATHOLOGIC ANATOMY

The morphologic features of ovarian tumors are extremely diverse, since a variety of fundamentally different neoplasms may develop in the female gonad (see : sections dealing with specific tumor types). Their size varies from minute microscopic growths to the largest tumors ever seen in man. Sometimes the cystic tumors are heavier than the patient herself and ovarian neoplasms weighing up to 300 pounds have been recorded. These "mammoth tumors" immobilize the patient and may cause severe dyspnea, vascular disturbances (varicose veins, edema of the thighs, "*caput medusæ*") or prolapse of the uterus, merely because of their extraordinary size. Sometimes they are attached to the ovarian surface or the ligaments by long pedicles which permit the tumors to migrate from their original position towards Douglas' space, where they may become impacted, or towards the abdominal cavity, where there is more space for expansion.

Ovarian tumors are often bilateral. Originally benign neoplasms may undergo malignant transformation or degenerative changes such as liquefaction-necrosis, calcification, edema with hemorrhages, etc.

INCIDENCE

The GENERAL INCIDENCE of ovarian tumors (of any type) among gynecologic patients admitted to hospitals for various reasons, is estimated to be about 3%.

The percentual incidence of the various types in a series of more than 1,000 ovarian tumor-bearers (*Bernstein*) was as follows:

Follicle cysts	36
Dermoid cysts	16
Serous papillary cystadenocarcinomas	14
Corpus luteum cysts	11
Pseudomucinous papillary cyst-adenomas	6
Endometriosis of the ovary	4
Serous papillary cystadenomas	3
"Tubo-ovarian cysts"	2.6
Fibromas	2

Pseudomucinous papillary cystadenocarcinomas	1
Solid, medullary and undetermined carcinomas	1
Sarcomas	0.8
Malignant teratomas	0.7
Krukenberg tumors	0.5
Squamous carcinomas in dermoid cysts	0.2
Dysgerminomas	0.2
Carcinosarcomas	0.1

The AGE INCIDENCE of the various ovarian tumor types differs (see : each specific type), but about 40% of all ovarian tumors occur in women 30-40 years of age.

The PREGNANCY INCIDENCE among women, while under observation for various types of ovarian tumors, is estimated to be about 3.5%. Conversely, the ovarian tumor incidence among pregnant women is about 1%. Many investigators believe that pregnancy predisposes to ovarian tumor formation, perhaps due to the pelvic hyperemia, the increased folliculoid hormone production or other metabolic changes accompanying gestation.

PATHOGENESIS

The HISTOGENESIS of ovarian tumors has been touched upon in the chapter on the embryology of the ovaries; it will receive further attention in connection with the various specific tumor types.

Little was known until recently about the FUNCTIONAL PATHOGENESIS of ovarian tumors, that is to say, the factors likely to promote tumor formation in the ovary. Statistical studies suggest that repeated pregnancies and hereditary factors may have a predisposing effect. Experimental findings indicate that ovarian neoplastic disease is related to a breakdown of the normal ovarian-hypophyseal relationship. Follicular and luteal cysts, and hemorrhagic luteal cysts have been reported in animals after extensive partial ovariectomy (see p. 376). Recently, true ovarian neoplasms have been noted when the ovarian control of the hypophysis was deranged by intrasplenic ovarian grafting (see p. 354) in castrates. Here, the ovarian hormones pass through the liver before they reach the general circulation. Hence the ovarian graft is over-stimulated by the uncontrolled hypophysis. There is, in the rat, excessive luteinization of the enormously enlarged ovary, but also production of non-luteinized granulosa cell tumors (*Biskind et al.*) In the guinea pig, cystic growth of mostly hemorrhagic follicles prevails, but later luteomas also develop (*Lipschutz*). In mice these tumors metastasize

and are transplantable (*Li and Gardner; Furth and Sobel*). These tumors in rats and mice are similar to those which can be produced in mice by X-rays (see p. 384).

CLINICAL COURSE

The clinical course of ovarian tumors in general — disregarding the symptoms of the various specific types — is mainly characterized by the manifestations due purely to the physical presence of a growth in the pelvis. The position and shape of the uterus and oviduct can be influenced by pressure or adhesions. Ascites can develop with practically any type of ovarian tumor and is estimated to be present in about 20% of all cases. It is particularly frequent with malignant neoplasms, papillomas and fibromas. Pleural fluid accumulations are present almost only with fibromas.

Mammary gland engorgement, sometimes with tenderness and lactation is rare among patients with ovarian tumors, except with hormone-producing neoplasms.

Fertility is not necessarily influenced even by large bilateral tumors, as long as an adequate amount of normal gonadal tissue remains.

COMPLICATIONS

The clinical course of ovarian tumors is very frequently altered by a variety of complications among which torsion of the pedicle, rupture of cystic tumors with massive hemorrhages, infection of the tumors (especially following perforation to the outside or into the intestinal tract), incarceration within the pelvis (usually due to immobilization by intraligamentous growths or adhesions), the development of hernias (due to increased intra-abdominal pressure), ectopic pregnancy, intestinal obstruction and circulatory disturbances are the most important.

Very large tumors may cause disturbances owing to vertical displacement of the heart, pressure upon the stomach or compression of the urinary bladder and ureters.

Table illustrating our classification of ovarian tumors
 (After H. Selye: "Ovarian Tumors," Encyclopedia of Endocrinology, 1946.)

BENIGN	MALIGNANT
POTENTIALLY ENDOCRINE NEOPLASMS	POTENTIALLY ENDOCRINE NEOPLASMS
<i>Follicle cysts</i> (persistent follicles) <i>Small cystic degeneration</i> <i>Folliculomas</i>	<i>Malignant folliculomas</i> (granulosa cell carcinoma)
<i>Lipid cell tumors</i> Corpus luteum cysts, corpus luteum adenomas, Leydig cell adenomas Adrenal rest adenomas Testoid hyperthecosis	<i>Malignant lipid cell tumors</i> Corpus luteum carcinomas (and sarcomas?) Leydig cell carcinomas Malignant hypernephromas
<i>Tubular adenomas</i> (arrhenoblastomas) <i>Thyroid tumors</i> (struma ovarii)	<i>False Seminomas</i> (dysgerminomas) <i>Chorioneitheliomas</i> (Chorio ⁿ carcinomas)
NON-ENDOCRINE TUMORS	NON-ENDOCRINE TUMORS
PREPONDERANTLY EPITHELIAL NEOPLASMS	PREPONDERANTLY EPITHELIAL NEOPLASMS
<i>Common Cysts</i> Serous Macrocystic Microcystic Tubular adenomas Papillomas Racemose cystomas Peritoneal papillomatosis (Mesonephromas)	<i>Common Carcinomas</i> Serous Primary solid carcinomas Carcinomatous serous cystomas Metastatic serous carcinomas (Malignant mesonephromas)
<i>Pseudomucinous</i> Macrocystic Microcystic Tubular adenomas Papillomas Racemose cystomas Pseudomyxoma peritonei (Brenner tumors)	<i>Pseudomucinous</i> Carcinomatous pseudomucinous cystomas Metastatic pseudomucinous carcinomas (including Krukenberg tumors)
<i>Dermoids and embryomas</i>	<i>Solid teratomas</i> (carcinomatous or sarcomatous dermoids)
<i>Benign nerve tissue tumors</i>	<i>Malignant nerve tissue tumors</i>
NON-EPITHELIAL NEOPLASMS	NON-EPITHELIAL NEOPLASMS
<i>Fibromas</i>	<i>Common sarcomas</i> (spindle and round cell)
<i>Myxomas</i>	<i>Myxosarcomas</i>
<i>Lipomas</i>	<i>Melanosarcomas</i>
<i>Chondromas</i>	<i>Lymphosarcomas</i>
<i>Osteomas</i>	<i>Myelomas and chloromas</i>
<i>Leiomyomas</i>	<i>Liposarcomas</i>
<i>Rhabdomyomas</i>	<i>Chondrosarcomas</i>
<i>Hemangiomas</i>	<i>Osteosarcomas</i>
<i>Lymphangiomas</i>	<i>Leiomyosarcomas</i>
<i>ENDOMETRIOSIS</i>	<i>Rhabdomyosarcomas</i> <i>Hemangiosarcomas</i> <i>Lymphangiosarcomas</i> (<i>Endotheliomas</i>) (<i>Peritheliomas</i>) MALIGNANT ENDOMETRIOSIS

DIAGNOSIS

The diagnosis of ovarian tumors is usually made on the basis of the above-mentioned CLINICAL MANIFESTATIONS in conjunction with the past history. The diagnosis of accompanying hormonal disturbances will be discussed with the individual tumor types most likely to produce them.

Irrespective of possible endocrine derangements, the diagnosis rests chiefly upon PALPATION of the tumor region. Whenever possible, it should be performed bimanually after evacuation of the bladder and rectum, the patient being placed in a position suitable for palpation of the particular region in which the tumor is felt. Sometimes palpation may have to be performed under light anesthesia or, in the event of considerable ascites, after evacuation of the free peritoneal fluid by tapping.

X-RAY examination is especially useful in calcified tumors, embryomas and dermoids in which concretions, teeth and bones may be visible. The definite diagnosis, however, is usually possible only after morphologic examination of the tumor tissue itself, obtained by EXPLORATORY COLPOTOMY OR LAPAROTOMY.

Differential Diagnosis. — The most common source of error in the diagnosis of ovarian and para-ovarian tumors are INFLAMMATORY PROCESSES (e.g., salpingitis, perinephritis, pelvic abscesses), HYDROSALPINX (past history, usually bilateral tender tumors); ectopic or normal PREGNANCIES, (gonadotrophins in urine) and UTERINE FIBROIDS (connection with uterus, menorrhagias, frequent association with submucous fibroids). Tumors of the intestinal tract, "fecal tumors," a distended bladder, ascites or general obesity are less likely to be confused with ovarian tumors. In general, it is helpful in the diagnosis of large ovarian tumors, to remember as likely possibilities : "The Six "F-s" (Fat, Fetus, Fluid, Feces, Flatus, Fibroids).

PROGNOSIS

The prognosis of the various ovarian tumor types depends largely upon their benign or malignant nature, and in the latter case, upon their tendency to infiltrate and metastasize. In any case, if no treatment is applied, the course of true ovarian neoplasms is progressive. Spontaneous regression (e.g., rupture and subsequent involution of cysts, torsion with self-amputation and absorption of neoplasms) is extremely rare. If complete surgical removal is possible, the prognosis of most ovarian tumors is favorable, while X-ray or radium treatment rarely lead to permanent cures.

THERAPY

In spite of many enthusiastic statements concerning other types of treatment, the best therapy for any type of ovarian tumor is still its complete SURGICAL removal, whenever technically feasible. The operation should be performed as soon as possible, since it is almost never certain that the growth is not malignant or at least in the process of malignant transformation. It should be remembered, furthermore, that by postponing the operation the likelihood of complications (adhesions, hemorrhage, torsion, etc.) is increased. Of course, if a woman is in labor when the ovarian tumor is diagnosed, its removal may be postponed as long as the neoplasm does not mechanically interfere with delivery. During the first three months of gestation, ovariectomy entails the danger of abortion and if the tumor is not malignant, it may be advisable to postpone intervention until after delivery. Generally speaking, ovarian tumors should be removed by the abdominal and not by the vaginal route.

X-RAY and RADIUM therapy should be reserved for cases in which radical removal is technically impossible, because of widespread infiltrations and adhesions, metastases or great debility of the patient.

PARA-OVARIAN TUMORS IN GENERAL

In this book, the designation "para-ovarian tumors" is used to include all neoplasms situated in the immediate vicinity of the ovary, within its ligaments.

Many of these growths are described in the literature under such terms as tumors of the broad ligament, Wolffian body, parovarium (epooophoron), paroöphoron, rete ovarii, pronephric remnants, Müllerian or Wolffian duct remnants, Gartner's duct remnants, etc. The most common designations are "parovarian tumors" and "broad ligament tumors." While many, if not all, the embryologic vestiges mentioned above may be the site of tumor formation, it is very questionable whether in the majority of the tumors described in the literature, the exact derivation from any one of these cell types could be proven with certainty.

The non-committal, rather all-inclusive term of para-ovarian tumors (which only implies that the neoplasm or cyst is in the immediate vicinity of the ovary), is therefore recommended for all those relevant blastomas in which the histogenetic mechanism is in doubt. It is particularly objectionable to use the term "parovarian" as synonymous with "para-ovarian" neoplasm, since the parovarium is a well defined morpho-

logic structure derived from the Wolffian body and no neoplasm should be described as parovarian unless it demonstrably originates in this organ.

Although the histogenesis of para-ovarian tumors is often difficult to prove, the following types are theoretically possible :

A. PRIMARY PARA-OVARIAN TUMORS :

- (1) Hydatid of Morgagni (pronephric?).
- (2) Kobelt's tubules (pronephric?).
- (3) The Wolffian body, also known as parovarium, epooophoron or organ of Rosenmüller (mesonephric).
- (4) The Wolffian duct (cranial part of mesonephric duct).
- (5) Gartner's duct (caudal, usually obliterated, part of mesonephric duct).
- (6) The junctional or rete tubules (usually obliterated connecting tubules between ovary and parovarium).
- (7) The paroöphoron (mesonephric).
- (8) Accessory Fallopian tubes (Müllerian).
- (9) Diverticula of the Fallopian tubes (Müllerian).
- (10) Independent tumors of the connective tissue elements in the broad ligaments (e.g., fibromas, lipomas, sarcomas, lymphangiomas).
- (11) Tumors of accessory adrenal-cortical tissue.

B. SECONDARY PARA-OVARIAN TUMORS :

- (1) Metastases from distant malignant neoplasms.
- (2) Transplants of self-amputated benign neoplasms (usually of ovarian origin).
- (3) Endometriosis (uterine mucosa transplants).
- (4) Endosalpingiosis (tubal mucosa transplants).

For a discussion of the embryologic structures mentioned above, the reader is referred to the brief summary in this book (see : "Embryology") and to textbooks of Embryology.

The para-ovarian tumors are not sufficiently common nor of such endocrinologic interest as to justify a more lengthy description here.

HYPERFOLLICULOIDISM ASSOCIATED WITH OVARIAN GROWTHS (FOLLICLE CYSTS, SMALL-CYSTIC DEGENERATION OF THE OVARIES, FOLLICULOMAS)

DEFINITION

Hyperfolliculoidism in its purest form, is produced by various ovarian growths; only some of these are true neoplasms, but all of them will be discussed conjointly in this chapter because they represent closely allied conditions from the endocrinologist's viewpoint, although morphologically they are unrelated.

The principal relevant conditions are the following :

- (1) FOLLICLE CYSTS (simple cysts of the follicles, retention cysts, persistent follicles, hydrops of the follicles) are enlarged and usually abnormally persistent Graafian follicles. There are transitional types between multiple follicle cysts and so-called "small-cystic degeneration of the ovaries."

- (2) SMALL-CYSTIC DEGENERATION OF THE OVARIES (sclerocystic ovaries, polycystic ovaries, fibrocystic ovaries, oöphoritis follicularis, follicular hypertrophy of Ziegler, "ovaire à petits kysts de Trélat") is a condition in which the connective tissue stroma of the ovary and its capsule is increased and many small follicle cysts develop throughout the glandular parenchyme. In contradistinction multiple, simple follicle cysts tend to develop in the ovarian cortex and are usually unaccompanied by any marked degree of fibrosis. Corpus luteum formation is deficient in both these conditions and both types may lead to the same clinical picture of hyperfolliculoidism.
- (3) OVARIAN FOLLICULOMAS (granulosa cell tumors, basal cell tumors of the ovary, Kahlden's tumors, thecoma, fibroma thecocellulare xanthomatodes ovarii, Löffler-Priesel tumor) are true tumors of the ovarian follicle, which may produce excessive amounts of folliculoids. It has been customary to distinguish sharply between granulosa and theca cell tumors on the basis of purely morphologic characteristics. It is true that some folliculomas resemble the granulosa and others the theca of a normal follicle, but mostly there are granulosa and theca elements, as well as transition forms between the two in the same tumor.

PATHOLOGIC ANATOMY

OVARIAN FOLLICLE CYSTS rarely possess a diameter of more than 5 cm. They are lined by a histologically characteristic granulosa layer, which is surrounded by the theca interna. Occasionally, pressure atrophy may affect certain areas of the lining, but widespread atresia of the granulosa is rare.

SMALL-CYSTIC DEGENERATION leads to ovarian enlargement. The albuginea

is almost invariably greatly thickened and scar-like; the whitish-grey surface is smooth and has a fascia-like appearance. The cysts are scattered throughout the parenchyme and do not tend to protrude beyond the surface level. They represent persistent Graafian follicles. There is little evidence of any inflammatory change in the sclerotic stroma. Corpora lutea are usually absent.

OVARIAN FOLLICULOMAS vary from a few mm. to several inches in diameter. They can be solid or cystic and are occasionally bilateral.

Microscopically, the structure of folliculomas is extraordinarily variable. They can consist predominantly of granulosa or of theca-like cells, and sometimes imitate immature testicular trabeculae and tubules.

The term "folliculome lipidique de Lécène" has been employed to describe folliculomas in which the granulosa-like cells contain many lipid granules. They resemble corpus luteum tumors (luteomas) although their cells are usually smaller.

Functionally, these tumors certainly belong to the folliculomas, since they produce cystic-glandular endometrial changes.

Although many transitional types are known, the following are the principal histologic forms of the folliculomas :

- (1) Cystic; (a) microcystic, (b) macrocystic.
- (2) Diffuse.
- (3) Tubular.
- (4) Trabecular (often "gyriform").
- (5) Papillomatous.
- (6) Carcinomatous.
- (7) Theca cell type.
- (8) Sarcomatoid type.
- (9) Mixed folliculomas (in which no constituent is particularly predominant).
- (10) Various types of folliculomas combined with other ovarian tumors (e.g., teratomas, cystic adenomas).

