Steroids of Unnatural Configuration. The Absence of Long-Range Conformational Effects in Ring A Modified 20-Ketopregnanes1a,b

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The mother liquor solids consisted of about 60% product and 40% starting material.

In another experiment, the sulfur dioxide solution was added to the mixture of steroid, dimethylformamide, and collidine prior to the addition of benzoyl chloride. Only unchanged starting material was recovered. This observation has not been checked.

E. *p*-Nitrobenzoyl Chloride and Sulfur Dioxide.—A mixture of 16.7 g. (0.04 mole) of 16α-methyl-1,4-pregnadiene-11β,17α,21-triol-3,20-dione 21-acetate, 33 ml. of collidine, and 90 ml. of dimethylformamide at 10° was treated with a solution of 23.7 g. (0.128 mole) of *p*-nitrobenzoyl chloride in 10 ml. of dimethylformamide which also contained about 3% sulfur dioxide by weight. After 5 min. at 25–45° (initial heat evolution quite pronounced), the reaction mixture was worked up in the manner described for the benzoyl chloride run. The gum, which separated when the batch was added to water, solidified upon standing over the weekend. It was collected, washed with water, and recrystallized still wet, from 75 ml. of ethanol (hot filtration). There was obtained 6.55 g. (*ca.* 41%) of 16α-methyl-1,4,9(11)-pregnatriene-17α,21-diol-3,20-dione 21-acetate which melted at 212–215.5°. Paper-strip analysis of this product and its mother liquor revealed that essentially quantitative conversion had taken place.

4,9(11)-Pregnadiene-17α,21-diol-3,20-dione 21-Acetate [11-(9)-Anhydrocortisol Acetate].—A change of 60.0 g. (0.148 mole) of 4-pregnene-11β,17α,21-triol-3,20-dione 21-acetate (cortisol acetate) was slurried with 122 ml. of natural collidine, and then 370 ml. of dimethylformamide was added. This sequence of addition allows the cortisol acetate to dissolve momentarily and then quickly separate as fine crystals of the dimethylformamide complex. Good stirring is essential to keep the resulting thick slurry mobile. The mixture was cooled to 10° and treated in about 2 min. with 37 ml. of methanesulfonyl chloride containing 3.2% sulfur dioxide. The batch was allowed to stir at 25–35° for 10 min. and then excess reagent was destroyed by the gradual addition (1 min.) of 60 ml. of water. Despite ice-bath cooling the temperature of the reaction mixture rose to 59°. The thin slurry was cooled to room temperature and added gradually to 3700 ml. of hot (80–90°) water with good agitation.[5] This mixture was stirred at 85–90° for 1 hr., cooled to room temperature, and filtered. The product was washed several times with water and dried in air at 60°. There was obtained 56.0 g. (97.7%) of cream-colored powder which melted at 226–228.5°, contained 0.6% water (by Karl Fischer titration), and possessed a specific rotation (*c* 0.5) in chloroform of +131.8°. Treatment of this product with ten parts of refluxing methanol gave a recovery of about 92% of 11(9)-anhydrocortisol acetate which melted at 234–237°; lit.[1a] m.p. 236–237°, [α]D (*c* 1, chloroform) +117°; lit.[6] m.p. 231.5–234.5°, [α]D (*c* 1.04, chloroform) +124°; lit.[1g] m.p. 232.5–236.5°.

The above procedure in the absence of sulfur dioxide gave only unchanged starting material.

1,4,9(11)-Pregnatriene-17α,21-diol-3,20-dione 21-Acetate.—A charge of 16.2 g. (0.04 mole) of 1,4-pregnadiene-11β,17α,21-triol-3,20-dione 21-acetate (prednisolone acetate) was dehydrated by the procedure described for 11(9)-anhydrocortisol acetate. The crude product was obtained in a yield of 15.85 g. (103%), m.p., 154–205°. Paper-strip analysis showed complete dehydration had occurred. Refluxing the product with five parts of acetone permitted a recovery of 46.8% of triene which melted at 220–222°, lit.[7] m.p. 223–226°.

Acknowledgment.—We wish to express our appreciation to Mr. Charles B. Muchmore for the preparation and help in the interpretation of the vapor phase chromatograms. We also wish to thank Dr. Erwin Schoenewaldt of the Merck Sharp and Dohme Research Laboratories for his valuable suggestions and information. It was he who proposed the mechanism which we have described here.

(5) Precipitation of the product by hot water allows the isolation of a partially hydrated material which is easily freed of water at moderate temperatures. Precipitation by cold water furnished the dimethylformamide complex of the product which requires vigorous drying conditions in order to be rid of the solvent or a slurry treatment with hot water, in which case the hydrate is obtained.

(6) R. P. Graber, A. C. Haven, Jr., and N. L. Wendler, *J. Am. Chem. Soc.*, **75**, 4722 (1953).

(7) J. A. Hogg, F. H. Lincoln, A. H. Nathan, A. R. Hanze, W. D. Schneider, P. F. Beal, and J. Korman, *ibid.*, **77**, 4438 (1955).

Steroids of Unnatural Configuration. The Absence of Long-Range Conformational Effects in Ring A Modified 20-Ketopregnanes[1a,b]

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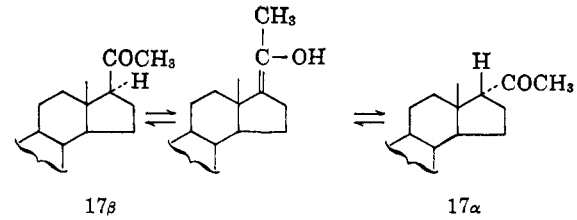
Syntheses of 5α,17α-pregnane-3,20-dione, 5β,17α-pregnane-3,20-dione, and 17α-1-dehydroprogesterone are described. No detectable long-range conformational effects were observed in the n.m.r. spectra or relative stabilities (*vs.* 17β-isomers) of these three compounds as well as 17α-pregnenolone and 17α-progesterone.

A large variety of 20-ketopregnanes, unsubstituted at C-17, have been described. Although the two C-17 isomers of these ketones are interconvertible through a common enol (or enolate ion), only the 17β-epimers are naturally occurrring.[3] We have undertaken a systematic study of the unnatural (17α) isomers of these ketones[4a] with the objectives of evaluating the role of C-17 configuration in biological activity and investigating the operation of a variety of steric effects in fused ring systems. This report describes the first stage of this investigation and is concerned with the



17β 17α

(1) (a) This research was supported in part by a Public Health Service Research Grant, A-3943, from the National Institute of Arthritis and Metabolic Diseases; (b) presented in part at the 145th National Meeting of the American Chemical Society, New York, N. Y., Sept., 1963.

