was packed on top of a column prepared from 500 g. of Celite and 250 ml. of the lower phase and the column (5.5 × 60 cm.) was developed with the upper phase (800 cc. h.b.v.). The effluent stream was monitored at 240 mμ, and the large peak eluted in the 2nd and 3rd h.b.v. was collected by evaporation of relevant fractions. The material so obtained (386 mg.) was still impure and was recolumned in the same manner on 300 g. of Celite from the system cyclohexane–dioxane–methanol–water (10:8:2:2).[37] The column (3.8 × 66 cm.; 410 ml. h.b.v.) was developed with the upper phase of the solvent system and there was isolated from a major peak in the 4th h.b.v. 246 mg. of solid which after several recrystallizations from ether–methylene chloride afforded 126 mg. (6.7%) of product; m.p. 211–213°; $[\alpha]_D$ + 62.4° (*c*, 1.01), MD + 281.7, ΔMD (product-parent) − 345; λ_{max} 230 mμ (ε 13,860); 5.85 (s), 5.94 (s), 5.99 μ (shoulder). The corresponding 6α-epimer[25] has $[\alpha]_D^{CH_3OH}$ + 120°, MD + 545; compound III has $[\alpha]_D^{CH_3OH}$ + 129°, MD + 563; ΔMD (6α epimer-III) − 18.

Anal. Calcd. for $C_{24}H_{32}F_2O_6$: C, 63.41; H, 7.10; F, 8.36. Found: C, 63.62; H, 7.29; F, 8.65.

(37) In subsequent experiments it was found that this solvent system could be used directly on crude material to give the desired product in a single chromatogram.

Synthesis of Certain 16α-Substituted Derivatives of 9α-Fluoro-11-dehydrocorticosterone, Progesterone and Deoxycorticosterone

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Received April 26, 1962

The various 16-substituted derivatives were prepared by 1,4-addition to the appropriate 16-dehydro-20-keto steroids of certain mercaptans, thioacetic acid, hydrogen chloride, nitromethane, methanol, and certain primary and secondary amines.

The introduction of new groups into the 16-position of steroids having glucocorticoid activity has often resulted in compounds with significantly enhanced biological utility. In fact, substitution at C-16 has been one of the most fruitful approaches to corticoid analog research in recent years. Among the groups which have been introduced at this position and which have exerted a favorable biological

effect are the 16α-hydroxy,[1] 16α- and 16β-methyl[2] and 16α-fluoro[3] groups. Fully elaborated glucocorticoids having the 16β-hydroxy,[4] 16β-fluoro,[5] 16α-methoxy,[6] 16β-methoxy,[5b] or 16β-chloro[7] substituents present have also been prepared but substitution by these groups at best seems to afford no important biological advantage.

In the course of a research program carried out with the purpose of discovering new structure-activity principles in the glucocorticoid field it was therefore of considerable interest to investigate the effect on glucocorticoid activity that other substituents at C-16 might have. However, in general the construction of 16-substituted (particularly 16α-) fully elaborated glucocorticoid derivatives present a synthetic problem of considerable magnitude, one major difficulty being the introduction of the 17α-hydroxyl group. In order to minimize the potential synthetic difficulties, we have taken advantage of the fact that, at least in the several reported instances,[8] the 17-deoxy-11-oxygenated corticoids appear to respond to the introduction of new groups in a manner roughly parallel to the response of the corresponding 17α-hydroxy derivatives when this response is measured by thy-

(1) S. Bernstein, R. H. Lenhard, W. S. Allen, M. Heller, R. Littell, S. M. Stolar, L. I. Feldman and R. H. Blank, *J. Am. Chem. Soc.*, **78**, 5693 (1956); **81**, 1689 (1959).

(2) G. E. Arth, D. B. R. Johnston, J. Fried, W. W. Spooncer, D. R. Hoff and L. H. Sarett, *J. Am. Chem. Soc.*, **80**, 3160 (1958); (b) G. E. Arth, J. Fried, D. B. R. Johnston, D. R. Hoff, L. H. Sarett, R. H. Silber, H. Stoerk and C. A. Winter, *J. Am. Chem. Soc.*, **80**, 3161 (1958); (c) E. P. Oliveto, R. Rausser, A. L. Nussbaum, W. Gebert, E. B. Hershberg, S. Tolksdorf, M. Eisler, P. L. Perlman, and M. M. Pechet, *J. Am. Chem. Soc.*, **80**, 4428 (1958); (d) E. P. Oliveto, R. Rausser, L. Weber, A. L. Nussbaum, W. Gebert, C. T. Coniglio, E. B. Hershberg, S. Tolksdorf, M. Eisler, P. L. Perlman and M. M. Pechet, *J. Am. Chem. Soc.*, **80**, 4431 (1958); (e) D. Taub, R. D. Hoffsommer, H. L. Slates and N. L. Wendler, *J. Am. Chem. Soc.*, **80**, 4435 (1958); (f) D. Taub, R. D. Hoffsommer, H. L. Slates, C. H. Kuo and N. L. Wendler, *J. Am. Chem. Soc.*, **82**, 4012 (1960); (g) E. P. Oliveto, R. Rausser, H. L. Herzog, E. B. Hershberg, S. Tolksdorf, M. Eisler, P. L. Perlman and M. M. Pechet, *J. Am. Chem. Soc.*, **80**, 6687 (1958).

(3) B. J. Magerlein, R. D. Birkenmeyer, and F. Kagan, *J. Am. Chem. Soc.*, **82**, 1252 (1960).

(4) S. Bernstein, M. Heller and S. M. Stolar, *J. Am. Chem. Soc.*, **81**, 1256 (1959).

(5) (a) D. E. Ayer and W. P. Schneider, *J. Am. Chem. Soc.*, **82**, 1249 (1960); (b) W. T. Moreland, R. G. Berg, D. P. Cameron, C. E. Maxwell III, J. S. Buckley and G. D. Laubach, *Chem. & Ind. (London)*, 1084 (1960).

