TABLE III
ANTITUMOR ACTIVITY OF URETHAN-TYPE NITROGEN MUSTARD DERIVATIVES FROM ANDROGENIC HORMONES (I AND II)

No.	Tumor	Dose, mg/day I	Dose, mg/day II	Period of treatment, days I	Period of treatment, days II	Inhibition I T/C	Inhibition I %	Inhibition II T/C	Inhibition II %
1	Walker 256 carcinosarcoma	8[a]	10[a]	17[b]	17[b]	19.9 ± 2.63/39.2 ± 2.29	49	15.8 ± 3.67/25.3 ± 2.05	35
2	Ehrlich carcinoma	0.2	0.5	16[c]	16[c]	3.1 ± 0.48/5.5 ± 1.34	43	3.0 ± 0.40/5.5 ± 1.34	45
3	Mammary adenocarcinoma	0.2[a]	0.5[a]	17[d]	17[d]	2.0 ± 0.77/3.7 ± 0.52	45	2.2 ± 0.44/3.7 ± 0.52	40
4	Sarcoma 180	0.2	0.5	8[e]	8[e]	5.3 ± 0.74/11.3 ± 1.21	55	3.4 ± 0.82/11.3 ± 1.21	69
5	Carcinoma O-Ya	...	5	...	8[f]	...	...	16.4 ± 2.08/46.6 ± 3.20	64

[a] Given every 2 days. [b] Treatment begins 7 days after tumor transplantation in rats. [c] Treatment begins 8 days after tumor transplantation in mice. [d] Treatment begins 3 days after tumor transplantation in mice. [e] Treatment begins 24 hr after tumor transplantation in mice. [f] Treatment begins 24 hr after tumor transplantation in rats.

as a gum, purified by chromatography on alumina column. After placing the material on a column in benzene solution, the column was eluted with benzene, benzene–absolute ethanol mixtures (99:1, 98:2, 95:5, 50:50), and absolute ethanol. Compound III was isolated from the 99:1 benzene–absolute ethanol fraction and recrystallized from toluene.

17α-Ethynylestradiol 3-Chloroformate.—To 2 g (0.007 mole) of 17α-ethynylestradiol and 3 ml of triethylamine in 20 ml of dioxane, 4 g (0.016 mole) of phosgene in 25 ml of anhydrous benzene at 0° was added with stirring. After standing overnight at room temperature, the triethylamine hydrochloride was removed by filtration and the solution was concentrated under vacuum. The resulting oil, triturated with petroleum ether, gave 2.2 g (85.3%) of the chloroformate, mp 138–139°.

3-Benzylestradiol 17β-chloroformate (IV), mp 104–106° (89.4%), was prepared similarly but without an HCl acceptor (Et_3N).

17α-Ethynylestradiol 3-[N,N-Bis(2-chloroethyl)]carbamate (VI).—N,N-Bis(2-chloroethyl)carbamoyl chloride (2.4 g, 0.012 mole) was added to a solution of 3 g (0.01 mole) of 17α-ethynylestradiol in 30 ml of pyridine. After 3 days of standing at room temperature, the reaction mixture was poured onto ice–water containing a small quantity of methanol. VI (73.4%, 3.45 g) crystallized on standing. A recrystallization from methanol gave an analytical sample, mp 141–142°.

17β-Estradiol 3-[N,N-Bis(2-chloroethyl)]carbamate (V).—Estradiol (1.5 g, 0.0055 mole) was added to 10 ml of anhydrous ethanol in which 0.13 g (0.0056 g-atom) of Na had been dissolved. After removal of the solvent under vacuum the solid was suspended in 20 ml of toluene and 1.2 g (0.006 mole) of nitrogen mustard chloroformate was added with stirring. The mixture was refluxed for 4 hr. After filtration from NaCl and removal of the solvent under reduced pressure, the residue was recrystallized from benzene–petroleum ether, giving 1.6 g (72.7%) of V, mp 104–105°.