(2) National Institutes of Health Predoctoral Fellow, 1961–1962.

(3) Two exceptions to this generalization have been reported: (a) H. I. Calvin and S. Lieberman [*Biochemistry*, **1**, 639 (1962)] have isolated tritiated IIα from human urine after ingestion of tritiated 16-dehydroprogesterone. Earlier isolations of IIα from human urine were explicable on the basis of isomerization of 17β-isomer during vigorous acid hydrolysis involved in the isolation procedures [S. Lieberman, K. Dobriner, B. R. Hill, L. F. Fieser, and C. P. Rhoads, *J. Biol. Chem.*, **172**, 263 (1948); G. Birke, C. A. Gemzell, L. O. Plantin, and H. Robbe, *Acta Endocrinol.*, **27**, 389 (1958)]; (b) P. D. Meister, D. H. Peterson, H. C. Murray, S. H. Eppstein, L. M. Reineke, A. Weintraub, and H. M. Leigh [*J. Am. Chem. Soc.*, **75**, 55 (1953)] isolated 25% of 11α-hydroxy-17α-progesterone from incubation of 16-dehydroprogesterone with *Rhizopus nigricans*.

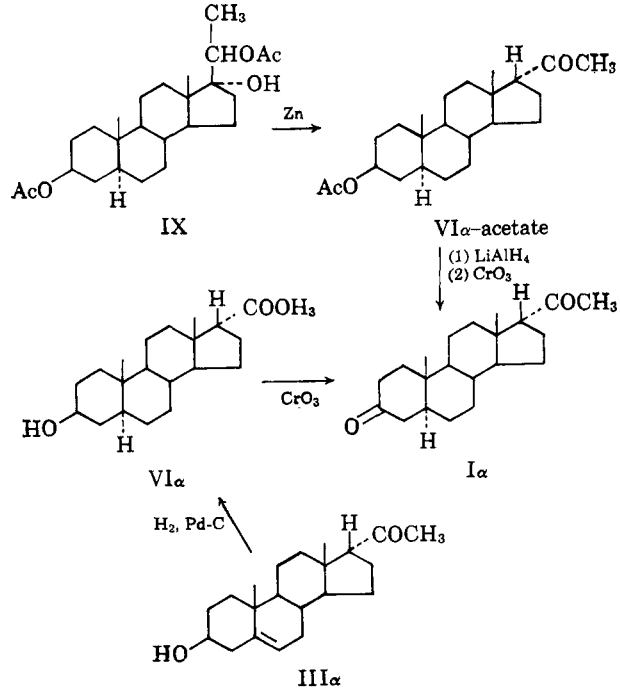
(4) (a) A review of 17α-20-ketopregnanes has recently appeared [M. B. Rubin, *Steroids*, **2**, 561 (1963)]. (b) In this report 20-ketopregnanes (unsubstituted at C-17) are designated by a Roman numeral followed by α or β to indicate the stereochemistry at C-17.

following 17α-20-ketopregnanes[4b] in which the nature of ring A has been varied: 5α,17α-pregnane-3,20-dione (Iα), 5β,17α-pregnane-3,20-dione (IIα), 17α-pregnenolone (IIIα, 17α-Δ⁵-pregnen-3β-ol-20-one), 17α-progesterone (IVα, 17α-Δ⁴-pregnene-3,20-dione), and 17α-1-dehydroprogesterone (Vα, 17α-Δ¹,⁴-pregnadiene-3,20-dione).

Shortly before the inception of this work, Barton and co-workers[5] described a "conformational transmission effect" in 3-keto steroids and 3-ketotriterpenes. They observed variations in the rate of base-catalyzed aldol condensation at C-2 as unsaturation in rings B, C, and D was changed, and attributed these variations to distortions transmitted through the fused ring system to the reaction site.[6] The two extremes of reaction rate differed by a factor of 43. Since Barton's findings were published, other workers have described similar effects on reaction rate as a function of remote structural changes.[7] In an attempt to observe the effect of conformational transmission on equilibrium composition, Allinger and Greenberg[8] examined the mixtures obtained by base-catalyzed equilibration of A-nor-androstane-3,20-dione and A-nor-D-homoandrostane-3,17a-dione but observed no effect of change in size of ring D on composition of equilibrated mixtures. A particular point of interest in the present work lay in the possibility of conformational transmission from ring A to C-17 which might be detected by examination of the variation in relative stabilities of 17α- *vs.* 17β-isomers as a function of ring A structure.

As we have reported recently,[9] modification of the Serini–Logemann reaction provides a convenient entry into the 17α-20-ketopregnane system. Appreciable quantities of IIIα-acetate and IVα could be prepared without difficulty from commercially available 16α,17α-epoxypregnenolone and 17α-hydroxyprogesterone. The hydrolysis of IIIα-acetate to IIIα (without isomerization at C-17) has been described,[10] and proceeded in quantitative yield in our hands. It had been anticipated that IVα would serve as starting material for synthesis of both 5α,17α- and 5β,17α-pregnane-3,20-diones. In model experiments, hydrogenation of progesterone (IVβ) over 10% palladium on charcoal in the presence of a trace of potassium hydroxide[11] yielded a 3:2 mixture of allopregnanedione (Iβ) and pregnanedione (IIβ) which could be separated without difficulty by chromatography on Florisil.[12] However, when the same procedure was applied to IVα, a mixture was obtained from which no pure products could be isolated by repeated chromatography on Florisil or silica gel.[13] In view of these results it was necessary to prepare Iα and IIα by independent procedures.

