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POLYCYSTIC OVARIAN DISEASE*

BY

N. M. DUIGNAN†

Department of Clinical Endocrinology
Birmingham and Midland Hospital for Women

Summary

Sex hormone binding globulin (SHBG) capacity was reduced in 9 of 31 patients with polycystic ovarian (PCO) disease and the mean level in PCO patients was significantly less ($p<0\cdot001$) than normal. Serum testosterone levels were elevated in 21 of 32 PCO patients and the mean level was significantly elevated ($p<0\cdot001$). Serum androstenedione values were raised in 17 of 31 patients and the mean value was also significantly raised ($p<0\cdot001$). Serum dehydroepiandrosterone sulphate (DHAS) concentrations were elevated in only 2 of 14 patients. Urinary 17-oxo and 17-oxogenic steroids were normal in all patients studied. Basal follicle-stimulating hormone (FSH) and luteinizing hormone (LH) levels were normal but LH release following injection of luteinizing hormone-releasing hormone (LH-RH) was enhanced. A highly significant negative correlation ($r=-0\cdot449$; $p<0\cdot01$) was found between the logarithm of testosterone and the logarithm of LH levels. Serum prolactin concentrations were elevated in 4 of 21 PCO patients. Thyroid-stimulating hormone (TSH) values were normal. Eighteen of 20 patients ovulated following treatment with clomiphene and nine became pregnant. Five of 12 of patients treated with oestrogen/progesterone preparations noticed an improvement in their hirsutism. It is suggested that the normal cyclical release of LH is inhibited in PCO disease by a negative feedback by androgens to the hypothalamus or the pituitary, and that wedge resection should be reserved for patients in whom other forms of treatment have failed.

THE pathogenesis of polycystic ovarian (PCO) disease is far from clear. For one thing, histological changes similar to those found in polycystic ovaries have been produced in the gonads of animals treated with androgens (Barraclough, 1961); for another, polycystic ovaries have been found in some cases of adrenogenital syndrome, in Cushing's disease, and in women with virilizing adrenal or ovarian tumours (Abu-Haydar *et al*, 1954; Sizonenko *et al*, 1972; Goldzieher, 1973).

The majority of patients with PCO disease have been found to have increased androgen secretion from the ovaries or the adrenal glands and also to have abnormal gonadotrophin release. It is uncertain, however, whether these clinical abnormalities are primarily due to abnormal steroidogenesis, which produces elevated levels of circulating androgens along with secondary changes in gonadotrophin secretion, or to a primary hypothalamic disorder producing abnormal gonadotrophin release which, in turn,

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† Present address: Consultant Obstetrician and Gynaecologist, Coombe Lying-in Hospital, Dublin 8.

causes both cystic changes in the ovaries and abnormal steroidogenesis.

In the present investigation, a study was made of circulating androgens with particular regard to the effect which abnormal androgen secretion might exert on hypothalamic-pituitary function; in addition, the effect of different forms of therapy was assessed. Thirty-four patients, in whom the diagnosis of PCO disease was confirmed by the macroscopic appearance of the ovaries at laparoscopy, were studied.

Androgens

The androgens secreted in PCO disease were examined by estimating serum sex hormone binding globulin (SHBG) capacity determined by the method of Rudd *et al* (1974), and serum levels of testosterone, androstenedione and dehydroepiandrosterone sulphate (DHAS) measured by radioimmunoassays (Duignan *et al*, 1975; Smith *et al*, 1975), as well as by the estimation of urinary 17-oxo and 17-oxogenic steroids (Gray *et al*, 1969) and urinary 11-deoxy and 11-oxy-17-oxogenic steroids (Few, 1968). In addition, a short-term dexamethasone suppression test, using 0·5 mg six hourly for 48 hours, was performed on 24 of the 34 patients.

Sex hormone binding globulin (SHBG) capacity

Between 1 and 3 per cent of testosterone circulates in the free, and biologically active state. The major fraction of the remainder is bound to SHBG (Mercier *et al*, 1966), which is a plasma protein having a high affinity with, but a low capacity for, testosterone, oestradiol-17β and closely related steroids with a 17-β ol configuration (Mercier-Bodard *et al*, 1970).