17β-Estradiol 17β-[N,N-Bis(2-chloroethyl)]carbamate (III).—A solution of 3 g (0.0056 mole) of IV in 20 ml of absolute ethanol was hydrogenated in the presence of 2 g of 5% Pt–C. The theoretical amount of H_2 was absorbed after 2 hr. After filtration from the catalyst and removal of the solvent, 2.2 g (88.3%) of crystalline III was obtained; mp 128–130°, after recrystallization from toluene, mp 130–132°.

17α-Ethylestradiol 3-[N,N-Bis(2-chloroethyl)]carbamate (VII).—Compound VI (2 g, 0.0044 mole) and 1 g of 5% Pt–C in 20 ml of EtOH were shaken under hydrogen at atmospheric pressure. The theoretical amount of H_2 was absorbed after 1.5 hr. The catalyst was removed by filtration, and the solvent was distilled under vacuum giving 1.6 g (79.6%) of VII, mp 115–117° after recrystallization from petroleum ether.

Acknowledgment.—We thank Mrs. G. Botez and Mr. F. Chiraleu for carrying out the spectrophotometric measurements, Mrs. E. Creangă for optical rotatory determinations, and Mrs. M. Ionescu for the purification of certain compounds. The authors are also indebted to Mr. V. Feyns for his help in the purification of compound IV and his review of the manuscript, as well as to Mr. V. Dobre for making the screening data available to us.

Histamine Releasers. III. Dibasic Acid Amides of 4-Phenyl-4-aminomethylpiperidines

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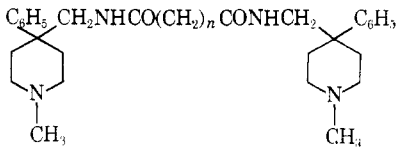
Received May 12, 1966

A series of 1-alkyl-4-phenyl-4-aminomethylpiperidine amides of various dibasic acids were found to have histamine-releasing activity in dogs. The most potent compound was 4,4′-dimethyl-N,N′-4-phenyl-4-piperidylmethylterephthalamide (XIII). An exploration of the structure–activity relationship in this area is described.

In 1955, Chiavarelli, *et al.*,[1] in quest of compounds with curare-like activity, discovered a new series of hypotensive agents (later known to be histamine releasers), which were amides derived from 1-methyl-4-phenyl-4-aminomethylpiperidine and aliphatic dibasic acids of formula I. They found that a peak in activity occurred when n equaled 8. It was also reported that



I

quaternization of the piperidine nitrogen or reduction of the carbonyl groups caused a loss of activity. With these data we began an exploration of the structure–

(1) S. Chiavarelli, O. Corvillon, and G. B. Marini-Bettolo, *Rend. Ist. Super. Sanità*, **18**, 1023 (1955).

activity requirements in order to find a still more potent histamine releaser.

Our first thought was to replace the 4-phenyl group with hydrogen as in II (Table III). This structural change caused a complete loss of activity, so the phenyl group was henceforth left undisturbed. The biological activity of this and subsequent compounds is summarized in Table I. The next grouping to be varied

TABLE I

BLOOD PRESSURE AND PLASMA HISTAMINE LEVELS

Compd[a]	Time after admin, min	Dog 1 Blood pressure	Dog 1 Plasma histamine, μg %	Dog 2 Blood pressure	Dog 2 Plasma histamine, μg %
b	0	140	1.0	115	3.0
	2	40	36.9	80	46.0
	30	120	4.1	60	2.9
III	0	130	1.6	175	1.4
	2	70	8.8	160	2.1
	30	130	1.5	170	1.4
IV	0	150	1.0	175	1.5
	2	75	6.7	160	5.1
	30	150	1.5	170	1.5
VIII	0	200	0.6	175	1.7
	2	125	3.5	175	2.4
	30	190	0.7	185	1.6
IX	0	140	2.0	160	1.1
	2	125	1.2	60	6.0
	30	125	1.0	155	0.8
XII	0	185	2.4	215	0.9
	2	150	8.1	200	1.6
	30	165	1.2	200	1.2
XIII	0	160	2.7	175	0.6
	2	60	182.0	135	53.7
	30	95	31.2	125	5.5
XVIII	0	165	0.8	200	1.3
	2	35	36.0	125	20.4
	30	120	2.1	175	2.1

