in absolute EtOH using W-5 Raney nickel[17] catalyst. The catalyst was removed by filtration and the EtOH was evaporated at reduced pressure. The residue was recrystallized from petroleum ether (60–70°) affording 0.5 g (42%) of desired amino alcohol **3**: mp 149–151°; ir ($CHCl_3$), 2.78, 2.96, 3.33, 3.41, 3.50, 6.25, 6.35, 6.70, 6.92, 7.40, 9.85, 10.66 $\mu$; nmr ($CDCl_3$), $\delta$ 7.70 (multiplet, aromatic *ortho* protons), 7.34 (multiplet, aromatic *meta* and *para* protons), 3.00 ($W_{1/2}$ = 19 cps, axial methine proton at C-3). *Anal.* ($C_{16}H_{23}NO$) C, H, N.

**2(a)-Phenyl-*trans*-decalin-2(e),3(e)-diol 3-Tosylate (12).**—To **9** (1.05 g, 0.0043 mole), dissolved in 20 ml of anhydrous pyridine, was added *p*-toluenesulfonyl chloride (2.0 g, 0.01 mole) and the solution was allowed to stand at room temperature for 48 hr. $H_2O$ was added and the resulting oil was scratched with a glass rod to promote crystallization. The solid was removed by filtration and recrystallized from petroleum ether (60–70°) affording 1.0 g (58%) of tosylate **12**: mp 99–100°; ir ($CHCl_3$), 2.78, 3.42, 3.50, 6.27, 6.70, 6.92, 7.40, 8.55, 9.13, 9.74, 10.33, 10.82, 11.25, 11.62, 11.93 $\mu$; nmr ($CCl_4$), $\delta$ 7.2–8.0 (multiplet, aromatic protons), 4.80 (quartet, $J_{aa}$ = 11 cps, $J_{ae}$ = 6 cps, axial methine proton at C-3), 2.43 (singlet, $ArCH_3$). *Anal.* ($C_{23}H_{28}SO_4$) C, H.

**3(a)-Amino-2(a)-phenyl-*trans*-2(e)-decalol (2).**—Compound **12** (1.0 g, 0.0025 mole) was placed in a steel bomb and the bomb was cooled in Dry Ice–$Me_2CO$. To the bomb was added *ca.* 100 ml of liquid $NH_3$. The bomb was sealed and heated at 120° for 24 hr. The pressure was released and the residue was dissolved in $CHCl_3$. The $CHCl_3$ solution was filtered and the solvent was evaporated. The residue was recrystallized from petroleum ether (60–70°) affording 0.30 g (50%) of amino alcohol **2**: mp 116–117°; ir ($CHCl_3$), 2.79, 2.97, 3.34, 3.42, 3.51, 6.25, 6.35, 6.71, 6.92, 7.40, 9.85, 10.01, 10.28, 10.67 $\mu$; nmr ($CDCl_3$), $\delta$ 7.43 (multiplet, aromatic protons), 3.83 ($W_{1/2}$ = 6 cps, equatorial methine proton at C-3). *Anal.* ($C_{16}H_{23}NO$) C, H, N.

**3(e)-Amino-2(e)-phenyl-*trans*-2(a)-decalol (4).**—Compound **15** (1.0 g 0.004 mole) was dissolved in 100 ml of absolute EtOH saturated with $NH_3$ and material was hydrogenated under 70 kg/cm² of $H_2$ using W-5 Raney Ni catalyst.[17] The catalyst was removed by filtration and the solvent was evaporated at reduced pressure. The residue was chromatographed on silica gel (Merck 0.05–0.20 mm) eluting with cyclohexane–EtOAc (1:1) affording 0.5 g (50%) of desired amino alcohol **4**: mp 146–148°; ir ($CHCl_3$), 2.78, 2.95, 3.34, 3.52, 3.51, 6.25, 6.34, 6.70, 6.92, 7.63, 8.60, 9.51, 9.66, 10.00, 10.26 $\mu$; nmr ($CDCl_3$, $CF_3CO_2H$), $\delta$ 7.46 (broad singlet, aromatic protons), 3.55 ($W_{1/2}$ = 10 cps, methine proton at C-3). *Anal.* ($C_{16}H_{23}NO$) C, H, N.

**Acknowledgment.**—The authors gratefully acknowledge support of this project by the National Institutes of Health Grant He-08555. The authors wish to express their appreciation to Drs. J. B. LaPidus and P. N. Patil and Mr. S. Hetey for performing the vas deferens assays.

(17) H. R. Billica and H. Adkins in "Organic Syntheses," Coll. Vol. III, E. C. Horning, Ed., John Wiley and Sons, Inc., New York, N. Y., 1955, p 180.

# Synthesis and Myotrophic–Androgenic Activity of Substituted 2α,3α-Methano-5α-androstane Derivatives[1]

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*Received February 17, 1968*

The preparation and androgenic–myotrophic testing of analogs of **17** having substituents on the cyclopropyl ring were undertaken in an effort to obtain information regarding the steric and electronic requirements in the A ring of anabolic–androgenic androstanes. Treatment of 17β-hydroxyandrost-2-ene acetate with ethyl diazoacetate in the presence of anhydrous $CuSO_4$ gave 2α,3α-(β-carbethoxymethano)-5α-androstan-17β-ol acetate which was converted to a variety of substituted cyclopropane derivatives. The most potent is the aldehyde **15** which is more active than testosterone propionate in the myotrophic test and is much less androgenic.