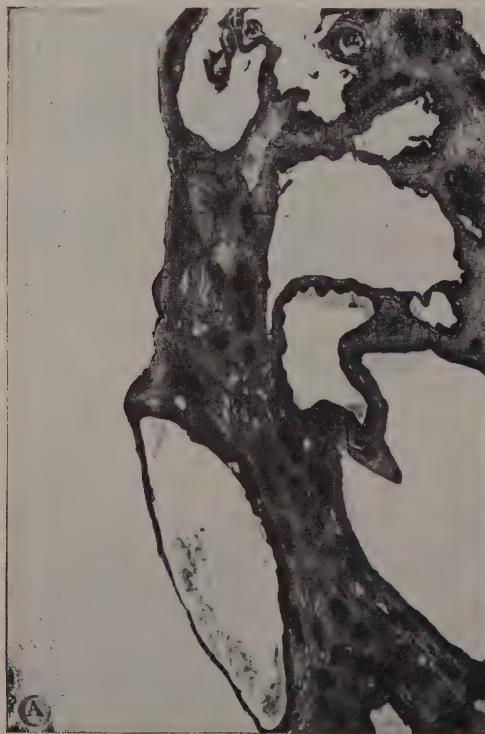


Polymorph granulosa-cell tumor. Within the same field we see tubular (center), diffuse (right) and microcystic (left and upper sides) cell arrangements.

(After H. Selye: "Ovarian Tumors," Encyclopedia of Endocrinology, 1946.)

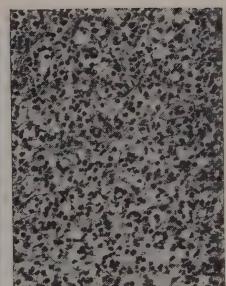
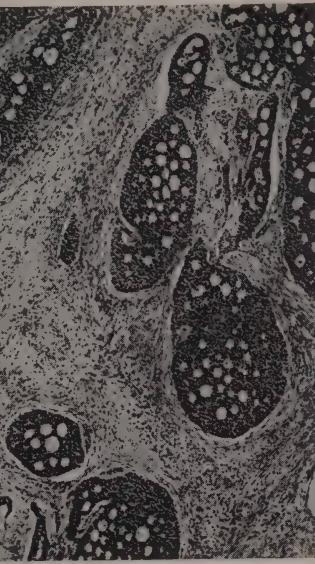
"Gyriform" granulosa-cell tumor. High magnification of characteristic "pseudotubular" cell arrangement, columns consist of single cell layers arranged around a central axis without lumen.

(Courtesy of Dr. P. Masson.)



Cystic granulosa-cell tumor. — A. Folliculoma with comparatively large cystic cavities. — B. Metropathia hemorrhagica (typical "Swiss Cheese" aspect) in patient whose cystic folliculoma is illustrated in Fig. A.

(Courtesy of Dr. P. Masson.)



Folliculoid granulosa cell tumor. — A. Characteristic folliculoid pattern in a prevailingly epithelial field. Some small cavities contain coagulated fluid remotely resembling degenerating ova. — B. Another field of tumor shown in Fig. A.; here epithelial and stroma elements are about equally represented.

(Courtesy of Dr. H.-T. Karsner.)

"Folliculome lipidique" of Lecene. Partially-luteinized region in a granulosa-cell tumor. Histologically this type is difficult to differentiate from luteomas; indeed, it may represent a transition to the luteoma.

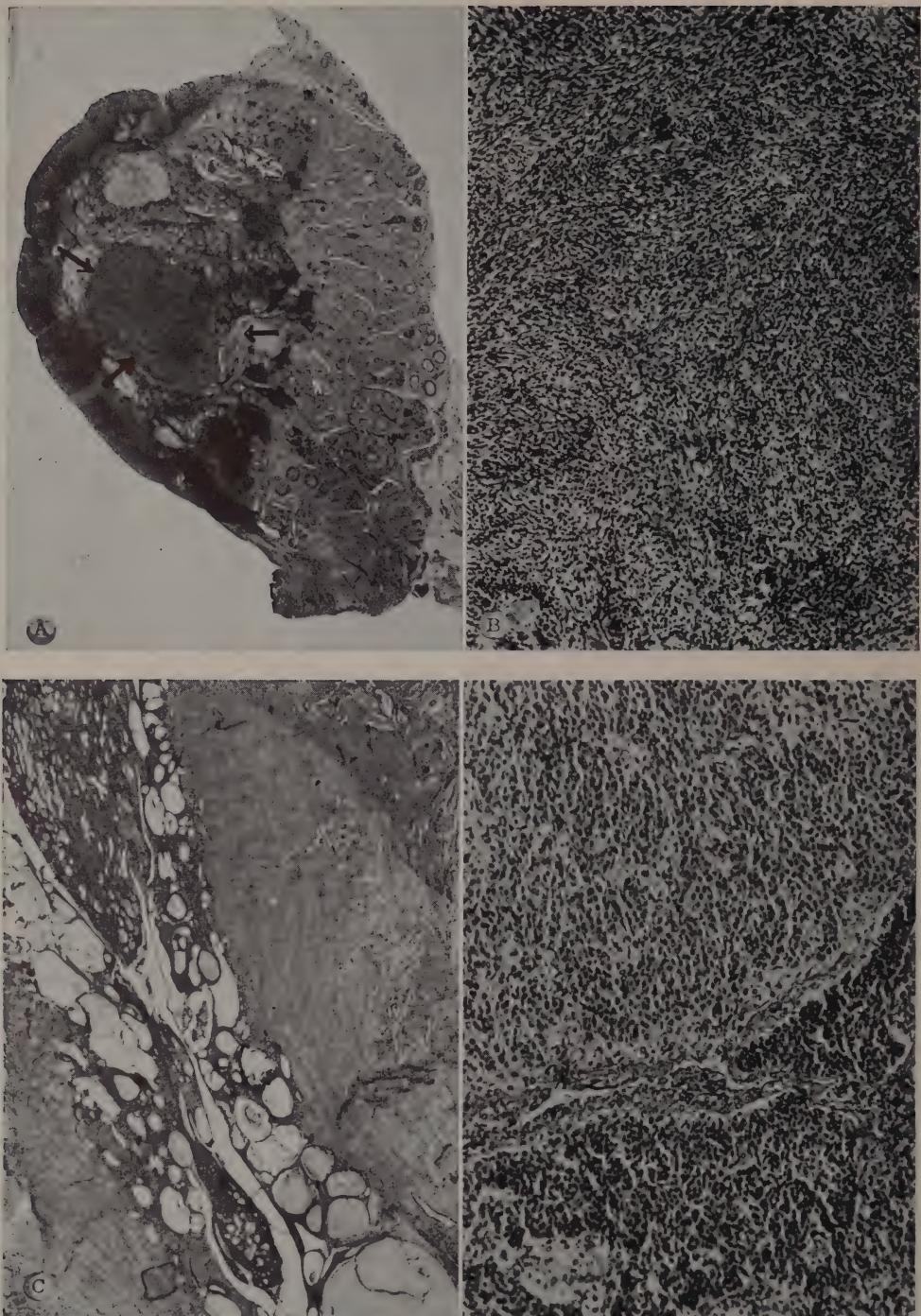
(Courtesy of Dr. H.-T. Karsner.)



"Gyriform" granulosa-cell tumor. Note the characteristic gyriform arrangement of the granulosa cells. The epithelial columns are almost invariably two cells deep and thus resemble narrow tubules without central lumen.

(Courtesy of Dr. L.-C. Simard.)

Hyalinized thecoma with epithelioid cells. This roundish tumor (about 2 inches in diameter) exhibited the same pattern throughout. Dense fibrous-tissue stroma with irregular islets of "epithelioid" cells. The 42-year-old patient suffered from polymenorrhea (bleeding every 18 days) and hypermenorrhea (duration of bleeding 8 days). Epithelioid cells were rich in fat granules. (Courtesy of Dr. L. Berger.)



Thecoma. — A. General view of a cross-section through ovary with small thecoma in the middle of the organ. 66-year-old patient suffered from slight postmenopausal metrorrhagia (with passing of blood clots). — B. Higher magnification of a small area in same tumor. Note partly "epithelioid" aspect of the cells (lower part of field) among spindle-shaped theca-cells. — C. Metropathia hemorragica. "Swiss cheese" endometrium, small polypoid excrescences and some fairly large endometrial cysts in same patient. (After H. Selye: "Ovarian Tumors," Encyclopedia of Endocrinology, 1946.)

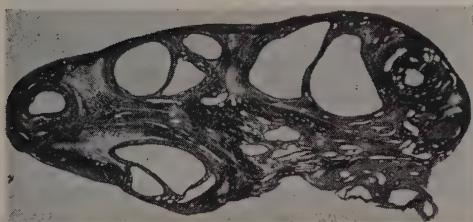
Partially luteinized granulosa cell tumor. Typical diffusely arranged granulosa cells (lower field) and partially luteinized cells (upper field) are seen side by side. 54-year-old patient who never underwent a typical menopause but had irregular menses which lasted 15 to 21 days as a rule.

(Courtesy of Dr. L. Berger.)



Small-cystic degeneration of the ovaries. — **A.** Naked eye appearance of ovarian surface in a case of pronounced small-cystic degeneration. Note the smooth surface devoid of protruding cysts. The patient was 16 years old and suffered from a hydrocephalus internus due to a glioma. — **B.** Cross-section through the ovaries represented in Fig. A. Note the numerous small cystic cavities, the dense connective tissue stroma and the thickened capsule.

(After E.-J. Kraus: Arch. f. Gynäk. 152, 383, 1933.)



Small-cystic degeneration of the ovaries. Very low magnification of a histologic section through the ovary, showing cystic cavities of varying size, dense connective tissue stroma and thick capsule. The patient was 20 years old and suffered from a glioma of the brain.

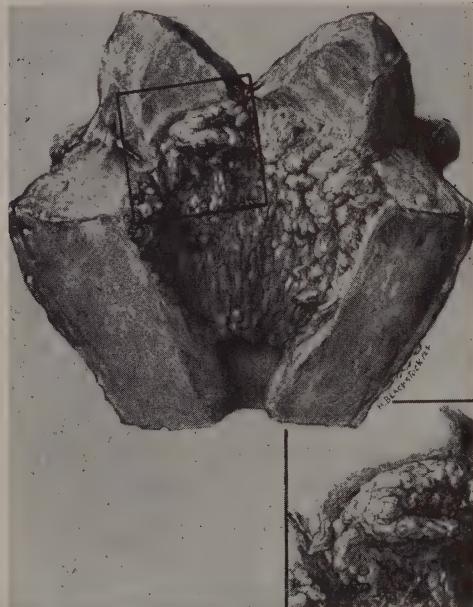
(After E.-J. Kraus: Arch. f. Gynäk. 152, 383, 1933.)

Malignant folliculomas may metastasize into lymph nodes, uterus, bones, etc.

INCIDENCE

Ovarian FOLLICLE CYSTS are very common, but since only few of them are removed for histologic study, the exact incidence is difficult to estimate. However, the vast majority of all cases of metropathia hemorrhagica (one of the most common disorders found in gynecologic practice) are undoubtedly due to follicular cysts.

Follicle cysts can occur in newborn infants, in whom they are probably caused by "synkainogenesis," the tran-



Gross appearance of benign polypoid hyperplasia of endometrium. Note physiologic line of demarcation at the internal os. The area in the square is shown under higher magnification in lower right corner.

(After E. Novak: Textbook of Gynecology, Williams & Wilkins, 1944.)

sit of maternal (in this case gonadotrophic) hormones to the embryo.

The occasional development of follicle cysts during pregnancy and under

the influence of chorionepitheliomas will be discussed elsewhere.

METROPATHIA HEMORRHAGICA and the causative follicle cysts have a double-peaked age-incidence curve, with a first, small maximum at or immediately after puberty, followed by a minimum between 31-35 years and a second, very high maximum at or after the menopause, between 45 and 65 years. It is very probable that disturbances in the hormonal interrelations between the pituitary and the ovary are responsible for the high incidence of these cysts (and the resulting metropathia) at the onset and cessation of normal menstrual life.

SMALL-CYSTIC DEGENERATION OF THE OVARIES also belongs to the more common gynecologic diseases, although here again the exact incidence is difficult to calculate. Supposedly relevant cases have been described in children, but the condition usually occurs only in adults.

OVARIAN FOLLICULOMAS are comparatively rare, but not as exceptional as had been thought. It is now estimated that about 10% of all primary ovarian carcinomas belong to the malignant granulosa-cell-tumor type. Benign granulosa cell tumors and thecomas are less common.

Folliculomas may occur at any age, but the vast majority develop prior to or during the menopause and about $\frac{1}{3}$ of the cases are seen after the menopause.

PATHOGENESIS

OVARIAN FOLLICLE CYSTS merely represent persistent, but otherwise normal, Graafian follicles. They are probably due to a derangement of the normal sequence in the elaboration of FSH and LH. This view is supported by the fact that metropathia hemorrhagica occurs most frequently at the beginning and end of normal menstrual life as well as



Small-cystic degeneration of ovaries. Appearance of typical, small-cystic ovaries as seen at operation. Note great enlargement of the glands, shiny tendon-like surface. The cysts are all within the parenchyme and do not bulge out through the capsule.

(Courtesy of Drs. R.-W. Schneider and E.-P. McCullagh.)

in women who have had many children. Presumably, under these conditions FSH formation tends to be excessive and continuous while LH is produced in subnormal amounts.

METROPATHIA HEMORRHAGICA is due to the persistent action of folliculoids upon the endometrium. It is independent of the nature of the underlying ovarian lesion as long as the latter occasions an excessive or prolonged production of folliculoids. The manifestations of metropathia hemorrhagica produced by endogenous folliculoids, are identical with those seen in castrate women following continuous, excessive folliculoid hormone treatment. In this connection, it is of interest that partial

ovariectomy frequently causes persistent follicle-cyst formation in the remaining ovarian tissue; this also leads to metropathia hemorrhagica, due to hyperfolliculoidism. (See : p. 376.)

SMALL-CYSTIC DEGENERATION OF THE OVARIES has been ascribed to various types of inflammatory lesions conducive to an excessive development of connective tissue, especially in the ovarian capsule. It is thought that this capsular fibrosis may mechanically interfere with follicle rupture and thus cause the persistence of follicles.

OVARIAN FOLLICULOMAS could perhaps arise from "Walthard's cell nests" (granulosa-like formations originating in the germinal epithelium), or from mature granulosa and theca cells.

As regards the functional pathogenesis of folliculomas, the high incidence at, or just prior to the menopause, and the occasional occurrence of theca-adenomas during pregnancy, imply that pronounced changes in the hormonal equilibrium (increased gonadotrophin production ?), such as occur at these times, play an etiologic rôle.

It is of interest that folliculomas have been produced by X-ray treatment in a certain strain of mice. These tumors appear to develop from surviving granulosa cells and are true, transplantable neoplasms. They produce folliculoids as judged by the resulting estrous changes and cystic hyperplasia of the endometrium. (See : p. 384.)

Racial factors presumably are also of importance in the development of folliculomas. In women, these growths tend to occur in several members of the same family and certain strains of mice have been observed to possess a special tendency to develop spontaneous, transplantable, malignant papillomatous granulosa cell tumors which produce signs of hyperfolliculoidism.

CLINICAL COURSE

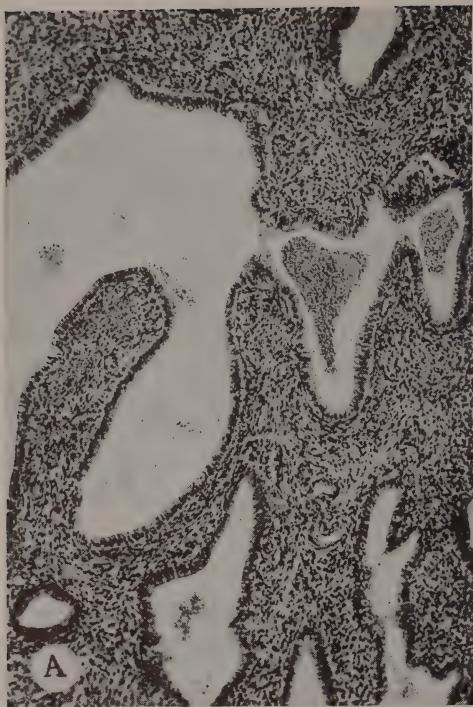
The most prominent clinical manifestation of all types of ovarian growths accompanied by hyperfolliculoidism (follicle cysts, small-cystic degeneration, folliculomas), is the development of METROPATHIA HEMORRHAGICA (synonyms : cystic-glandular hyperplasia or "Swiss cheese" endometrium, endometrial hyperplasia, functional uterine bleeding, polypoid hyperplasia of the endometrium, etc.). This is a condition in which the *endometrial changes characteristic of the follicular phase of the menstrual cycle* are excessively developed. Usually, the total height of the endometrium is far above normal, many glands are cystically dilated ("Swiss cheese" appearance), mitotic figures in the epithelial cells are plentiful and there is a great tendency to irregular, more or less continuous, uterine bleeding. Not infrequently, the uterus also shows other changes presumably due to hyperfolliculoidism, such as endometrial polyps, epithelial metaplasia, fibroids or endometriosis. (See also : Endometriosis on p. 467.)

Among the clinical manifestations of metropathia hemorrhagica, persistent *uterine bleeding* (rarely accompanied by pain), *sterility* and a *follicular-phase type of vaginal smear* are especially characteristic.

Postmenopausal bleeding is very common in older women with hyperfolliculoidism. It frequently gives rise to confusion with uterine carcinomas.

Pregestational changes are absent in typical instances of metropathia hemorrhagica, since corpus luteum formation is suspended.

