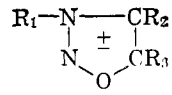
TABLE II

3-SUBSTITUTED SYDNONES AND SYDNONE IMINES



R_1	R_2	R_3	Recrystn. solvents	Yield, %	M.p., °C.	λ_{max}^{EtOH}, mμ	ϵ_{max}	Calcd., % C	Calcd., % H	Calcd., % N	Found, % C	Found, % H	Found, % N
3,4-$CH_2O_2C_6H_3$	H	O	*a*	41.5[b]	165–166	308	9,500	52.5	2.92	13.6	52.3	3.21	13.3
3,4-$CH_2O_2C_6H_3CH_2$	H	O	*c*	72.4	158–159	288	11,450	54.5	3.63	12.7	54.7	3.98	12.6
3,4-$CH_2O_2C_6H_3CH_2$	CH_3	O	*c*	58.8[b]	138–141	291	12,650	56.4	4.28	12.0	56.6	4.53	11.8
3-CH_3O-4-$C_2H_5OC_6H_3CH_2$	H	O	*d, e*	98.0	115–119	285	10,000	57.6	5.60	11.2	57.3	5.46	11.2
3,4-$CH_2O_2C_6H_3C_2H_4$	H	O	*d, f*	36.0[b]	122–125	287	12,200	56.4	4.28	12.0	56.1	4.20	11.7
3,4-$CH_2OC_6H_3CH_2$	Cl	O	*d*	13.2	110–111 dec.	292	10,700	47.2	2.75	11.0	47.2	3.00	11.0
3,4-$CH_2O_2C_6H_3CH_2$	H	NH·HCl	*f, g*	42.6	97 dec.	291	9,500	46.9	3.91	16.4	46.7	4.13	16.1
3,4-$CH_2O_2C_6H_3CH_2$	CH_3	NH·HCl	*f, g*	71.1	111 dec.	294	11,600	49.0	4.45	15.6	49.1	4.71	15.4
3,4-$CH_2O_2C_6H_3CH_2$	H	*p*-$(NSO_2C_6H_4)NHCOCH_3$	*f*	36.9	111 dec.	262 317	23,500 11,700	49.8[h,i]	4.15	12.9	49.6	4.20	12.9

[a] Butanol. [b] Yield calculated from the corresponding glycine. [c] Toluene. [d] Heptane. [e] Ethyl acetate. [f] Ethanol. [g] Petroleum ether (b.p. 35–60°). [h] Monohydrate. [i] Karl–Fischer determination of water: calcd., 4.15; found, 4.11.

and sydnone, and the latter could only be isolated after repeated recrystallizations.

Experimental[18]

N-Nitroso-N-piperonylglycine (III).—To a solution of 24.4 g. of N-piperonylglycine hydrochloride[9] in 250 ml. of water cooled to 0° was added dropwise, with stirring, during a period of 30 min., a concentrated aqueous solution of 7.5 g. of $NaNO_2$. After the addition was complete the reaction mixture was stirred at 0° for an additional 3 hr. and extracted with six 200-ml. portions of ether. The ether extract was treated with decolorizing charcoal, dried (Na_2SO_4), filtered, and evaporated to a sirup under reduced pressure. The sirup was triturated with a small amount of petroleum ether (b.p. 35–60°) and the resulting solid was recrystallized from a mixture of ethyl acetate and heptane to give 17 g. of analytically pure product, m.p. 114–116° (see Table I).

α-Methyl-N-nitroso-N-piperonylglycine, N-nitroso-N-piperonylglyconitrile, and other related compounds (see Table I) were prepared by essentially the same procedure.

3-Piperonylsydnone (I).—To a warm (70°) solution of 4 g. of III in 150 ml. of dry benzene was added a warm (70°) solution of N,N′-dicyclohexylcarbodiimide[16] in 50 ml. of dry benzene. N,N′-Dicyclohexylurea precipitated immediately. The mixture was stirred at 50–60° for 2 hr. and filtered while hot. The filtrate was evaporated to dryness *in vacuo*, and the residue was recrystallized from 75 ml. of toluene to give 2.7 g. of I as shiny needles, m.p. 158–159° (see Table II).

In a parallel run, a comparable yield of the same purity was obtained when acetic anhydride was used as the cyclizing agent.[2] Products prepared by both methods had identical antimalarial activity. Other 3-substituted sydnones were prepared by the carbodiimide procedure.