Studies in this laboratory have resulted in the proposal[2] that anabolic–androgenic androstanes are bound to their receptor by a β-face π-bond to an $sp^2$ system in the A ring. The pronounced anabolic–androgenic activity of 2α,3α-methano-5α-androstan-17β-ol (**17**)[2] was taken as evidence for this hypothesis. Recent work[3a] on steroidal episulfides, bioisosteric with these methano steroids, has shown that the 2α,3α isomers indeed have high parenteral activity, whereas the 2β,3β isomers are essentially inactive. This is in harmony with our proposal.[3b] On the other hand, 2α,3α- and 2β,3β-steroidal difluorocyclopropanes have similar activity.[4]

To gain further information in this area, the preparation of analogs of **17** having altered electron density at the $sp^2$ centers was undertaken. If a π bond is important, the strength of the binding would be different in such analogs. Since β-face binding is assumed to be involved, the preparation of substituted cyclopropane analogs of **17** should be feasible since the substituent groups should not interfere sterically with drug receptor binding.

The synthetic plan involved the preparation of 2α,3α-carbethoxymethano-5α-androstan-17β-ol acetate (**7**) as a common intermediate for the other derivatives. This material was prepared by the reaction of 17β-hydroxyandrost-2-ene acetate[5] (**2**) with ethyl diazoacetate. Although carbene intermediates have been proposed in the reaction of diazo compounds with olefins,[6] the reaction failed when the reagents were heated at 120–180°, or were irradiated in toluene or hexane solution with a medium-pressure mercury arc. On the other hand, a 45% yield of **7** was realized when the reagents were heated in the presence of anhydrous $CuSO_4$. These results point to

(1) (a) This investigation was supported in part by a Public Health Service research grant (AM-05016) from the National Institute of Arthritis and Metabolic Diseases, U. S. Public Health Service. (b) Portions of this work are taken from the Ph.D. thesis of S.-Y. Cheng, University of California, San Francisco, 1966.

(2) M. E. Wolff, W. Ho, and R. Kwok, *J. Med. Chem.*, **7**, 577 (1964).

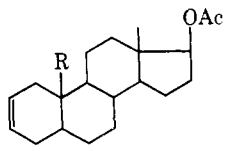
(3) (a) P. D. Klimstra, E. F. Nutting, and R. E. Counsell, *ibid.*, **9**, 693 (1966). (b) The Searle group did not interpret their data in this way.

(4) (a) L. H. Knox, E. Velarde, S. Berger, D. Cuadriello, P. W. Landis, and A. D. Cross, *J. Am. Chem. Soc.*, **85**, 1851 (1963); (b) F. A. Kincl and R. E. Dorfman, *Steroids*, **3**, 109 (1964).

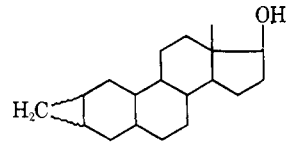
(5) A. Bowers, A. D. Cross, J. H. Edwards, H. Carpio, M. Calzada, and E. Denot, *J. Med. Chem.*, **6**, 156 (1963).

(6) J. Hine, "Divalent Carbon," The Ronald Press Co., New York, N. Y., 1964, p 122.

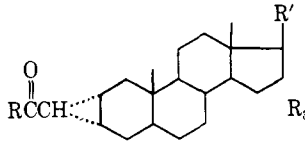
the intervention of an organocopper intermediate rather than a carbene, but further research is needed to clarify this point.



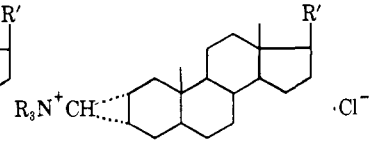
**1**, R = H
**2**, R = $CH_3$



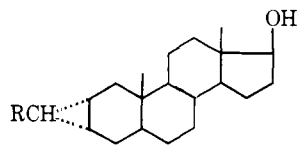
**3**, isomer A
**4**, isomer B



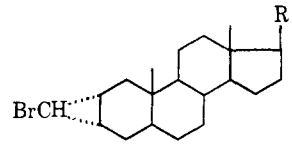
**5**, R = OH; R′ = OH
**6**, R = OH; R′ = O
**7**, R = $OC_2H_5$; R′ = OAc
**8**, R = $OCO_2C_2H_5$; R′ = O
**9**, R = $C_7H_4N$; R′ = O
**10**, R = $C_7H_4N$; R′ = OH



**11**, R = H; R′ = OH
**12**, R = H; R′ = O
**13**, R = $CH_3$; R′ = OH
**14**, R = $CH_3$; R′ = O



**15**, R = CHO
**16**, R = $CH_2OH$
**17**, R = H



**18**, R = OAc
**19**, R = O

The configuration of the cyclopropane ring and substituent in **7** was established in the following manner. The presence of a single C-19 methyl resonance in the nmr spectrum indicated that only one isomer was at hand. Moreover, the expected position of this isomer could be calculated by use of the Zürcher values[7] for substituents in 5α-androstane and by the Tori equation[8] for calculating the anisotropic shielding effect of the cyclopropane ring.