(6) (a) S. Bernstein, M. Heller and S. M. Stolar, *Chem. & Ind. (London)*, 516 (1961); (b) G. R. Allen, Jr. and N. A. Austin, *J. Org. Chem.*, **26**, 4574 (1961).

(7) R. E. Beyler and F. Hoffman, *J. Org. Chem.*, **21**, 572 (1956).

(8) 1-Dehydrocorticosterone is about 2.5 times as active as corticosterone.[9] 9α-Fluorocorticosterone and 9α-fluoro-11-dehydrocorticosterone show enhanced liver glycogen and sodium retention activity.[10] Moreover, 16α-hydroxylation of 1-dehydro-9α-fluorocorticosterone abolishes the sodium-retaining property and decreases, but does not eliminate, the glucocorticoid activity of the parent compound.[11] Note should be taken of the fact that, although 12α-fluoro and 12α-chlorocorticosterone derivatives show increased glycogenic action, the corresponding 17α-hydroxy derivatives are relatively inactive.[12]

(9) S. Tolksdorf, M. L. Battin, J. W. Cassidy, R. M. MacLeod, F. H. Warren and P. L. Perlman, *Proc. Soc. Exptl. Biol. Med.*, **92**, 207 (1956).

(10) J. Fried, J. E. Herz, E. F. Sabo, A. Borman, F. M. Singer and P. Numerof, *J. Am. Chem. Soc.*, **77**, 1068 (1955).

(11) C. E. Holmlund and co-workers, private communication.

(12) J. Fried and A. Borman, *Vitamins and Hormones*, **XVI**, 366 (1958).

molytic, liver glycogen and electrolyte assays in the rat. Since it could reasonably be assumed that 16α-substituted 17-deoxycorticoids would be readily available by the addition of various nucleophiles to an appropriate 16-dehydro-20-keto derivative,[13] it appeared to us that considerable preliminary information concerning the effect of a particular group on at least certain of the important parameters of biological activity could be gained conveniently by the synthesis of appropriate 16α-substituted corticosterone derivatives. For our study, we chose to prepare derivatives of 9α-fluoro-11-dehydrocorticosterone since this compound has relatively high glucocorticoid activity and is also a strong salt retainer.[10] It was our thought that we would therefore be able to detect not only those groups which enhanced glucocorticoid activity but also those groups which might cause a reversal of 9α-fluorine-induced salt retention even at the expense of a significant decrease in glucocorticoid-type activity.

The requisite 11,16-bisdehydro-9α-fluorocorticosterone (II) and its acetate I are available from 9α-fluorocortisone by 3,20-bisketalization, 21-acetylation, 17α-hydroxyl elimination,[14] ketal hydrolysis (to give II) and finally reacetylation (to give I). 1,4-Addition to the 16-dehydro-20-keto moiety of I and II (and related compounds) was carried out with mercaptans, thioacetic acid and hydrogen chloride under conditions of acid catalysis and with nitromethane, methanol and certain amines under conditions of base catalysis. Thus, acid-catalyzed condensation of I with methyl mercaptan, ethyl mercaptan and thioacetic acid gave the corresponding 16α-substituted derivatives III–V. With isopropyl mercaptan, this procedure afforded the 16α-isopropylthio-3-thioenol ether, which on acid hydrolysis gave the desired 16α-isopropylthio-Δ^4-3-ketone VII. Stepwise oxidation with monoperphthalic acid[15] of the 16α-methylthio derivative III afforded the corresponding methylsulfinyl and methylsulfonyl derivatives VIII and IX, respectively. An attempt to prepare the 16α-sulfhydryl derivative X by treatment of the acetylthio

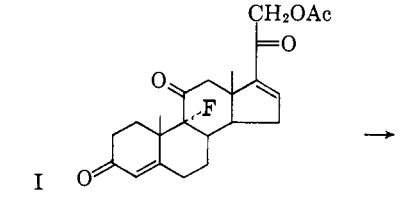
(13) The condensation of 16-dehydro-20-keto steroids with a variety of nucleophiles to give 16-substituted-20-ketones has been reported. For examples see the following: (a) with thioacetic acid and hydrogen sulfide: R. M. Dodson and P. B. Sollman, U. S. Patent 2,912,443 (Nov. 10, 1959); (b) with hydrogen chloride: R. M. Dodson and P. B. Sollman, U. S. Patent 2,708,201 (May 10, 1955); (c) with amines: D. Gould, E. L. Shapiro, L. E. Finckenor, F. Gruen and E. B. Hershberg, *J. Am. Chem. Soc.*, **78**, 3158 (1956); (d) with nitromethane: R. M. Dodson, U. S. Patent 2,913,466 (Nov. 17, 1959); (e) with alcohols: D. K. Fukushima and T. F. Gallagher, *J. Am. Chem. Soc.*, **73**, 196 (1951); D. Gould, F. Gruen and E. B. Hershberg, *J. Am. Chem. Soc.*, **75**, 2510 (1953).

(14) The intermediate 21-acetoxy-3,20-bisethylenedioxy-9α-fluoropregna-5,16-dien-11-one has been described by J. Fried and G. H. Thomas, U. S. Pat. 2,963,496 (Dec. 6, 1960) and W. S. Allen, H. M. Kissman, S. Mauer, I. Ringler and M. J. Weiss, *J. Med. and Pharm. Chem.*, **5**, 133 (1962).

(15) R. E. Schaub and M. J. Weiss, *J. Org. Chem.*, **27**, 2221 (1962)

derivative V with methanolic perchloric acid resulted in preferential de-*O*-acetylation to give the 16α-acetylthio-21-ol VI. However, brief methanolic methoxide treatment of the diacetate V gave the desired sulfhydryl (21-OH) derivative X in 38% yield. From this experiment there was also isolated in 25% yield a product which appears to be the bis-steroidal sulfide XVII, presumably formed by hydrogen sulfide elimination followed by addition of a 16α-sulfhydryl molecule to the resulting 16-dehydro-20-ketone. Continuing our investigation, the 16α-chloro derivative XI was prepared by reaction of I with hydrogen chloride in dioxane. However, an attempt to bring about the addition of hydrogen fluoride failed, starting material being recovered.