3-Piperonylsydnone Imine Hydrochloride.—Dry HCl was bubbled at a moderate rate into a solution of 6.6 g. of N-nitroso-N-piperonylglyconitrile in 150 ml. of dry methylene chloride cooled at −50°. After 10 min. the gas inlet was disconnected and the reaction mixture was evaporated below 0° under high vacuum. To the resulting residue there was added a small amount of petroleum ether (b.p. 35–60°), and the solid product which separated was filtered. It was recrystallized from a mixture of ethanol and petroleum ether to give 3.3 g. of analytically pure product (see Table II). **4-Methyl-3-piperonylsydnone imine hydrochloride** was prepared by essentially the same procedure.

3-Piperonyl-4-chlorosydnone was prepared by chlorination of I, according to the method of Baker, Ollis, and Poole[19] (see Table II).

3-(N-Exo-*p*-acetylaminobenzenesulfonyl)piperonylsydnone imine was prepared by treating 3-piperonylsydnone imine hydrochloride with *p*-acetylaminobenzenesulfonyl chloride in pyridine, according to the procedures of Yashunskii and Ermolaeva[20] and Daeniker and Druey[21] (see Table II).

Acknowledgment.—The authors wish to express their appreciation to Mr. Hal P. Van Fossen, Mr. John R. Gravatt, and Mrs. Margaret Rounds for the analytical and instrumental measurements.

(18) All melting points (corrected) were taken on a Thomas–Hoover melting point apparatus. The ultraviolet absorption spectra were determined with a Beckman DK-2 spectrometer.

(19) W. Baker, W. D. Ollis, and V. D. Poole, *J. Chem. Soc.*, 1542 (1950).

(20) V. G. Yashunskii and V. G. Ermolaeva, *Zh. Obshch. Khim.*, **32**, 186 (1962).

(21) H. U. Daeniker and J. Druey, *Helv. Chim. Acta*, **45**, 2462 (1962).

Thyroxine Analogs. XV.[1] Synthesis and Antigoitrogenic Activity of the 3′-*t*-Butyl Analog of 3,5,3′-Triiodo-L-thyronine and Its O-Methyl Ether

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Analog studies carried out to define structural features necessary for thyroid hormonal activity have led to active compounds in which the iodine atoms of thyroxine (Ia) and 3,5,3′-triiodothyronine (Ib) have been replaced by other halogen atoms[2] and by alkyl groups.[3–6] Iodine has proved to be the most effective

(1) (a) Paper XIV: E. C. Jorgensen and J. A. W. Reid, *Endocrinology*, **76**, 312 (1965). This investigation was supported by Research Grant AM-04223 from the National Institute of Arthritis and Metabolic Diseases, U. S. Public Health Service. (b) The synthesis and biological evaluation in the tadpole of 3-[4-(3-*t*-butyl-4-hydroxyphenoxy)-3,5-diiodophenyl]-L-alanine hydrochloride (compound Va hydrochloride) is reported independently by C. M. Buess, T. Giudici, N. Kharasch, W. King, D. D. Lawson, and N. N. Saha, *J. Med. Chem.*, **8**, 469 (1965).

(2) (a) M. V. Mussett and R. Pitt-Rivers, *Metab. Clin. Exptl.*, **6**, 18 (1957); (b) W. F. Cuthbertson, P. V. Elcoate, D. M. Ireland, D. C. Mills, G. S. Boyd, and M. F. Oliver, *J. Clin. Endocrinol. Metab.*, **21**, 1579 (1961).

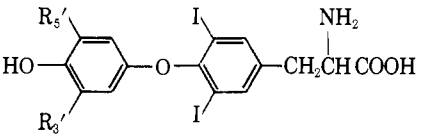
(3) (a) T. C. Bruice, R. J. Winzler, and N. Kharasch, *J. Biol. Chem.*, **210**, 1 (1954); (b) C. S. Pittman, H. Shida, and S. B. Barker, *Endocrinology*, **68**, 248 (1961).

(4) E. C. Jorgensen, P. A. Lehman, C. Greenberg, and N. Zenker, *J. Biol. Chem.*, **237**, 3832 (1962).

(5) (a) H. J. Bielig and G. Lützel, *Ann. Chem.*, **608**, 140 (1957); (b) E. C. Jorgensen and R. A. Wiley, *J. Med. Pharm. Chem.*, **5**, 1307 (1962).