$$\Delta\delta \text{ (ppm)} = -6.67 \times 10^{-30} \text{ cm}^3 \sum_{i=1}^{3} \frac{3\cos^2\theta_i - 1}{R_i^3}$$

where $R_i$ is the distance in Å between the midpoint of a C–C bond of the cyclopropane ring and a C-19 proton and $\theta_i$ is the acute angle which the line $R_i$ makes with the C–C bond. An expected shielding of +0.273 ppm was calculated by the Tori equation for 2β,3β-methano-5α-androstane, whereas the corresponding effect in the 2α,3α isomer was calculated to be only +0.010 ppm. Experimentally, in compounds **6**–**8**, **10**, and **16**, the effect of the cyclopropane ring was found to differ by only −0.049 to +0.011 ppm from the Zürcher values.[1b] These results are compatible only with a 2α,3α fusion.

Chemical studies were in harmony with this assignment. Thus a modified Hunsdiecker reaction[9,10] on **6** gave a mixture of the epimeric bromides **19** as shown by the nmr spectrum. The formation of two bromides is not unexpected in view of the probable free-radical mechanism of this reaction.[11] Reduction of **19** with lithium tri-*t*-butoxyaluminohydride followed by reductive dehalogenation of the mixture of halides in the presence of Raney nickel gave **17** identical in all respects with an authentic sample,[2] together with a *single* isomer of starting material. Thus it is clear that the least hindered cyclopropane is formed in the addition reaction, and therefore we assign the β configuration to the carboxyl group as well.[12] The α position is hindered by interaction with protons at the 1α and 5α positions. It is also evident that these same factors are involved in the difference in stability of the two bromides toward catalytic reductive dehalogenation, but it is not possible to specify which isomer reacts preferentially.

Hydrolysis of **7** gave the acid **5** which was oxidized to the ketone **6** with Jones reagent. This was subjected to a modified Curtius[13] reaction in order to obtain the corresponding amine. The mixed anhydride **8** could be isolated and upon treatment with sodium azide smoothly gave the amine **12**. Reduction at C-17 with $NaBH_4$ gave the 17β-alcohol **11**.

One of the goals of this work was the formation of a cyclopropene ring fused to C-2 and C-3, in order to study the consequences of increasing the p character of the 2 and/or 3 carbon. Possible routes to the desired cyclopropenes included dehydrohalogenation of halocyclopropanes[14] and Hofmann elimination of cyclopropylamines.[15,16] The required quaternary amine **14** was prepared by quaternization of **12** with methyl iodide and alkali. However, no cyclopropenes were obtained from either method of preparation, in spite of very careful work-up procedures.

The reaction of the mixed anhydride **8** with ethylenimine gave the amide **9**. Reduction with $LiAlH_4$ gave the aldehyde **15** and the alcohol **16**. The last compound was also obtained by reduction of **15** with $NaBH_4$. The preparation of both isomers of 2,3-methano-5α-estran-17β-ol **3** and **4** by means of the Simmons–Smith reaction is included in the Experimental Section.

## Discussion

The data from the pharmacological testing[17,18] are displayed in Table I. The most active compound is the cyclopropyl aldehyde **15**, which is more active than testosterone propionate or the corresponding unsubstituted cyclopropane derivative **17** in the myotrophic test. At the same time, the compound has low androgenic action.

The next ranking compound in activity is the hydroxymethylene derivative **16** which has more than 10% the activity of testosterone propionate in the myotrophic test and less than 10% in the androgenic assay. Compound **16** has roughly one-tenth the activity of **15** in both tests.

(7) N. S. Bhacca and D. H. Williams, "Applications of Nmr Spectroscopy in Organic Chemistry," Holden-Day, Inc., San Francisco, Calif., 1964, p 19.
(8) K. Tori and K. Kitahonoki, *J. Am. Chem. Soc.*, **87**, 386 (1965).
(9) J. Cristol and W. C. Firth, *J. Org. Chem.*, **26**, 280 (1961).
(10) J. A. Davis, S. Carroll, J. Bunds, and D. Johnson, *ibid.*, **30**, 415 (1965).
(11) R. H. Johnson and R. K. Ingham, *Chem. Rev.*, **56**, 219 (1956).
(12) The usual α,β dotted-solid-line convention is employed to describe the cyclopropane substituent. The β substituent is attached to the uppermost bond in the planar structural formula.
(13) J. Weinstock, *J. Org. Chem.*, **26**, 3511 (1961).
(14) C. L. Osborn, T. C. Shields, B. A. Shoulders, J. F. Krause, H. V. Cortezand, and P. D. Gardner, *J. Am. Chem. Soc.*, **87**, 3158 (1965).
(15) N. Y. Demyanov and N. N. Doyarenko, *Ber.*, **56**, 2200 (1923).
(16) N. J. Schlatter, *J. Am. Chem. Soc.*, **63**, 1733 (1941).
(17) Pharmacological tests were performed at The Endocrine Laboratories, Madison, Wis.
(18) L. G. Hershberger, E. G. Shipley, and R. K. Meyer, *Proc. Soc. Exptl. Biol. Med.*, **83**, 175 (1953).