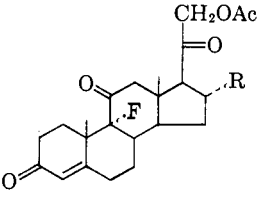
It was also possible to effect the addition of certain primary and secondary amines to the 16-dehydro-20-keto system in II. This condensation was carried out by a procedure previously described by Gould and co-workers[13c] in which the catalyst is an anion-exchange resin such as Amberlite IRA-400 resin. In this manner we were able to prepare the 16α-methylamino (XII), 16α-dimethylamino (XIII), 16α-piperidino (XIV) and 16α-anilino (XV) derivatives of 9α-fluoro-11-dehydrocorticosterone. The 16α-methoxy derivative XVI of 9α-fluoro-11-dehydrocorticosterone was obtained (5% yield) in the course of a methanolic methoxide deacetylation of the 16-dehydro-21-acetate I.



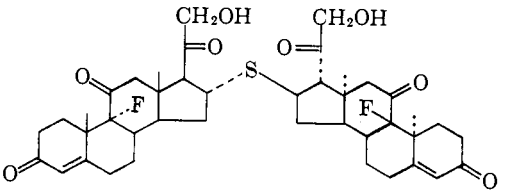
I

II, 21-OH

III, R = SCH_3
IV, R = SC_2H_5
V, R = $SCOCH_3$
VI, R = $SCOCH_3$ (21-OH)
VII, R = $SCH(CH_3)_2$
VIII, R = $SOCH_3$
IX, R = SO_2CH_3

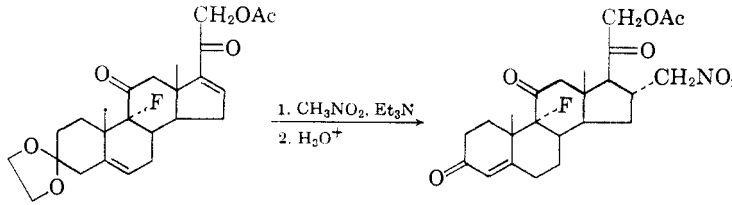


X, R = SH (21-OH)
XI, R = Cl
XII, R = $NHCH_3$ (21-OH)
XIII, R = $N(CH_3)_2$ (21-OH)
XIV, R = N⟨⟩ (21-OH)
XV, R = NHC_6H_5 (21-OH)
XVI, R = OCH_3 (21-OH)



XVII

The 16α-nitromethyl derivative (XIX) of 9α-fluoro-11-dehydrocorticosterone was also prepared. *A priori*, we considered that the preparation of this compound *via* the base-catalyzed addition of nitromethane to a 16-dehydro-20-ketone would require blocking of the 3-keto group.[13d] The 16-dehydro-3-ketal XVIII was therefore prepared by preferential ketalization.[16] Nitromethane addition to XVIII proceeded smoothly and the desired XIX was then readily obtained on ketal hydrolysis. The nitromethyl derivatives are of considerable additional interest as possible precursors for the preparation of various monosubstituted 16α-methyl derivatives. At a later date we hope to report on our efforts in this area.



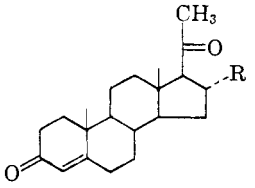
XVIII XIX

Finally, we wish to report the preparation by analogous procedures of certain 16α-substituted derivatives of progesterone and of deoxycorticosterone. Thus, condensation of 16-dehydroprogesterone with hydrogen chloride gave 16α-chloroprogesterone (XX)[17] and with methyl mercaptan 16α-methylthioprogesterone (XXI). Starting with the latter compound, stepwise oxidation with monoperphthalic acid gave the corresponding sulfoxide XXII and sulfone XXIII. Similarly from 16-dehydrodeoxycorticosterone or its 21-acetate, the 16α-chloro,[17] 16α-piperidino, 16α-acetylthio, 16α-methylthio, 16α-methylsulfinyl and 16α-methylsulfonyl derivatives (XXIV–XXIX) were prepared. A by-product, obtained in substantial yield, of the methylthio preparation was a compound which is apparently the 16α-methylthio-20-thioketal XXX.

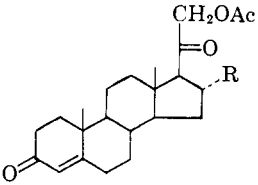
Most of the various 16α-substituted derivatives reported in this paper are listed in Table I. We have assigned the α-configuration to these derivatives on grounds previously reviewed by Gould and coworkers.[13c] In support of this designation, the rotatory contribution of the various 16-substituents, with two exceptions, (the nitromethyl derivative XIX and the methylsulfinyl derivatives VIII and XXII)

(16) H. J. Dauben, Jr., B. Löken and H. J. Ringold, *J. Am. Chem. Soc.*, **76**, 1359 (1954).

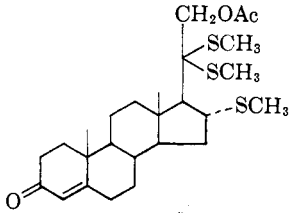
(17) The preparation of 16-chloroprogesterone and 16-chlorodeoxycorticosterone acetate by the addition of hydrogen chloride to 16-dehydropregnenolone and 21-acetoxy-16-dehydropregnenolone, respectively, followed by Oppenauer oxidation of the resulting mixture of 16-chloro and 5,16-dichloro derivatives has been reported.[13b]



XX, R = Cl
XXI, R = SCH_3
XXII, R = $SOCH_3$
XXIII, R = SO_2CH_3



XXIV, R = Cl
XXV, R =  (21-OH)
XXVI, R = $SCOCH_3$
XXVII, R = SCH_3
XXVIII, R = $SOCH_3$
XXIX, R = SO_2CH_3



XXX

is substantially negative (see Table I).[18] However, it should be noted that in the case of the two 16-methylsulfinyl derivatives VIII and XXII, the corresponding precursor methylthio and succeeding methylsulfonyl derivatives have normal ΔMD values.[19]

Biological Evaluation.—Of the various 16-substituted-9α-fluoro-11-dehydrocorticosterone derivatives, the most interesting was the 16α-acetylthio derivative V and the related deacetylated compounds VI and X. Although V, with an activity in a thymolytic assay[20] 0.33 (95% confidence limits: 0.23–0.46) times hydrocortisone, was substantially less active than the parent 9α-fluoro-11-dehydrocorticosterone[10] [2.5(1.8–3.6)], it showed a reversal of the strong salt-retaining properties[21] of the latter compound. Similar observations were made with the 16α-acetylthio-21-ol VI and the 16α-sulfhydryl-21-ol X.