Two syntheses of Iα in low yield have been described in the literature. In the first of these,[14] 5α,17α-pregnan-3β-ol-20-one (VIα), obtained by base-catalyzed equilibration of the acetate of the 17β-isomer, was oxidized with chromium trioxide in acetic acid to give a mixture of C-17 epimers from which reported Iα, m.p. 134–135°, $[\alpha]_D$ −15°, was isolated. In the second synthesis, Shoppee[15] prepared the 3-acetate of VIα by Serini–Logemann reaction of 5α-pregnane-3β,17α,20-triol 3,20-diacetate (IX), hydrolyzed the acetate with aqueous methanolic potassium bicarbonate, and oxidized the resultant VIα as described by the earlier workers[14] to obtain Iα, m.p. 148–149°, $[\alpha]_D$ −50°. In our hands, the 3-acetate of VIα, obtained in 79% yield by Serini–Logemann reaction of IX, either was not hydrolyzed to VIα (potassium carbonate in aqueous methanol) or yielded a mixture containing appreciable amounts of allopregnanolone (VIβ, 1 *N* potassium hydroxide in methanol). This difficulty could be circumvented by lithium aluminum hydride reduction of VIα-acetate to the 3,20-diol followed by oxidation with chromium trioxide–pyridine complex[16] to yield Iα, m.p. 145–147°, $[\alpha]_D$ −44°, in reasonable agreement with the properties described by Shoppee. A much simpler



procedure which furnished Iα in 81% yield from IIIα consisted of catalytic hydrogenation of IIIα to VIα followed by oxidation with chromic acid in acetone.[17]

Since reductive methods were unpromising, we turned to the Serini–Logemann reaction for the preparation of 5β,17α-pregnanedione (IIα). Lithium aluminum hy-

(5) D. H. R. Barton and A. J. Head, *J. Chem. Soc.*, 932 (1956); D. H. R Barton, A. J. Head, and P. J. May, *ibid.*, 935 (1957); D. H. R. Barton, F McCapra, P. J. May, and F. Thudium, *ibid.*, 1297 (1960).

(6) It should be noted that other possible explanations, such as classical conformational effects or inductive effects, did not explain satisfactorily the observed variations. Theoretical treatments of conformational transmission in simple systems have recently appeared [R. Bucourt, *Bull. soc. chim. France*, 1983 (1962); 1262 (1963)].

(7) T. L. Kim-Phuong and H. B. Kagan, *Compt. rend.*, **256**, 4036 (1963), and references contained therein.

(8) N. L. Allinger and S. Greenberg, *J. Org. Chem.*, **25**, 1399 (1960).

(9) M. B. Rubin and E. C. Blossey, *Steroids*, **1**, 453 (1963).

(10) A. Butenandt, J. Schmidt-Thomé, and H. Paul, *Ber.*, **72**, 1112 (1939).

(11) W. S. Johnson, E. R. Rogier, J. Szmuszkovicz, H. I. Hadler, J. Ackerman, B. K. Bhattacharya, B. M. Bloom, L. Stalmann, R. A. Clement, B. Bannister, and H. Wynberg, *J. Am. Chem. Soc.*, **78**, 6289 (1956).

(12) A 2:3 mixture of the same products was obtained by reduction of 1-dehydroprogesterone under the same conditions.

(13) The mild conditions and low concentration of base in the hydrogenation reaction seem to preclude the possibility that purification was complicated by partial conversion to 17β-isomers.

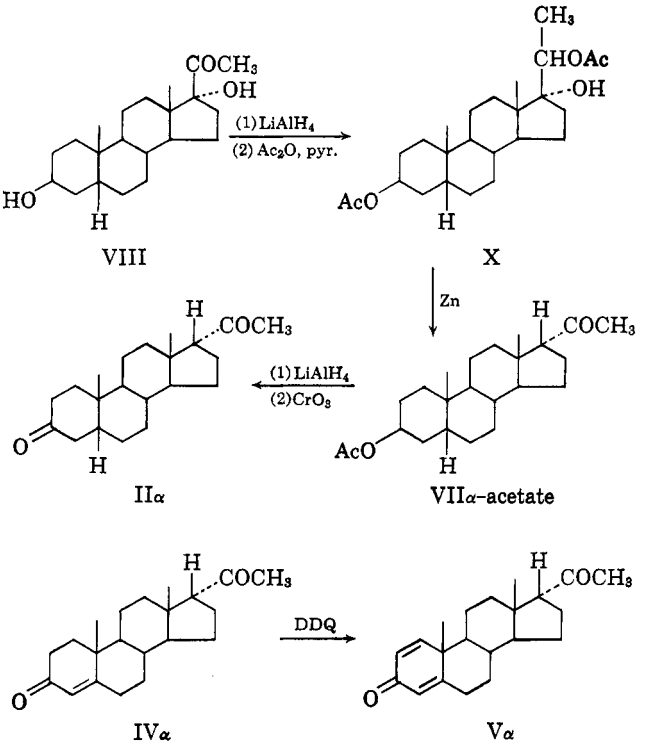
(14) A. Butenandt and L. Mamoli, *Ber.*, **68**, 1847 (1935).

(15) C. W. Shoppee, *J. Chem. Soc.*, 1671 (1949).

(16) G. I. Poos, G. E. Arth, R. E. Beyler, and L. H. Sarett, *J. Am. Chem. Soc.*, **75**, 422 (1953).

(17) K. Bowden, I. M. Heilbron, E. R. H. Jones, and B. C. L. Weedon, *ibid.*, 39 (1946); C. Djerassi, R. R. Engle, and A. Bowers, *J. Org. Chem.*, **21**, 1547 (1956).

dride reduction of commercially available 5β-pregnane-3β,17α-diol-20-one (VIII) followed by acetylation and Serini reaction (86% yield) gave 5β,17α-pregnan-3β-ol-20-one acetate (VIIα-acetate). The difficulties encountered in the hydrolysis of VIα-acetate were also observed with the 5β-epimer necessitating use of the hydride reduction and chromic acid oxidation sequence for conversion of VIIα-acetate to IIα.



The synthesis of 17α-1-dehydroprogesterone (Vα) was achieved in one step from IVα. After the selenium dioxide dehydrogenation[18] of IVα was shown to yield a difficultly resolvable mixture containing about 50% of the desired product, the use of 2,3-dichloro-5,6-dicyanoquinone[19] (DDQ) was investigated. Reaction of this quinone with IVα in refluxing benzene gave the desired Vα in 79% yield.

Ultraviolet and infrared spectra of the various compounds described were in agreement with the structures proposed. As would be expected, comparisons of infrared spectra of the 17α-ketones with those of the corresponding 17β-isomers showed significant differences in the "fingerprint" region although no assignment of a band characteristic of the 17α-acetyl group could be made.