TABLE I

ANDROGENIC–MYOTROPHIC ASSAY

| Compd (total dose, mg) | Wt, mg[a] Ventral prostate | Seminal vesicle | Levator ani | Act.[d] Androgenic | Myotrophic |
|---|---|---|---|---|---|
| | | Series A | | | |
| Castrate control | 14.0 ± 0.70 | 11.5 ± 0.71 | 28.2 ± 1.37 | | |
| Testosterone propionate (0.3) | 35.4 ± 2.00 | 24.4 ± 1.81 | 40.0 ± 1.85 | | |
| *p* | <0.001 | <0.01 | <0.01 | | |
| 3 (3.0) | 18.5 ± 0.46 | 16.5 ± 1.05 | 35.8 ± 1.13 | <0.1 | <0.1 |
| *p* | 0.02 | 0.02 | 0.02 | | |
| 4 (3.0) | 20.9 ± 2.70 | 15.5 ± 1.48 | 32.8 ± 2.25 | 0 | 0 |
| *p* | NS[c] | NS | NS | | |
| | | Series B | | | |
| Castrate control | 14.1 ± 0.95 | 11.8 ± 0.31 | 27.3 ± 0.75 | | |
| Testosterone propionate (0.3) | 44.7 ± 3.31 | 31.6 ± 1.89 | 41.0 ± 1.11 | | |
| *p* | <0.001 | <0.001 | <0.001 | | |
| 5 (3.0) | 17.3 ± 2.64 | 12.6 ± 0.74 | 23.3 ± 1.77 | 0 | 0 |
| *p* | NS | NS | >0.05 | | |
| 11 (3.0) | 19.2 ± 0.98 | 19.6 ± 0.39 | 31.1 ± 0.81 | <0.1 | <0.1 |
| *p* | <0.02 | <0.001 | <0.02 | | |
| | | Series C | | | |
| Castrate control | 16.1 ± 1.05 | 11.5 ± 0.37 | 24.2 ± 0.83 | | |
| Testosterone propionate (0.3) | 41.5 ± 4.62 | 27.4 ± 3.79 | 42.1 ± 3.17 | | |
| *p* | <0.01 | <0.01 | <0.01 | | |
| 5 (6.0) | 12.5 ± 0.05 | 9.8 ± 0.14 | 26.0 ± 0.78 | 0 | 0 |
| *p* | 0.02 decrease | 0.01 decrease | NS | | |
| 7 (6.0) | 15.0 ± 0.89 | 12.2 ± 1.09 | 36.0 ± 2.31 | 0 | <0.05 |
| *p* | NS | NS | 0.01 | | |
| | | Series D | | | |
| Castrate control | 18.0 ± 0.67 | 12.1 ± 1.01 | 26.5 ± 1.66 | | |
| Testosterone propionate (0.3) | 34.8 ± 4.06 | 16.8 ± 1.17 | 32.4 ± 1.07 | | |
| *p* | <0.01 | <0.02 | <0.02 | | |
| 10 (3.0) | 16.0 ± 0.70 | 14.4 ± 1.70 | 25.7 ± 3.92 | 0 | 0 |
| *p* | NS | NS | NS | | |
| 13 (3.0) | 14.2 ± 0.49 | 12.1 ± 0.54 | 19.9 ± 3.21 | 0 | 0 |
| *p* | <0.01 decrease | NS | NS | | |
| 15 (0.3) | 22.2 ± 1.25 | 21.8 ± 0.27 | 53.1 ± 5.07 | | |
| *p* | <0.02 | <0.001 | *Ca.* 0.001 | ≪1.0 | >1.0 |
| 16 (3.0) | 24.8 ± 3.76 | 19.3 ± 1.02 | 49.2 ± 1.49 | | |
| *p* | NS | *Ca.* 0.001 | <0.001 | <0.1 | >0.1 |
| 18[b] | 16.0 ± 1.77 | 12.6 ± 0.44 | 24.4 ± 2.94 | 0 | 0 |
| *p* | NS | NS | NS | | |
| | | Series E | | | |
| Castrate control | 14.2 ± 0.42 | 11.4 ± 0.63 | 26.5 ± 1.32 | | |
| Testosterone propionate (0.3) | 49.6 ± 3.11 | 31.4 ± 1.82 | 37.3 ± 1.74 | | |
| *p* | <0.001 | <0.001 | <0.001 | | |
| 17 (0.3) | 21.2 ± 3.34 | 14.1 ± 0.70 | 35.4 ± 1.00 | ≪1.0 | 1.0 |
| *p* | NS | <0.05 | <0.01 | | |

[a] Mean ± standard error. [b] Free alcohol. [c] NS = not significant. [d] *Vs.* testosterone propionate.

Compounds of still lower activity are **3**, **7**, and **11**, which have less than 5–10% the potency of the standard. Finally, **4**, **5**, **10**, **13**, and **18** are inactive. Thus, in the substituted cyclopropane, the order of activity is CHO > H ≫ $CH_2OH$ > $CO_2Et$, $H_2N$ >>> $CO_2H$, $CONCH_2CH_2$, $Me_3N^+$, Br.

Two major conclusions can be drawn from these data. First, activity can be retained or enhanced in substituted compounds. This would not be expected on steric grounds if $\alpha$-face adsorption were involved, but is in harmony with adsorption on the $\beta$ face of the steroid.