[a] The dosages were 100 μg/kg iv. Compounds II, V–VII, X, XI, XIV, and XXIX were found to be completely inactive in tests involving one animal per compound. [b] *p*-Methoxyphenethylamine–formaldehyde condensation product.

was the substituent on the piperidine nitrogen. A comparison of the unsubstituted, methyl, ethyl, benzyl, and *p*-methoxyphenethyl compounds (III–VII) indicated that alkylation of the piperidine nitrogen with a group larger than methyl was detrimental to activity. The unsubstituted compound (III) and the N-methyl derivative (IV) were of a comparable order of activity. Compound VII was prepared since other *p*-methoxyphenethylamine derivatives possess histamine-releasing ability.[2] However, it proved to be inactive as a histamine releaser.

We then turned our attention to another variable, the nature of the dibasic acid moiety (R_4) which presumably serves as a "spacer" between the piperidine rings. Since the variation in alkyl chain length had already been investigated,[1] we decided to insert aromatic residues between the amide groups. The terephthalic acid amides (VIII and IX) were about as active as their corresponding analogs (III and IV) in the sebacamide series. While keeping the piperidine portion constant, the isophthalic acid (X) and 4,4′-diphenylcarboxylic acid (XI) amides were prepared and both were found to be inferior to the terephthalic amide (VIII). The 1,4-phenylenediacetic acid amide (XII) had approximately the same potency as VIII.

Before proceeding to a study of our last variable, substitution on the amide nitrogen, we wished to know whether an amide function was truly essential. Though it was shown that reduction of the carbonyl destroyed activity, this did not prove that insertion of a different type of carbonyl function would be inappropriate. To demonstrate this point we prepared the terephthalic acid ester (XXIX) of 1-methyl-4-hydroxymethyl-4-phenylpiperidine[3] and found it to be inactive. Although it is possible that some esterase in the blood stream could have rapidly hydrolyzed the ester, this experiment discouraged us from substituting other carbonyls for the amide group.

Replacement of the hydrogen on the amide nitrogen by a methyl group (XIII and XVIII) gave surprising results. These compounds were far more active and exhibited longer duration than the other members of the series. The activities of XIII and XVIII as measured by intravenous injection in dogs were compared with that of the potent *p*-methoxyphenethylamine–formaldehyde condensation product.[2] In two runs compound XIII showed about 1.3 and 4.5 times the activity of the standard, while XVIII had about 0.5 and 0.9 times the potency of the standard.

The chemical synthesis of the compounds was very straightforward and involved the treatment of 2 equiv of an appropriate piperidine compound with 1 equiv of a bis(acid chloride) in an inert solvent. For preparation of the 1-unsubstituted piperidines, 1-benzyl-4-aminomethyl-4-phenylpiperidine was acylated and the benzyl group was removed by hydrogenolysis. 1-Methyl-4-aminomethyl-4-phenylpiperidine (XXII) used for the preparation of II, IV, IX, and XI was obtained by the $LiAlH_4$ reduction of 1-carbomethoxy-4-cyano-4-phenylpiperidine (XIX). 1-Ethyl- (XXIII) and 1-(*p*-methoxyphenethyl)-4-aminomethyl-4-phenylpiperidine (XXIV) were prepared by hydride reduction of the appropriate amides (XX and XXI, respectively). 1-Benzyl-4-methylaminomethyl-4-phenylpiperidine (XXVI) used for preparation of XIII was obtained by hydride reduction of the methyl urethan of 1-benzyl-4-aminomethyl-4-phenylpiperidine (XXV). 1-Methyl-4-methylaminomethyl-4-phenylpiperidine (XXVIII) used for the preparation of XVIII was similarly obtained from the urethan (XXVII).