Serum SHBG capacity was determined in normal men and women, in pregnant and postmenopausal women and in patients with idiopathic hirsutism or PCO disease (Fig. 1). The mean value found in ten normal males was $5{\cdot}0\times10^{-8}$ mol/l with a narrow range between 4·5 and $5{\cdot}5\times10^{-8}$ mol/l. The levels found in 27 normal, regularly menstruating women ranged from 4·2 to $9{\cdot}2\times10^{-8}$ mol/l and the mean level of $6{\cdot}9\times10^{-8}$ mol/l was significantly higher ($p<0{\cdot}001$) than that of normal males. Nine prepubertal children of both sexes were found to have widely varying levels but the mean level

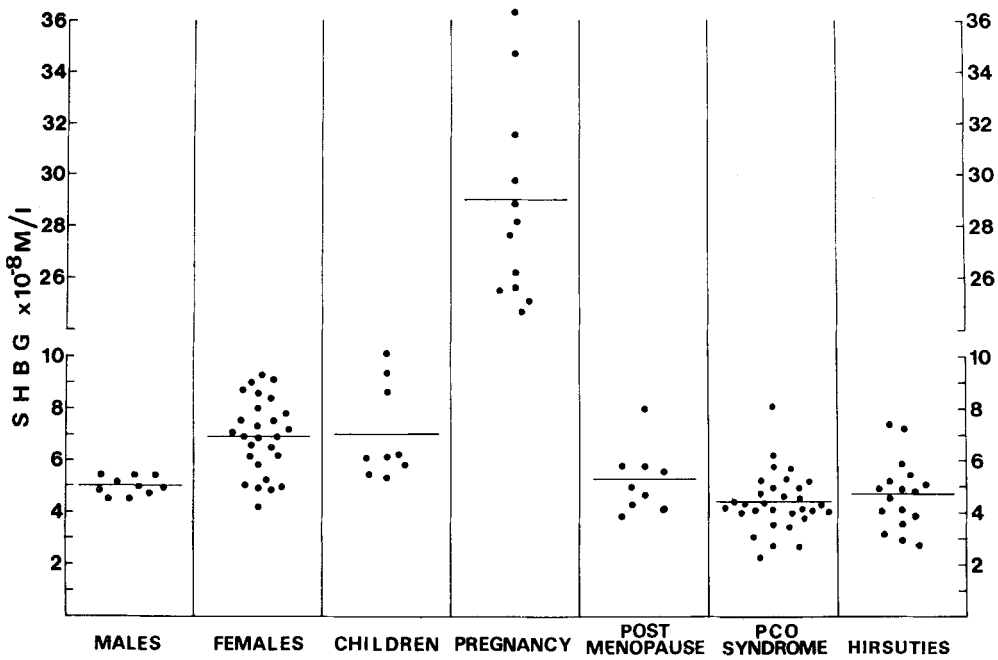

FIG. 1

Serum SHBG capacity in normal subjects as well as in patients with idiopathic hirsutism and PCO disease.

was similar to that of adult females. Eleven women examined during the third trimester of pregnancy were found to have values more than four times greater than non-pregnant women. Nine postmenopausal women, aged between 53 and 76 years, were found to have values significantly lower ($p<0{\cdot}01$) than those of premenopausal women, but similar to those of normal males. Nine of 31 patients with PCO disease had values more than 2SD below the mean of normal women and the mean level of the PCO patients was significantly less ($p<0{\cdot}001$) than that of normal women; similarly, the mean level in patients with idiopathic hirsutism was significantly reduced ($p<0{\cdot}001$).

The low SHBG capacity in PCO disease may have been due to increased androgen production as it has been shown that testosterone diminishes the synthesis of SHBG in the liver (Vermeulen *et al*, 1969). Burke and Anderson (1972) suggested that SHBG played a specific biological role in regulating the ratio of unbound testosterone to unbound oestradiol: if this hypothesis is correct then the PCO patients had a disproportionate amount of free and biologically active testosterone in their circulation.