The standard method[4] for assignment of 17α-configuration to one of a pair of C-17 epimeric 20-ketones has been based on the fact that the 17α-isomer is the more levorotatory of the two. The values for the molecular rotation difference between C-17 epimers ($M_D^\beta - M_D^\alpha$) obtained in this work were the following: I, 543; II, 563; III, 560; IV, 580; V, 635; VI, 535; and VII-acetate, 570. These are in agreement with the configurations assigned although it might be noted that the values for IV and V, both of which possess a conjugated carbonyl function in ring A, lie outside the range of 550 ± 20 which has been observed[15,20] with a variety of 20-ketopregnanes which are not substituted in the vicinity of the 20-keto group.[21]

More recently, the application of optical rotatory dispersion studies has provided a superior method for assignment of C-17 configuration since both isomers need not be available. In all reported cases, the 17α-20-ketones have exhibited strong negative Cotton effect curves in contrast to the positive Cotton effects observed with their 17β-isomers.[22a-c] The constancy of molecular rotation difference observed at 589 mμ has been shown by Struck and Houtman[22b] to extend throughout the spectrum. These workers derived an "average difference curve" which, by algebraic addition to the curve of a 17β-20-ketopregnane, allowed prediction of the curve of its 17α-isomer. The rotatory dispersion data for Iα–Vα are presented in the Experimental section; in all cases the expected negative Cotton effect was observed. The results for compounds Iα, IIα, and IIIα, which exhibited single Cotton effects, fit the average difference curve reasonably well. However, the steep slope of this curve in the region of 310–380 mμ where the fine structure of the multiple Cotton effect curve of IVα is observed allowed only approximate agreement between calculated and observed values. Interestingly, only single Cotton effect curves were exhibited by Vα and Vβ.

Preliminary comparisons of the n.m.r. spectra of 17α-steroids with spectra of their 17β-isomers indicated appreciable variations in the chemical shifts of methyl protons as a function of C-17 configuration. Accordingly, the spectra were carefully determined on a field-frequency controlled instrument (Varian A-60) and checked on two other instruments[23] (A-60 and HR-60); the maximum observed deviation from the average for three scans of each spectrum was 0.4 c.p.s. The results, expressed in c.p.s. from tetramethylsilane, are presented in Table I.

TABLE I

CHEMICAL SHIFTS OF METHYL PROTONS[a]

	C-18			C-19		
	17β	17α	ΔC-18[b]	β	α	C-21
I	38.1	55.7	17.6	60.9	59.9	
II	38.5	55.8	17.3	62.1	60.2	
III	38.1	55.8	17.7	60.6	60.0	127.8 ± 0.5
IV[c]	40.2	57.8	17.6	71.4	70.6	(τ 8.87)
V	41.9	59.3	17.4	74.2	73.2	

[a] In c.p.s. from tetramethylsilane. Spectra were determined on 0.32 *M* solutions in deuteriochloroform with internal tetramethylsilane using a Varian Associates A-60 spectrometer. Reported values are averages of three scans; maximum observed deviation was 0.4 c.p.s. [b] Chemical shift of 17α-isomer minus shift of 17β. [c] Identical results were obtained with 0.08 and 0.16 *M* solutions.

Comparison of the shift of the C-18 protons for each pair of 17-epimers indicates a marked downfield shift

(18) J. A. Edwards, H. J. Ringold, and C. Djerassi, *J. Am. Chem. Soc.*, **82**, 2318 (1960).

(19) D. Burn, D. N. Kirk, and V. Petrow, *Proc. Chem. Soc.*, 14 (1960).

(20) C. W. Marshall and T. F. Gallagher, *J. Biol. Chem.*, **179**, 1265 (1949).

(21) An even larger deviation is observed with the Δ4,6-pregnadiene-3,20-diones where ΔMD = 812 (J. E. Vaux, Jr., unpublished results).

(22) (a) C. Djerassi, *Bull. soc. chim. France*, 741 (1957); *cf.* also C. Djerassi, "Optical Rotatory Dispersion," McGraw-Hill Book Co., Inc., New York, N. Y., 1960, p. 52; (b) W. A. Struck and R. L. Houtman, *J. Org. Chem.*, **26**, 3883 (1961); (c) P. Crabbé, *Tetrahedron*, **19**, 51 (1963).

(23) We wish to acknowledge the cooperation of Dr. E. Legoff of the Mellon Institute and Mr. P. Yajko of NMR Specialties, Inc., in the determination of these spectra.

for the 17α-isomer in each case.[24] Although the position of the resonance varied from compound to compound in both the 17α- and 17β-series (presumably due to long-range shielding by ring A functionality), the magnitude of the difference between pairs of isomers (ΔC-18) was remarkably constant, the average value being 17.5 ± 0.2 c.p.s. This difference is undoubtedly due to long-range shielding by the 20-ketone; its magnitude must reflect the spatial relationship between the ketone and C-18 methyl groups and might allow calculation of molecular geometry. The constant value of ΔC–18 strongly suggests that changes in ring A do not lead to appreciable change in dihedral angle between C-13 and C-17 substituents.

The chemical shifts of C-19 protons varied as expected[25] with changes in ring A. Interestingly, however, the chemical shifts of C-17 epimers were not identical. A slight upfield change in the position of the resonance of the 17α-isomer, varying from 0.5 c.p.s. for III to 1.9 c.p.s. for II, was observed. This again may be attributed to long-range shielding by the 20-ketone. No variations in chemical shift of the C-21 protons were observed; the value for the ten compounds was 127.8 ± 0.5 c.p.s. (τ 8.87 ± 0.01) independent of C-17 configuration or ring A structure. The remaining features of the n.m.r. spectra were in agreement with the structures assigned.

The fact that 17β is the more stable of the two possible configurations in most 17-substituted steroids has been attributed[26] to the pseudo-equatorial character of the 17β-bond as opposed to the pseudo-axial nature of the 17α-bond. Distortions in the steroid skeleton which lead to changes in conformation at C-17 should then be reflected in changes in the relative stabilities of C-17 epimers. Since acid- or base-catalyzed enolization of the 20-ketone towards C-17 provides a reversible path for interconversion of 17α- and 17β-20-ketopregnanes, their relative stabilities can be determined by measurement of the compositions of equilibrated mixtures. This has been done with each of the 5 pairs of isomers (I–V) available in this work using optical rotation as a convenient method for analysis of mixtures. Preliminary experiments indicated a reduction in the ultraviolet absorption of progesterone upon standing in acid medium, so that equilibration studies were confined to alkaline solution. The standard procedure used in all cases involved measurement of the optical rotation of a 1% solution of an individual steroid in 1 *N* methanolic potassium hydroxide at room temperature in a nitrogen atmosphere until no further change was observed (12–24 hr.).[27] The product mixture then was isolated, its spectra were determined to ensure that no appreciable side reactions had occurred, and the optical rotation was determined in chloroform solution. Since the rotations of the pure 17-epimers were known, the composition of the equilibrated mixture could be calculated by a simple proportionality.[28] In all cases the equilibrium compositions obtained from 17α- and 17β-isomers were in agreement within the estimated error of ±2% composition.