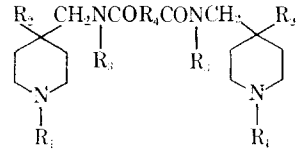
Biological Evaluation.—For tests of histamine release, healthy mongrel dogs were anesthetized,[4] the left femoral arteries were surgically cannulated, and arterial blood pressures were measured directly by means of a Statham P-23 pressure transducer connected to a recording polygraph (Gilson Medical Electronics). The compounds were administered intravenously (100 μg/kg) as aqueous solutions of their hydrochloride salts. Blood (5 ml) was withdrawn immediately before and at 2- and 30-min intervals after administration, and heparinized plasma was obtained in the usual manner. Histamine content was measured by the

(2) R. Baltzly, J. S. Buck, E. J. DeBeer, and F. J. Webb, *J. Am. Chem. Soc.*, **71**, 1301 (1949).

(3) B. Elpern, *ibid.*, **76**, 281 (1954).

(4) In conducting the research reported herein, the investigators adhered to the "Principles of Laboratory Animal Care" as established by the National Society for Medical Research.

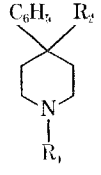
Table II: Dibasic Acid Amides



Compd	R_1	R_2	R_3	R_4	Mp, °C	Recryst solvent	Method	Formula	Calcd, % C	Calcd, % H	Calcd, % N	Found, % C	Found, % H	Found, % N
II	CH_3	H	H	1,4-C_6H_4	243–245	*i*-PrOH	A	$C_{22}H_{34}N_4O_2 \cdot 0.5H_2O$	66.8	8.68	14.2	66.8	8.78	14.4
III	H	C_6H_5	H	$(CH_2)_8$	187–189	*i*-PrOH–EtOAc	B	$C_{34}H_{52}N_4O_2 \cdot H_2O$	72.3	9.22	9.93	72.3	9.28	9.92
IV	CH_3	C_6H_5	H	$(CH_2)_8$	143–144.5[a]	EtOAc	A							
V	C_2H_5	C_6H_5	H	$(CH_2)_8$	129–131	EtOAc	A	$C_{38}H_{58}N_4O_2$	75.7	9.70	9.29	75.6	9.65	9.15
VI	$C_6H_5CH_2$	C_6H_5	H	$(CH_2)_8$	92–95	C_6H_6–C_6H_{12}	A	$C_{48}H_{62}N_4O_2$	79.3	8.60	7.71	79.6	8.99	7.30
VII	4-$CH_3OC_6H_4(CH_2)_2$	C_6H_5	H	$(CH_2)_8$	118–121	C_6H_6–C_6H_{12}	A	$C_{52}H_{70}N_4O_4$	76.7	8.60	6.88	76.9	8.27	7.10
VIII	H	C_6H_5	H	1,4-C_6H_4	258–259.5	$CH_3OCH_2CH_2OH$	B	$C_{32}H_{38}N_4O_2 \cdot H_2O$	72.7	7.57	10.6	72.9	7.57	10.7
IX	CH_3	C_6H_5	H	1,4-C_6H_4	>300	*b*	A	$C_{34}H_{42}N_4O_2 \cdot 2HCl \cdot 0.5H_2O$	65.8	7.26	9.03	66.0	7.08	9.00
X	H	C_6H_5	H	1,3-C_6H_4	200–205	MeOH–EtOH	B	$C_{32}H_{38}N_4O_2 \cdot H_2O \cdot CO_2$	69.2	6.99	9.79	69.2	7.04	9.78
XI	CH_3	C_6H_5	H		214–217	*i*-PrOH–MeOH	A	$C_{36}H_{36}N_4O_2$	78.1	7.54	9.11	78.0	7.38	9.01
XII	H	C_6H_5	H	1,4-$CH_2C_6H_4CH_2$	202–204	*i*-PrOH–EtOAc	B	$C_{34}H_{42}N_4O_2$	75.8	7.86	10.4	75.7	7.91	10.2
XIII	H	C_6H_5	CH_3	1,4-C_6H_4	190–194	*i*-PrOH–EtOAc	B	$C_{34}H_{42}N_4O_2 \cdot 0.5H_2O$	74.6	7.86	10.2	74.8	7.75	9.99
XIV	$C_6H_5CH_2$	C_6H_5	H	1,4-C_6H_4	236–238	$CH_3OCH_2CH_2OH$	A	$C_{46}H_{50}N_4O_2 \cdot 0.5H_2O$	79.0	7.30	8.01	79.0	7.46	7.93
XV	$C_6H_5CH_2$	C_6H_5	CH_3	1,4-C_6H_4	207–211	C_6H_5	A	$C_{48}H_{52}N_4O_2$	80.2	7.57	7.79	79.9	7.57	7.75
XVI	$C_6H_5CH_2$	C_6H_5	H	1,3-C_6H_4	213–218	MeOH	A	$C_{46}H_{50}N_4O_2 \cdot 2HCl \cdot 0.25H_2O$	71.5	6.86	7.25	71.4	6.80	7.22
XVII	$C_6H_5CH_2$	C_6H_5	H	1,4-$CH_2C_6H_4CH_2$	266–275	MeOH–EtOH	A	$C_{48}H_{54}N_4O_2 \cdot 2HCl \cdot 0.25H_2O$	72.4	7.15	7.03	72.3	7.20	7.06
XVIII	CH_3	C_6H_5	CH_3	1,4-C_6H_4	234–237	C_6H_6–MeOH	A	$C_{28}H_{36}N_4O_2$	76.3	8.18	9.89	76.3	8.22	9.63