Testosterone, androstenedione and dehydroepiandrosterone sulphate (Fig. 2)

Serum testosterone concentrations ranged from 0·4 to 6·1 nmol/l. Sixty-five per cent of the PCO patients at some time had elevated values, and the mean level of the group was significantly greater ($p<0{\cdot}001$) than that of normal women. When repeated samples were collected from individual patients, at intervals of between three days and three months, the levels were found to fluctuate considerably in 66 per cent of the patients. Serum androstenedione levels also varied considerably ranging from 1·1 to 17·0 nmol/l. Fifty-four per cent of the patients had elevated values and the mean level was significantly greater ($p<0{\cdot}001$) than that of normal women. Serum DHAS concentrations were elevated in only 2 of 14 patients and the mean value was no different to that of normal women. This last finding is at variance to that reported by others (DeVane *et al*, 1975), but suggests that the adrenal glands were not the major source of excess androgen secretion in those particular patients.

Neither the raised testosterone nor the raised androstenedione levels correlated with the degree of hirsutism. This is not surprising because the concentrations measured by radioimmunoassay did not take account of the amount of bound and free androgen, nor did they account for the peripheral interconversion of androgens which occurs mainly in the liver, skin and blood. This interconversion accounts for between 33 and

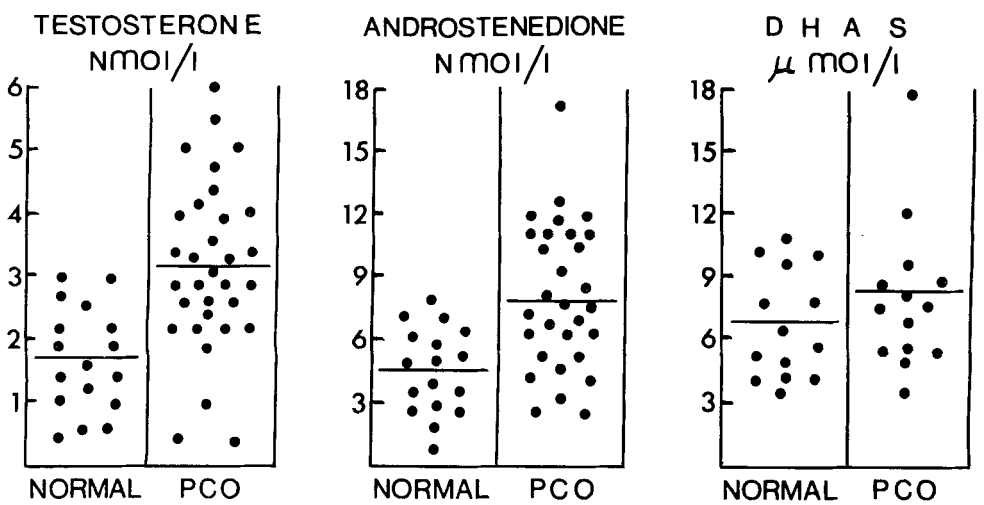

FIG. 2

Serum testosterone, androstenedione and DHAS levels in normal women and in PCO disease.