The values for per cent of 17α-isomer at equilibrium were the following: I, 23%; II, 23%; III, 21%; IV, 25%; and V, 22% (average 23 ± 2%). It might be noted that these compositions correspond to free-energy differences of about 1 kcal. per mole. The only other ring A modified 20-ketopregnanes whose base-catalyzed[29] equilibrations have been reported are 5β,17α- and 5β,17β-pregnan-3α-ol-20-one[32] where the value of 23% of 17α-isomer at equilibrium[33] is in agreement with the results obtained in this work. The commonly accepted value of 30% of 17α-isomer at equilibrium[34] is based on the results of Moffett and Hoehn[32] and of Butenandt, *et al.*,[14,35] whose 17α-20-ketones were later[15,20] shown to have been impure.

The equilibration results described above suggest no appreciable conformational change at C-17 upon introduction of unsaturation in ring A. Combined with the interpretation of n.m.r. spectra suggesting no change in the C-13, C-17 dihedral angle, the results lead to the conclusion that no significant conformational transmission effect is operative from ring A to ring D. The possibility that variations in the steroid molecule closer to C-17 might produce an observable effect is being investigated.

The five 17α-20-ketopregnanes were examined in a general endocrine screening program and were uniformly inactive[36] in contrast to some of the 17β-epimers.

Experimental[37]

5α-Pregnane-3β,17α,20-triol 3,20-Diacetate.—Hydrogenation of 1.20 g. of Δ⁵-pregnene-3β,17α,20-triol 3,20-diacetate[9] in 50 ml. of ethyl acetate over 0.12 g. of 10% palladium on charcoal at atmospheric pressure, followed by filtration and concentration, afforded an oil, $[\alpha]^{30}D$ −3 ± 2°. Crystallization from isopropyl ether gave 0.66 g. (55%) of mixed C-20 epimers, m.p. 147–195°, $[\alpha]^{30}D$ −3 ± 2°; $\lambda_{max}^{CH_2Cl_2}$ 2.75, 5.75, 8.1 μ [lit.[15] for 20β-isomer,

(24) This shift provides a convenient method for establishing configuration of the side chain; *cf.* also W. J. Wechter and H. C. Murray, *J. Org. Chem.*, **28**, 755 (1963).

(25) J. N. Shoolery and M. T. Rogers, *J. Am. Chem. Soc.*, **80**, 5121 (1958); G. Slomp and B. R. McGarvey, *ibid.*, **81**, 2200 (1959); J. S. G. Cox, E. O. Bishop, and R. E. Richards, *J. Chem. Soc.*, 5118 (1960).

(26) D. H. R. Barton, *Experientia*, **6**, 316 (1950); L. J. Chinn, *J. Org. Chem.*, **27**, 54 (1962).

(27) It is noteworthy that half-lives were approximately 2 hr. under these mild conditions. Similar reaction times have been observed in equilibrations of 16β-methyl-5α-Δ⁹⁽¹¹⁾-pregnen-3β-ol-20-ones (17α and 17β) by E. J. Bailey, D. H. R. Barton, J. Elks, and J. F. Templeton [*J. Chem. Soc.*, 1578 (1962)].

(28) Partial resolution of synthetic mixtures of Iα and Iβ, IIIα- and IIIβ-acetates, and IVα and IVβ could be achieved by gas chromatography (50-μg. samples, 1.5% SE-30 on silanized Chromosorb W, 225°, 75 ml. of He/min.); in these instances the 17α-isomer had the shorter retention time [*cf.* W. J. Wechter and H. C. Murray, *J. Org. Chem.*, **28**, 755 (1963)]. Because of the considerable overlap of the two peaks, unsatisfactory results were obtained upon attempted analysis of synthetic mixtures of known composition. No appreciable amounts of impurities were detected by gas chromatographic examination of equilibrated mixtures. Modifications which might afford improved resolution are being investigated for application in future work.

(29) Acid-catalyzed equilibrations of pregnenolones[30] (18% of 17α) and pregnenolone methyl ethers[31] (15% of 17α) have been reported. The reasons for the slightly lower per cent of 17α-isomer at equilibrium are not clear.

(30) J.-F. Biellman, D. Kucan, and G. Ourisson, *Bull. soc. chim. France*, 337 (1962).

(31) O. R. Rodig, P. Brown, and P. Zaffaroni, *J. Org. Chem.*, **26**, 2431 (1961).

(32) R. B. Moffett and W. M. Hoehn, *J. Am. Chem. Soc.*, **66**, 2098 (1944).

(33) The original workers reported 29% of 17α-isomer. It was later shown[20] that their 17α-steroid was impure; the value of 23% presented above has been recalculated from the original data using $[\alpha]D$ −50° instead of $[\alpha]D$ −41° for the 17α-isomer.

(34) L. F. Fieser and M. Fieser, "Steroids," Reinhold Publishing Corp., New York, N. Y., 1959, p. 566.

(35) A. Butenandt and G. Fleischer, *Ber.*, **70**, 96 (1937).

(36) These tests were performed under the supervision of Dr. L. J. Lerner at the Squibb Institute for Medical Research.

(37) Melting points are corrected. Optical rotations were determined in 1% chloroform solutions and ultraviolet spectra were determined in 95% ethanol.

m.p. 161–162°, $[\alpha]^{19}D$ −25 ± 1 (acetone); for 20α-isomer, m.p. 250–251°, $[\alpha]^{20}D$ −39° (acetone)].

5α,17α-Pregnan-3β-ol-20-one Acetate (VIα-Acetate).—Reaction of 0.64 g. of mixed C-20 epimers described above for 26 hr. under the standard Serini-Logemann reaction conditions[9] furnished 0.61 g. of oil, $[\alpha]^{29}D$ −64 ± 1°. Crystallization from isopropyl ether furnished, in two crops, 0.43 g. (79%) of product, m.p. 97–112°, $[\alpha]^{20}D$ −75 ± 2°. Recrystallization from methanol raised the m.p. to 113–114°, $[\alpha]^{30}D$ −75 ± 1°; lit.[15] m.p. 119–122°, $[\alpha]^{21}D$ −75° (alcohol).