(18) However, see ref. 6b where little difference in rotatory contribution was observed in the case of two epimeric pairs of 16-methoxy derivatives.

(19) The rotatory contribution of several 7α-methylsulfinyl steroids is similar to that made by the corresponding methylthio and methylsulfonyl derivatives (see ref. 15).

(20) Subcutaneous assay carried out by the procedure of I. Ringler and R. Brownfield, *Endocrinology*, **66**, 900 (1960).

(21) The mineralocorticoid activity was determined in adrenalectomized male rats and is a measure of the urinary response to 16 mcg. of steroid administered subcutaneously following a 5 ml. oral dose of tap water and 1 hr. later a 5 ml. saline load I.P. (unpublished procedure of S. Mauer and I. Ringler of these laboratories).

TABLE I

VARIOUS 16-SUBSTITUTED

Compound[a]	Yield % (m.p., °C.)	Anal. purity m.p., °C.	$[\alpha]_D$ ($CHCl_3$)	M_D	ΔM_D[a]	CH_3OH λ_{max},mμ (ε)	Formula
9α-Fluoro-16α-methylthio-AA, III	77(160–175)	182–184	+142	+640	−169	234(18,000)	$C_{24}H_{31}FO_5S$
16α-Ethylthio-9α-fluoro-AA, IV	30(113–120)	118–120	+137	+635	−174	235(17,150)	$C_{25}H_{33}FO_5S$
16α-Acetylthio-9α-fluoro-AA,V	67(143–149)	159–163	+127	+606	−203	234(22,000)	$C_{25}H_{31}FO_6S$
9α-Fluoro-16α-isopropylthio-AA, VII	45(166–177)	180–184	+112	+536	−273	234(14,800)	$C_{26}H_{35}FO_5S$
9α-Fluoro-16α-methylsulfinyl-AA, VIII	64(166–170)	171–174	+235	+1100	+291	236(17,200)	$C_{24}H_{31}FO_6S$
9α-Fluoro-16α-methylsulfonyl-AA, IX	82(208–211)	208–211	+160	+772	−37	233(17,300)	$C_{24}H_{29}FO_7S$
9α-Fluoro-16α-mercapto-A, X	38(178–193)	183–188	+162	+640	−169	235(18,500)	$C_{21}H_{27}FO_4S$
16α-Chloro-9α-fluoro-AA, XI	63(95–98	90–96 176-179	+163	+715	−94	234(16,700)	$C_{23}H_{28}ClFO_5$[b]
9α-Fluoro-16α-methylamino-A, XII	48(150–162 dec.)	152–158 dec.	+132			235(18,100)	$C_{22}H_{30}FNO_4$
16α-Dimethylamino-9α-Fluoro-A, XIII[c]	31(151–4)	160–162				234(18,100)	$C_{23}H_{32}FNO_4$
9α-Fluoro-16α-piperidino-A, XIV	31(165–170 dec.)	171–174 dec.	+145			234(17,000)	$C_{26}H_{36}FNO_4$
16α-Anilino-9α-fluoro-A, XV[c]	63(180–183)	180–183					$C_{27}H_{32}FNO_4$
9α-Fluoro-16α-nitromethyl-AA, XIX	67(234–237)	239–242	+188	+870	+61	233(17,600)	$C_{24}H_{30}FNO_7$
16α-Chloro-P, XX	63(178–190)	191–200 dec.[d]	+150	+524	−79	240(16,200)	$C_{21}H_{29}ClO_2$[e]
16α-Methylthio-P, XXI	58(142–149)	146–150	+118	+424	−179	240(14,400)	$C_{22}H_{32}O_2S$
16α-Methylsulfinyl P, XXII	80(143–153)	164–168	+198	+745	+142	240(16,200)	$C_{22}H_{32}O_3S$
16α-Methylsulfonyl P, XXIII	79(209–215)	229–232	+125	+491	−112	239(17,400)	$C_{22}H_{32}O_4S$
16α-Chloro-DOCA, XXIV	37(165–175)	181–183[f]	+122	+497	−195	240(16,700)	$C_{23}H_{31}ClO_4$[g]
16α-Piperidino-DOC, XXV	36(88–100)	160–169	+78	+322	−290	240(18,200)	$C_{26}H_{39}NO_4$
16α-Acetylthio-DOCA, XXVI[c]	50(150–153)	153–155	+71	+446	−246	238(20,100)	$C_{25}H_{34}O_5S$
16α-Methylthio-DOCA, XXVII[c,h]	34(157–162)						
16α-Methylsulfinyl-DOCA, XXVIII[c,i]	84(146–153)						
16α-Methylsulfonyl-DOCA.[c] XXIX	65(173–178	177–178	+116	+518	−174	240(17,800)	$C_{24}H_{34}O_6S$

[a] A = 21-Hydroxypregn-4-ene-3,11,20-trione;
AA = 21-Acetoxypregn-4-ene-3,11,20-trione [M_D + 809 ($CHCl_3$)];
DOCA = 21-Acetoxypregn-4-ene-3,20-dione [M_D + 692 ($CHCl_3$). G. Rosenkranz, O. Mancera and F. Sondheimer, *J. Am. Chem. Soc.*, **77**, 145 (1955)];
DOC = 21-Hydroxypregn-4-ene-3,20-dione [M_D + 612 ($CHCl_3$)];
P = Pregn-4-ene-3,20-dione [M_D + 603 ($CHCl_3$). W. Dirscherl and F. Hanusch, *Z. physiol. Chem.*, **252**, 49 (1938)];