[a] Lit.[3] mp 146°. [b] Triturated with ethanol.

Table III: Substituted 4-Phenylpiperidine Intermediates



No.	R_1	R_2	Method	Yield, %	Mp, °C	Recryst solvent	Formula	Calcd, % C	Calcd, % H	Calcd, % N	Found, % C	Found, % H	Found, % N
XIX	$COOCH_3$	CN	C	91	103–105	Cyclohexane	$C_{13}H_{16}N_2O_2$	68.8	6.60	11.5	68.6	6.55	11.7
XX	CH_3CO	CN	*a*	78	96–98	Cyclohexane	$C_{13}H_{16}N_2O$	73.6	7.06	12.3	73.7	6.83	12.4
XXI	*p*-$CH_3OC_6H_4CO$	CN	*b*	29	135.5–137	MeOH	$C_{20}H_{22}N_2O_2$	75.4	6.63	8.38	75.2	6.47	8.58
XXII	CH_3	CH_2NH_2	D	80	245–248	2-Methoxyethanol	$C_{25}H_{33}N_5O_{16}$[c]	45.5	4.64	15.2	45.6	4.94	15.3
XXIII	C_2H_5	CH_2NH_2	D	98	226–228	2-Methoxyethanol–H_2O	$C_{26}H_{36}N_8O_{16}$[c]	46.3	4.82	14.9	46.3	4.99	15.1
XXIV	*p*-$CH_3OC_6H_4CH_2$	CH_2NH_2	D	56	263–266	2-Propanol–ethanol	$C_{21}H_{28}N_2O \cdot 2HCl$	63.5	7.69	7.05	63.2	7.68	6.97
XXV	$C_6H_5CH_2$	$CH_2NHCOOCH_3$	C	93	204–207	Et_2O–EtOAc	$C_{19}H_{26}N_2O_2 \cdot HCl \cdot 0.25H_2O$	66.5	7.25	7.38	66.7	7.27	7.47
XXVI	$C_6H_5CH_2$	CH_2NHCH_3	D	83	71–73	Petr ether (bp 30–60°)	$C_{20}H_{26}N_2$	81.6	8.90	9.52	81.9	8.97	9.23
XXVII	CH_3	$CH_2NHCOOCH_3$	C	92	89–91	Cyclohexane	$C_{15}H_{22}N_2O_2$	68.7	8.45	10.7	68.9	8.29	10.6
XXVIII	CH_3	CH_2NHCH_3	D	70	246–248	5% Acetone	$C_{28}H_{38}N_8O_{14}$[d]	46.2	4.17	16.6	45.7	4.29	16.6