Attempted Hydrolyses of VIα-Acetate. A.—A solution of 0.9 g. of potassium carbonate in 25 ml. of water was added to a solution of 173 mg. of VIα-acetate in 50 ml. of methanol. After 1 hr. at room temperature, the pH was adjusted to 2 by addition of concentrated hydrochloric acid and the solution was concentrated under reduced pressure. The residue was taken up in ethyl acetate, washed with water and saturated salt solution, dried over anhydrous sodium sulfate, and concentrated to give 136 mg. of solid, m.p. 95–123°. The infrared spectrum showed weak absorption at 2.75 μ and strong absorption at 5.76, 5.85, and 8.09 μ.

B.—Twenty milliliters of 1 *N* methanolic potassium hydroxide containing 104 mg. of VIα-acetate was stirred at room temperature for 1 hr. After work-up as described in A, 103 mg. of solid, m.p. 105–133°, was obtained. This was chromatographed on Florisil. Elution with 1 and 2% ethyl acetate in benzene gave 25 mg. (24%) of VIα, m.p. 115–134°, $[\alpha]^{30}D$ −73 ± 1°; lit.[15] m.p. 139°, $[\alpha]^{22}D$ −78° (alcohol). Elution with 2, 3, and 4% ethyl acetate in benzene gave 28 mg. (28%) of VIβ, m.p. 145–187°, $[\alpha]^{30}D$ 87 ± 1°; lit.[35] m.p. 194.5°, $[\alpha]D$ 91° (alcohol).

5α,17α-Pregnane-3β,20-diol.—A solution of 0.53 g. of VIα-acetate in 20 ml. of anhydrous ether was added to 0.3 g. of lithium aluminum hydride in 20 ml. of ether. After refluxing for 1.5 hr. the solution was cooled, excess hydride was decomposed with wet ether, and the solid was filtered. The filtrate, after washing with water, drying over anhydrous sodium sulfate, and concentration, yielded 0.46 g. of white solid, m.p. 120–160°, $\lambda_{max}^{CH_2Cl_2}$ 2.75–3.0 μ. This was used without further purification.

5α,17α-Pregnane-3,20-dione (Iα). A. From 17α-Pregnenolone.—Hydrogenation of 1.18 g. of 17α-pregnenolone, $[\alpha]^{31}D$ −149° (*c* 1, $CHCl_3$), over 0.10 g. of 10% palladium on charcoal in 50 ml. of ethyl acetate at atmosphere pressure and room temperature resulted in absorption of 1.0 equiv. of hydrogen after 2 hr. when reaction ceased. Filtration and evaporation afforded 1.13 g. (95%) of 5α,17α-pregnan-3β-ol-20-one (VIα), m.p. 120–136°, $[\alpha]^{29}D$ −74°; lit.[15] m.p. 139°, $[\alpha]D$ −78° (alcohol).

A solution of 1.00 g. of the above in 100 ml. of acetone under nitrogen was treated with 0.91 ml. of 8 *N* chromic acid solution[17] at 10–14° for 5 min. with stirring. The mixture was poured into 500 ml. of water and the precipitated solid was filtered to give 0.85 g. (85%) of Iα as glistening plates, m.p. 140–146°, $[\alpha]^{29}D$ −45°, λ_{max}^{KBr} 5.85 μ; lit.[15] m.p. 148–149°, $[\alpha]^{21}D$ −49.5° (alcohol). Crystallization from petroleum ether (b.p. 68–75°) raised the m.p. to 145–147°, specific rotation unchanged.

B. From 5α,17α-Pregnane-3β,20-diol.—A solution of 0.45 g. of the crude diol in 5 ml. of dry pyridine was added to the reagent[16] prepared from 0.85 g. of chromium trioxide and 9 ml. of pyridine. After 17 hr. at room temperature the mixture was worked up in the usual manner to give 382 mg. of solid which was chromatographed on 37 g. of silica gel. Elution with 3 to 5% ethyl acetate in benzene gave 189 mg. (41% over-all from VIα-acetate) of material, m.p. 120–148°. The infrared spectrum was identical with that of the product obtained by procedure A.

5β-Pregnane-3β,17α,20-triol 3,20-Diacetate.—A solution of 4.00 g. of 5β-pregnane-3β,17α-diol-20-one[38] in 100 ml. of tetrahydrofuran (THF) was added slowly with stirring to a slurry of 1.4 g. of lithium aluminum hydride in 200 ml. of THF at 0°. After 1.5 hr. at reflux, the solution was cooled in an ice bath and excess hydride decomposed by careful addition of a solution of 5 ml. of water in 5 ml. of THF. Anhydrous sodium sulfate then was added, the supernatant was decanted, the solid was washed with THF, and the combined solutions were evaporated to give 4.00 g. of 5β-pregnane-3β,17α,20-triol as a white solid, m.p. 180–190°. Repeated crystallization from acetone–petroleum ether afforded the analytical sample as needles which melted at 170° and resolidified to fine needles melting at 190°, $[\alpha]^{30}D$ −15°.

Anal. Calcd. for $C_{21}H_{36}O_3$: C, 74.95; H, 10.78. Found: C, 75.22; H, 10.72.

(38) Intermediates Incorporated, Joliet, Ill.

Reaction of 2.58 g. of the triol with 10 ml. of acetic anhydride and 20 ml. of pyridine overnight at room temperature afforded, after the usual work-up procedure, 3.66 g. of oil which was chromatographed on 183 g. of Florisil. Elution with 1 through 5% ethyl acetate in benzene gave a total of 3.28 g. of white solid. The analytical sample of triol-diacetate X was obtained by crystallization from petroleum ether as white needles, m.p. 146–148°, changing to plates, m.p. 164°; λ_{max}^{KBr} 2.80, 5.78, 5.87, 8.00 μ; $[\alpha]^{29}D$ 36 ± 1°.

Anal. Calcd. for $C_{25}H_{40}O_5$: C, 71.39; H, 9.59. Found: C, 71.28; H, 9.52.