(6) (a) B. Blank, F. R. Pfeiffer, C. M. Greenberg, and J. F. Kerwin, *ibid.*, **6**, 554 (1963); (b) C. M. Greenberg, B. Blank, F. R. Pfeiffer, and J. F. Pauls, *Am. J. Physiol.*, **205**, 821 (1963); (c) S. B. Barker and M. Shimada, *Proc. Staff Meetings Mayo Clinic*, **39**, 609 (1964); (d) A. Wahlborg, C. Bright, and E. Frieden, *Endocrinology*, **75**, 561 (1964).

halogen substituent. In the ring bearing the alanine side chain ("inner ring"), alkyl replacements have been restricted to the methyl group[5]; replacements in the



Ia, $R_3' = R_5' = I$
b, $R_3' = I$; $R_5' = H$
c, $R_3' = CH_3$; $R_5' = H$
d, $R_3' = CH(CH_3)_2$; $R_5' = H$
e, $R_3' = R_5' = H$

phenolic "outer ring" have included methyl, ethyl, isopropyl, isobutyl, and cyclohexyl.[3-6] The L-3′-methyl analog (Ie) was twice as active as thyroxine (Ia) and 44% as active as L-triiodothyronine (Ib) in oxygen consumption studies,[3b] while the L-3′-isopropyl analog (Id) possessed 1.8 times the antigoitrogenic activity and 1.3 times the plasma cholesterol-lowering activity of L-triiodothyronine in the rat.[6b] Barker and Shimada[6c] have estimated the biological activity of the 3′-isopropyl analog, as 500–750% that of thyroxine. In the tadpole metamorphosis assay, Frieden and co-workers[6d] found that the 3′-methyl, 3′-ethyl, and 3′-isopropyl analogs were all more active than thyroxine when administered either by injection or immersion.

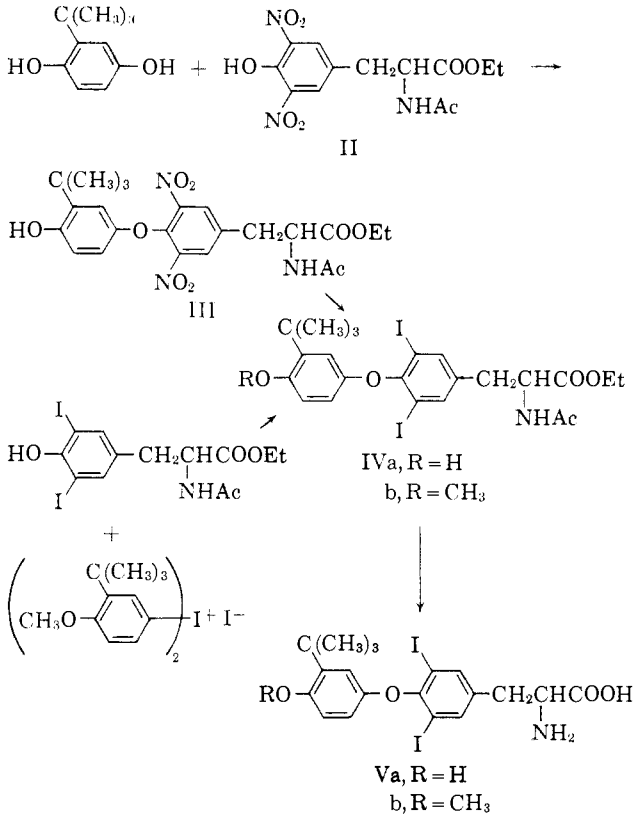
In connection with a correlation of the physical-chemical characteristics of "outer ring" substituents, Hansch and Fujita[7] have predicted that lipophilic electron-releasing groups such as alkyl should be superior to halogen atoms in promoting thyroxine-like activity. The ideal group was predicted to be the *t*-butyl, assuming the absence of steric effects. The high activities already demonstrated for other 3′-alkyl analogs and the prediction by Hansch led us to synthesize and biologically evaluate the L-3′-*t*-butyl analog (Va) and its O-methyl ether (Vb).

An unsuccessful attempt at the synthesis of Va has been reported,[6a] in which the protected amino acid (IVb) was prepared by condensation of N-acetyl-3,5-diiodo-L-tyrosine ethyl ester with di(3-*t*-butyl-4-methoxyphenyl)iodonium iodide (Chart I). Treatment with hydriodic acid gave Ie, hydrolysis and demethylation being accompanied by loss of the *t*-butyl group. We confirmed these results and attempted the O-methyl ether cleavage with hydrobromic acid or with a mixture of hydrochloric and hydriodic acids (see Experimental). Model studies on *o*-*t*-butylanisole with various combinations of aqueous hydrochloric, hydrobromic, and hydriodic acids in glacial acetic acid were also carried out, using thin layer chromatography to follow the reactions. When the conditions were such that cleavage of the O-methyl ether was obtained, the *t*-butyl group was also lost. However, hydrolysis of IVb with aqueous hydrochloric acid gave the O-methyl ether amino acid (Vb) which was desired for biological testing.