Secondly, activity of a given analog is determined by the structure of the substituent on the cyclopropane ring. Therefore, it is now established that in principle such a series of analogs could shed light on the electronic requirements in the A ring, as outlined previously. However, the small number of active compounds makes the present series inadequate for this purpose, and a further discussion must await the completion of another series of compounds, currently being carried out in our laboratory.

## Experimental Section[19]

The method[18] employed in the androgenic–myotrophic assay has been discussed previously.[2]

**2ξ,3ξ-Methano-5α-estran-17β-ol (Isomer A) (3).**—A stirred mixture of 8.5 g (0.10 mole) of Zn–Cu couple, 30 g of $CH_2I_2$, and 60 mg of $I_2$ in 200 ml of anhydrous $Et_2O$ was heated under reflux for 1 hr. Then 3.0 g (0.009 mole) of **1**[2] dissolved in anhydrous $Et_2O$ was added. The mixture was heated under reflux for 90 hr and then filtered through alumina, washed with dilute HCl solution and $H_2O$, and dried ($Na_2SO_4$). After evaporation of the solvent

the residue was dissolved in 100 ml of 5% methanolic KOH solution. It was refluxed for 0.5 hr and the product was obtained by evaporation of the MeOH and addition of $H_2O$. It was recrystallized from MeOH to afford 1.2 g of material, mp 105–110°. Further recrystallization gave the analytical sample, mp 114–115°, $[\alpha]^{28}D$ +77° (*c* 1, $CHCl_3$). *Anal.* ($C_{19}H_{30}O$) C, H.

From the mother liquid there was obtained 0.4 g of **isomer B (4)**, mp 96–102°. Recrystallization from aqueous MeOH gave the analytical sample, mp 104–105°, $[\alpha]^{28}D$ +62° (*c* 1, $CHCl_3$). *Anal.* ($C_{19}H_{30}O$) C, H.

**2α,3α-(β-Carboxymethano)-5α-androstan-17β-ol (5).**—A solution of 1.0 g (0.0025 mole) of **7** in 100 ml of 5% methanolic KOH was refluxed for 30 min, concentrated *in vacuo*, and poured into 300 ml of ice–water. Upon acidification to pH 1 with 20% HCl crystalline product precipitated. It was filtered and dried to afford 0.8 g (96%) of **5**, mp 258–260°. Several recrystallizations from MeOH furnished the analytical sample, mp 265–266°, $[\alpha]^{20}D$ +14° (*c* 0.4, dioxane). *Anal.* ($C_{21}H_{32}O_3$) C, H.

**2α,3α-(β-Carboxymethano)-5α-androstan-17-one (6).**—A solution of 1.20 g of **5** in 200 ml of acetone was allowed to react with 2 ml (excess) of 8 *N* $CrO_3$ solution at 27°. After 20 min, the excess reagent was destroyed with *i*-PrOH and the mixture was filtered. After the addition of a small quantity of $H_2O$, the mixture was evaporated under reduced pressure to give 0.98 g (82%) of product, mp 280–281°. Several recrystallizations from MeCN furnished the analytical sample, mp 282–283°, $[\alpha]^{20}D$ +76° (*c* 1, $CHCl_3$), nmr 0.851 ppm (19-H). *Anal.* ($C_{21}H_{30}O_3$) C, H.

**2α,3α-(β-Carbethoxymethano)-5α-androtan-17β-ol Acetate (7).**—A mixture of 4.0 g (0.0126 mole) of **2**[5] and 0.4 g of anhydrous $CuSO_4$ was heated to 120° and 7.0 g (0.06 mole) of ethyl diazoacetate was added dropwise. The mixture was kept at 120° for 15 min under stirring and 10 ml of 10% HOAc was added to decompose the excess reagent. The resulting mixture was cooled and extracted with $Et_2O$ and the extracts were combined, washed with 5% $NaHCO_3$ solution, and dried ($Na_2SO_4$). The $Et_2O$ solution was evaporated *in vacuo* and the oily residue was treated with 2 ml of cold MeOH to afford, after filtration, 2.3 g (46%) of the crude product, mp 120–125°. Several recrystallizations from MeOH gave the analytical sample, mp 148–150°, $[\alpha]^{20}D$ −15° (*c* 1, $CHCl_3$), nmr 0.778 ppm (19-H). *Anal.* ($C_{25}H_{38}O_4$) C, H.

**Ethylcarbonic 2α,3α-(β-Carboxymethano)-5α-androstan-17-one Anhydride (8).**—A solution of 0.40 g (0.0012 mole) of **6** in 60 ml of $Me_2CO$ and 6 ml of $H_2O$ was cooled to 0° and 0.15 g (0.0014 mole) of $Et_3N$ in 2 ml of $Me_2CO$ was added. While maintaining the temperature at 0°, a solution of 0.144 g (0.0013 mole) of ethyl chloroformate in 2 ml of $Me_2CO$ was added slowly. The mixture was stirred at 0° for 1 hr and the solvent was evaporated giving 0.33 g (69%) of crude product, mp 143–144°. Several recrystallizations from MeCN furnished the analytical sample, mp 145–147°, $[\alpha]^{20}D$ +173° (*c* 1, $CHCl_3$), nmr 0.820 ppm (19-H). *Anal.* ($C_{24}H_{34}O_5$) H; C: calcd, 71.61; found; 71.20.