5β,17α-Pregnan-3β-ol-20-one Acetate (VIIα-Acetate).—Reaction of 4.54 g. of triol-diacetate X described above for 20 hr. using the standard Serini-Logemann reaction procedure[9] afforded 3.69 g. of oil, $[\alpha]^{31}D$ −42°, which was chromatographed on 185 g. of Florisil collecting 2-l. fractions. Elution with 1 and 2% ethyl acetate in benzene gave 1.953 g. (49%) of crude VIIα-acetate. Crystallization of 1.14 g. from petroleum ether gave 0.76 g. of white plates, m.p. 119–123°. The analytical sample was obtained by crystallization from methanol as white plates, m.p. 120–123°; λ_{max}^{KBr} 5.77, 5.85, 8.00 μ; $[\alpha]^{31}D$ −78 ± 1°.

Anal. Calcd. for $C_{23}H_{36}O_3$: C, 76.62; H, 10.07. Found: C, 76.54; H, 9.91.

Further elution of the column with 2, 3, 4, and 5% ethyl acetate in benzene gave 1.24 g. (37%) of VIIα, m.p. 155–171°; $\lambda_{max}^{CH_2Cl_2}$ 2.71, 2.84, and 5.87 μ; $[\alpha]^{31}D$ −70 ± 1°. Acetylation with acetic anhydride and pyridine at room temperature yielded material identical with the acetate obtained from the earlier fractions.

5β,17α-Pregnane-3,20-dione (IIα).—A solution of 1.03 g. of VIIα-acetate in 50 ml. of dry ether was added with stirring to a slurry of 0.87 g. of lithium aluminum hydride in 50 ml. of ether at 0°. After 1 hr. at room temperature, the excess hydride was decomposed with wet ether and anhydrous sodium sulfate. The solvent was evaporated after filtration and washing, the residue was taken up in ethyl acetate, washed with water, dried over anhydrous sodium sulfate, and the solvent was evaporated to give 0.87 g. (100%) of white solid, m.p. 100–120°, λ_{max}^{KBr} 2.85 μ. This diol was oxidized without further characterization.

A solution of 0.80 g. of diol in 110 ml. of acetone under nitrogen was treated with 1.41 ml. of 8 *N* chromic acid solution[17] at 10° for 5 min. The mixture was poured into water which then was extracted with ethyl acetate, and the extracts were washed with water, dried over anhydrous sodium sulfate, and evaporated to give 0.75 g. (94%) of faintly yellow crystals, m.p. 85–100°, $\alpha^{29}D$ −46°. This was chromatographed on 21 g. of Florisil collecting 200-ml. fractions. Elution with benzene and 1% ethyl acetate benzene gave 0.63 g. of solid, m.p. 80–104°, which crystallized from petroleum ether to yield 0.33 g. (41%) of IIα, m.p. 107°. The analytical sample crystallized from petroleum ether as prisms, m.p. 107°, $[\alpha]^{32}D$ −65°.

Anal. Calcd. for $C_{21}H_{32}O_2$: C, 79.70; H, 10.19. Found: C, 80.02; H, 10.08.

17α-$\Delta^{1,4}$-Pregnadiene-3,20-dione (Vα).—A solution of 1.26 g. of 17α-progesterone and 1.22 g. of freshly crystallized DDQ[19] in 200 ml. of dry benzene was refluxed with stirring under nitrogen for 18 hr. The solution was cooled, filtered, and evaporated to dryness. The red residue was dissolved in ethyl acetate, washed with 5% sodium hydroxide solution and water, dried over anhydrous sodium sulfate, and concentrated under reduced pressure on the steam bath to give 1.02 g. of red tar. This was washed through 20 g. of alumina with 200 ml. of benzene to yield 1.00 g. (79%) of white solid, m.p. 113–136°. Recrystallization from isopropyl ether and then acetone–petroleum ether gave the analytical sample as white needles, m.p. 146–147°, λ_{max} 244 mμ (ε 18,600); (KBr) 5.85, 6.00, 6.13, 6.22 μ; $[\alpha]^{30}D$ −60°.

Anal. Calcd. for $C_{21}H_{28}O_2$: C, 80.73; H, 9.03. Found: C, 80.88; H, 9.48.

Rotatory dispersions, in methanol, gave the following results: Iα, methanol (*c* 0.147), $[\alpha]_{400}$ −180, $[\alpha]_{305}$ −1000, $[\alpha]_{255}$ +710, $[\alpha]_{245}$ +530; IIα, methanol (*c* 0.147), $[\alpha]_{400}$ −240, $[\alpha]_{309}$ −1990, $[\alpha]_{261}$ +2160, $[\alpha]_{240}$ +1550; IIIα-acetate, methanol (*c* 0.084), $[\alpha]_{400}$ −400, $[\alpha]_{308}$ −1790, $[\alpha]_{261}$ +710, $[\alpha]_{250}$ +500. In dioxane, the values were IVα, dioxane (*c* 0.145), $[\alpha]_{400}$ −160, $[\alpha]_{367}$ −593, $[\alpha]_{364}$ −578; (*c* 0.0483), $[\alpha]_{354}$ −694, $[\alpha]_{344}$ −383, $[\alpha]_{338}$ −472, $[\alpha]_{328}$ −74, $[\alpha]_{332}$ −203, $[\alpha]_{310}$ −20 (inflection); (*c* 0.029), $[\alpha]_{250}$ +4500; Vα, dioxane (*c* 0.115), $[\alpha]_{400}$ −450, $[\alpha]_{310}$ −1900, $[\alpha]_{265}$ +660, $[\alpha]_{250}$ +210; Vβ, dioxane (*c* 0.26), $[\alpha]_{400}$ +320, $[\alpha]_{325}$ +1900; (*c* 0.087), $[\alpha]_{310}$ +3000; (*c* 0.029), $[\alpha]_{276}$ −240, $[\alpha]_{225}$ +2100°.

Base-Catalyzed Equilibrations.—A 1% solution of each ketone in 1 *N* methanolic potassium hydroxide was introduced into a polarimeter tube of appropriate capacity which was then flushed with nitrogen and sealed. Optical rotations were measured periodically until no further change was observed. The solution then was recovered from the tube and neutralized with acetic acid, and the solvent was removed by evaporation under reduced pressure. The residue was taken up in ethyl acetate, washed twice with water, dried over anhydrous sodium sulfate, and evaporated to dryness under reduced pressure. The specific rotation of the residue was then determined in 1% chloroform solution and the infrared and ultraviolet (for IV and V) spectra were determined. The following rotational values were observed.