Metropathia hemorrhagica may eventually lead to *uterine cancer*. Although this is uncommon, endometrial carcinomas appear to be somewhat more frequent with various types of hyperfolliculoidism than in otherwise

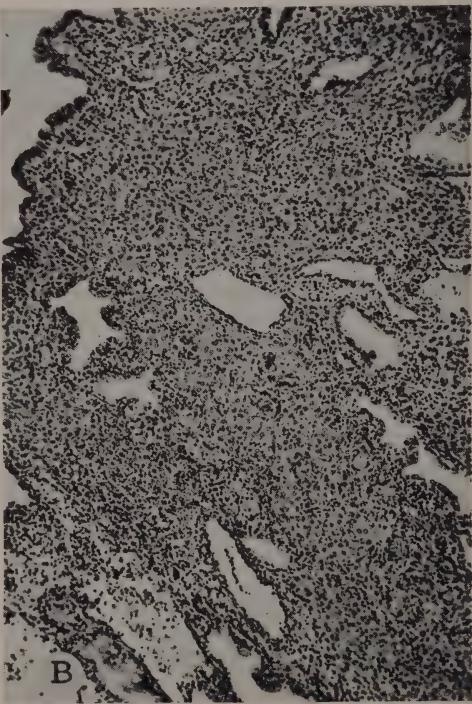


A

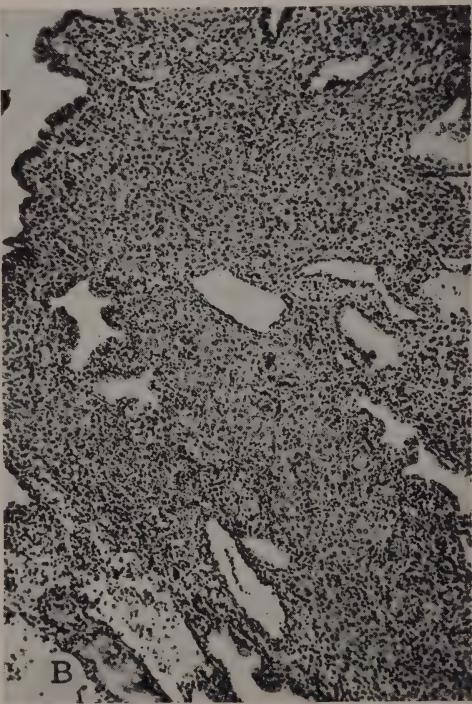


B

Metropathia hemorrhagica. — A. Specimen obtained by curettage for profuse uterine bleeding. Note widely dilated cystic glandular spaces, some filled with blood. — B. Recurrence of metropathia in same patient, following discontinuation of successful progesterone therapy.



A



B

Sterility treated with progesterone and LH. — A. 9th day of luteal phase after 7 daily injections of 10 mg. of progesterone. Note cystic glandular endometrium, which here did not respond well to progesterone. — B. Same patient on 12th day of luteal phase, after 12 daily injections of 1000 I.U. of LH. Note marked progestational (decidual) proliferation, apparently mediated by the patient's own ovary.

(Courtesy of Drs. G.-S. Henry and J.-S.-L. Browne.)



Precocious pseudopuberty due to granulosa cell tumor. — A. 4-year-old girl exhibiting precocious genital development. Menstruation began at age of 3 years. Granulosa cell cyst of ovary found. — B. Bone age $8\frac{1}{2}$ years (although actual age 4 years). Accelerated somatic development due to granulosa cell cyst.

(Courtesy of Dr. R.-C. Grauer.)

normal women. This is of interest in view of the carcinogenic effect of folliculoids.

Oligomenorrhea or amenorrhea are rare in women with persistent follicle cysts, small-cystic degeneration or ovarian folliculomas.

In children, follicle cysts or folliculomas often cause precocious PSEUDOPUBERTY, with the occurrence of uterine bleeding (not menstruation, since

there is no progestational phase), premature development of the breasts, pubic hair, clitoris, labia, adult female body contours, and sexual drive.

NERVOUS MANIFESTATIONS are especially common in women suffering from hyperfolliculoidism (irrespective of the underlying ovarian lesion). Among these, excessive libido (nymphomania) is particularly prominent. Hysteria, gastrointestinal spasms, ovaralgia (pain in the ovarian region), and what the French investigators designate as "toux utérine" or "uterine cough" may perhaps also be related to certain types of hyperfolliculoidism, but a causal connection with the hormonal derangement is less clearly demonstrable as

regards these manifestations. Pigmentation of the skin, "chloasma uterinum," is not very frequent in hyperfolliculoidism.

Other clinical manifestations are chiefly due to the mere mechanical presence of the ovarian neoplasm (see: Ovarian Tumors in General on p. 417).

DIAGNOSIS

In addition to the LOCAL SIGNS characteristic of an ovarian tumor (see: Ovarian Tumors, p. 419), the growths conducive to hyperfolliculoidism manifest themselves by the specific signs of hormone overdosage (precocious pseudopuberty, metropathia hemorrhagica, postmenopausal bleeding, increased folliculoid and decreased gonadotrophin elimination in the urine).

From the differential diagnostic point of view, the ADRENOGENITAL SYNDROME deserves attention. It will be remembered, that unlike folliculomas,



A



B



C



D

Extreme hirsutism with sclerocystic ovaries. — A, B, C and D. 28-year-old woman, height 64", weight 211 lbs., who suffered from obesity, amenorrhea, endometrial polyps, multiple bilateral follicle cysts, and extreme hirsutism. There was no evidence of adrenal tumor at surgical exploration.

(Courtesy of Dr. E.-P. McCullagh.)

adrenal-cortical tumors tend to produce virilism accompanied by increased 17-KS excretion.

In postmenopausal women, continuous bleeding must always raise the question of a possible UTERINE CARCINOMA, POLYPS, submucous FIBROMA, SENILE ENDOMETRITIS, etc. The diagnosis of these conditions is facilitated by endometrial biopsies, hystero-salpingography and the general clinical history.

It is also noteworthy that hyperfolliculoidism caused by any type of ovarian growth leads to the same clinical symptomatology, and hence the differential diagnosis between ovarian folliculomas, persistent follicle cysts, and small-cystic degeneration may not be possible without an exploratory laparotomy. Very atypical granulosa cell carcinomas often cause no endocrine disturbance because their cells are too undifferentiated to elaborate hormones.

Differentiation of the ovarian growths which produce hyperfolliculoidism from ectopic or normal PREGNANCY (increased gonadotrophin excretion), and APPENDICITIS is rarely difficult.

Metropathia hemorrhagica is frequently confused with CARCINOMA OF THE UTERUS, UTERINE POLYPS, INCOMPLETE ABORTION, SUBMUCOUS MYOMAS, etc., but here again endometrial biopsies and hormone analyses of blood and urine usually permit a definite diagnosis.

SYSTEMIC DISEASES CONDUCIVE TO BLEEDING (e.g., blood dyscrasias, hemophilia) should also be considered.

PROGNOSIS

The OVARIAN FOLLICLE CYSTS, especially those occurring at puberty, often regress spontaneously. In the adult, the prognosis is also favorable, although occasionally, intractable bleeding may result in fatal anemia unless

suitable therapy is instituted. Even in adults, spontaneous cures do occur. Approximately the same may be said about the prognosis of metropathia hemorrhagica as such, whether due to follicle cysts or small-cystic degeneration of the ovaries.

OVARIAN FOLLICULOMAS show no tendency to regress spontaneously. The rate of their growth and development is very variable. The highly differentiated granulosa and theca cell tumors grow slowly and have little tendency to metastasize, while malignant granulosa carcinomas and theca sarcomas grow rapidly, metastasize and infiltrate neighboring organs. Following the extirpation of malignant folliculomas, survivals of 10 to 20 years have repeatedly been reported.

THERAPY

Follicle cysts. — In children, after removal of individual persistent follicle cysts, the accessory sex organs return to the infantile state; in adults, the normal menstrual rhythm reappears in the majority of the cases. Puncturing of the follicles and partial ovariectomy have occasionally been reported as bringing temporary relief, but are less satisfactory. Complete OVARIECTOMY is unjustified, except in menopausal cases. X-RAY treatment may be beneficial, but is indicated chiefly in postmenopausal cases, since in young women the danger of subsequent sterility is too great.

With comparatively small persistent follicles, the cyclic administration of LUTEOID HORMONES should be attempted; sometimes it may even lead to a permanent cure.

Metropathia hemorrhagica. — The metropathic endometrium should first be removed by CURETTAGE. Usually, this in itself brings only temporary relief, but it is necessary for diagnostic

purposes and increases the efficacy of subsequent endocrine therapy. It also diminishes the likelihood of the otherwise often alarming, profuse withdrawal-bleeding which tends to occur in these patients after a course of ovarian hormone injections. Sometimes however, even mere curettage effects a permanent cure.

HISTERECTOMY or "DEFUNDATION OF THE UTERUS" should not be performed to check metropathia hemorrhagica except where there are intractable post-menopausal hemorrhages, or when the possibility of cancer can not be ruled out.

PARTIAL RESECTION OF THE OVARY has been practiced in order to remove the source of excess folliculoids. In the light of contemporary knowledge, this intervention is not advisable, since we know that even in normal individuals, the ovarian remnant tends to become cystic after partial ovariectomy.

X-RAY treatment of the pituitary has been suggested as a means of inhibiting hypophyseal hormone production and its effect upon the ovary. Since the optimal dosage is difficult to determine, this therapy is fraught with the danger of eliciting permanent damage. X-ray castration on the other hand is not dangerous, and yields almost 100% cures, hence this technic is highly advisable if the patient is past the menopause or if for other reasons, her future sterility is not a deterring factor.

The RADIUM TREATMENT of metropathia hemorrhagica is less popular, although it has been claimed that with intra-uterine application of 200-400 mg. hours there is a satisfactory reduction of the bleeding without premature menopause in 80% of the adult patients.

The method of choice is ENDOCRINE THERAPY. In young girls, whenever possible, this should be given without

preliminary curettage, since such operations often cause severe mental upset at this age. In adult women, on the other hand, it is advisable to remove the pathologic endometrium. This precaution is particularly indicated whenever polypoid excrescences assume large dimensions. If curettage is to be avoided, it may be well to give *testoids* for a short period with the view of obtaining involution of the endometrium instead of its mechanical removal. It should be kept in mind, however, that some prominent gynecologists are strongly opposed to the use of *testoids* in females for any reason. In any case, such therapy must be limited to the absolutely necessary minimum (about 5 mg./day), in order to avoid virilization.

The most advisable procedure is to administer *progesterone* in repeated courses, each consisting of four doses of 5-10 mg. given every other day, in order to substitute for the lack of endogenous corpus luteum hormone production. These *progesterone* treatment periods must be separated by about 18 days of rest, during which the endogenous folliculoids have time to develop a proliferative endometrium. It is true that even this therapy is merely symptomatic, since it deals only with the result of the ovarian derangement. However, for reasons which are not yet quite clear, in many instances intermittent *progesterone* therapy re-establishes the normal interrelations between the ovary and pituitary and thus results in a more or less permanent cure.

"Metropathia non-hemorrhagica" that is, cystic-glandular hyperplasia of the endometrium with amenorrhea, is less common and probably due to chronic folliculoid hormone action of a very even degree. Unlike in amenorrhea due to hypofolliculoidism, bleeding may be obtained by treatment with *progesterone* alone. Alternate folliculoid and

luteoid treatment has also been recommended. 5.0 mg. of stilbestrol daily for 10 days supplemented with 5.0 mg. of progesterone during the last 5 days is usually followed by uterine bleeding within 3-4 days after discontinuation of treatment. This procedure may be repeated once monthly to imitate a cycle, but often spontaneous menses are at least temporarily re-established.

Short treatment with very high doses of *folliculoids* — the so-called "folliculin shock treatment" — may also be attempted but has not yet been tried on a large scale.

A truly rational therapy will only be possible when the various *gonadotrophins* become available in a pure form. In the past, these have not given very encouraging results. More recently, it has been claimed that *luteotrophin* is beneficial in "functional bleeding."

Ovarian Small-Cystic Degeneration. — In most cases of small-cystic degeneration of the ovaries, the condition is resistant to all efforts of internal therapy.

Complete **BILATERAL OVARIECTOMY** was the treatment of choice towards the end of the last century, but except in postmenopausal women, such a radical intervention is hardly ever advisable.

IGNIPUNCTURE of the follicles or **LIGATION OF THE OVARIAN ARTERY** may give at least temporary relief in some cases, but the method of choice is **PARTIAL RESECTION** or **DECORTICATION** of the ovary. For this, a variety of technics

have been devised, among which bilateral wedge-excision, with subsequent union of the ovarian wound by suture, is generally considered to be the best. Another modification of this procedure consists in turning the ovary inside out after the wedge-excision, and stabilizing it in this position so as to bury the sclerotic serosal surface and expose the presumably normal follicles of the medulla.

The ovariangia which frequently accompanies small-cystic degeneration can often be completely cured by bilateral **DENERVATION OF THE OVARIES** and this intervention may also have a beneficial effect upon the ovarian lesion itself.

Among other therapeutic methods, **X-RAY** or **RADIUM** treatment of the ovaries have been claimed to be effective. In view of the danger of destroying too much ovarian tissue however, it appears inadvisable to resort to such drastic means as a routine procedure. Considering our deep ignorance of the underlying endocrine disturbance, it is not surprising that **HORMONAL THERAPY** has so far proved rather ineffective.

Ovarian Folliculomas. — Here, whenever possible, the therapy of choice is the complete surgical removal of the ovary which bears the neoplasm. Radiation therapy should not be employed except in patients with widespread, inoperable, malignant folliculomas or in those in whom metastases and recurrences following operation make additional surgical interventions impossible.

HYPRLUTEOIDISM AND CORPUS LUTEUM CYSTS

DEFINITION

Corpus luteum cysts are transformation products of corpora lutea resulting from the excessive accumulation of fluid in the central cavity. Their life span is usually longer than that of the normal corpus luteum of menstruation, but the

luteal tissue which lines their wall is not capable of the unlimited growth characteristic of true blastomas. They are usually simple cysts and should be distinguished from the multiple cystic corpora lutea found in the ovaries of women who suffer from hydatidiform moles or chorionepitheliomas.



Corpus luteum cyst. — **A.** Low magnification of an ovary containing a medium-sized corpus luteum cyst. The cyst wall is lined by an irregular, but in general fairly high, corpus luteum cell border. The latter is thinnest on the left side and thickest in the lower right section of the cyst. The mesosalpinx and a cross-section through the oviduct are seen below the cyst. — **B.** Higher magnification of the wall of the corpus luteum cyst shown in Fig. A. Note the typical corrugated appearance of the corpus luteum lining which forms a stratified, fairly thick epithelial layer. The cell borders and nuclei are of normal appearance.

(Courtesy of Dr. L.-C. Simard.)

Corpus luteum cysts are the most common cause of hyperluteoidism, since most of the true blastomas of the corpus luteum (luteomas) are not hormone-producing. It is, therefore, in connection with these cystic and persistent corpora lutea that we shall discuss the syndrome of hyperluteoidism.

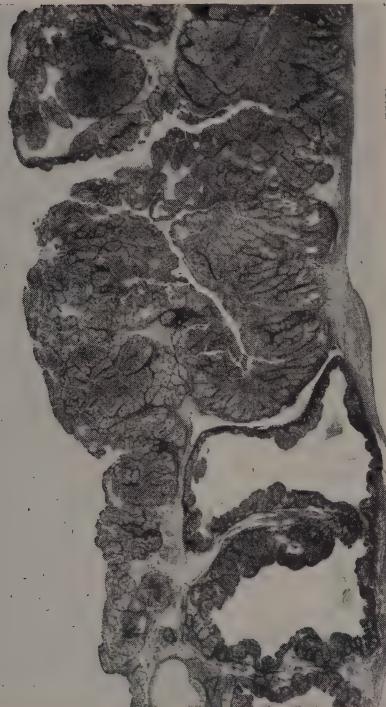
PATHOLOGIC ANATOMY

The histologic structure of corpus luteum cysts is comparatively simple. It resembles that of a large corpus luteum whose cavity is greatly distended by fluid. The wall consists of more or less typical, lipid-containing (macroscopically yellow) corpus luteum cell tissue, which may be either in direct contact with the central fluid accumulation,

or separated from it by a simple epithelial lining or a band of connective tissue. It has been claimed that the persistent corpus luteum resembles that of gestation inasmuch as it is comparatively poor in lipid granules and rarely contains a central hemorrhage. Lipid-containing "pseudoxanthoma" cells in the walls of endometriomas, abscesses or infected ovarian cysts are often mistaken for corpus luteum cysts.

INCIDENCE

Persistent corpus luteum cysts are not very commonly observed, although persistently pregestational endometria can hardly be ascribed to any other ovarian lesion. One worker, who examined 100 pairs of ovaries gathered



Carcinoma of the corpus luteum? Very low magnification of the tumor wall. Note greatly corrugated surface which resembles the wall of a corpus luteum cyst. In left lower corner several independent cystlets developed; their walls are similar to that of the main cavity but somewhat thinner.

(Courtesy of Dr. P. Masson.)

from unselected autopsy material, found corpus luteum cysts in 9% of them. However, it is sometimes difficult to differentiate with certainty between normal and persistent cystic corpora lutea, there being many transitional stages.

PATHOGENESIS

The etiology of corpus luteum cysts is still insufficiently understood. It is known that in women suffering from gonadotrophin-producing tumors, the ovaries almost invariably contain a large number of cystic corpora lutea (see: Ovarian changes in chorionepitheliomas, hydatidiform moles and pituitary tumors). Furthermore, treatment

with gonadotrophins produces "mulberry ovaries" in various laboratory animals. Unusually large, cystic corpora lutea can be produced in the rat by simultaneous treatment with gonadotrophins and folliculoids. Similar hormonal factors are likely to be the cause of multiple corpus luteum cysts in patients suffering from tumors of placental origin. This view is consonant with the comparative frequency of persistent corpora lutea and corpus luteum cysts in normal and in ectopic pregnancy as well as immediately following incomplete abortion (see: Ovarian changes in pregnancy). In all these instances, the simultaneous production of gonadotrophins and folliculoids may well be the cause of the ovarian change. It remains to be shown, however, whether the typical, usually single, corpus luteum cyst is due to a similar pathogenic mechanism, and if so, what determines the number of corpora lutea which undergo cystic change.

Some authors claim that pseudopregnancy, elicited by sexual intercourse or nursing, may also be the cause of persistent corpora lutea in women as it undoubtedly is in animals.

CLINICAL COURSE

Women with corpus luteum cysts frequently believe themselves to be pregnant. The outstanding clinical manifestations of persistent corpora lutea are those of pseudopregnancy. There is amenorrhea, with PROGESTATIONAL TRANSFORMATION of the endometrium, which fails to break down at monthly intervals. Occasionally, severe metrorrhagias occur after a period of amenorrhea, presumably due to a variation in the ovarian hormone production or the eventual complete breakdown of the corpus luteum cyst.

It has been claimed that in 3.2% of the cases, persistent corpora lutea are associated with internal or external ENDOMETRIOSIS and that sometimes they

may even cause METROPATHIA-HEMORRHAGICA-LIKE CHANGES in the endometrium. One author goes so far as to claim that 14% of his patients with "functional uterine bleeding" exhibit a secretory, not a proliferative, endometrium. Among these, corpus luteum cysts were commonly demonstrable. The pathogenesis of these changes is not fully understood, although certain animal experiments indicate that progesterone may produce a "Swiss cheese" endometrium if the uterus is subjected to trauma while it is under the influence of the hormone.