**2α,3α-[β-Carbo(1-aziridyl)methano]-5α-androstan-17-one (9).**—To a stirred mixture of 0.10 g (0.0024 mole) of ethylenimine and 0.125 g (0.0012 mole) of $MeNH_2$ in 5 ml of $C_6H_6$ was added 0.46 g (0.0012 mole) of **8** in 3 ml of $C_6H_6$ during 1 hr at 0° and the reaction was stirred for an additional 1 hr at 0°. The solvent was evaporated *in vacuo* and the gummy residue was treated with a small amount of cold hexane. The precipitated product (0.23 g) (52%) was recrystallized from hexane to furnish the analytical sample, mp 142–144°, $[\alpha]^{20}D$ +91° (*c* 1, $CHCl_3$). *Anal.* ($C_{23}H_{33}NO_2$) C, H, N.

**2α,3α-[β-Carbo(1-aziridyl)methano]-5α-androstan-17β-ol (10).**—A solution of 0.100 g of **9** and 0.2 g of LiAlH $(t\text{-BuO})_3$ in 10 ml of anhydrous THF was kept at 0° for 1 hr. The product was isolated by $Et_2O$ extraction and recrystallized from MeCN to give the analytical sample, mp 175–176°, $[\alpha]^{20}D$ −16° (*c* 1, $CHCl_3$), nmr 0.788 ppm (19-H). *Anal.* ($C_{23}H_{35}NO_2$) C, H, N.

**2α,3α-(β-Aminomethano)-5α-androstan-17β-ol Hydrochloride (11).**—The crystalline free amine derived from 1.50 g (0.0044 mole) of **12** was dissolved in MeOH and reduced with 0.6 g of $NaBH_4$. The excess $NaBH_4$ was decomposed by addition of 5% HCl and the mixture was concentrated *in vacuo* and poured into ice water. The aqueous solution was made alkaline and extracted with $Et_2O$. The product was rather insoluble in $Et_2O$ and partially precipitated. The 0.85 g (63%) of crude amine obtained was dissolved in anhydrous $Et_2O$ and acidified with saturated ethereal HCl solution. The precipitated salt was filtered and recrystallized from EtOH–$Et_2O$ to give the analytical sample, mp 279–280°, $[\alpha]^{20}D$ +22° (*c* 0.5, $CH_3OH$). *Anal.* ($C_{20}H_{34}ClNO$) C, H, N.

**2α,3α-[β-Aminomethano]-5α-androstan-17-one Hydrochloride (12).**—Compound **6** (2.50 g, 0.0076 mole) was converted to **8** as described. To the resulting reaction mixture was added a solution of 0.815 g (0.012 mole) of $NaN_2$ in 6 ml of $H_2O$. The mixture was stirred for 1 hr and poured into 500 ml of ice water. The white precipitate was collected by filtration and dried to afford 2.42 g (73%) of the crude azide, mp 122° (evolution of $N_2$).

This azide (2.42 g) was dissolved in 12 ml of toluene and heated on a steam bath until no more nitrogen was evolved (1 hr). Removal of toluene *in vacuo* afforded a yellowish oil which was shown to be almost pure isocyanate by its infrared spectrum ($\nu_{max}^{neat}$ 2230 $cm^{-1}$). The isocyanate was suspended in 16 ml of 20% aqueous HCl and the mixture was heated under reflux for 1 hr, during which time the amine HCl precipitated. Recrystallization from 1% aqueous HCl gave 2.00 g (78%) of colorless needles. Further recrystallization from $CH_3OH$–$Et_2O$ gave the analytical sample $[\alpha]^{20}D$ +92° (*c* 1, $CH_3OH$). *Anal.* ($C_{20}H_{32}ClNO$) C, H, N.

**2α,3α-(β-Dimethylaminomethano)-5α-androstan-17β-ol methochloride (13)** was obtained from **11** with MeI and $K_2CO_3$ in MeOH. A solution of 0.36 g of the methiodide in 5 ml of MeOH was passed through 10 g of IRA-400 resin which was washed with MeOH previously. The eluates were collected until neutral and evaporated to dryness. Several recrystallizations from EtOH–$Et_2O$ gave the analytical sample, mp 280–283° dec, $[\alpha]^{20}D$ −31° (*c* 1, $CH_3OH$). *Anal.* ($C_{30}H_{40}ClNO\cdot H_2O$) C, H, N.

**2α,3α-(β-Dimethylaminomethano)-5α-androstan-17-one methochloride (14)** was obtained from **12** with MeI and KOH in MeOH solution. The crude product was recrystallized from $H_2O$ to give 0.43 g (60%) of the quaternary iodide, mp 272–276°. One additional recrystallization from EtOH–$Et_2O$ raised the melting point to 280–283°.

A solution of the iodide in MeOH was passed through IRA-400 resin previously washed with MeOH. Recrystallization from EtOH–$Et_2O$ gave the analytical sample, mp 274–275° dec, $[\alpha]^{20}D$ +58° (*c* 0.5, $CH_3OH$). *Anal.* ($C_{23}H_{38}ClNO$) C, H, N.