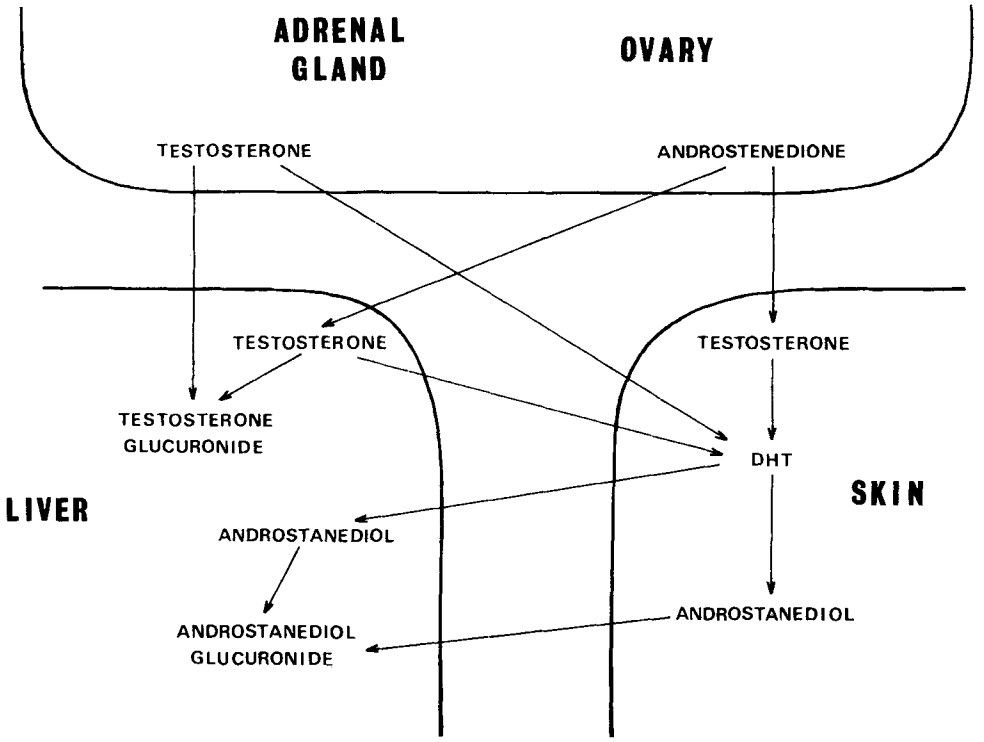

FIG. 3
Main pathways of peripheral interconversion of testosterone and androstenedione.

80 per cent of testosterone production (Rivarola *et al*, 1967; Abraham *et al*, 1969) and involves not only the transformation of weak androgens into more active substances but also the breakdown of inactive androgens into active ones.

The main pathways of peripheral metabolism of testosterone and androstenedione, secreted by the adrenal glands or the ovaries, are summarized in Figure 3. Testosterone may be converted in the liver to testosterone glucuronide (Gordon *et al*, 1971), or it may be transformed into the much more active androgen, dihydrotestosterone (DHT), in the skin (Wilson and Walker, 1969), and be subsequently excreted as a glucuronide of androstanediol. Similarly, androstenedione—a biologically weak androgen—may be transformed in the liver or converted to the highly active DHT in the skin. Thus, measurement of urinary or serum androstanediol levels would probably have afforded a more accurate index of androgenicity (Mauvais-Jarvis *et al*, 1973).

Dexamethasone suppression test

A dexamethasone suppression test was performed on 24 patients (Table I). Testosterone concentrations fell after dexamethasone in 71 per cent of patients and the mean degree of suppression among these subjects was 50 per cent. Similarly, androstenedione levels were reduced in 77 per cent of patients and the mean degree of suppression was 41 per cent. These results, at first, suggest that the adrenal glands were the major source of androgen secretion in those patients whose levels were lowered, but this inference is not necessarily correct. It has been stated that short term changes in androgen levels are largely independent of ACTH secretion (Ettinger *et al*, 1973); furthermore, the catheterization studies of Kirschner and Jacobs (1971) suggested that the ovaries could have been the main source of androgen secretion in some patients whose androgen levels were suppressed by dexamethasone.

TABLE I

Cortisol, testosterone and androstenedione concentrations in PCO disease before and after dexamethasone 0·5 mg 6-hourly for 48 hours

Patient No.	Cortisol (nmol/l)		Testosterone (nmol/l)		Androstenedione (nmol/l)	
	Before	After	Before	After	Before	After
1	303	30	5·7	6·9	12·0	–
2	140	30	3·6	2·1	6·0	14·9
3	303	145	1·5	2·2	5·5	8·0
4	552	–	2·4	1·3	4·2	1·2
5	585	385	6·2	2·8	4·8	1·8
6	571	276	5·5	1·2	7·5	–
7	378	82	2·7	3·1	11·7	8·4
8	634	30	2·1	1·4	2·4	3·6
9	214	30	1·3	0·5	10·0	4·1
10	568	30	0·9	1·5	3·0	1·4
11	530	33	3·5	1·5	11·0	4·1
12	318	41	4·7	3·8	4·8	3·5
13	432	30	2·2	2·4	7·9	3·4
14	386	110	4·1	1·5	9·1	7·2
15	358	55	0·9	0·4	7·1	1·8
16	304	30	2·5	0·9	10·4	4·9
17	248	30	3·4	2·2	6·6	4·8
18	620	247	3·3	2·1	9·7	8·0
19	358	82	4·1	1·3	6·1	5·0
20	745	82	2·0	0·4	6·9	1·7
21	254	30	4·0	4·9	15·0	34·0
22	598	64	0·4	1·7	2·3	3·9
23	–	–	4·0	1·3	5·6	4·5
24	248	138	2·3	1·6	11·6	9·0

Cortisol: nmol/l ÷ 27·6 = μg/dl. Testosterone: nmol/l ÷ 0·035 = ng/dl. Androstenedione: nmol/l ÷ 0·035 = ng/dl.