As in pregnancy, the BREASTS are enlarged and may secrete colostrum.

COMPLICATIONS

Since the walls of corpus luteum cysts are thin and fragile, they tend to RUPTURE easily. Even careful pelvic examination may suffice to rupture corpus luteum cysts and to elicit an intra-peritoneal hemorrhage. There is reason to suspect, however, that in many allegedly pertinent cases the patient actually suffered from an ectopic gestation.

DIAGNOSIS

It is rarely possible to diagnose a corpus luteum cyst, without exploratory laparotomy.

Upon PELVIC EXAMINATION, a corpus luteum cyst reveals itself as an approximately walnut-sized, smooth, somewhat mobile, adnexal tumor which may be painful. The uterus is often enlarged. It should be kept in mind that these cysts rarely reach a diameter of more than about $2\frac{1}{2}$ inches and many of the large tumors described as corpus luteum cysts are in reality abscesses with a lipid-containing lining.

Most relevant cases are operated upon under the diagnosis of ECTOPIC GESTATION because almost all the symptoms and signs of these two conditions are identical. Even the gonadotrophin

excretion in the urine may be augmented in the presence of corpus luteum cysts as it is in pregnancy. This fact gives further support to the view that hypophysoid hormones play an important rôle in the pathogenesis of these cysts. Since, on the other hand, ectopic pregnancy often fails to elicit a marked increase in urinary gonadotrophins, even a negative Aschheim-Zondek test is of little diagnostic value. Systematic pregnanediol determinations in the urine of women with corpus luteum cysts have not yet been made but they would probably give helpful information.

PROGNOSIS

In many cases corpus luteum cysts persist for a very long time and tend to recur, as judged by the menstrual histories. Permanent, spontaneous cures may occur. The otherwise favorable prognosis is somewhat darkened by the ever-existing possibility of a severe, potentially fatal, peritoneal hemorrhage.

THERAPY

The usual therapy of corpus luteum cysts is SURGICAL excision of the cyst itself, leaving the remaining ovarian tissue intact. This leads to a "withdrawal hemorrhage" after which the menstrual cycles may become normal owing to the re-establishment of a physiologic relationship between the ovaries and the pituitary. If the primary cause of the condition is entirely extra-ovarian, however, it tends to recur following removal of the cyst.

Could the condition be recognized with certainty, the advisability of a surgical intervention would be very doubtful, especially since complications, such as serious ovarian or uterine hemorrhages, are rare. Most cases, however, are recognized only on the operating table and then, removal of the cystic corpus luteum is certainly justified.

"HYPERNEPHROMAS," "LUTEOMAS" AND OTHER "LIPID CELL TUMORS" OF THE OVARY

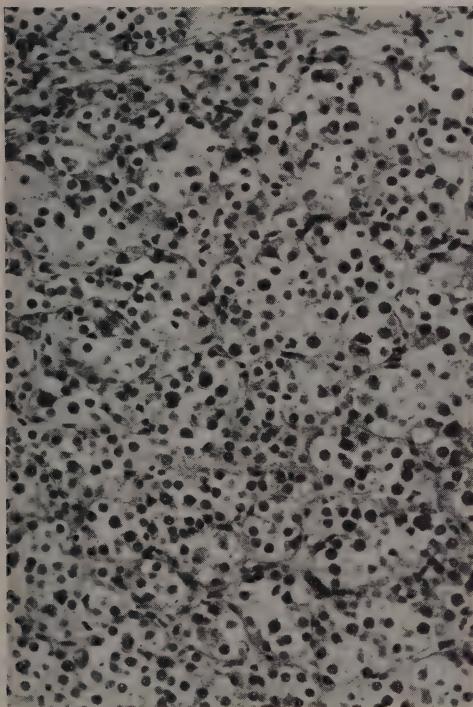
DEFINITION

The "lipid cell tumors" constitute a group of ovarian neoplasms characterized mainly by their lipid-granule-containing endocrine cells which resemble those of the corpus luteum, the adrenal cortex, the theca folliculi or the Leydig cells. Most of them are virilizing growths. With the methods now available it is impossible to determine the origin of these tumors with certainty. Therefore, without excluding the possibility that they are morphogenetically heterogeneous, they will be discussed conjointly under the non-committal heading of lipid cell tumors. The common corpus luteum cysts, and the aden-

omatous proliferation of theca lipid cells or ovarian Leydig cells are discussed in special chapters, merely because their identity is more clearly established. The term "hypernephromas" of the ovaries is to be avoided, since this designation has been reserved for the so-called Grawitz tumors of the kidney (see: p. 191).

PATHOLOGIC ANATOMY

MACROSCOPICALLY, the lipid cell tumors of the ovary are usually more or less irregularly shaped, solid growths which can be readily differentiated from the corpus luteum cysts (see above). They rarely reach a size of

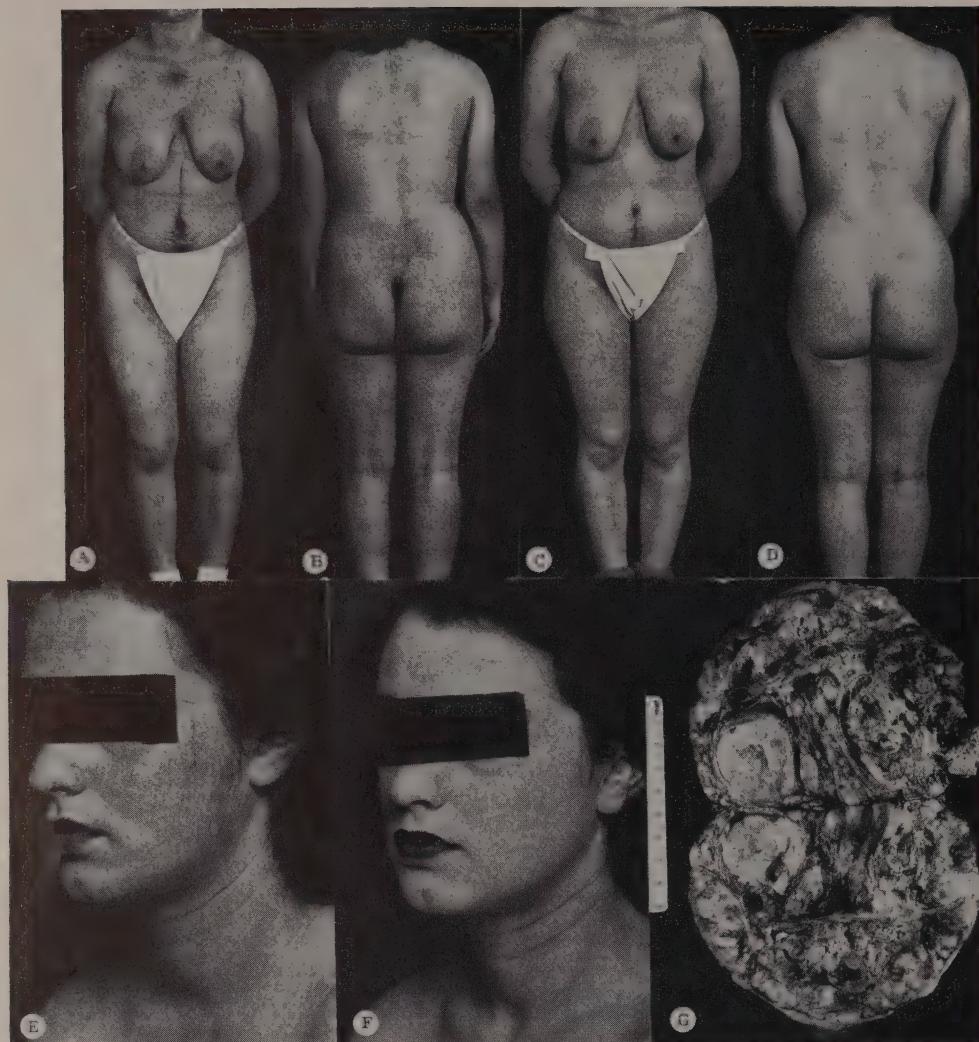


Hypernephroma of the ovary. Adrenal-cortical cells similar to those seen in accessory adrenals. At this high magnification the cell details are evident. The tumor appears to be benign and would be difficult to differentiate from an ordinary accessory adrenal.

(Courtesy of Dr. H.-T. Karsner.)

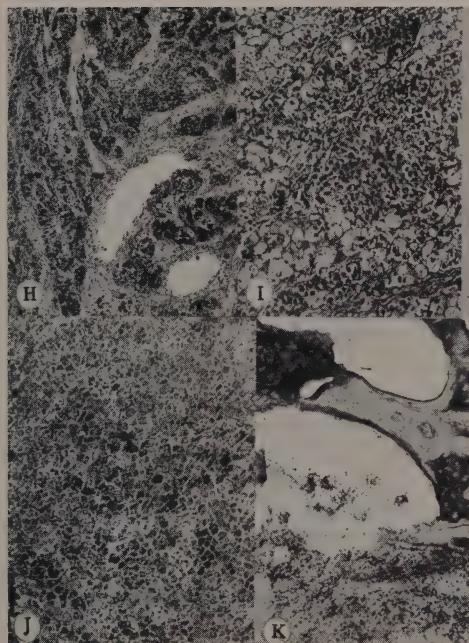


Leydig cell hyperplasia. Note the extensive hyperplasia of epithelial cells within the nerve tracts. These ovarian Leydig cells may contain Reinke crystalloids and resemble in every respect the Leydig cells of the testis. In this particular woman of 45 years, no obvious clinical manifestations resulted from this comparatively small cell proliferation. (Courtesy of Dr. L. Berger.)



Ovarian lipid-cell tumor. — A. and B. Appearance of patient's body before operation: The patient suffered from hypertension (160/110), hirsutism, acne, clitoris enlargement, secondary amenorrhea, obesity, and a palpable ovarian tumor. She excreted 55 mg. of 17-KS/day (about 5 times normal). Weight 141 pounds (64 Kg.) — C. and D. Appearance of body five months after ablation of ovarian tumor. By this time her virilism had vanished, her blood pressure was normal (139/90), the urinary 17-KS 2.6mg/day, her body weight 132 pounds (59.9 Kg.) and her previously virile attitude became feminine. — E. Appearance of face before operation (hirsutism, acne). — F. Five months after operation. — G. Cut surface of tumor removed from patient shown in previous figures. The dark areas represent hemorrhages within the tumor substance; remaining portion was of bright orange-yellow color. (Cont'd.)

(After E.-J. Kepler et al.: Am. J. Obst. & Gynec. 47, 43, 1944.)



— H. The high lipid content of the tumor cells is shown in this section stained with Sudan III. — I. Glycogen is indicated by the small, dark tumor cells, stained with carmine. — J. Darkened cells indicate a positive reaction in section stained with Ponceau-fuchsin. All these histochemical reactions are characteristic of adrenal-cortical tumors. — K. Nodule from center of tumor showing well-formed bony trabeculae and marrow elements; stained with hematoxylin and eosin.

more than about 6 inches in diameter and the smallest examples are minute nodules, hardly distinguishable from the non-blastomatous adrenal rests. Perhaps their most characteristic macroscopic feature is their bright yellow color which strikingly resembles that of the corpus luteum or adrenal cortex.

Often, lipid cell tumors develop bilaterally. They are most frequent within the mesovarium or the broad ligament, where accessory adrenocortical tissue and nests of ovarian Leydig cells are common.

Microscopically, they usually consist of more or less irregular bands or islets of vacuolated, large, polyhedral cells which imitate the structure of the corpus luteum or adrenal cortex. In vivo,

the vacuoles are filled with lipid granules and sometimes considerable quantities of glycogen. The latter is allegedly characteristic of tumors derived from the adrenal cortex, permitting their differentiation from growths of the corpus luteum. Fuchsinophilic granules, supposedly characteristic of the adrenals in the adrenogenital syndrome, are present only in some of the ovarian lipid-cell-tumors.

These tumors are often polymorph and certain portions can exhibit the aspect of an alveolar sarcoma, a medullary, papillary or a cirrhotic carcinoma. Without any very definite reason, some lipid cell tumors have been described as CORPUS LUTEUM TUMORS or "luteomas", others as CARCINOMAS OF THE CORPUS LUTEUM and still others as OVARIAN HYPERNEPHROMAS OR SARCOMAS.

As emphasized in the chapter on folliculomas, certain granulosa cell tumors may become partially luteinized and assume the aspect of the "FOLLICULOME LIPIDIQUE" of Lecène. It is obvious that if this process of luteinization goes far enough, transitional types will result which are difficult to classify. Luteinized granulosa-cell-tumors and corpus luteum tumors are, however, so closely related that it would be difficult to make even a theoretic distinction between them.

INCIDENCE

Lipid cell tumors are very rare and can appear at any age. Most frequently they occur in adult women, often after the menopause.

PATHOGENESIS

It is debatable whether solid lipid cell tumors can develop from fully-formed corpora lutea, in spite of the many reports on so-called "luteomas." It must be clearly stated that in most cases no reason is given by authors for their designation of a growth as "luteoma" rather than adrenal-rest tumor

and vice versa. Probably the majority, if not all, of these growths are derived from adrenal-cortical remnants (Marchand's rests). This view is supported by the previously mentioned fact that many lipid cell tumors occur in the broad ligament where Marchand's rests are particularly common.

CLINICAL COURSE

In addition to the usual local manifestations of an ovarian tumor, the lipid cell growths — if hormonally active — are mainly characterized by signs of VIRILIZATION, which frequently are so pronounced that one may well speak of pseudohermaphroditism. There can be marked hirsutism of the trunk and extremities, beard growth, enlargement of the clitoris, deepening of the voice, amenorrhea, and atrophy of the breasts.

In children, lipid cell tumors cause PRECOCIOUS PSEUDOPUBERTY with growth of the breasts (unlike in adults!), pubic hair and libido. At this early age, however, pseudohermaphroditism has not been noted with these ovarian growths.

In many bearers of such lipid cell tumors, there are also other symptoms of the adrenogenital syndrome (or Cushing's syndrome) such as HYPERTENSION, POLYCYTHEMIA, OBESITY, CUTANEOUS STRiae and DIABETES. These manifestations are clearly reminiscent of adrenal hyperfunction and strongly suggest that the ovarian lipid cell tumors can imitate adrenal growths in producing gluco-corticoids and mineralo-corticoids as well as sex hormones.

DIAGNOSIS

The diagnosis of lipid cell tumors is extremely difficult, as shown by the fact

that only in one case was at least a probable diagnosis of "adrenal-cortical tumor of the ovary" made on the basis of clinical evidence. Such a diagnosis would appear warranted in patients in whom signs resembling the adrenogenital syndrome or Cushing's syndrome develop simultaneously with an ovarian tumor.

Cushing's disease and primary adrenal-cortical tumors have to be eliminated on the basis of the usual local symptoms pointing to a tumor in the adrenal or pituitary region. It is well to remember that hypophyseal growths can secondarily cause luteinization of the ovaries and subsequently lead to virilization of the testoid hyperthecosis type. (See patients on pp. 446, 447.)

HORMONAL studies can also be of some diagnostic value. There is some indication that 17-KS excretion is increased, but comparatively few relevant data are available as yet.

PROGNOSIS AND THERAPY

Lipid cell tumors do not regress spontaneously, but following surgical ablation the results are very encouraging. If the patient survives the complete removal of the growth, the symptoms almost invariably vanish.

There is, however, a comparatively high mortality rate postoperatively, perhaps because a sudden drop in the corticoid hormone content of the blood and tissues such as is occasioned by extirpation of an adreno-cortical tumor, is likely to produce a transitory corticoid deficiency. Hence, the same pre-operative measures (corticoid and NaCl treatment) are recommended as with orthotopic cortical neoplasms.

TESTOID HYPERTHECOSIS AND LEYDIG CELL TUMORS OF THE OVARY

Experimental work and clinical case reports indicate that hyperplasia and hypertrophy of luteinized theca cells can cause a type of virilism which has

hitherto not been clearly recognized as a special disease entity. Stein *et al.* (1935-9) in their study of the "polycystic ovary" incidentally mention that

in some women, this condition is accompanied by signs of virilism and especially hirsutism. At the same time, the ovaries reveal a tendency toward theca luteinization.

Under the name of "hyperthecosis," *Fraenkel* (1941) described a syndrome of virilization elicited by diffuse hyperplasia of the theca cells. He apparently considered the theca proliferation as truly blastomatous, since occasionally he referred to it as "thecomatosis." In order to distinguish these cases from thecomas (which cause hyperfolliculoidism) the term "TESTOID HYPERTHECOSIS" will be used here.

In most relevant cases, ovariectomy is followed by at least partial disappearance of the pseudohermaphroditic traits. Yet the ovarian origin of the clinical symptoms is sometimes subject to doubt. In one bearded woman with normal menses there was a typical Cushing's syndrome. The ovaries were greatly enlarged, due to small-cystic degeneration with theca luteinization. She died from pulmonary embolism shortly after removal of her ovaries and autopsy revealed a basophilic pituitary adenoma and adrenal-cortical hyperplasia. In another case, the clinical manifestations and the ovarian changes were very similar. Ovariectomy led only to temporary regression of the virilism (depression of testoid production during postoperative alarm reaction?). She suffered from severe headaches but since she survived, the presence of a primary pituitary lesion could not be checked.

Further work will have to be done before the pathogenesis of this interest-

ing syndrome is understood, but it is suggestive that theca luteinization induced by gonadotrophins can cause virilism in animals. Thus we have experimental, as well as clinical, evidence indicating that the theca can produce both folliculoid and testoid hormones. In cases with primary hypophyseal hyperfunction the possibility of simultaneous ovarian and adrenocortical hyperactivity must also be kept in mind.

In connection with testoid hyperthecosis, it is pertinent to mention that hyperplasia and the formation of small ADENOMAS IN LEYDIG CELLS OF THE HILUS are also conducive to virilism. These cells are not known to give rise to large or malignant tumors and the resulting virilism is generally moderate. There is usually some growth of the clitoris, deepening of the voice, hirsutism and breast atrophy, but the menstrual cycle may continue normally. Frequently the cause of such changes remains unknown. Perhaps hilus-cell hyperplasia would prove to be more common if these cells were examined in all cases of inexplicable virilism.