**2α,3α-(β-Aldehydomethano)-5α-androstan-17β-ol (15).**—Reduction of 0.68 g (0.0019 mole) of **10** in 180 ml of $Et_2O$ with 0.07 g (0.0019 mole) of $LiAlH_4$ at 0° for 1 hr, decomposition with 10 ml of cold 5 *N* $H_2SO_4$, and work-up using preparative tlc on silica gel gave **16** (27%) and **15** (55%). The product **15** had mp 158–160°, $[\alpha]^{20}D$ +30° (*c* 1, $CHCl_3$). *Anal.* ($C_{21}H_{32}O_2$) C, H.

**2α,3α-(β-Hydroxymethylmethano)-5α-androstan-17β-ol (16).**—A solution of 0.100 g of **9** in 15 ml of EtOH was treated with 0.04 g of $NaBH_4$. The crude product (98%) was crystallized from MeCN–MeOH (7:3) to give the analytical sample, mp 217–218°, $[\alpha]^{20}D$ + 24° (*c* 0.5, dioxane), nmr 0.762 ppm (19-H). *Anal.* ($C_{21}H_{34}O_2$) C, H.

**2α,3α-(ξ-Bromomethano)-5α-androstan-17β-ol Acetate (18).**—Compound **19** was reduced with LiAlH $(t\text{-BuO})_3$ in THF solution in the usual way. Acetylation of the resulting alcohol with $Ac_2O$ in pyridine solution gave **18**. Recrystallization from EtOH gave a sample, mp 159–160°, which was a mixture of bromo epimers as shown by a multiplet at 146–157 Hz in the nmr spectrum. *Anal.* ($C_{22}H_{33}BrO_2$) C, H, Br.

**2α,3α-Methano-5α-androstan-17β-ol.**—To a mixture of approximately 2.0 g of Raney Ni and 0.2 g of the product of the reduction of **19** with LiAlH $(t\text{-BuO})_3$ in 150 ml of MeOH was added 1.0 g of KOH in 5 ml of $H_2O$. The resulting mixture was shaken under $H_2$ at 2.1 $kg/cm^2$ for 6 hr. The catalyst was removed, the filtrate was concentrated *in vacuo* and $H_2O$ was added giving 0.2 g of crude product. Upon separation by preparative tlc (35%

(19) Melting points were determined with a Thomas–Hoover apparatus equipped with a corrected thermometer. Ir spectra were obtained with a Beckman IR-8 or Perkin-Elmer 337 instrument. Microanalyses were performed by the Microanalytical Department, University of California, Berkeley, Calif. Nmr spectra were obtained at a field strength of 60 MHz on samples in $CDCl_3$ solutions on a Varian A-60A instrument, using TMS as internal standard. When only small amounts of sample were available, a Varian C-1024 computer was used for time averaging. Optical rotations were obtained in a 0.5-dm tube with a Rudolph photoelectric polarimeter. Glpc was carried out using a Barber-Coleman Model 5000 system employing 1.83-m U-tube columns of 2% SE-30 on Gas Chrom Q or Z, He carrier, $H_2$ flame detection, column temperatures of 220–240°, "on column" injection at 290°, and detector temperatures of 250°. Where analyses are indicated only by symbol of the elements or functions, analytical results obtained for those elements or functions were within ±0.4% of the theoretical values.

petroleum ether (bp 30–60°) in $Et_2O$ was used in the development), there was obtained 0.076 g of 2α,3α-methanol-5α-androstan-17β-ol, mp 129–130° after recrystallization from MeOH. Ir and nmr spectra and mixture melting point were indistinguishable from those of the authentic sample.[2]

The compound with lower $R_f$ value was isolated in a yield of 0.093 g and was shown to be a single isomer of **2α,3α-(bromomethano)-5α-androstan-17β-ol**. One recrystallization from EtOH gave colorless crystals: mp 145–146°; nmr 0.72 (C-19 methyl), 0.75 (C-18 methyl), 2.55 (triplet, bromo proton) 3.73 (triplet, 17α-H) ppm.

**2α,3α-(ξ-Bromomethano)-5α-androstan-17-one (19).**—To a refluxing mixture of 1.50 g (0.0045 mole) of **6** and 2.00 g of red HgO in anhydrous $CCl_4$ was added 0.72 g (0.0045 mole) of $Br_2$ in 5 ml of anhydrous $CCl_4$. The resulting mixture was refluxed gently for 1.5 hr and was filtered after cooling. The clear yellow filtrate was washed ($H_2O$, 5% NaOH, $H_2O$), dried ($Na_2SO_4$), and evaporated *in vacuo*. The oily residue was treated with petroleum ether (bp 30–60°) to give 0.60 g (36%) of crystals. Several recrystallizations from $Me_2CO$ gave the analytical sample: mp 159–160°; nmr 0.77, 0.78 (C-19 $CH_3$) ppm. *Anal.* ($C_{20}H_{29}BrO$) C, H.

# Synthesis of 6,7-Difluoromethylene Corticoids[1]

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*Received March 12, 1968*

The synthesis of 6α,7α- and 6β,7β-difluoromethylene corticoids by addition of "difluorocarbene" to selected $\Delta^{4,6}$-3-keto steroids is described. The observed potentiation of corticoid activity by both α- and β-face difluoromethylene adducts is inconsistent with an antiinflammatory receptor site which requires binding to rings A and B of the steroid molecule.