Urinary steroids

Urinary 17-oxo and 17-oxogenic steroids, measured in 20 of the 34 patients, were normal in all instances even though 13 patients had elevated serum androgen levels. Urinary pregnanediol levels were elevated in only 3 of the 20 patients, and dexamethasone lowered serum androgen levels in these patients.

HYPOTHALAMIC-PITUITARY FUNCTION

The effect that the raised androgen levels might exert on hypothalamic-pituitary function was studied in detail:

(1) by measuring basal serum FSH and LH concentrations,
(2) by determining FSH and LH release following injection of synthetic luteinizing hormone-releasing hormone (LH-RH),
(3) by observing the effect of pretreatment with oestradiol and progesterone on the subsequent response of patients to LH-RH,
(4) by studying the oestrogen feedback mechanisms,
(5) by measuring prolactin levels, and
(6) by estimating concentrations of thyroid stimulating hormone (TSH) both before and after injection of 200 μg of thyrotrophin-releasing hormone (TRH).

Basal FSH and LH levels (Fig. 4)

Serum FSH and LH concentrations were determined by specific radioimmunoassay (Duignan *et al*, 1975). Basal FSH levels were similar to those of normal women while only 2 of the 34 patients were found to have elevated LH values. This latter finding is at variance with the report of Yen *et al* (1970) but may partly be due to the fact that the concentrations measured

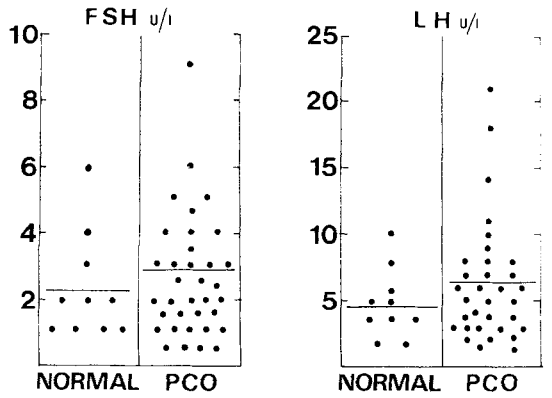

FIG. 4

Basal serum FSH and LH levels in normal women and in PCO disease.

in the present study were isolated values which may have been subject to the day-to-day variation noted by Yen *et al* (1970).

No correlation was found between any of the sex steroids and FSH levels, nor between oestradiol-17β, progesterone or androstenedione and LH levels. However, when testosterone and LH concentrations were simultaneously measured on 33 samples from 26 patients, a highly significant negative correlation ($r = -0{\cdot}449$; $p<0{\cdot}01$) was found between the logarithm of testosterone and the logarithm of LH concentrations (Duignan, 1974). This suggests that the elevated testosterone values found in PCO disease might be interfering with the normal release of LH by exerting a negative feedback either on the hypothalamus or the pituitary gland (Duignan *et al*, 1975).

FSH and LH release following LH-RH

The release of FSH and LH following injection of 100 μg LH-RH, has previously been reported (Duignan *et al*, 1974 and 1975). Although FSH release was similar to that of normal women, the patients with PCO disease showed an enhanced LH response to LH-RH when compared with normal controls studied in the early follicular phase of the cycle (Fig. 5).