The DIAGNOSIS of testoid hyperthecosis or Leydig cell hyperplasia is impossible with our present day methods except by biopsy. From the differential diagnostic viewpoint arrhenoblastomas, adrenal tumors, congenital true- and pseudohermaphroditism and virilizing folliculomas must be considered. (See patients on pp. 446, 447.)

The THERAPY is ovariectomy whenever the symptoms are sufficiently severe to justify it.

OVARIAN TUBULAR ADENOMAS (ARRHENOBLASTOMAS)

In this book, the term "tubular adenoma" is used to designate any ovarian tumor whose epithelial components consist prevailingly of gland-like tubules free of mucin.

An attempt has been made to define as "arrhenoblastomas" or "andreiblastomas" all those neoplasms which are "masculinity tumors" (*Männlichkeitsgeschwülste*), without distinguishing

ing between growths which are "masculine" because of testoid production and those which morphologically imitate the male gonad. Other investigators regard all clinically masculinizing tumors as arrhenoblastomas irrespective of their histologic structure. Such definitions are confusing, since they group together a number of essentially different tumors (e.g., tubular adenomas, lipid cell tumors, etc.).

Some authors strictly separate the tubular adenoma without virilization from the arrhenoblastoma, defining the latter as histologically similar to the former, but always associated with pseudohermaphroditic traits. This purely functional separation is likewise not acceptable, at least until much more is known about the pathologic physiology of these neoplasms. Such a distinction would be no more justifiable than the assumption that a struma ovarii, which produces Graves' disease, is a neoplasm essentially different from one which does not elaborate enough thyroid hormone to cause detectable signs of hyperthyroidism. It is quite possible that the degree of Leydig cell development within the tubular adenomas could account for the virilization seen in some cases, but this has not yet been proven.

Some of the rete adenomas appear to be merely a slight exaggeration of the normal "male" part of the ovary, while in other cases the tubular structures imitate testis tissue so closely that the ovaries, which contain it, were described as true ovotestes. The difficulty of accurate differentiation is well illustrated by the fact that one of the famous relevant cases was first described as an ovotestis and only later reclassified as an arrhenoblastoma.

The "false seminoma" or dysgerminoma, is listed as a separate blastoma type because its characteristic morphologic appearance clearly distinguishes it from the tubular adenoma.

CLASSIFICATION

According to their FUNCTION we distinguish between :

- (1) *Virilizing tubular adenomas*
(which usually contain Leydig cells).
- (2) *Hormonally inert tubular adenomas.*

On a purely HISTOLOGIC BASIS we differentiate between :

- (1) *Rete adenomas.*
- (2) *Typical tubular adenomas.*
- (3) *Partly tubular and partly diffuse adenomas.*
- (4) *Entirely atypical tubular adenomas.*

(For their histologic characteristics, see : Pathologic Anatomy, below.)

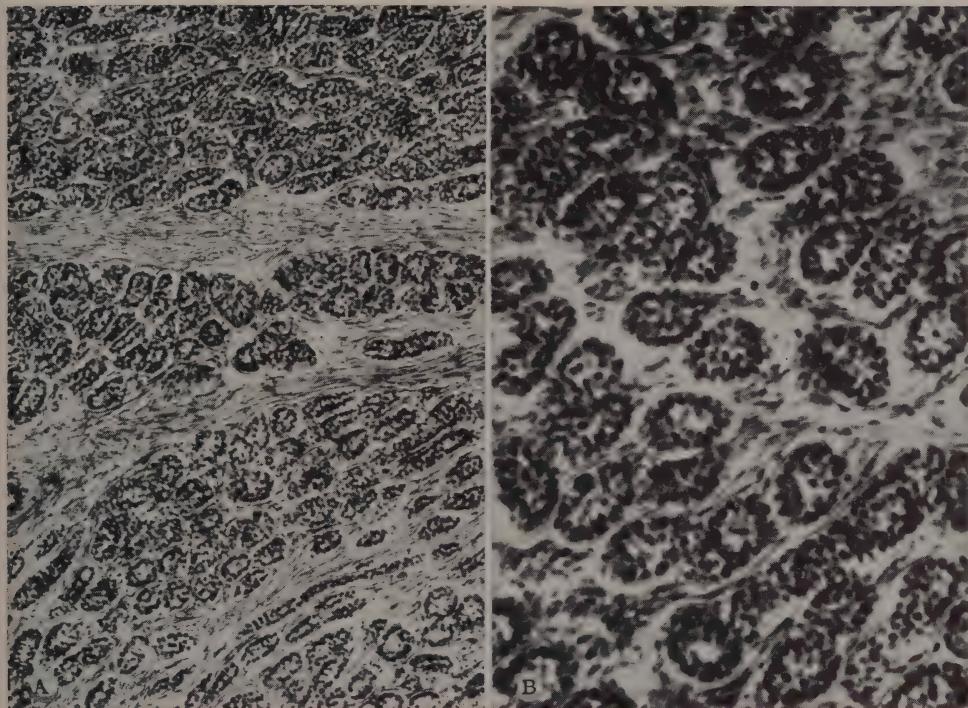
PATHOLOGIC ANATOMY

Upon naked eye inspection, most tubular adenomas prove to be medium-sized, round, or ovoid solid growths. Their color as viewed from the surface is greyish-white, their consistency rather firm. On the cut surface, some of them (especially those containing many Leydig cells) are bright yellow.

The microscopic characteristics of the various types may be summarized as follows :

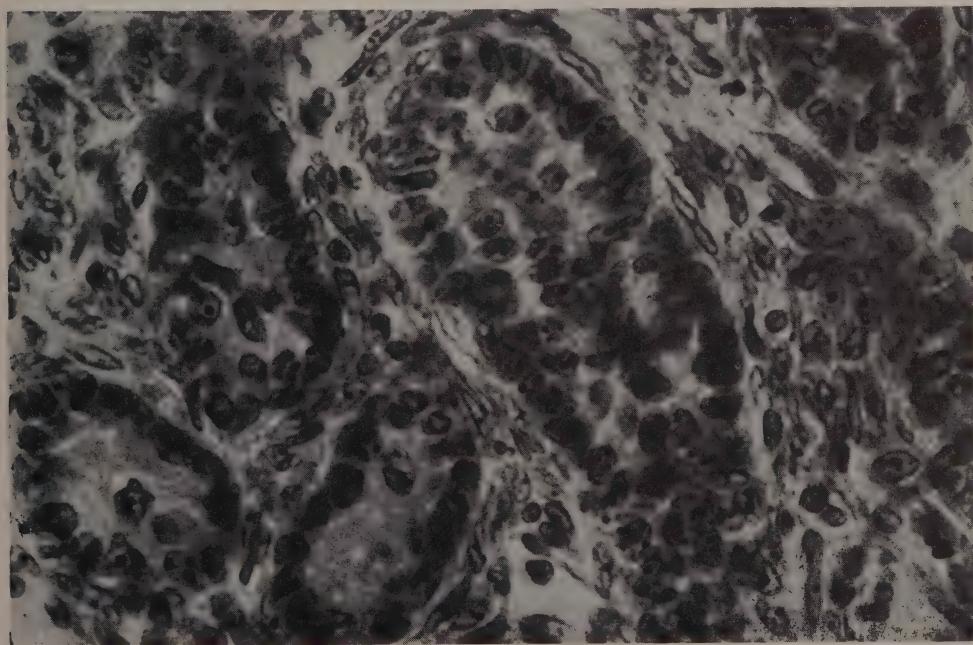
(1) **RETE ADENOMAS** consist mainly of rather regular, narrow tubules, localized in the rete region and resembling normal rete tubules. Such adenomas are comparatively common, usually of small size, and asymptomatic.

(2) **TYPICAL TUBULAR ADENOMAS** are often difficult to distinguish from the rete adenomas. They are claimed to originate, not from the rete tubules but from medullary cords, that is, structures corresponding to the seminiferous tubules of the testis. In these neoplasms, the stroma and the interstitial cells tend to be more developed than in the rete adenomas and they often reach a greater size. Frequently the epithelial components have no lumen and represent trabeculae rather than



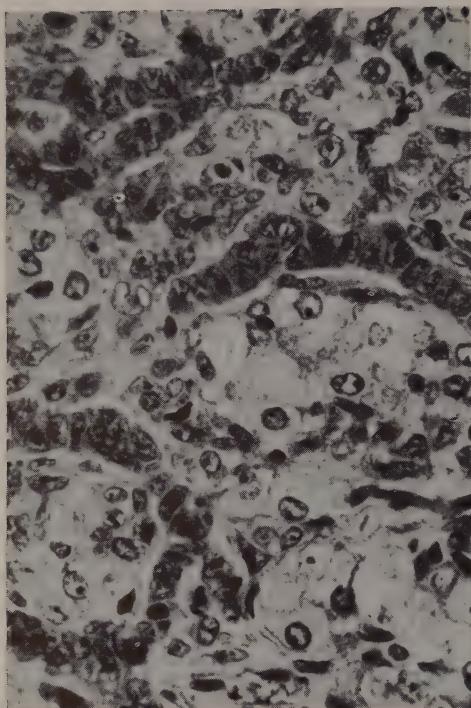
Tubular adenoma. — A. Under low magnification note tubules separated by connective tissue septa. — B. Higher magnification of same tumor, note the resemblance to fetal testicular tissue. In this region, the stroma consists of ordinary connective tissue without well-defined Leydig cells.

(Courtesy of Dr. H.-T. Karsner.)



Tubular adenoma. At high magnification note absence of tubular lumina and resemblance to tubular granulosa-cell tumor.

(Courtesy of Dr. P. Masson.)

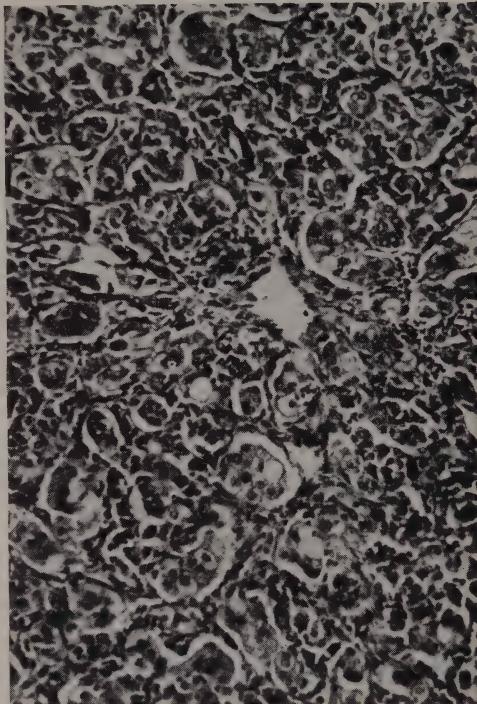


Arrhenoblastoma with Leydig cells? This tumor was originally described as a "luteinoma" (by Cosacesco et al). However, regions such as this, are definitely suggestive of the trabecular type of arrhenoblastoma or tubular adenoma (dark cells) with lipid-containing (light) Leydig cells between the cords.

(Courtesy of Dr. P. Masson.)

tubules. A very large number of these neoplasms are distinctly virilizing.

(3) In ENTIRELY ATYPICAL TUBULAR ADENOMAS, the epithelial elements are so irregular that usually they cannot be distinguished with certainty and the diagnosis must rest partly upon the clinical manifestations of virilization. Many relevant cases have been mistaken for solid carcinomas or sarcomas of the ovary. Probably most, if not all ovarian solid carcinomas, carcinosarcomas, and sarcomas purported to have elicited signs of pseudohermaphroditism, actually belong to this group. The same may be said about the malignant or carcinomatous tubular adenomas although, in some of these,



Tubular adenoma. The tubular arrangement of epithelial cells is extremely indistinct in this neoplasm but large, epithelioid Leydig cells are clearly distinguishable among the narrow and irregular tubules.

(Courtesy of Dr. H.-T. Karsner.)

tubular formations remain fairly distinct, at least in certain areas.

Transitional types between these main groups are common. Special emphasis must be laid upon the fact that in the case of group 3, the co-existence of diffusely arranged cells and tubulo-alveolar formations may give the growth an appearance very similar to that of granulosa cell tumors.

The interstitial cells in the stroma of tubular adenomas are often in close contact with nerves. In this they resemble sympatheticicotropic cells of the normal ovary and the extra-testicular Leydig cells in the male.

In cases of unilateral tubular adenomas, the contralateral ovary is fre-

quently atrophic, perhaps as part of the "virilization" syndrome.

INCIDENCE

Tubular adenomas are very rare. Among solid ovarian tumors, their incidence is approximately 1%.

They can occur at any age, but are most common in women between 20-35 years. In spite of their virilizing effect, they are compatible with pregnancy and tend to induce pseudohermaphroditic traits both in mother and offspring.

PATHOGENESIS

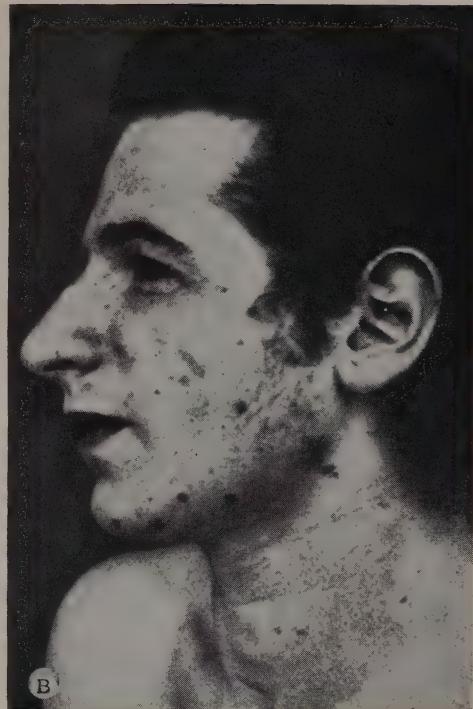
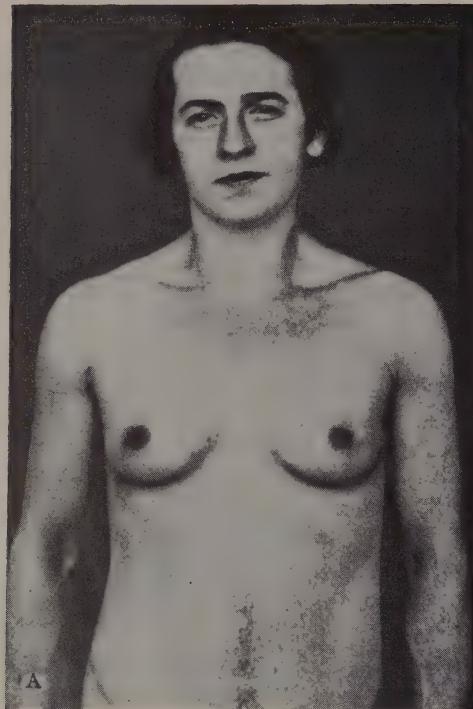
The striking resemblance of ovarian tubular adenomas to the corresponding testis tumors, their histologic structure, their frequent occurrence in the "male" hilus region of the ovary, and their ability to produce testoids, has led to the almost universal acceptance of the theory that they are derived from "MALE" GONADAL ELEMENTS IN THE OVARY. There are minor differences of opinion concerning the specific point of origin within this region. Some derive them from the rete, others from the medullary cords or even from the seminiferous tubules of a congenitally true-hermaphroditic gonad. The existence of intermediate types between dysgerminomas and tubular adenomas suggests close pathogenic correlations between these two "male" ovarian tumors.

In connection with such histogenetic considerations, surprisingly little attention has been given to the common occurrence of INTERSTITIAL CELLS within the stroma of tubular adenomas. These "Leydig cells" are strikingly similar to those found in the normal testis and in the hilus region of the normal ovary. In tubular adenomas, they can appear not only in the form of individual cell groups between tubules, but even as distinct lipid cell adenomas. Such cases may be regarded as intermediate between the pure (interstitial-cell-free) tubular adenomas

and the pure Leydig cell tumors. (See : Testoid Hyperthecosis on p. 440.)

HEREDITARY FACTORS appear to play an important rôle in the causation of tubular adenomas, since they have repeatedly been observed in several members of the same family, or in combination with congenital malformations. It is probable, however, that even in hereditarily predisposed individuals, an additional growth impulse is necessary to elicit neoplastic proliferation in the ovary some time during post-embryonic life. Unlike "false seminomas" the tubular adenomas are rarely associated with congenital pseudohermaphroditism. Signs of mild virilism are common in normal women during pregnancy and after the menopause, when the ovarian rete tubules and Leydig cells tend to proliferate. The endocrine change responsible for "pseudohermaphroditic traits" during pregnancy and old age is still unknown, but it is rather tempting to look upon the virilizing tubular adenoma as an abnormal exaggeration of the same processes.

It is of interest in this connection that in female birds during embryonic life, treatment with TESTOID HORMONES can cause almost complete sex reversal with the development of functional seminiferous tubules, (see : Stimuli Influencing Ovary on p. 379). In mammalian embryos, such marked degrees of virilization cannot be obtained. However, during early post-embryonic life, overdosage with testoids causes pronounced inhibition in the development of the ovary, uterus and vagina, as well as adenomatous proliferation of the rete tubules. It is possible therefore that in women it is also a hormonal stimulus which elicits both the formation of "defeminizing and virilizing" tubular adenomas and the occasionally co-existent malformations of the Müllerian duct derivatives.



Virilization due to lipid-cell tumor (hypernephroma?). — A. Note masculine facial expression and body build, atrophy of breasts, excessive hair growth on abdomen and arms. The 32-year-old patient suffered from secondary amenorrhea, polycythemia (6,660,000 erythrocytes), slight hypertension, slight hyperglycemia and excessive development of the clitoris. She lost much of her scalp hair, especially in the temporal region but grew a light beard. After removal of a unilateral, lipid-cell tumor (interpreted as a lutein-cell tumor), the menses returned and all other symptoms and signs disappeared. — B. Same patient as that shown in Fig. A. Note acne, beard growth, slight temporal baldness and resemblance to patient on p. 447.

(After J. Novak and O. Wallis: Arch. f. Gynäk. 164, 543, 1937.)