[a] Prepared by treatment of 4-cyanophenylpiperidine hydrochloride with Ac_2O–pyridine at room temperature. [b] Prepared by treatment of 4-cyano-4-phenylpiperidine (free base) with *p*-methoxyphenylacetyl chloride in $CHCl_3$ at room temperature. [c] Obtained as dipicrate solvated by 1 mole of 2-methoxyethanol. Attempts at crystallization from another solvent or to free the solvate were unsuccessful. [d] Dipicrate.

method of Shore, *et al.*,[5] employing a later modification[6] where one-tenth the usual amount of *o*-phthalaldehyde was used. Duplicate 1-ml aliquots of plasma were analyzed; fluorescence of the final solutions was measured on a Turner fluorometer[7] equipped with 7-60 primary and 2-A secondary filters. The lower limit of sensitivity was 0.005 μg of histamine/ml of initial sample, and the precision of reading samples was ±0.0005 μg. Recoveries of added histamine from plasma averaged 92% (range 90–96%). Average deviations of single determinations from their means did not exceed 4%. Serotonin did not contribute in this assay.

Experimental Section

Physical and analytical data are listed in Tables II and III.

A. N,N′-(1-Benzyl-4-phenyl-4-piperidylmethyl)terephthalamide Dihydrochloride (XIV).—To a chilled mixture of 2.0 g (7.1 mmoles) of 4-phenyl-4-aminomethyl-1-benzylpiperidine (Aldrich Chemical Co.) in 15 ml of anhydrous methylene chloride was added (dropwise) a mixture of 0.73 g (3.6 mmoles) of terephthaloyl dichloride in 10 ml of anhydrous CH_2Cl_2. The mixture was stirred at room temperature 1 hr, then allowed to stand at room temperature overnight. The white solid was collected by filtration and recrystallized from absolute methanol, giving 1.83 g (67%) of white crystals, mp 283–290°. A sample was converted to the free amine (partitioned between $CHCl_3$ and 10% NaOH) and recrystallized from 2-methoxyethanol.

B. N,N′-(4-Phenyl-4-piperidylmethyl)terephthalamide (VIII).—To 500 mg of 100% Pd catalyst were added 20 ml of glacial acetic acid and 1.3 g (1.7 mmoles) of XIV. The mixture was hydrogenated at 70° and atmospheric pressure, taking up the theoretical amount of hydrogen in 1 hr. The mixture was diluted with 25 ml of water, the catalyst was removed by filtration, and the filtrate was evaporated *in vacuo* to dryness. The gummy material was taken up in water and alkalized (pH 10) with 10% NaOH to precipitate a white solid. The solid was collected by filtration and washed thoroughly with water to leave 0.74 g. Recrystallization from 2-methoxyethanol afforded 0.56 g (65%) of white crystals.

C. 1-Carbomethoxy-4-cyano-4-phenylpiperidine (XIX).—To a mixture of 5.0 g (22.5 mmoles) of 4-phenyl-4-cyanopiperidine hydrochloride, 100 ml of water, 2.2 g (54 mmoles) of NaOH, and 50 ml of $CHCl_3$ was slowly added 2.0 ml (27 mmoles) of methyl chloroformate. The mixture was vigorously stirred at room temperature for 2 hr, chilled, and acidified to pH 2 with 6 *N* HCl. The mixture was stirred (chilled) for 1 hr, and the chloroform layer was removed and washed with water. The $CHCl_3$ extract was dried ($MgSO_4$) and evaporated *in vacuo* to dryness to give an orange syrup which crystallized upon standing to yield 5.0 g (91%) of white crystals. An analytical sample was obtained from cyclohexane.