5α-Pregnane-3,20-diones (I): Iβ, $[\alpha]^{32}D$ 113° ($CHCl_3$); after 2 hr. in 1 *N* methanolic potassium hydroxide, 92°; 12 hr., 80°; Iα, $[\alpha]^{30}D$ −45° ($CHCl_3$); after 2 hr., −7°; 24 hr., +76°; recovered product, $[\alpha]^{32}D$ +77° ($CHCl_3$).

5β-Pregnane-3,20-diones (II): IIβ, $[\alpha]^{32}D$ 116° ($CHCl_3$); after 2 hr., 103°; 24 hr., 80°; IIα, $[\alpha]^{32}D$ −64°; after 2 hr., −2°; 24 hr., 79°; recovered product, $[\alpha]^{34}D$ +75° ($CHCl_3$).

Pregnenolones (III): IIIβ, $[\alpha]^{32}D$ 24° ($CHCl_3$); after 2 hr., 14°; after 12 hr., −3°; IIIα, $[\alpha]^{32}D$ −150° ($CHCl_3$); after 2 hr., −70°; 12 hr., −4°; recovered product, $[\alpha]^{34}D$ −10° ($CHCl_3$).

Progesterones (IV): IVβ, $[\alpha]^{30}D$ 192° ($CHCl_3$); after 4 hr., 151°; 22 hr., 142°; IVα, $[\alpha]^{30}D$ 10° ($CHCl_3$); after 4 hr., 107°; 22 hr., 144°; recovered product $[\alpha]^{32}D$ 140° ($CHCl_3$), λ_{max} 242 mμ (ε 16,000).

1-Dehydroprogesterones (V): Vβ, $[\alpha]^{36}D$ +131 ($CHCl_3$); after 2 hr., 107°; 22 hr., 92°; 17α, $[\alpha]^{36}D$ −60 ($CHCl_3$); after 2 hr., +41°; 22 hr., +86°; recovered product, $[\alpha]^{32}D$ +87 ($CHCl_3$), λ_{max} 244 mμ (ε 18,000).

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A Study of Isobutylene–Nitric Oxide Reaction Products

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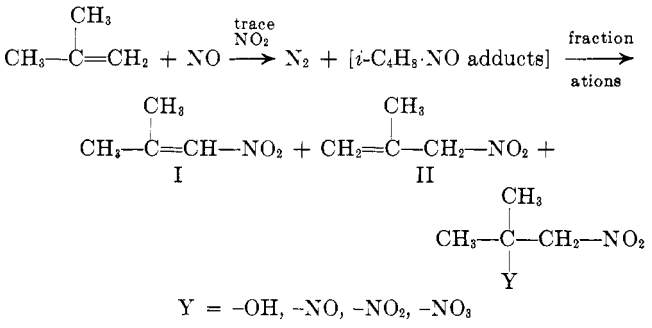
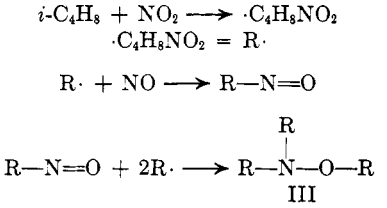
The products of the reaction of nitric oxide with isobutylene were investigated and a synthesis of methyl methacrylate precursors was developed. A previously unreported compound, tris(nitro-*t*-butyl)hydroxylamine, was found to be a major component of the isobutylene–nitric oxide reaction product. This compound decomposed readily to O,N-bis(nitro-*t*-butyl)hydroxylamine and a mixture of α- and β-nitroisobutylene. The α- and β-nitroisobutylenes were found to equilibrate in the presence of a variety of basic catalysts.

A previous investigation[1] has shown that nitric oxide reacts with liquid isobutylene in the presence of traces of nitrogen dioxide to give nitrogen and an unstable liquid mixture of nitro compounds. A series of fractionations of the liquid product under mild conditions was reported to yield a mixture of α-nitroisobutylene (I), β-nitroisobutylene (II), and small quantities of substances having nitro-*t*-butyl structures.



In addition to the products reported earlier,[1] we have found that the isobutylene–nitric oxide reaction product contains 34–45% by weight of crystalline tris(nitro-*t*-butyl)hydroxylamine (III). This new compound probably arises through formation of nitro-*t*-butyl radicals, as described by Brown,[1] followed by their reaction with nitric oxide. Hoffmann and coworkers[2] recently reported a similar preparation of tris-*t*-butylhydroxylamine by reaction of *t*-butyl radicals with *t*-nitrosobutane. (See col. 2.)

We were able to isolate pure tris(nitro-*t*-butyl)-hydroxylamine only by recrystallization below room



temperature. The trishydroxylamine was found to decompose rapidly at moderate temperatures to form O,N-bis(nitro-*t*-butyl)hydroxylamine (IV), and a mixture of nitroisobutylenes accompanied by trace quantities of acetone, nitromethane, and O-(nitro-*t*-butyl)-acetoxime. Crude or solvent-wet tris(nitro-*t*-butyl)-hydroxylamine decomposes completely at room temperature within a few days. O,N-Bis(nitro-*t*-butyl)-hydroxylamine decomposes slowly upon heating to give a mixture of nitroisobutylenes, nitromethane, acetoxime, and O-(nitro-*t*-butyl)acetoxime. N-(Nitro-*t*-butyl)hydroxylamine decomposes rapidly to acetoxime and nitromethane under similar conditions. These decomposition products suggest that O,N-bis(nitro-*t*-butyl)hydroxylamine first decomposes to nitroisobutylenes and N-(nitro-*t*-butyl)hydroxylamine, which then immediately undergoes decomposition to acetone and nitromethane.

It appears likely that thermal decomposition of the three nitro-*t*-butylhydroxylamines is promoted by transition states involving quasi six-membered ring intermediates. According to this mechanism, the nitro groups of the tris and bis compounds would be in the *aci* form, and the nitro group of the mono compound in the normal form. A representative structure for decomposition of tris(nitro-*t*-butyl)hydroxylamine is illustrated.

(1) J. F. Brown, Jr., *J. Am. Chem. Soc.*, **79**, 2480 (1957).

(2) (a) A. K. Hoffmann and A. T. Henderson, *ibid.*, **83**, 4671 (1961); (b) A. K. Hoffmann, W. G. Hodgson, and W. H. Jura, *ibid.*, **83**, 4675 (1961).