CLINICAL COURSE

In the majority of women with tubular adenomas of the ovary, LOCAL SIGNS are not particularly prominent (see: "Diagnosis" below).

The most striking manifestations of these neoplasms are due to the production of excessive quantities of testoid hormones, so that in the so-called "silent" varieties of these tumors, there are no characteristic clinical features. The sexual development and menstrual cycles are usually normal until the beginning of the illness when VIRILIZATION and DEFEMINIZATION become conspicuous. Gradually, there appears excessive hair growth on the upper lip,

chin, arms, legs and buttocks; the scalp hair tends to fall out, so that these bald women resemble middle-aged men. Acne may appear, the voice assumes a low pitch and is often hoarse, the clitoris is enlarged, there is amenorrhea and the uterus as well as the breasts, become atrophic. There is often a marked loss of libido. Usually neither the blood pressure nor the blood count show any conspicuous change, but in some cases there is a definite tendency to put on weight as the tumor develops.

Whether tubular adenomas can be the cause of METRORRHAGIAS is somewhat doubtful. Seemingly pertinent cases could belong to the granulosa cell group.

Tubular adenomas can undergo carcinomatous transformation in which case the usual manifestations of an ovarian cancer are added to those of a virilizing neoplasm.

DIAGNOSIS

In the diagnosis of tubular adenomas, LOCAL SIGNS are rarely helpful, since these blastomas are usually of moderate size and exhibit no great tendency towards invasive growth. Sometimes however, pelvic examination furnishes valuable data. If the tumor is virilizing, one should try to differentiate between ovarian and adrenal-cortical neoplasms and if there are no pseudohermaphroditic traits, the local signs may be the only detectable mani-

festations of the tubular adenoma. Hence the preoperative diagnosis of these tumors is rarely possible. During the operation, the often (but not invariably) bright, yellow color of the cut surface can help the recognition of the neoplasm, but other virilizing tumors (especially the hypernephromas and Leydig cell tumors), as well as corpus luteum tumors and lipid-containing folliculomas may have the same color.

The signs of DEFEMINIZATION (atrophy of breasts and uterus, amenorrhea, loss of libido, etc.) and VIRILIZATION (hirsutism, growth of the clitoris, deepening of the voice, loss of scalp hair, etc.) should always raise the suspicion of a tubular adenoma. From a DIFFERENTIAL DIAGNOSTIC viewpoint, adren-



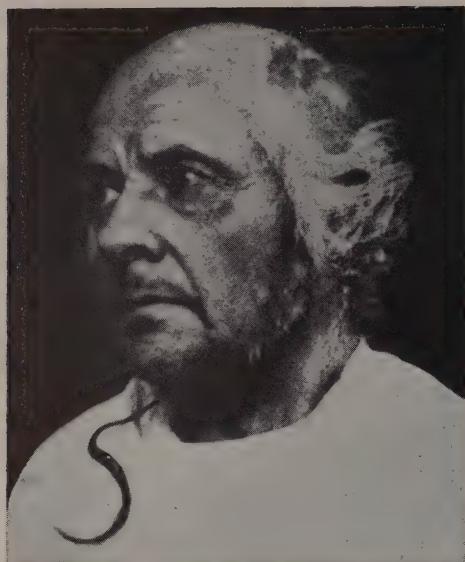
A



B

Virilization due to tubular adenoma. — A. This 24-year-old patient suffered from amenorrhea with an abdominal tumor, erroneously diagnosed as pregnancy. Her facial expression became masculine and she developed some beard and moustache growth so that she had to shave every three days. Her libido was increased. Following removal of a roundish tumor (about 6 inches in diameter) the symptoms disappeared and the menses reappeared. Histologically, the tumor was a tubular adenoma with sarcomatoid stroma. — B. Same patient as that shown in Fig. A, but following ablation of her virilizing tumor. The facial appearance of the patient became feminine again, the menses reappeared and two years after ablation of the tumor, she became pregnant. She is shown here with her child.

(After E. Strassman: Ztschr. f. Geburtsh. u. Gynäk., 99, 368, 1938.)



Virilization caused by arrhenoblastoma. 66-year-old woman with marked virilization. Note pronounced beard growth, baldness and coarse, masculine facial expression. The ovary contained a partly tubular, partly papillary tumor with occasional giant cells. The tubular portion resembled the common, tubular adenoma but the neoplasm was not very typical.

(After A. Büttner: *Virchow's Arch. f. Path. Anat.* 289, 452, 1933.)

ad-cortical tumors (of the adrenal itself or of adrenal-rests in the ovary) and Cushing's disease can often be excluded, since in most tubular adenomas, there is no special tendency towards the development of plethora, hypertension, diabetes, abdominal striae or the characteristic "moon face." However, a search for specific local signs in the adrenal and pituitary region is always indicated in such cases.

Urinary HORMONE DETERMINATIONS (17-KS, folliculoids or gonadotrophins) have not been carried out in a sufficiently large number of cases to be of real diagnostic value. The few reported data do not reveal any great deviation from the normal.

PROGNOSIS

The prognosis of tubular adenomas is favorable, if the tumors are completely removed. Since these neoplasms tend to occur in young women, before or during the child-bearing period, and since they are usually not malignant or bilateral, conservative operations are justified. Following removal of the growth, the menstrual cycles tend to reappear in previously amenorrheic patients, while most of the pseudohermaphroditic traits disappear. The loss of the excessive body and facial hair is particularly striking and becomes noticeable a few weeks after the operation. The libido also tends to return within a short period, and the previously atrophic breasts become full again. The enlarged clitoris usually regresses more slowly and some virilization may persist.

After the tumor is removed and the patient "refeminized," normal pregnancies occur in a fairly large number of cases.

Recurrences after the operation are comparatively rare, since most of these tumors are benign or of a low-grade malignancy.

THERAPY

The therapy of choice is the surgical removal of the tubular adenomas. No data are available concerning the efficacy of X-ray therapy and in view of the excellent results obtained by conservative surgery, experimentation along these lines is hardly justified.

In exceptional cases, following removal of the (usually atrophic) ovary containing the tubular adenoma, severe menopause-like deficiency symptoms are noted. This could be due to withdrawal of the hormones produced by the tumor itself and should be treated with folliculoids in the usual manner.

OVARIAN FALSE SEMINOMAS (DYSGERMINOMAS) OR EMBRYONIC CARCINOMAS OF THE OVARY

DEFINITION

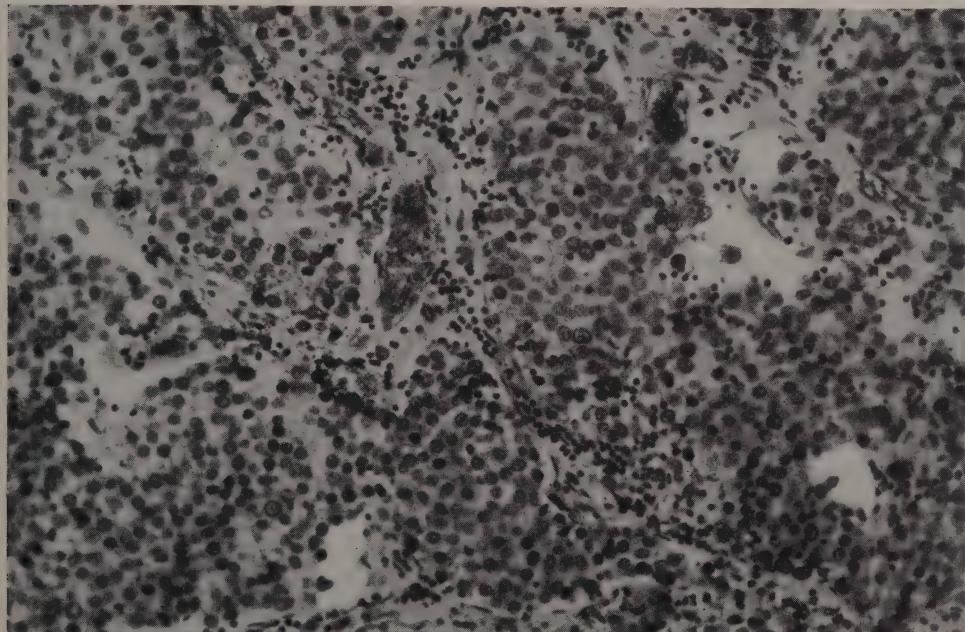
The false seminoma of the ovary is a malignant neoplasm characterized by the presence of medium-sized, round or polyhedral, lightly-staining cells, diffusely arranged in a loose stroma. The epithelial elements sometimes contain large amounts of glycogen and tend to form multinuclear giant-cells; the stroma is infiltrated with lymphocytes. This tumor often develops in children and in already pseudohermaphroditic individuals. Histologically, it strikingly resembles the false seminomas of the testis, but tends to be less malignant. It is not known to produce any hormones or obvious endocrine disturbances, although it can cause increased urinary elimination of gonadotrophins.

PATHOLOGIC ANATOMY

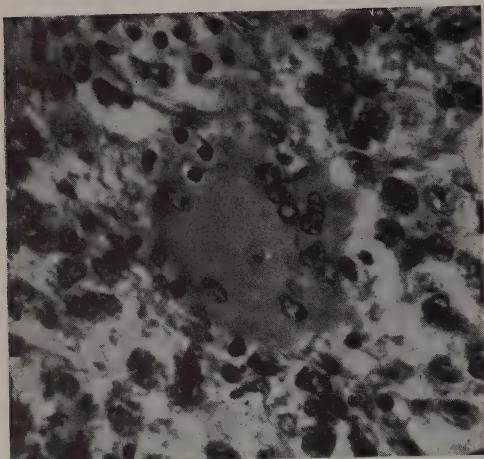
Seminomas are yellowish-grey, often encapsulated tumors of moderate size. They may cause death due to metastases, before the primary tumor reaches extensive proportions. On the cut surface, they exhibit a testis-like appearance. About 35% of them are bilateral.

The microscopic appearance of the seminomas has been briefly outlined above in connection with their definition. Contrary to common belief, they are not large-cellular cancers, since the average diameter of the epithelial cells usually varies between 12-18 μ .

They often contain polynuclear giant cells which have been mistaken for the Langhans cells of tubercles. These



False Seminoma. Note lymphocytic infiltration, in the connective tissue septa, between the epithelial cells. (Courtesy of Dr. P. Masson.)



Dysgerminoma giant cell. Note structure of typical large giant cell. There are several entirely independent nuclei. The outlines of the cytoplasm are indistinct and merge with the surroundings. A few lymphocytes appear to be engulfed by the giant cell.

(Courtesy of Dr. L. Berger.)

gave rise to much speculation concerning a possible causal relationship between tuberculosis and seminoma formation. They have also been claimed to represent the same type of giant cell which is formed from normal seminiferous epithelium, under the influence of various damaging agents. A possible relationship with placental giant-cells must also be considered.

The lymphocytic infiltration of the stroma (mentioned above) is one of the most characteristic features of seminomas.

Metastases are infrequent, but can occur in almost any organ.

INCIDENCE

False seminomas are rare. They hardly represent more than 1-2% of all solid ovarian tumors. They may occur at any age, but are most common during the second decade of life. Their incidence in pregnancy is comparatively high.

PATHOGENESIS

The false seminomas have been claimed to develop from *remnants of the*

medulla of the embryonic ovary. The extra-ovarian false seminomas, which are apparently incompatible with this theory, have been derived from undifferentiated ectopic gonadal parenchyme.

Another theory holds that these growths arise from abnormal *placental Langhans cells*, which tend to form syncytia. This interpretation is consonant with the increased gonadotrophin production of some false-seminoma-bearers.

The attempted derivation of these cells from the *Langhans cells of tubercles* is entirely unfounded.

Heredity factors certainly have an important part in the pathogenesis of false seminomas. They frequently occur in sisters, in true hermaphrodites, and in conjunction with other congenital malformations. The often pronounced hypoplasia of the female accessory sex organs (e.g., uterus, vagina, breasts) may also be interpreted as due to a hereditary stigmatization. In some cases, there is even a complete or partial aplasia of Müllerian duct derivatives.

CLINICAL COURSE

False seminomas cause no typical endocrine disorder but they frequently occur in pseudohermaphrodites and in women with genital hypoplasia (see : Pathogenesis). Humoral influences probably play a rôle in the pathogenesis of these associated sexual manifestations.

In cases with pronounced uterine hypoplasia or aplasia, there is primary AMENORRHEA. In a large number of patients, the menstrual periods remain normal and pregnancy may occur.

In general, it can be said that the clinical course is notably devoid of any specific characteristics other than those due to the presence of an unusually rapidly growing, ovarian neoplasm

which frequently shows malignant properties and tends to develop in young or pseudohermaphroditic individuals.

Women with false seminomas usually exhibit pseudohermaphroditic traits or genital hypoplasia from birth, or at least a long time before the neoplasm is noted (unlike patients with virilizing, testoid-hormone-producing ovarian tumors).

DIAGNOSIS

The preoperative diagnosis of ovarian false seminomas is next to impossible. The only clinical signs which may justify the suspicion of this tumor are its tendency to develop at an early age and often in individuals with marked signs of genital hypoplasia or pseudohermaphroditism.

False seminomas tend to be mobile and rarely become adherent to the adjacent tissues until late in their development. The frequently rapid growth rate of the neoplasm can be verified by repeated palpation, but none of these physical criteria are very characteristic. The use of lipiodol to obtain a good X-ray picture of the internal sex organs may be of some value, especially in recognizing associated abnormalities in the Müllerian derivatives.

Bioassays sometimes reveal increased gonadotrophin elimination, but the pertinent results are contradictory.

PROGNOSIS

False seminomas are rapidly-growing-tumors; they can fill the whole pelvis a few months after being barely detectable by palpation. Although some of them are benign, in most cases, their progress is rapid and lethal.

THERAPY

With ovarian false seminomas, the therapy of choice is complete SURGICAL removal of the ovaries and adnexa, combined with panhysterectomy. The frequency of recurrence is too great to warrant conservative interventions. This is true not only of large infiltrating false seminomas, but even of apparently circumscribed growths, especially if the patients are pseudohermaphrodites or if they suffer from malformations of the internal sex organs which are incompatible with normal reproduction. Yet in a fairly large number of patients, conservative removal of the tumor (leaving the contralateral ovary with the uterus and tube intact) has corrected previously existing menstrual anomalies. Hence unilateral ovariectomy may be considered if the neoplasm is very small and the accessory sex organs not severely deformed. In each case the risk entailed must be weighed against the desirability of maintaining the function of the contralateral gonad.

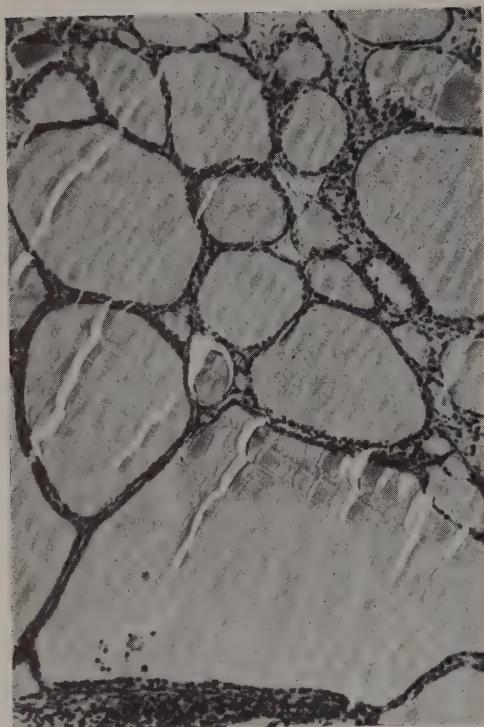
The great X-RAY sensitivity of the testicular false seminomas is apparently shared by the corresponding ovarian tumors. Under the influence of deep X-ray treatment, these tumors simply melt away and huge growths can almost disappear within a short time. With inoperable false seminomas, especially if they have produced widespread metastases, roentgen therapy is therefore the treatment of choice. It also has its place following surgical removal of the neoplasms as a prophylactic measure against recurrences. However, in spite of the usually impressive initial results, the neoplasms tend to become X-ray resistant and recur after some time.

STRUMA OVARI

DEFINITION

The struma ovarii is an ovarian teratoid which consists exclusively, or

at least predominantly, of thyroid tissue. It can resemble the normal thyroid or the various forms of goiter;



Struma ovarii. Typical ovarian-thyroid tumor containing no other tissue. This field illustrates a macrofollicular region.

(Courtesy of Dr. T.-R. Waugh.)

in some instances, it undergoes malignant transformation. Because of its potentially endocrine nature and histologic uniformity, this growth is discussed separately from other teratoids. It will be kept in mind, however, that small nodules of thyroid tissue can appear in any teratoid although usually their size is approximately in proportion to that of the other organs in the embryo.

PATHOLOGIC ANATOMY

Macroscopically, the struma ovarii is usually an irregularly-shaped, nodular, brownish growth. The cut surface is honey-combed by numerous small follicles, sometimes even by large cystic cavities filled with colloid.

Microscopically, the growth consists of colloid-containing follicles, or

tubules and trabeculae similar to those in diffuse or nodular micro- or macrocystic cervical goiters.

It has been emphasized, however, that not all the colloid-containing ovarian growths which resemble goiter tissue are actually derived from a thyroid primordium. Some so-called "thyroid tumors" of the literature, greatly resemble ordinary cystadenomas. Hence it has even been claimed that all these tumors are merely modified papillary or pseudomucinous cystadenomas. This interpretation is untenable, especially because the struma ovarii may copy any type of the struma colli and does not give a positive mucicarmine reaction; it contains iodine and thyroid hormone and in this respect, also resembles thyroid tissue.

It is estimated that about 5% of the ovarian strumas become malignant.

INCIDENCE

Struma ovarii is a rare neoplasm, its general incidence among ovarian teratomas being about 2.7%. Small, organoid thyroid inclusions however are detectable in about 10% of all ovarian embryomas.

Struma ovarii is about equally frequent at any age.

PATHOGENESIS

The true thyroid nature of these growths appears to be well established. Histologically, especially with regard to their staining reactions, and chemically, with regard to their iodine and thyroid hormone content, these growths resemble thyroid tissue. As we shall see below they may even produce clinical manifestations of Graves' disease. It remains to be explained however, why thyroid tissue develops and even predominates in so many ovarian teratoids.