The enormous effort expended in the synthesis and modification of cortisone has led to the accumulation of a considerable body of empirical knowledge relating structure to antiinflammatory activity. Within the last two decades every position of the cortisone molecule has been subject to scrutiny and chemical modification. These efforts have resulted in the discovery of a number of activity-enhancing and activity-modifying groups which alone or in combination have led to the development of several clinically useful corticoids.

Although the primary locus of corticoid action is unknown, the hypothesis is generally accepted that biological action is the result of an interaction with a complementary receptor site. Considerable speculation as to the nature and geometry of the receptor site has led to the suggestion that corticoids interact with a surface complementary to a portion of the β face of the steroid molecule.[3-6]

Sarett[3,4] has further defined the properties of the receptor by suggesting rigid geometry with provisions for specific binding to the 11β-hydroxyl and to the 3- and 20-keto groups of hydrocortisone.[7] Additional interactions are provided by the summation of London forces over the total of the β face of the steroid molecule.

Alternatively, Bush[6] envisages little if any binding to rings A and B with the major interactions being provided by the 11β-hydroxyl, rings C and D, and the side chain. With this hypothesis, the requirements of the receptor would not be inconsistent with axial β-face B-ring substituents. The inactivity of 6β-halo and 6β-methyl corticoids, an important consideration in the previous proposal,[3,4] was rationalized by the suggestion that a general distortion of the steroid molecule due to intramolecular interaction with the axial 6-substituent interfered with binding.[6,8]

Clearly a critical evaluation of these hypotheses must be based on biologically active compounds.[9] In particular, an active corticoid substituted with bulky β-face B-ring substituents would provide evidence in support of the proposal that rings A and B are not involved in binding to the complementary surface of the receptor.

We have recently reported an efficient method for the preparation of 6,7-difluoromethylene steroids.[10,11] The application of these findings to the corticoid series provided an opportunity to further evaluate the requirements for biological activity.

Addition of "difluorocarbene" to the dienones **1a**, **b** gave a mixture of products from which the 6α,-7α-difluoromethylene adducts **2a**, **b** were isolated after

(1) Publication 340 from the Syntex Institute of Steroid Chemistry. For publication 339 see P. H. Nelson, J. W. Murphy, J. A. Edwards, and J. H. Fried, *J. Am. Chem. Soc.*, in press. This publication is also part VI of the series, Methylenation of Unsaturated Ketones. Part V: G. Tarzia, N. H. Dyson, I. T. Harrison, J. A. Edwards, and J. H. Fried, *Steroids*, **9**, 387 (1967). A portion of this material was presented at the symposium on Antiinflammatory Agents sponsored by the Medicinal Chemistry Section at the 154th National Meeting of the American Chemical Society, Chicago, Ill., Sept 1967.

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(3) L. H. Sarett, *Ann. N.Y. Acad. Sci.*, **82**, 802 (1959).

(4) L. H. Sarett, A. A. Patchett, and S. L. Steelman, *Progr. Drug Res.*, **5**, 13 (1963).

(5) J. Fried, "Mechanism of Action of Steroid Hormones," The MacMillan Co., New York, N. Y., 1961, p 232.

(6) I. E. Bush, *Pharmacol. Rev.*, **14**, 447 (1963).

(7) The 3-ketone can be replaced by a 2,3-fused heterocyclic ring with a considerable enhancement of biological activity; *cf.* R. Hirschmann, N. G. Steinberg, P. Buchschacher, J. H. Fried, G. J. Kent, M. Tishler, and S. L. Steelman, *J. Am. Chem. Soc.*, **85**, 120 (1963); J. H. Fried, P. Buchschacher, and H. Mrozik, *Steroids*, **2**, 391 (1963); H. Mrozik, P. Buchschacher, J. Hannah, and J. H. Fried, *J. Med. Chem.*, **7**, 584 (1964); P. DeRuggieri, C. Candolfi, H. Guzzi, D. Chiaiamonte, and C. Ferrari, *Farmaco*, **20**, 280 (1965).

(8) The contrasting antiinflammatory activities of 6β- and 6α-fluoro corticoids can also be attributed to a more rapid metabolic reduction of the C-3 ketone in the 6β-fluoro series; *cf.* H. J. Ringold, S. Rainachandran, and E. Forchielli, *Biochim. Biophys. Acta*, **82**, 143 (1964).

(9) Since the biological action of hormones depends upon numerous biochemical and physicochemical equilibria involved in the process of absorption, distribution, and metabolism, absence of biological activity cannot be safely ascribed to the lack of binding and, therfore, inferences as to the geometry of the receptor site based on inactive compounds are open to question.

(10) C. Beard, N. H. Dyson, and J. H. Fried, *Tetrahedron Letters*, 3281 (1966); C. Beard, I. T. Harrison, L. Kirkham, and J. H. Fried, *ibid.*, 3287 (1966).

(11) G. Tarzia, N. H. Dyson, I. T. Harrison, J. A. Edwards, and J. H. Fried, *Steroids*, **9**, 387 (1967); C. Beard, B. Berkoz, N. H. Dyson, I. T. Harrison, P. Hodge, L. Kirkham, G. Lewis, D. Giannini, J. A. Edwards, and J. H. Fried, to be published.