D. 1-Methyl-4-aminomethyl-4-phenylpiperidine Dipicrate (XXII).—To a cold suspension of 7.7 g (0.2 mole) of $LiAlH_4$ in 175 ml of anhydrous tetrahydrofuran was slowly added 5.0 g (22.3 mmoles) of XIX. The mixture was refluxed 15 hr. Excess $LiAlH_4$ was decomposed by the careful addition of absolute ethanol. The reaction mixture was then treated with water, stirred briefly, and evaporated *in vacuo* to near dryness. The pasty material was extracted with 1-butanol, which was dried ($MgSO_4$) and evaporated *in vacuo* to yield 3.64 g (80%) of orange syrup. A sample was converted to the picrate and recrystallized from 2-methoxyethanol.

1-Methyl-4-hydroxymethyl-4-phenylpiperidine Terephthalate (XXIX).—To a chilled mixture of 1.02 g (5 mmoles) of 1-methyl-4-phenyl-4-hydroxymethylpiperidine[3] in 10 ml of anhydrous CH_2Cl_2 was added a suspension of 0.505 g (2.5 mmoles) of terephthaloyl chloride in 10 ml of of anhydrous CH_2Cl_2. The mixture was stirred at room temperature (25°) for 18 hr, and the white crystals were collected by filtration. Trituration with acetone gave 1.25 g of crude material. Recrystallization from 2-propanol–absolute methanol yielded 0.292 g (11%), mp 276–279°.

Anal. Calcd for $C_{34}H_{40}N_2O_4 \cdot 2HCl \cdot H_2O$: C, 64.6; H, 7.03; N, 4.43. Found: C, 64.6; H, 7.09; N, 4.50.

(5) P. A. Shore, A. Burkhalter, and V. H. Cohn, Jr., *J. Pharmacol. Exptl. Therap.*, **127**, 182 (1959).

(6) D. von Redlich and D. Glick, *Anal. Biochem.*, **10**, 459 (1965).

(7) Model 110 was equipped with a high-sensitivity conversion kit, No. 110-865, and a microcuvette adaptor, No. 110-66.

16-Aza Steroids

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Received October 3, 1966

The preparation of several 16-azaandrostenes is described, utilizing 3β-hydroxy-16,17-secoandrost-5-ene-16,17-dioic acid (I) as starting material. The latter compound was transformed, *via* its diester IIa, half-ester IIb, acid chloride IIc, and isocyanate III, into 3β-hydroxy-16-azaandrost-5-en-17-one (V). In a similar manner 16-azaestrone was prepared from the methyl ether of marrianolic acid. The results of some preliminary biological tests are reported.

The introduction of a nitrogen atom into the steroid nucleus has led to compounds exhibiting diverse biological activity.[1] Although most of the ring D aza steroids previously described[2] are analogs of D-homosteroids, the preparation of various 17-aza steroids has been reported[3] in which the nitrogen is incorporated in a five-membered D ring. Studies on the corresponding 16-aza series were initiated by Bachmann and Ramirez, who described[4] the synthesis of *dl*-16-azadeoxyisoequilenin and *dl*-16-azadeoxyequilenin. A recent report on the preparation of the 16-aza derivatives of estrone[5] prompts us to describe our related studies in this field.

In this connection, a diester of 3β-hydroxy-16,17-secoandrost-5-ene-16,17-dioic acid (I) appeared attractive as a starting material for the 16-azaandrostenes. Due to the steric hindrance of the tertiary carboxyl group, the diacid I has usually been converted to the dimethyl ester[6] with diazomethane. We have found that I was more conveniently esterified with the diethyl

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