Cases in which both the cervical and the ovarian struma became toxic (see below) indicate that the ectopic thyroid

is sensitive to the same stimuli which influence normal thyroid tissue. In some cases, the cervical thyroid became enlarged and toxic after removal of a struma ovarii. These observations suggest close (hormonal?) correlations between orthotopic and ectopic thyroids.

CLINICAL COURSE

Usually the symptomatology of struma ovarii does not differ from that of other ovarian teratoids, unless there is hyperthyroidism. Quite frequently, however, they produce ASCITES.

Manifestations of severe THYROTOXICOSIS, such as an increased B.M.R., tremor and tachycardia have been noted in about 5-6% of all struma ovarii cases. After ovariectomy, the thyrotoxicosis subsided.

DIAGNOSIS

The clinical diagnosis of struma ovarii is usually impossible if there is no hyperthyroidism. If however, an

ovarian tumor (especially a dermoid) appears in a woman who suffers from Graves' disease and yet has a normal thyroid, the diagnosis of struma ovarii must be considered. It is well to remember, furthermore, that several of the ovarian strumas which caused Graves' disease, developed in patients whose cervical thyroid also became toxic. In these, removal of the cervical thyroid produced but temporary improvement and final cure followed only after subsequent removal of the ovarian struma.

THERAPY

The therapy of choice in all cases of struma ovarii is REMOVAL OF THE GROWTH and this almost invariably necessitates complete ablation of the gonad itself. In view of the ever present danger of malignant or toxic transformation in ovarian thyroid tissue, conservative measures and delay are to be avoided.

OVARIAN CHORIONEPITHELIOMAS (CHORIONCARCINOMAS)

DEFINITION

The chorionepithelioma of the ovary is a carcinoma characterized by its great morphologic and functional resemblance to placental tissue. Its characteristics are :

(1) The tumor is composed of both syncytium and Langhans cells. These are arranged in masses or cords with Langhans cells inside and the syncytium peripherally.

(2) The tumor has no connective tissue stroma or vascular supply of its own, but its cells surround and penetrate maternal blood vessels.

(3) There is coagulation of tissue, hemorrhage and necrosis.

CLASSIFICATION

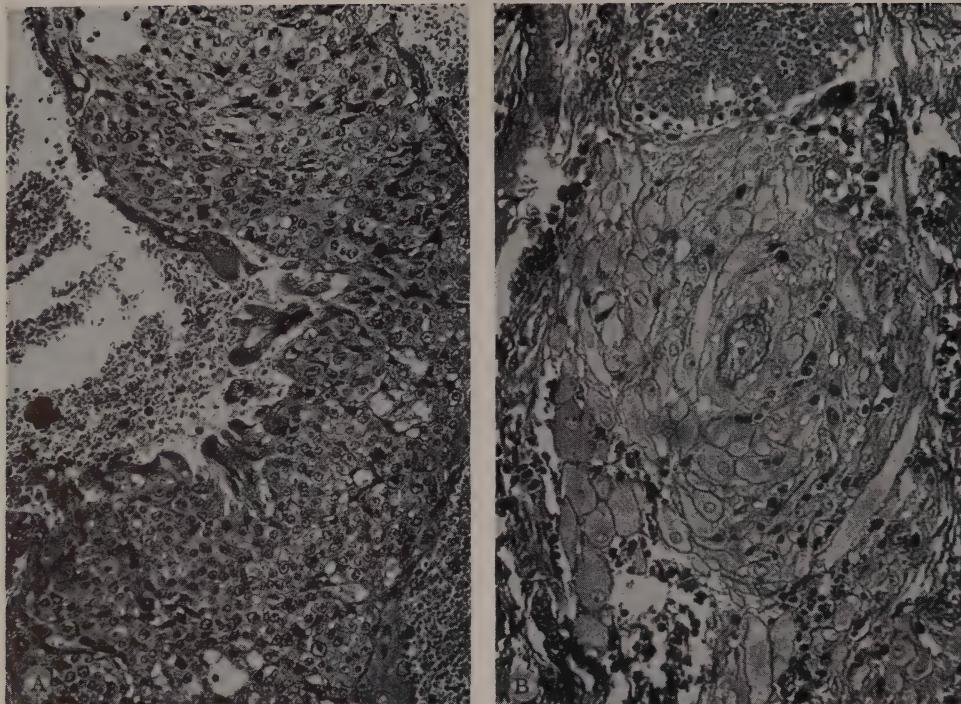
The chorionepitheliomas of the ovary are usually classified on the basis of their PATHOGENESIS as follows :

■ (1) OVARIAN CHORIONEPITHELIOMAS RESULTING FROM PREGNANCY.

(a) From an ovarian pregnancy.
(b) From a chorionepithelioma of the uterus which developed following pregnancy. In such cases, the original uterine tumor may be aborted before the ovarian neoplasm is noticed, thus rendering it difficult to recognize the secondary nature of the ovarian growth. A secondary chorionepithelioma may also arise from an ectopic pregnancy in the oviduct or the pelvic peritoneum.

(2) OVARIAN CHORIONEPITHELIOMAS WHICH ARE TERATOID TUMORS with one-sided development of chorionic elements, sometimes to the exclusion of other tissues.

(3) PRIMARY "COMMON" OVARIAN CARCINOMAS WITH TROPHOBlast-LIKE PROLIFERATIONS. It is doubtful whether these should be listed as a subdivision



Chorionepithelioma. — A. Part of a primary chorionepithelioma of the ovary. Note Langhans cells, bordered by large syncytial masses forming a villus. — B. Another region of the primary ovarian chorionepithelioma shown in Fig. A. Note perivascular decidual cells in the ovary.

(Courtesy of Dr. L.-C. Simard.)

of the true ovarian chorionepitheliomas, although some prominent investigators regard it as such.

PATHOLOGIC ANATOMY

The outstanding macroscopic characteristics of chorionepitheliomas are their red color and brittle or spongy texture. Their microscopic features have already been enumerated in connection with the definition of these neoplasms.

It is noteworthy that the contralateral ovary may be transformed into a mass of cystic corpora lutea, probably as a result of the excess gonadotrophin formed by the neoplastic cells.

Since chorionepitheliomas are highly malignant, they tend to produce numer-

ous metastases in various organs and to invade adjacent structures.

INCIDENCE

If secondary ovarian chorionepitheliomas are excluded, this is probably one of the rarest ovarian neoplasms. It tends to occur with approximately equal frequency in all age groups.

PATHOGENESIS

The various pathogenic possibilities which can lead to the formation of ovarian chorionepitheliomas, have been outlined in the part dealing with the classification of these tumors. There is, apparently, no single pathogenic mechanism which could explain all relevant cases. Some are evidently primary

growths originating in the ovary, either from the chorionic villi of an ovarian pregnancy, or from the teratomatous transformation of a parthenogenetic ovum in which chorionic elements grow to the partial or complete exclusion of all other tissues. In support of this view, it has been emphasized that in the guinea-pig, chorionepithelioma-like structures appear to arise from parthenogenetically developing ova. Chorionepitheliomas in prepubertal children and virgins show that this neoplasm can develop without a foregoing pregnancy.

CLINICAL COURSE AND COMPLICATIONS

Apart from the non-specific local manifestations of a rapidly growing malignant ovarian tumor, the clinical course of chorionepitheliomas is not very characteristic. In some cases, irregular and prolonged UTERINE HEMORRHAGES call attention to a possible endocrine disturbance, but since so often chorionepitheliomas develop following normal or ectopic pregnancy, the vaginal bleeding is usually ascribed to placental remnants.

In children, signs of PRECOCIOUS PUBERTY such as precocious menstruation and breast development, often result from the abnormal production of gonadotrophic and folliculoid hormones by the neoplastic, chorionic tissue.

In adult women, PREGNANCY CHANGES (e.g., secretion of colostrum, discoloration of the vaginal mucosa and enlargement of the uterus) ensue, due to hormones produced directly by the tumor, or by the corpus luteum cysts which are secondarily formed under the influence of the excess chorionic gonadotrophins.

The most important complications of chorionepitheliomas are hemorrhages from this highly vascular neoplasm or infiltration into adjacent organs, as well as metastases into distant structures.

DIAGNOSIS

The diagnosis of chorionepitheliomas is mainly based upon the local signs of an ovarian tumor in combination with evidence of excess sex-hormone production. The urine usually contains large amounts of chorionic gonadotrophin (LH) and folliculoids. However, urinary bioassays do not permit differentiation with certainty of the chorionepithelioma from hydatidiform moles and normal or ectopic gestation.

Unless pregnancy can be excluded with certainty, as in prepubertal children and virgins, ovarian chorionepitheliomas are clinically often indistinguishable from tubal or ovarian gestations. Since all these conditions require immediate surgical attention, such diagnostic errors are without serious consequences for the patient.

PROGNOSIS AND THERAPY

The PROGNOSIS of ovarian chorionepitheliomas is extremely grave, since this tumor is very malignant. Unless the neoplasm is surgically removed before it produces metastases, the outcome is invariably fatal within a short time. Even after radical surgical removal, fatal metastases and recurrences are common. The urinary gonadotrophin titer should always be checked repeatedly following the operation, since it is a good indicator of the continued presence of chorionepithelioma cells.

The THERAPY of choice is early radicle removal, followed by deep X-ray treatment, but even patients so treated can rarely be saved.

OVARIAN COMMON CYSTS

The common cysts of the ovary are of great practical importance for the gynecologist, but of comparatively little endocrinologic interest. They are not known to produce any specific hormones, nor is their development conditioned by endocrine stimuli. We mention them here only because they affect an endocrine gland, the ovary.

Most current CLASSIFICATIONS distinguish between the serous and the pseudomucinous ovarian cystomas, on the basis of the epithelium which lines the individual cyst cavities. The serous cysts or cystomas are lined by a low, cuboidal, or flattened epithelium, which produces a watery secretion. The pseudomucinous cystomas, on the other hand, possess high columnar "pseudomucin"-producing lining cells, similar to those of the large intestine. Hence,

their cavities are filled by a viscous mucin-like secretion. Both these types can be further subdivided according to the presence of various papillary excrescences in their cavity or on the outer cyst surface.

Little is known about the PATHOGENESIS of these neoplasms, but they are probably formed from embryonic remnants.

Their CLINICAL COURSE is not characterized by any endocrinologic abnormality. They rarely destroy the ovary sufficiently to cause signs of ovarian deficiency. They are important mainly because of the disturbances caused by their physical presence in the abdomen, and the ever-existing possibility of their malignant transformation. Some of the largest tumors occurring in man belong to this group.



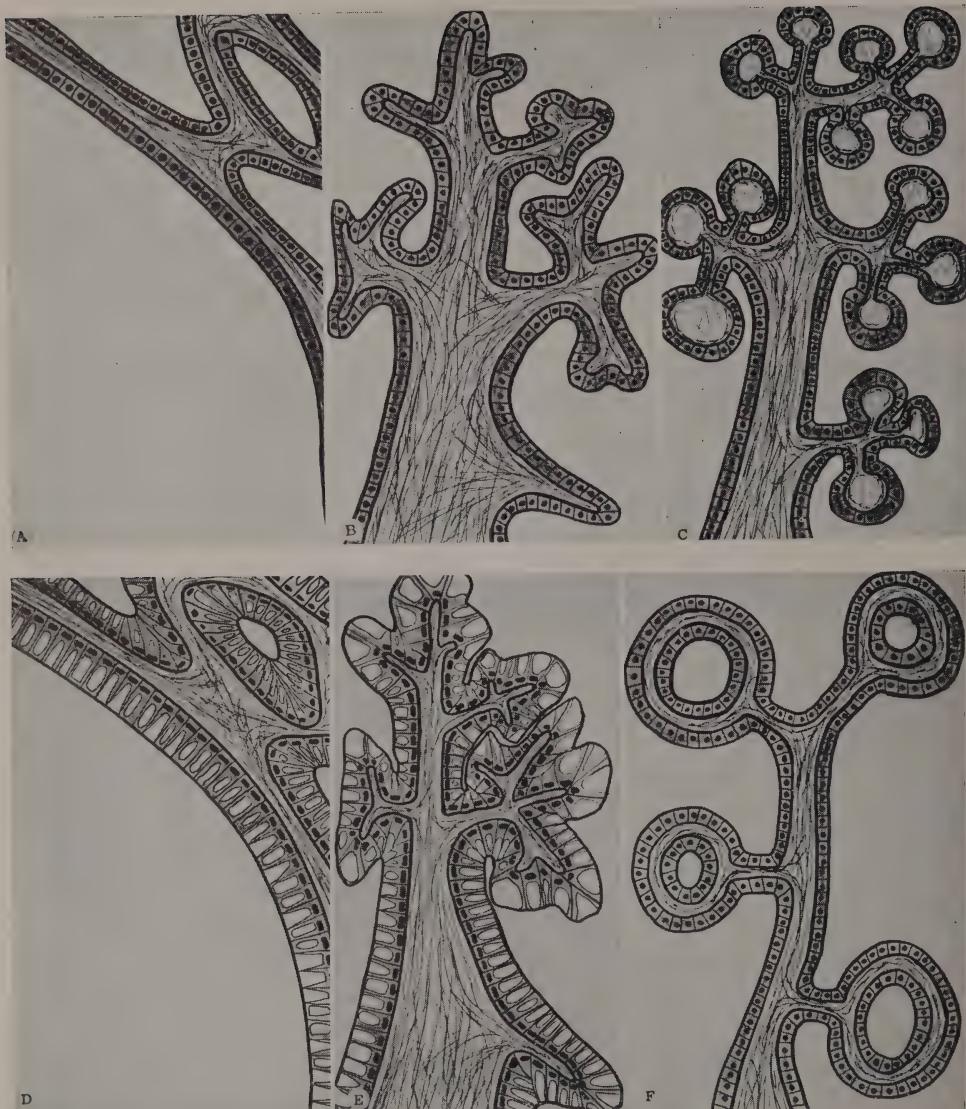
Internal, racemose, serous papilloma. Left side of field is occupied by normal ovarian tissue with follicles. On the right side there is a little, serous, unilocular cystoma from whose inner surface a racemose, small papilloma arises near the ovarian attachment. Note that each papillary excrescence is transformed into a grape-like structure. (Very low magnification.)

(Courtesy of Dr. L. Berger.)



Common cysts. — A. Simple unilocular cyst arising from surface of the ovary. In this, as well as in all subsequent illustrations on this plate, the ovary is indicated by a dotted area, the germinal epithelium as well as the surface lining of cysts is represented by a heavy line, the inner lining of cysts and the surface of papillomas by a thin line. — B. Multilocular, mainly macrocystic, common cystoma with a small microcystic area in the left part of the picture. Note that on this level there are three distinct large cystic cavities, some partially subdivided by cristae which are remnants of originally complete septa. The large cysts are formed due to rupture of smaller cavities into each other and subsequent disappearance of the septa. The small microcystic area contains gland-like acini and tubules, sometimes referred to as parvilocular or adenomatous formations. New macrocysts are formed owing to increasing fluid secretion and distension within the microcysts. — C. Unilocular cysts with an internal papilloma. Note that the connective tissue of the papilloma tree arises from the stroma of the cyst wall. The point of origin of such papillomas is subject to great variations and not necessarily located near the ovary proper. — D. Unilocular cyst with two external papillomas. Usually external and internal papillomas develop simultaneously within the same tumor and it is believed that most - if not all - external papillomas arise from the inner lining of the cyst, subsequent to perforation. In some cases, however, as in that illustrated here, external papillomas arise without any sign of internal papillomatous proliferation. — E. Two papillomas arising directly from the ovarian surface. Note also minute individual papillæ to the right of the small papilloma. Such excrescences give a characteristic "velvet-like" aspect to the ovarian surface and represent the earliest stages in surface papilloma formation. — F. Large adenofibroma. Note prevalence of dense, thick connective tissue fibers. The epithelium lined spaces are parvilocular tubules which frequently branch and sometimes contain "intracanalicular" papillomatous proliferations. This tumor type is always serous, never pseudomucinous.

(After H. Selye: "Ovarian Tumors," Encyclopedia of Endocrinology, 1946.)



Common cysts. — Schematic drawings illustrating microscopic structure of the most important forms. — A. Common serous, partly macrocystic (lower left), partly microcystic or adenomatous (upper right) cystoma. — B. Common serous papilloma. This type is found on inner or outer surface of serous ovarian cysts; sometimes it arises directly from the surface of the ovary. — C. False racemous serous cystoma. Note grape-like arrangement of small "cystlets". The formation of these false cysts is merely due to localized stroma edema at the tip of the papillæ. There is no actual cyst cavity but merely free fluid accumulation in the individual "grapes". — D. Common pseudomucinous, partly macrocystic (lower left), partly microcystic or adenomatous (upper right) cystoma. — E. Common pseudomucinous papilloma. This type differs from the corresponding serous papilloma merely by virtue of the pseudomucinous lining epithelium. — F. True racemous cystoma. This type differs from the corresponding false racemous cystoma by virtue of the fact that each "grape" actually contains a separate cyst cavity lined by the same type of serous epithelium which also covers the surface of the papillæ.

(After H. Selye: "Ovarian Tumors," Encyclopedia of Endocrinology, 1946.)



Large ovarian cyst causing cachexia.

Note enormous enlargement of the abdomen, dilatation of veins in abdominal wall, marked cachexia and characteristic "facies ovariana".

(After G. Winter: Lehrbuch der Gynäkologischen Diagnostik, Leipzig, 1897.)

Multilocular pseudomucinous cystoma.

At low magnification note numerous independent cyst cavities filled by pseudomucinous fluid. The partitions between some of the cysts are incomplete. Papillomatous excrescences are noticeable on their surface.

(Courtesy of Dr. L. Berger.)



Pseudomucinous papilliferous cystoma. Typically pseudomucinous lining with pseudomucin strands and cast-off cell debris between excrescences.

(After H. Selye: "Ovarian Tumors," Encyclopedia of Endocrinology, 1946.)