Analyses									
C		H		F		N		S	
Calcd.	Found	Calcd.	Found	Calcd.	Found	Calcd.	Found	Calcd.	Found
63.98	64.08	6.93	7.27	4.21	4.38			7.12	7.52
64.63	64.80	7.16	6.91	4.09	4.17			6.91	7.08
62.73	63.08	6.53	6.79	3.97	3.96			6.70	7.03
65.25	65.39	7.37	7.57	3.97	4.19			6.69	6.77
61.77	61.79	6.70	6.76	4.08	3.98			6.87	7.03
59.74	59.83	6.47	6.74	3.94	3.95			6.64	6.82
63.93	64.18	6.90	7.18	4.81	4.46			8.13	8.00
62.93	62.84	6.43	6.71	4.33	4.29				
67.49	67.39	7.72	7.81	4.85	4.89	3.57	3.36		
68.12	68.23	7.95	8.16	4.68	4.15	3.46	3.13		
70.07	70.11	8.14	8.17	4.26	4.11	3.14	3.42		
71.50	70.93	7.11	7.47	4.19	3.68	3.09	2.85		
62.19	62.30	6.52	6.60	4.10	4.13	3.02	2.93		
72.28	71.87	8.35	8.71						
73.29	72.95	8.95	9.06					8.89	8.84
70.17	69.99	8.57	8.54					8.52	8.55
67.31	66.95	8.24	8.43					8.17	8.31
67.86	68.01	7.68	7.70						
75.50	75.36	9.51	9.60			3.39	3.34		
67.23	66.97	7.67	8.03					7.18	6.89
63.98	63.94	7.61	7.56					7.12	7.19

ΔM_D = M_D (product) − M_D (parent). [b] Chlorine anal., calcd.: 8.8. Found: 8.13. [c] Preparation carried out by E. Ruckel and J. Nocera. [d] Lit.[17] m.p. 197–199°. [e] Chlorine anal. calcd.: 10.13. Found: 10.45. [f] Lit.[17] m.p. 179–181°. [g] Chlorine anal., calcd.: 8.71. Found: 8.89. [h] Satisfactory analytical data were not obtained for XXVII: the preparation was probably contaminated with XXX. [i] Satisfactory analytical data were not obtained; the preparation, as evidenced by analytical and infrared data, was contaminated with sulfone.

The 16-substituted progesterone derivatives XX–XXII when assayed by the Clauberg-MacPhail procedure (subcutaneously) appeared to be less active than progesterone.[22] The 16-substituted deoxycorticosterone derivatives XXIV and XXVII–XXIX were inactive in an anti-DOC assay.[23]

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Experimental

General.—Unless otherwise stated, *melting points* were determined on a Kofler micro-hot-stage and are corrected. *Infrared spectra* were determined in KBr disks with a Perkin-Elmer spectrophotometer (model 21). The infrared spectra for the compounds in Table I were consistent with the assigned structures. *Ultraviolet spectra* were determined in methanol solution with a Cary recording spectrophotometer. *Polarimetric* data were obtained in chloroform solution in a 1-dm. semimicro tube (*c* 0.5–1.6). It is preferable to carry out the ultraviolet and polarimetric determinations immediately on preparation of the necessary solution, since certain of these products in solution seem to have a tendency to decompose slowly. Solutions were *dried* over magnesium sulfate and *decolorized* with activated charcoal. All *evaporations* were carried out *in vacuo*.

9α-Fluoro-21-hydroxypregna-4,16-diene-3,11,20-trione (II).—A solution of 21-acetoxy-9α-fluoro-3,20-bisethylenedioxypregna-5,16-dien-11-one[14] (20.0 g., 0.041 mole) in methanol (500 ml.) containing 8% (v./v.) sulfuric acid (25 ml.) was stirred at reflux for 1 hr. The cooled reaction mixture was neutralized with 10% sodium hydroxide solution and the solvent was evaporated until all the methanol was removed. The residual aqueous suspension was poured into ice water, and this mixture was extracted with methylene chloride. The combined organic extracts were washed with water and saline solution, dried, decolorized, and evaporated. The residue was crystallized from methylene chloride-ether to yield 11.3 g. (77%) of II, m.p. 210–230°. The analytical sample was prepared by 3 recrystallizations from methylene chloride-ether and one from acetone; m.p. 225–245°; $[\alpha]^{25}D$ +172°; λ_{max} 2.95 (m), 5.79 (s), 5.96 (s, broad band), 6.13 (m), 6.29 μ (m); 236 mμ (ε 25,200).

Anal. Calcd. for $C_{21}H_{25}FO_4$: C, 69.98; H, 6.99; F, 5.27. Found: C, 69.91; H, 6.68; F, 5.15.

(22) Testing carried out by the Endocrine Laboratories, Madison, Wis.

(23) This assay is a modification (G. Fanelli and W. Sullivan of these laboratories) of one described by E. Rosemburg and I. Engel, *Endocrinology*, **69**, 496 (1961).

21-Acetoxy-9α-fluoropregna-4,16-diene-3,11,20-trione (I).—9α-Fluoro-21-hydroxypregna-4,16-diene-3,11,20-trione (II) (6.0 g., 16.6 mmoles), in pyridine (40 ml.) was acetylated with acetic anhydride (10 ml.) in the usual manner. The product (I) was crystallized from methylene chloride-ether to give 5.67 g. (84%); m.p. 208–216°. For analysis, the sample was recrystallized 3 times from methylene chloride-ether; m.p. 217-220°; $[\alpha]_D$ +175°; λ_{max} 5.71 (s), 5.78 (s), 5.93 (s, broad), 6.13 (m), 6.26 (m), 8.02 μ (s); 237 mμ (ε 25,800).

Anal. Calcd. for $C_{23}H_{27}FO_5$: C, 68.63; H, 6.76; F, 4.72. Found: C, 68.67; H, 6.92; F, 4.82.