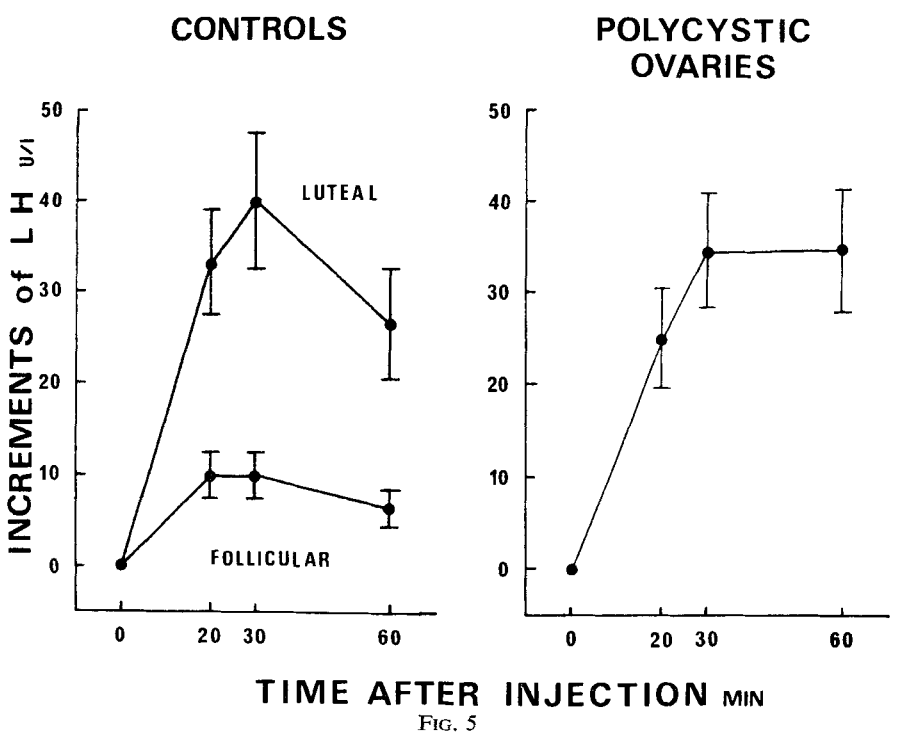

FIG. 5

Increments (Mean±SEM) of LH following injection of 100 μg LH-RH in normal women during the early follicular and luteal phases (N = 10) and in PCO disease (N = 23).

The release of LH after LH-RH was similar to that found in the normal luteal phase although oestradiol-17β and progesterone levels were akin to those found in the early follicular phase.

Modification of LH-RH response by pretreatment with oestradiol or progesterone

Pretreatment with oestradiol or progesterone altered the subsequent response of PCO patients to LH-RH although the modification was not always similar to that seen among normal women (Shaw *et al*, 1975*b*). The augmentative effect of pretreatment with oestradiol upon LH release was significantly less ($p<0{\cdot}02$) than that induced in normal women, whereas progesterone pretreatment produced a significantly greater ($p<0{\cdot}001$) LH release in PCO disease than in normal subjects during the early follicular phase (Shaw *et al*, 1976). These results suggest not only that the positive feedback mechanisms function normally in PCO disease but, also, that the system is already partly primed by oestradiol or some other oestrogen such as oestrone which has been found to be elevated in PCO disease (De Vane *et al*, 1975).

Oestrogen feedback mechanisms

The feedback mechanisms which help to control the midcycle surge of LH were further studied by measuring serum FSH and LH levels immediately before and 8, 24, 48, 56, 72, 86 and 96 hours after an intramuscular injection of 1 mg oestradiol benzoate (Shaw *et al*, 1975*a*). A positive feedback release of LH was found in 15 of 19 patients studied, thereby implying that the mechanisms responsible for the midcycle release of LH and FSH were functioning normally in the majority of patients with PCO disease.

Prolactin

Serum prolactin concentrations, measured by radioimmunoassay (Glass *et al*, 1975), were elevated in 4 of 21 patients (Fig. 6) but no correlation was found between oestradiol-17β, progesterone, testosterone, androstenedione or SHBG capacity and prolactin levels. In a previous study Talas *et al* (1968) reported elevated prolactin levels in PCO disease but the present results suggest that raised prolactin values do not play a major part in the pathogenesis of the syndrome.