In order to avoid the requirement for the selective removal of a phenolic protective group, a synthetic route to Va was devised in which this function was left unprotected. This involved the condensation in pyridine of 2-*t*-butyl-1,4-hydroquinone with the methanesulfonyl derivative of N-acetyl-3,5-dinitro-L-tyrosine ethyl ester

(7) C. Hansch and T. Fujita, *J. Am. Chem. Soc.*, **86**, 1616 (1964).

CHART I



(II) according to the method of Meltzer, *et al.*[8] It was assumed that condensation would occur with the less hindered hydroxyl (*i.e.*, *meta* to the *t*-butyl group), since 2-*t*-butyl-4-ethoxyphenol[9] did not condense with II under the usual reaction conditions,[10a] or in a higher boiling solvent.[10b] The dinitro diphenyl ether (III), obtained in low yield, was reduced, bis-diazotized, and converted to the diiodo ether (IVa). Hydrolysis with hydrochloric acid gave the optically active 3′-*t*-butyl analog (Va), which was shown to be free of 3,5-diiodothyronine (Ie) by paper chromatography.

Biological Results.—The 3′-*t*-butyl analog (Va) and its O-methyl ether (Vb) were tested for thyroxine-like activity by the rat antigoiter procedure as described previously.[11] The results and estimates of potency relative to L-thyroxine are presented in Table I. The analog Va was tested at molar ratios of 5:1, 1:1, and 0.25:1 with respect to a standard effective dose of 3 γ of sodium L-thyroxine pentahydrate for each 100 g. of body weight. On the same molar scale, Vb was tested at ratios of 5:1 and 1:1. At a molar ratio of 1:1, the O-methyl ether (Vb) produced an average thyroid weight which was significantly different ($P > 0.95$) from that produced by all dose levels of the thyroxine controls, but was not significantly different ($P > 0.95$) from the thiouracil controls. At a 5:1 molar ratio, Vb produced a response which was significantly different ($P > 0.95$) from the 3- and 4.5-γ dose levels of thyroxine but

(8) R. I. Meltzer, D. M. Lustgarden, and A. Fischmann, *J. Org. Chem.* **22**, 1577 (1957).

(9) Kindly provided by Universal Oil Products Company, Des Plaines, Ill.

(10) (a) N. Zenker, Ph.D. Dissertation, University of California, 1950; (b) S. J. Feinglass, Ph.D. Dissertation, University of California, 1964.

(11) (a) E. C. Jorgensen and P. Slade, *J. Med. Pharm. Chem.*, **5**, 729 (1962). (b) All compounds were dissolved in 0.9% aqueous NaCl containing 0.01 *N* NaOH, except Va which was dissolved using 0.02 *N* HCl. This was to avoid possible oxidation of Va under alkaline conditions.

TABLE I

RAT ANTIGOITER ASSAY OF THYROXINE ANALOGS[a]

Food	Compd. injected	Daily dose, γ/100 g.	Molar ratio	Thyroid wt., mg./100 g. ± S.D.	Approx. activity
Untreated	...	...	...	7.9 ± 1.5	...
Tu[b]	...	...	...	28.1 ± 8.3	...
Tu	Thyroxine[c]	2.0	0.67	14.6 ± 1.4	100
Tu	Thyroxine	3.0	1.0	8.7 ± 2.9	100
Tu	Thyroxine	4.5	1.5	6.1 ± 1.3	100
Tu	Va	0.49	0.25	20.4 ± 4.5	0
Tu	Va	1.99	1	9.9 ± 4.7	100
Tu	Va	9.95	5	6.3 ± 0.7	100
Tu	Vb	2.0	1	20.2 ± 2.5	0
Tu	Vb	10.0	5	18.2 ± 6.6	13

[a] Six rats were used at each control and dose level. [b] Thiouracil, 0.3%. [c] Sodium L-thyroxine pentahydrate.

was the same as the 2-γ dose level and the thiouracil controls. The O-methyl ether (Vb), therefore, has at most 13% of the activity of L-thyroxine. The low antigoitrogenic activity of Vb is typical of metabolic effects previously demonstrated for the O-methyl ether derivatives of this class of compound.[12] The biological data of compound Vb must be interpreted with some caution, since it was not completely soluble in the aqueous alkaline medium and was administered as a suspension.