General Procedures for the Preparation of the Various 16-Sulfur Derivatives. A. Addition of Sulfur Nucleophiles to Δ[16]-20-Keto Systems.—A solution of the appropriate 16-dehydro-20-ketone [21-acetoxy-9α-fluoropregna-4,16-diene-3,11,-20-trione (I), 16-dehydroprogesterone or 16-dehydrodeoxycorticosterone acetate] (500 mg.) in glacial acetic acid (25 ml.) was chilled, and concd. hydrochloric acid (1.0 ml.) was added. To this solution was added a 3- to 10-fold excess of the appropriate sulfur nucleophile (methyl mercaptan, ethyl mercaptan, isopropyl mercaptan, and potassium thioacetate). When potassium thioacetate was used the amount of hydrochloric acid was increased by an equivalent quantity. The reaction vessel was stoppered, stored at 5–7° for 4 days (on occasion the reaction solution froze under these circumstances, in which case the mixture was allowed to melt and then was again stored). The solvent was then evaporated at below 30° and the residue was partitioned between methylene chloride and water. The organic phase was washed with saturated sodium bicarbonate solution, water and saline solution, dried, decolorized and evaporated. The residual material was crystallized from ether, methylene chloride-ether or hexane. The compounds were then recrystallized for analysis from methylene chloride-ether. The yields, physical properties and analytical data for the various products are listed in Table I.

Condensation of **isopropyl mercaptan** with **21-acetoxy-9α-fluoropregna-4,16-diene-3,11,20-trione** (I) by the above procedure gave a 31% yield of 21-acetoxy-9α-fluoro-3,16-bisisopropylthiopregna-3,5-diene-11,20-dione; m.p. 105–109°; $[\alpha]^{25}D$ −38.6°; λ_{max} 5.70 (s), 5.89 (s), 6.06 (w), 6.25 (w), 8.21 μ (s); 235 (ε 11,700), 272 mμ (ε 19,200). The hydrolysis of this product to the 16α-isopropylthio-Δ[4]-3-ketone VII is described below.

Anal. Calcd. for $C_{29}H_{41}FO_4S_2$: C, 64.89; H, 7.70; F, 3.55; S, 11.95. Found: C, 65.22; H, 7.83; F, 3.54; S, 11.81.

With **16-dehydrodeoxycorticosterone acetate** (6.0 g.) and **methyl mercaptan,**[24] recrystallization from ether gave the desired **16α-methylthiodeoxycorticosterone acetate** (**XXVII**) (2.3 g., 34%) for which, however, fully satisfactory analytical data could not be obtained, probably because of contamination with XXX. Evaporation of the ether mother liquors from the above-mentioned recrystallization gave an amorphous product (3.1 g.) which could not be crystallized and which by analysis and spectroscopy is considered to be **21-acetoxy-16α-20,20-trismethylthiopregn-4-en-3-one** (**XXX**), $[\alpha]^{25}D$ +11; λ_{max} 5.77, 5.96, 6.18, 6.98, 8.06 μ; 241 mμ (ε 19,600).

Anal. Calcd. for $C_{26}H_{40}O_3S_3$: C, 62.89; H, 8.12; S, 19.37. Found: C, 63.24: H, 8.27; S, 19.20.

B. Oxidation of Methylthio Derivatives to Sulfoxides and Sulfones.—The stepwise oxidation of the 16α-methylthiosteroids to the corresponding sulfoxide and sulfone derivatives was carried out with monoperphthalic acid according to

(24) Experiment carried out by E. Ruckel and J. Nocera.

the procedures described previously by Schaub and Weiss.[15] Table I gives the physical characteristics of these compounds.

21-Acetoxy-9α-fluoro-16α-isopropylthiopregn-4-ene-3,11,20-trione (VII).—A suspension of 21-acetoxy-9α-fluoro-3,16-bisisopropylthiopregna-3,5-diene-11,20-dione (250 mg., 0.47 mmole) in ethanol (30 ml.) containing water (2 ml.) and 6 *N* hydrochloric acid solution (0.2 ml.) was refluxed for 2 hr. The reaction mixture was poured into ice water and extracted with ether. The combined ether extracts were washed with sodium bicarbonate solution, water and saline solution, dried, decolorized and evaporated. The residue was crystallized from ether to yield 100 mg. (45%) of VII with m.p. 165–180°. The analytical sample was prepared by 3 recrystallizations from ether, m.p. 180–184°. Physical properties and analytical data are given in Table I.

Hydrolysis of 21-Acetoxy-16α-acetylthio-9α-fluoropregn-4-ene-3,11,20-trione (V). A. With 72% Perchloric Acid.[25] Formation of 16α-Acetylthio-9α-fluoro-21-hydroxypregn-3-ene-3,11,20-trione (VI).—A solution of 21-acetoxy-16α-acetylthio-9α-fluoropregn-4-ene-3,11,20-trione (V), (410 mg., 0.86 mmole) in methanol (49 ml.) containing perchloric acid (72%, 1.25 ml.) was stirred at room temperature for 4 hr. The mixture was neutralized with 10% potassium acetate solution, and the resulting precipitate was removed by filtration and washed with methanol. The filtrate and methanol washings were combined, partially concentrated, and the residue was partitioned between methylene chloride and water. The organic phase was washed with water, 10% potassium acetate solution, water and saline solution, dried, decolorized and evaporated to leave 379 mg. of a white glass.

This glass (326 mg.) was dissolved in 5 ml. of the lower phase of the solvent system heptane–ethyl acetate–methanol–water (80:20:12:8) and 10 g. of Celite[26] diatomaceous earth was added to the solution. The mixture was placed on top of a column prepared from 100 g. of Celite diatomaceous earth which had been mixed thoroughly with 50 ml. of the lower phase of the above-described solvent system. The column [hold-back volume (h.b.v.)[27] = 128 ml.] was eluted with the upper phase of the above-described solvent system, and the effluent was allowed to pass through a recording spectrophotometer (set at 240 mμ). There were two major peaks of material with absorption at 240 mμ. The first peak occurred at the second and third h.b.v. and the crystalline material (60 mg.) isolated from this fraction gave the same infrared absorption spectrum as that of the starting material V. The second peak occurred at the seventh through eleventh h.b.v. and contained 192 mg. (51%) of crystalline 16α-acetylthio-9α-fluoro-21-hydroxypregn-4-ene-3,11,20-trione (VI), m.p. 136–143°. The analytical sample was obtained after 3 recrystallizations from acetone–hexane, m.p. 148–154°; $[\alpha]_D$ +123°; λ_{max} 2.90 (m), 5.75 (s), 5.90 (s, broad peak), 6.13 (m) (shoulder), 8.82 μ (s, broad); 235 mμ (ε 23,200).