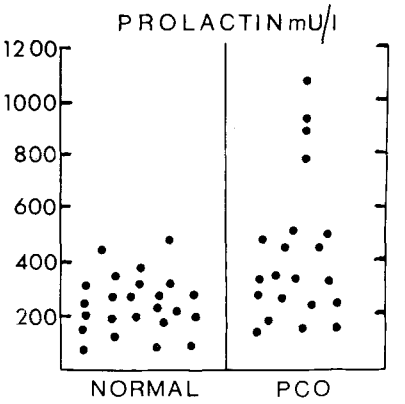

FIG. 6

Serum prolactin concentrations in normal women and in PCO disease.

Thyroid stimulating hormone

Basal TSH concentrations in 10 patients with PCO disease were similar to those of normal women, as was the release of TSH following injection of 200 μg TRH which suggest that thyroid function in PCO disease is normal.

These results imply that the abnormality of hypothalamic-pituitary function in PCO disease only affects gonadotrophin release. The negative correlation found between LH and testosterone suggests that the elevated androgens may exert a negative feedback effect to the hypothalamus or the pituitary, thereby interfering with the normal release of LH. Furthermore, the fact that the positive oestrogen feedback mechanism functions normally in the majority of patients suggests that the abnormality of gonadotrophin release must occur at an early stage of the cycle, thereby interfering with the normal maturation of ovarian follicles; this abnormality could be due to a malfunction of the cyclic centre within the hypothalamus thus leading to a failure of the normal stimulation of ovarian follicles.

TREATMENT

The treatment of PCO disease should aim at inducing ovulation in infertile patients and at reducing androgen production in those complaining of hirsutism. Ovulation can be induced

by wedge resection of the ovaries (Stein and Leventhal, 1935; Bailey, 1937); by administration of clomiphene (MacGregor *et al*, 1968) or, in some cases, by a combination of clomiphene and HCG (Shaw *et al*, 1975*a*); by treatment with FSH and HCG (Crooke *et al*, 1963) or sometimes by corticosteroids (Smith *et al*, 1965). Hirsutism may be improved by treatment with corticosteroids (Ettinger *et al*, 1973), oestrogen preparations (Ettinger *et al*, 1973) or antiandrogens (Ismail *et al*, 1974).

In a review of 1079 cases of PCO disease Goldzieher (1973) recorded that wedge resection induced ovulation in 80 per cent of patients and that 63 per cent became pregnant. This operation may, however, stimulate the formation of adhesions around the ovaries and tubes (Logan Edwards *et al*, 1975) and the procedure should be reserved for patients to whom medical treatment is either impossible or has failed. Thus, wedge resection was not used as a form of treatment in the present investigation.

Clomiphene is nowadays well established as the principal agent for inducing ovulation in PCO disease. A small percentage of patients, however, will not ovulate and Shaw *et al* (1975*a*) suggested that a failure of the positive oestrogen feedback mechanism may be present in those patients who do not respond to clomiphene; ovulation can be successfully induced in such patients by the additional administration of HCG, provided clomiphene has induced ovarian maturation as judged by a rise in urinary or plasma oestrogen levels. Twenty of the patients included in the present study were treated with clomiphene: ovulation was induced in 90 per cent and 45 per cent became pregnant. Furthermore, one of the two patients who did not respond to clomiphene was found to have an abnormality of the positive oestrogen feedback mechanism and to ovulate when HCG was given in addition to clomiphene (Shaw *et al*, 1975*a*).

Induction of ovulation by administration of FSH and HCG should be considered only when other forms of therapy have failed. These patients are sometimes highly sensitive to FSH and ovarian hyperstimulation may occur (Crooke *et al*, 1963). Only one of the 34 patients included in the present study was treated with FSH and HCG; she became pregnant following the third course of treatment.

Corticosteroids have the advantage of not only lowering ACTH-dependent androgen secretion (Ettinger *et al*, 1973), but also of stimulating gonadotrophin release (Butt *et al*, 1963), and Smith *et al* (1965) found the administration of steroids to induce ovulation as effectively as wedge resection of the ovaries. Hirsutism has been found to be reduced in 32 per cent of patients treated with dexamethasone alone, and in 50 per cent of patients treated with dexamethasone and a combination of ethinyl oestradiol and medroxyprogesterone (Ettinger *et al*, 1973). Three patients included in the present study were treated with corticosteroids; one noticed a slight improvement in hirsutism and another became pregnant.