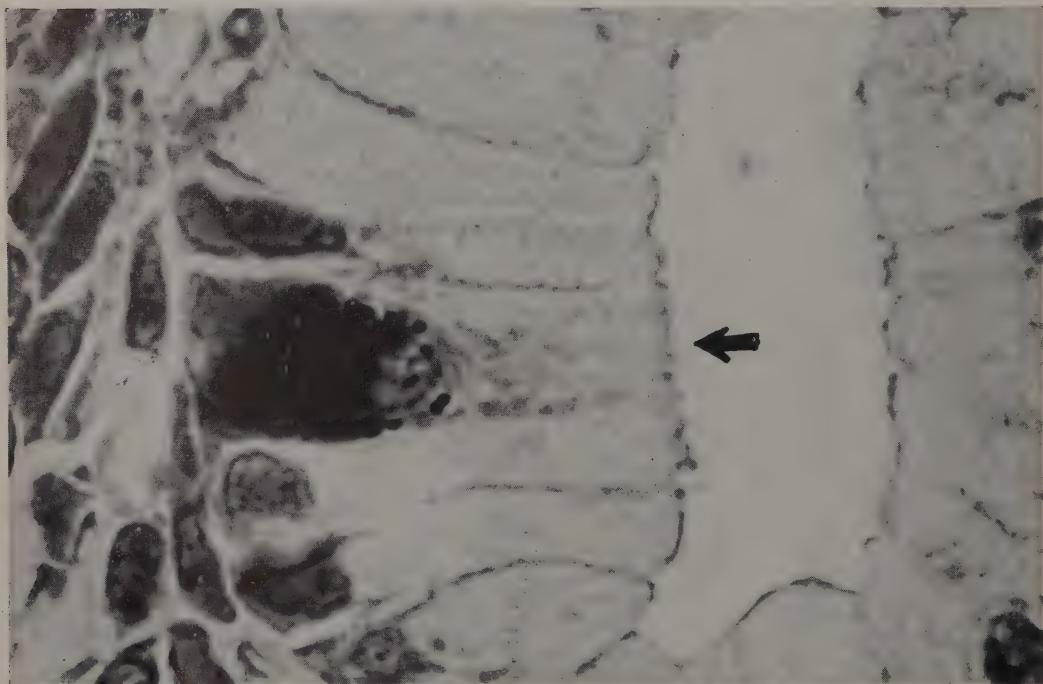


Pseudomucinous cystoma. Septum illustrating marked differences which may occur in the epithelium of two adjacent cavities. Cavity on left lined by cylindric epithelium containing practically no pseudomucin; on right high cylindric, pseudomucinous covering.

(Courtesy of Dr. P. Masson.)



Pseudomucinous cystoma. Most epithelial cells contain little or no pseudomucin but some are filled with secretion and assume a signet-ring like appearance. One cell (arrow) is in the process of discharging, several others (X) have been cast off into lumen.



Argentaffin cells in a pseudomucinous cystoma. Note the numerous argentaffin granules around and below the nucleus of one pseudomucinous cell (marked by arrow). (Courtesy of Dr. P. Masson.)

OVARIAN COMMON CARCINOMAS

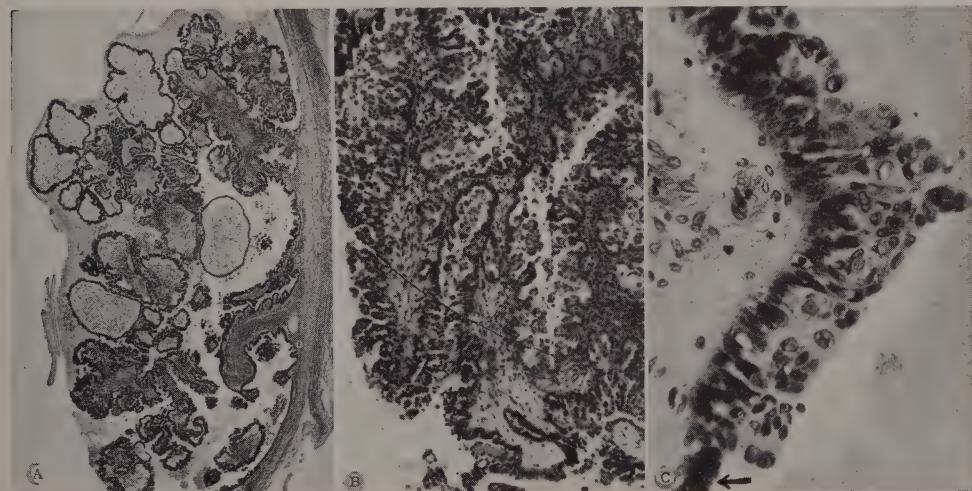
The common carcinomas — as the common cystomas — of the ovary are of little endocrinologic interest. They produce no hormones, nor are they due to endocrine abnormalities. However, they can completely destroy the gonad, causing signs of ovarian failure. In gynecology, the ovarian carcinomas are of great importance because of their comparative frequency and often great malignancy.

Most of the cystic PRIMARY OVARIAN CARCINOMAS are due to malignant transformation of originally benign cystomas. Correspondingly, we distinguish serous and pseudomucinous cystic ovarian carcinomas. A comparatively small percentage of the primary ovarian cancers are solid and these are probably due to direct malignant transformation

of epithelial ovarian components.

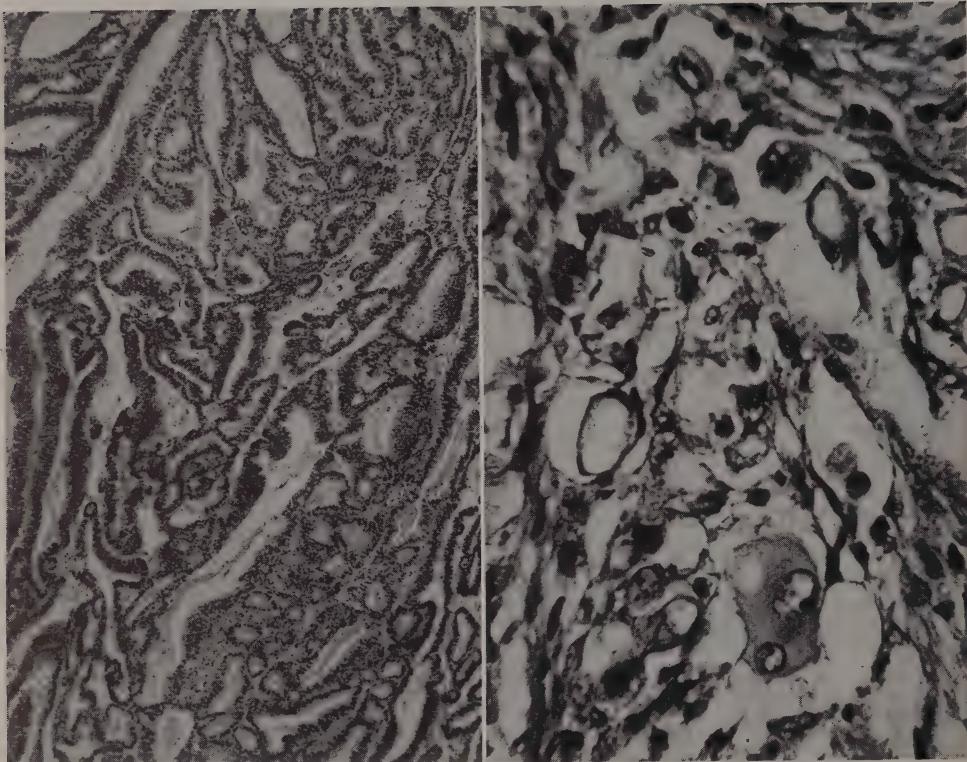
The ovary is also frequently the site of SECONDARY CARCINOMAS which may affect it by direct infiltration from adjacent organs, or due to deposition of metastatic tumor foci. Among the metastatic ovarian cancers, the so-called *Krukenberg tumors* are of special interest. These are characterized by typical "signet ring cells" which contain some mucous material. They are almost always bilateral and secondary to a mucous carcinoma of the gastrointestinal tract, usually the stomach.

The CLINICAL COURSE of ovarian carcinomas is chiefly dependent upon the rate of spread into adjacent organs, metastases, and in the case of secondary ovarian cancers, the progress of the primary neoplasm.



Serous, papillary carcinoma. — A. Small papillary excrescences on inner surface of a cyst wall. In this region, the tumor is remarkably typical. — B. Higher magnification of a region in the tumor shown in Fig. A. The invasive growth of the epithelium is clearly visible at this magnification. — C. A small field in the tumor shown in Figs. A and B. Note the regular and typical appearance of a few cells lining this excrescence (marked by arrow). The remainder of the lining is stratified, irregular and definitely of a malignant character. The shape and size of the nuclei are very irregular. (Oil immersion.)

(Courtesy of Dr. L. Berger.)

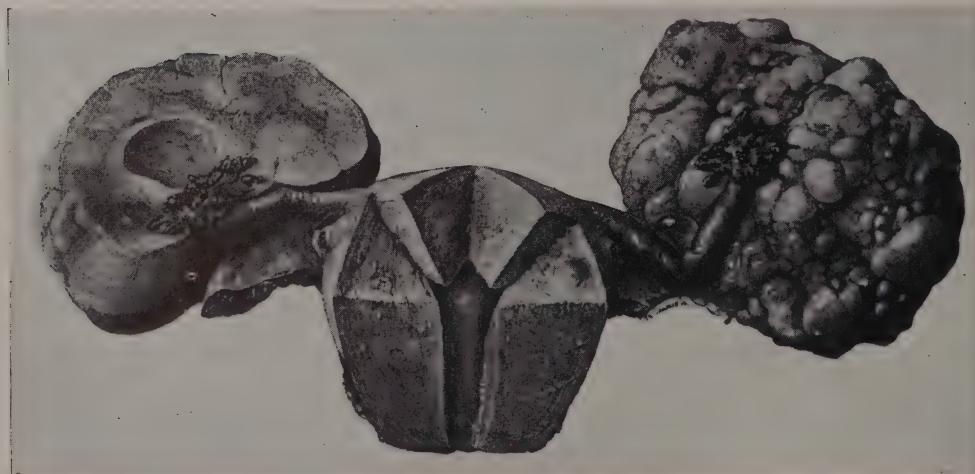


Adenocarcinoma. Purely gland-like solid carcinoma of the ovary.

(Courtesy of Dr. R.-T. Waugh.)

Krukenberg tumor. Signet-ring cells and one bi-nucleated giant cell in a Krukenberg tumor with a comparatively loose cell arrangement (as seen under very high magnification).

(After H. Selye: "Ovarian Tumors," Encyclopedia of Endocrinology, 1946.)



Gross appearance of a bilateral Krukenberg tumor together with the tubes and uterus. Note solid, nodular appearance characteristic of these tumors which are usually secondary to a carcinoma of the gastrointestinal tract.

(After E. Novak: Textbook of Gynecology, William & Wilkins, 1944.)

OVARIAN MESONEPHROMAS

The term "mesonephroma" is applied to ovarian tumors which resemble the embryonic mesonephros and have the following characteristics: (1) a glomerulus-like unit, consisting of a small cystic cavity which contains one capillary loop covered with columnar epithelial cells, whereas the cavity is lined with low endothelium-like cells; (2) solid areas consisting of proliferating, stellate endothelial cells, connected with each other by fine filiform projections. These two specific structures are definitely free from mucin. Limited

production of a mucinous secretion is occasionally found in the small cystic cavities, but the "mesonephroma" never shows mucinous secretion comparable to that of the pseudomucinous cystomas. The tumor may be benign or malignant.

Schiller (1939) who originally described this tumor, considered it as a special neoplastic entity but many pathologists doubt its mesonephric origin and regard it merely as one of the types of serous cystomas, or of teratoids. (See also: Teratoids of Testis.)

BRENNER TUMORS OR BRENNEROMAS

The Brenner tumor is a benign, partly fibrous and partly epithelial growth of the ovary. The relative proportions of epithelial and stroma tissue are extremely variable, but quite frequently the latter prevails. The epithelial component consists of solid or cystic aggregations of cells, not unlike those seen in the

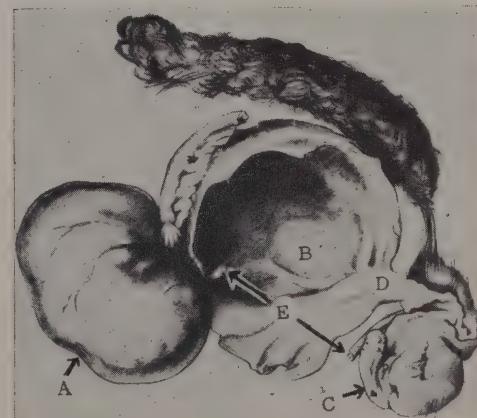
basal layers of stratified-squamous surface epithelia. If cystic cavities are formed, the innermost lining cells tend to produce pseudomucin.

They are comparatively rare neoplasms without known endocrinologic significance.

TERATOID TUMORS (DERMOIDS, SOLID TERATOMAS, EMBRYOMAS)

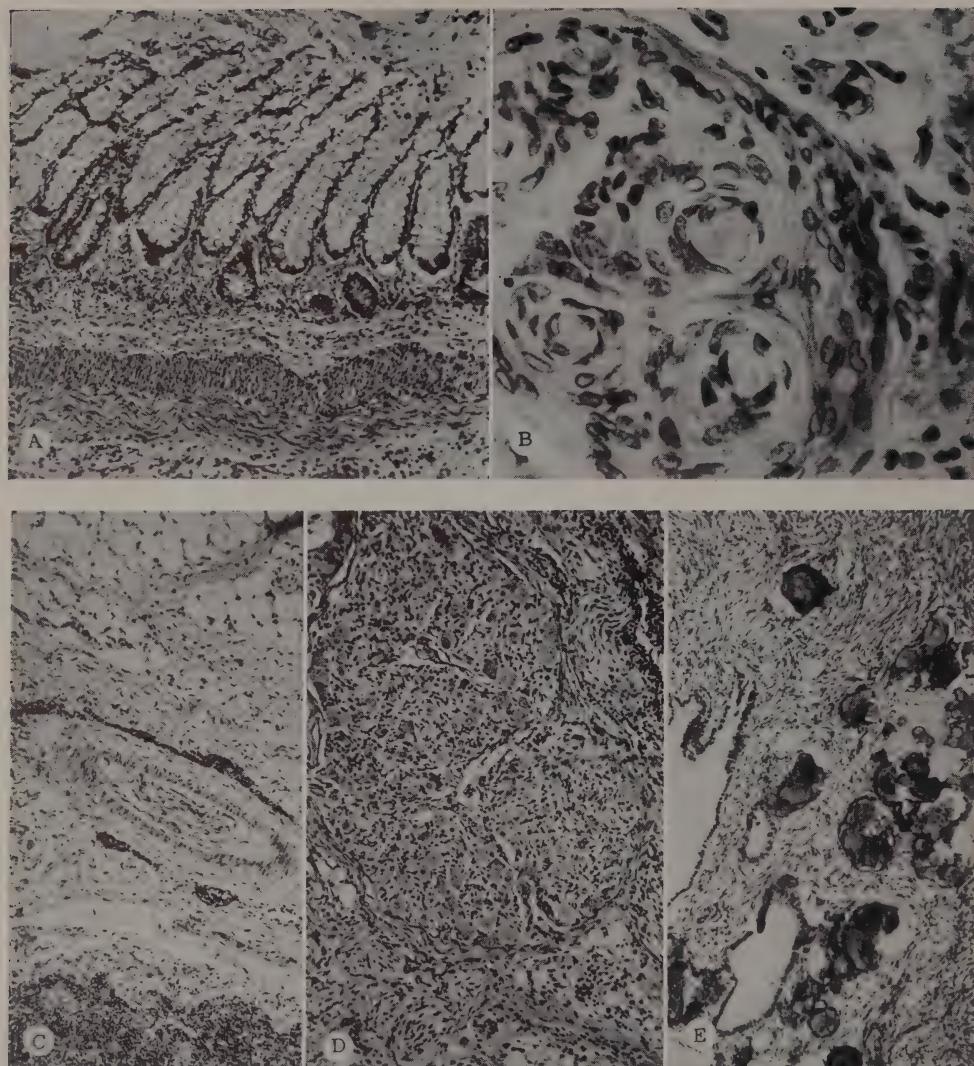
TERATOMAS are growths composed of tissues and organs of one, two or three germinal layers (monodermal, bidermal or tridermal types). The EMBRYOMA is composed of tissues derived from all three germinal layers, in more or less orderly imitation of a fetus. These growths differ therefore from simple, MIXED TUMORS which are composed of more than one type of irregularly arranged neoplastic tissue, named according to composition (e.g., "chondro-epithelioma," "osteochondro-myxosarcoma"). In the sense of these definitions, both the cystic and solid teratoid tumors of the ovary are teratomas. The DERMOID CYST deserves special consideration only because of its frequency.

Except when, in a complex embryo, one or the other endocrine tissue (e.g., thyroid, adrenal cortex) develops



Dermoid cyst with an almost completely formed fetus.

(A) "Enterocystoma", (B) dermoid cyst open, (C) embryo with female genital organs and long and abundant scalp hair. The embryo is connected with the wall of the dermoid by means of a stalk (D) but in addition to this an umbilical cord (E) connects it with the enterocystoma. Hence, the latter was interpreted as a yolk sac. The arrows (E) point to the stumps of the umbilical cord.
After G. Kaboth: Arch. f. Gynäk., 122, 803, 1924.)



Dysembryoma. — A. Portion of a particularly polymorph, microcystic teratoid. In this field a well-differentiated and regular intestinal wall is clearly distinguishable. Note numerous mucus-producing cells in intestinal crypts, lamina propria mucosæ and two layers of smooth muscle cells, the latter constituting the muscular wall of the intestinal segment. The region is highly reminiscent of large intestine. — B. Another field of the neoplasm illustrated in Fig. A. Note the presence of several structures reminiscent of Hassall's corpuscles. — C. Yet another field of the tumor represented in Fig. A. Note large lymph tissue accumulation in lower part of the field and infiltration of lymphatic elements in periarterial spaces surrounding the vessel in the center of the field. Above this, normal fat tissue is visible. — D. Yet another field of the tumor illustrated in Fig. A-C. Large ganglion cell accumulation with fat tissue in the lower and slight lymphatic infiltration in upper, right part of this field. — E. *Dermoid Cyst.* Part of a dermoid in which the epithelium lining the cyst cavity is serous and definite calcified psammoma bodies are visible within the stroma. This region is very reminiscent of serous psammomatous cystomas. The remainder of the cyst was lined by stratified, squamous epithelium, characteristic of dermoids.

(After H. Selye: "Ovarian Tumors," Encyclopedia of Endocrinology, 1946.)