Anal. Calcd. for $C_{23}H_{29}FO_5S$: C, 63.27; H, 6.69; F, 4.35; S, 7.34. Found: C, 63.65; H, 6.48; F, 4.28; S, 7.37.

B. With Sodium Methoxide. Formation of 9α-Fluoro-21-hydroxy-16α-mercaptopregn-4-ene-3,11,20-trione (X) and 16,16′-Thiobis(9α-fluoro-21-hy-

(25) J. Fried and E. F. Sabo, *J. Am. Chem. Soc.*, **79**, 1130 (1957).

(26) Celite® is the trademark of the Johns-Manville Corp. for diatomaceous earth products. The material used in this procedure was Celite 545, which had been washed with 6*N* hydrochloric acid solution, followed by distilled water to neutrality, and finally with methanol. The material was dried at 50°.

(27) Hold-back volume is the volume of solvent necessary to fill the packed column.

droxypregn-4-ene-3,11,20-trione) (**XVII**).—A solution of 21-acetoxy-16α-acetylthio-9α-fluoropregn-4-ene-3,11,20-trione (V) (239 mg., 0.5 mmole) in 0.09 *N* methanolic sodium methoxide solution (11 ml.) was magnetically stirred at room temperature for 20 min. The mixture was neutralized with acetic acid, evaporated, and the residue was partitioned between methylene chloride and water. The methylene chloride solution was washed with water and saline solution, dried, decolorized and evaporated to leave 180 mg. of colorless gum which on crystallization from methylene chloride–ether gave 23 mg. (6%) with m.p. 250–252°. (In a larger scale preparation 100 mg. (13%) of crystals with m.p. 234–242° and the same infrared spectrum were obtained.) The solid was recrystallized from methylene chloride-ether to give material with m.p. 264–274° (dec.); $[\alpha]_D$ +112°; λ_{max} 2.89 (m), 5.78 (s), 5.95 (s), 6.11 μ (m); 235 mμ (ε 37,750). This product appears to be **16,16′-thiobis(9α-fluoro-21-hydroxypregn-4-ene-3,11,20-trione**) (**XVII**).

Anal. Calcd. for $C_{42}H_{52}F_2O_8S$: C, 66.81; C, 6.94; F, 5.03; S, 4.25; mol. wt., 754.9. Found: C, 66.97; H, 7.65; F, 4.83; S, 4.39; mol. wt. (thermometric method, methanol), 730.

From the combined methylene chloride–ether mother liquors (above) there was obtained on evaporation 75 mg. (38% yield) of a second crystalline product (m.p. 178–193°). This substance was recrystallized from methylene chloride-ether to give **9α-fluoro-21-hydroxy-16α-mercaptopregn-4-ene-3,11,20-trione** (**X**), m.p. 183–188°. The physical constants and analytical data are given in Table I.

General Procedure for the Preparation of 16α-Chloro-Derivatives.[17]—A chilled dioxane solution (25 ml.) containing 1 mmole of the appropriate 16-dehydro-20-ketone [21-acetoxy-9α-fluoropregna-4,16-diene-3,11,20-trione (I), 16-dehydroprogesterone or 16-dehydrodeoxycorticosterone acetate] was saturated with hydrogen chloride. The reaction vessel was stoppered and stored at 5–7° for 4 days. The acid solution was neutralized by cautiously pouring it into a chilled mixture of aqueous saturated sodium bicarbonate solution and methylene chloride. The aqueous layer was extracted with methylene chloride, and the combined extracts were washed with water and saline solution, dried, decolorized and evaporated. The residues were crystallized from ether to give the various 16α-chloro derivatives described in Table I.

General Procedure for the Preparation of 16α-Amino Derivatives.[13c]—A 4- to 10-fold excess of the requisite amine was added to a tetrahydrofuran solution (5 ml.) containing 1 mmole of the appropriate 16-dehydro-20-ketone [9α-fluoro-21-hydroxypregna-4,16-diene-3,11,20-trione (II) or 16-dehydrodeoxycorticosterone]. (With *methylamine*, the steroid solution was saturated with the gaseous amine. With *aniline to give XV*, dioxane was used as solvent.) To the solution of steroid and amine was added Amberlite IRA-400[28] resin (OH) (400 mg.), and the suspension was stirred at room temperature under nitrogen (no nitrogen was used with methylamine) 1 to 4 days. (With *16-dehydrodeoxycorticosterone* and piperidine a 24 hr. reaction period gave results superior to those obtained with more prolonged reaction periods. Therefore, all amine additions to 16-dehydrodeoxycorticosterone were carried out for 24-hr. periods. With *21-acetoxy-9α-fluoropregna-4,16-diene-3,11,20-trione*, preliminary studies indicated the desirability of longer reaction periods, and condensations with this substrate were carried out for periods ranging from 2 to 4 days.) The mixture was then filtered, and the resin washed with tetrahydrofuran. The combined filtrate and washings

(28) Amberlite IRA-400® Rohm and Haas Company (synthetic anion exchange resin).

were evaporated (at below 30°), and the residue partitioned between methylene chloride and water. The organic phase was washed with water and saline solution, dried, decolorized and evaporated. The residual material was crystallized from ether or methylene chloride-ether to yield the 16α-amino derivatives described in Table I.