The administration of oestrogen/progestogen preparations provides a simple and effective method of inhibiting ovarian steroid production (Goldzieher, 1973); furthermore, oestrogens stimulate the synthesis of SHBG in the liver and also inhibit the enzyme 5α-reductase (Mauvais-Jarvis and Bercovici, 1967) which converts testosterone to the more biologically active DHT in the skin. Five of 12 patients treated with oestrogen/progestogen preparations noticed an improvement in their hirsutism; this improvement, however, did not correlate with the increase in SHBG capacity.

Antiandrogens have recently been found to be even more beneficial than oestrogen/progestogen preparations in reducing hirsutism (Ismail *et al*, 1974); however, more studies of their value are required and they were not used in the present investigation.

Conclusions

The majority of patients with PCO disease have elevated serum androgen levels as a result of increased secretion by the ovaries or the adrenal glands. These raised androgens may interfere with the normal cyclical release of LH by inhibition at either pituitary or hypothalamic level (Fig. 7), thereby blocking the proper maturation of ovarian follicles. The present study did not, however, differentiate between those patients in whom the adrenal glands were the major source of androgen production and those in whom the ovaries were largely responsible, though the normal DHAS levels suggest that the adrenal glands play a minor role. Ovulation can

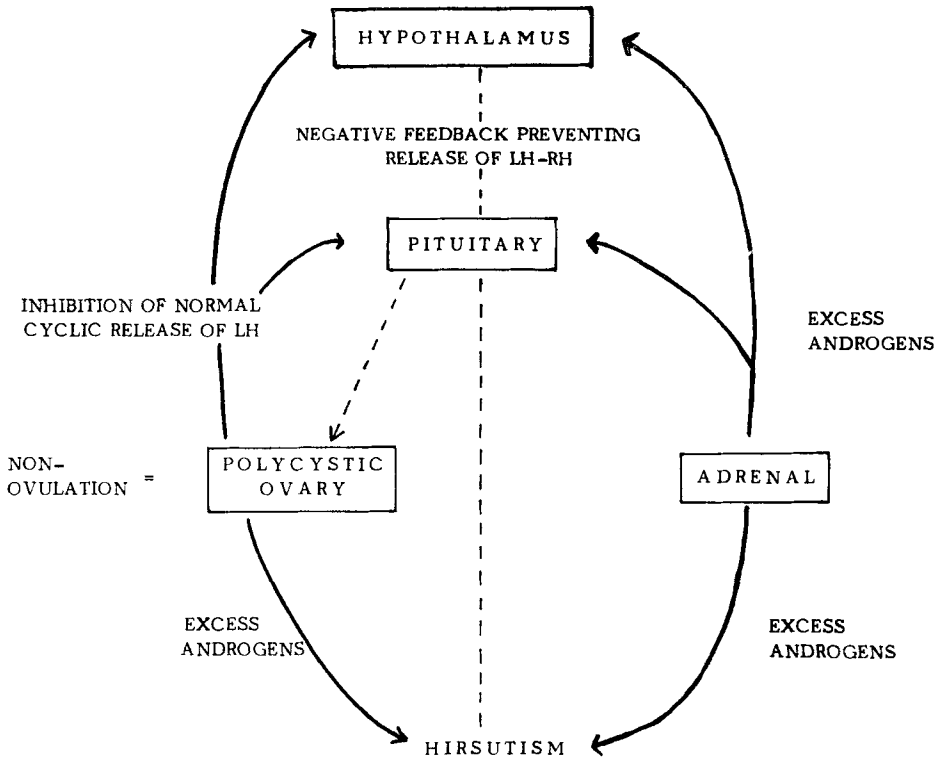

FIG. 7

Summary of effects of elevated androgens in PCO disease.

be satisfactorily induced by clomiphene or a combination of clomiphene and HCG in the majority of cases, and wedge resection should be reserved for patients in whom medical treatment has failed. On the other hand, patients complaining of hirsutism may well benefit from treatment with oestrogen/progestogen preparations or corticosteroids; the place for antiandrogen therapy remains to be resolved.

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