Methanolic Methoxide Treatment of 21-Acetoxy-9α-fluoropregna-4,16-diene-3,11,20-trione (I). Formation of 9α-Fluoro-21-hydroxypregna-4,16-diene-3,11,20-trione (II) and 9α-Fluoro-21-hydroxy-16α-methoxypregn-4-ene-3,11,20-trione (XVI).[24]—21-Acetoxy-9α-fluoropregna-4,16-diene-3,11,20-trione (6.0 g.) (I) was dissolved in reagent methanol (500 ml.) and to this solution was added 0.1 *N* methanolic sodium methoxide solution (23.5 ml.). After 30 min. at room temperature in a nitrogen atmosphere, the solution was neutralized with acetic acid. Evaporation *in vacuo* of solvent afforded a semisolid which was partitioned between water and methylene chloride. The methylene chloride extract was washed with saturated sodium bicarbonate solution and then with water. After drying over sodium sulfate, the solvent was evaporated and the residual material was recrystallized from methylene chloride-ether to give 3.2 g. of **9α-fluoro-21-hydroxypregna-4,16-diene-3,11,20-trione (II)**, m.p. 175–199°; recrystallization from the same solvent pair gave 1.6 g. with m.p. 215–222°.

Concentration of the recrystallization mother liquor and addition of ether gave **9α-fluoro-21-hydroxy-16α-methoxypregn-4-ene-3,11,20-trione (XVI)** (0.55 g., m.p. 170–174°). Recrystallization of this material from methylene chloride–ether gave product (0.393 g.) with m.p. 171–174°; $[\alpha]^{25}D$ +120°; λ_{max} 5.78, 5.82, 5.95, 6.20 μ; 234 mμ (ε 16,800).

Anal. Calcd. for $C_{22}H_{29}FO_5$: C, 67.32; H, 7.45; F, 4.84; methoxyl CH_3, 3.83. Found: C, 66.96; H, 7.69; F, 4.54; methoxyl CH_3, 3.84.

21-Acetoxy-3-ethylenedioxy-9α-fluoropregna-5,16-diene-11,20-dione (XVIII).—A solution of 21-acetoxy-9α-fluoropregna-4,16-diene-3,11,20-trione (I) (640 mg., 1.59 mmoles) and *p*-toluenesulfonic acid (15 mg.) in 2-ethyl-2-methyl-1,3-dioxolane (14 ml.)[16] was refluxed with partial distillation through a helices-packed column for 5 hr. Some 6 ml. of distillate was collected during this period. The cooled solution was diluted with benzene (30 ml.) and was washed with sodium bicarbonate solution and with water. The organic phase was dried over sodium sulfate and was evaporated to afford a residue which was crystallized from ether, 530 mg.; m.p. 165–169°. Material recrystallized several times from ether–methylene chloride (431 mg., 60%) showed m.p. 195–196°; $[\alpha]D$ −14.6°; λ_{max} 5.70 (s), 5.80 (s), 5.92 (s), 6.29 (m), 8.08 (s), 8.24 (s), 9.02 μ (s); 235 mμ (ε 11,150).

Anal. Calcd. for $C_{25}H_{31}FO_6$: C, 67.27; H, 7.00; F, 4.26. Found: C, 67.35; H, 7.15; F, 4.32.

21-Acetoxy-3-ethylenedioxy-9α-fluoro-16α-nitromethylpregn-5-ene-11,20-dione.[13d] A solution of 21-acetoxy-3-ethylenedioxy-9α-fluoropregna-5,16-diene-11,20-dione (XVIII) (223 mg., 0.5 mmole) in nitromethane (2.26 ml.) containing 0.61 ml. of triethylamine was allowed to stand at room temperature for 4 days. Solvent was then evaporated (at below 30°) to leave a crystalline residue which was triturated with ether to yield 190 mg. (75%) of product with m.p. 188–198°. The analytical sample was prepared by 3 recrystallizations from methylene chloride-ether, m.p. 188–205°; $[\alpha]^{25}D$ +20°; λ_{max} 5.70 (s), 5.79 (s), 6.40 (s), 8.12 μ (s) (broad peak); no significant ultraviolet absorption.

Anal. Calcd. for $C_{26}H_{34}FNO_8$: C, 61.53; H, 6.75; F, 3.75; N, 2.76. Found: C, 61.73; H, 6.98; F, 3.71; N, 2.94.

21-Acetoxy-9α-fluoro-16α-nitromethylpregn-4-ene-3,11,20-trione (XIX).—A suspension of 21-acetoxy-3-ethylenedioxy-9α-fluoro-16α-nitromethylpregn-5-ene-11,20-dione (250 mg., 0.5 mmole) in methanol (6 ml.) containing 8% (v./v.) aqueous sulfuric acid solution (0.6 ml.) was stirred at reflux for 2 hr. The white solid was collected by filtration, washed with methanol and dried *in vacuo* to yield 1.54 mg. (67%) of XIX, m.p. 234–237°. For analysis this material was recrystallized 3 times from methylene chloride-ether to give product with m.p. 239–242°. Physical properties and analytical data are given in Table I.

16-Alkylated Progesterones

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Comparison is made of the progestational activity of some 16-alkylated progesterones. The synthesis of these compounds is described.

Enhancement or transformation of activity by the modification of steroid structures represents a continuing effort on the part of the steroid chemist. Variations in activities, although often predictable, are, however, frequently surprising. In this connection, it seemed of particular interest to examine progesterone derivatives alkylated at carbon 16.

Accordingly, we have prepared: 16-methylene-17α-acetoxyprogesterone (IVa), 16,16-dimethylprogesterone (X), 16β-methyl-17α-acetoxyprogesterone (XVIIa), and 16α-methyl-17α-acetoxyprogesterone (XVIIb). The biological activities of some of them, as well as methods of synthesis, already have been reported. It seemed desirable, however, that the testing and comparison of the progestational activities be carried out by the same laboratory, and our results are presented below. We have included the older compounds, 16α- and 16β-methylprogesterone. In addition, because of certain differences both in procedures employed and in physical constants reported (both for end-products and intermediates[1]) we are presenting our methods of preparation.

(1) See experimental section, compounds XIVa, XVIb, XVIIa, XVIIb.