The free phenolic *t*-butyl analog (Va) produced a thyroid weight significantly lower ($P > 0.95$) than the thiouracil controls at molar ratios of 5:1 and 1:1, and not significantly different ($P > 0.95$) from the 3-γ dose of L-thyroxine. At a molar ratio of 0.25:1, Va was significantly different ($P > 0.95$) from the 2-, 3-, and 4.5-γ doses of L-thyroxine, and not significantly different ($P > 0.95$) from the thiouracil controls. The activity of Va is estimated at equal to or greater than 100%, and less than 400% that of L-thyroxine. This high activity for the 3′-*t*-butyl analog is at least qualitatively consistent with the predictions based on substituent constants assigned by Hansch.[7]

The importance of lipophilic character for substituents in the 3′-position of thyroxine analogs may be interpreted in terms of the contributions of such substituents in providing a balanced lipophilic–hydrophilic character to the molecule for transport processes in the intact animal involving passive diffusion through membranes. Of more importance, may be the role of such lipophilic substituents, if properly oriented sterically, in contributing to hydrophobic binding, both to transport protein and at the biological receptor sites. In addition, the electron-donating characteristics of alkyl substituents could contribute to a functional role for the "outer ring," such as its participation in electron transport systems *via* its hydroquinoid structure.[13]

Experimental[14]

Di(3-*t*-butyl-4-methoxyphenyl)iodonium Iodide.—Conditions similar to the general procedures described by Blank, *et al.*,[6a] were used.[15] To acetic anhydride (14 ml.) cooled to −15° was added dropwise 10 ml. of red fuming HNO_3 (*d* 1.60). The temperature was kept below 20° during this addition. Finely powdered iodine (5.0 g.) was added in one portion followed by trifluoroacetic acid (10 ml.). The temperature rose to 40° and was then maintained at 45° by intermittent heating on a steam bath for 20–25 min., or until all the iodine had dissolved. The solvents were removed under reduced pressure (1–2 mm.) with the bath temperature kept below 45°. The semicrystalline residue of iodine trifluoroacetate was allowed to react with 2-*t*-butylanisole (23.0 g., 0.14 mole) as described by Blank, *et al.*, to give the iodonium iodide (15.0 g., 37%), m.p. 180–181° (lit.[6a] m.p. 177–178°).

N-Acetyl-3-[4-(3-*t*-butyl-4-methoxyphenoxy)-3,5-diiodophenyl]-L-alanine Ethyl Ester (IVb).—N-Acetyl-4-hydroxy-3,5-diiodophenyl-L-alanine ethyl ester[16] (5.5 g., 0.011 mole) was condensed with di(3-*t*-butyl-4-methoxyphenyl)iodonium iodide as described by Blank, *et al.*,[6a] for the DL-compound. Trituration of the crude reaction residue with petroleum ether (b.p. 30–60°) (50 ml.) gave the ether as a white solid (4.7 g., 64%), m.p. 165–167°. Two crystallizations from ethanol raised the melting point to 170–171°; $[\alpha]^{25}D$ +40° (*c* 1, $CHCl_3$); λ_{max}^{KBr} 3.09, 5.78, and 6.1 μ. The n.m.r. spectrum, which has been published elsewhere,[17] confirms the structure.

Anal. Calcd. for $C_{24}H_{29}I_2NO_5$: C, 43.33; H, 4.39; I, 38.15. Found: C, 43.30; H, 4.30; I, 37.78.

3-[4-(3-*t*-Butyl-4-methoxyphenoxy)-3,5-diiodophenyl]-L-alanine (Vb).—A solution of IVb (0.3 g., 0.4 mmole) in glacial acetic acid (25 ml.), water (12 ml.), and concentrated HCl (12 ml.) was heated under reflux for 5 hr. Distillation reduced the volume of the reaction mixture by two-thirds and, on cooling to room temperature, the hydrochloride of Vb precipitated as white crystals (0.25 g.), m.p. 195–196°. The hydrochloride was dissolved in aqueous 2 *N* HCl–ethanol (1:1) by heating to 60°. The pH was adjusted to 4.5 with sodium acetate to give a white precipitate on cooling to 0°. This procedure was repeated to give the amino acid (Vb) (0.16 g., 61%), m.p. 243–244° dec., $[\alpha]^{25}D$ +25° (*c* 1, 1:3 aqueous *N* HCl–ethanol), R_f 0.88 (solvent system: isoamyl alcohol–2 *N* ammonium hydroxide). With the same solvent system, 3,5-diiodothyronine had R_f 0.67.

Anal. Calcd. for $C_{20}H_{23}I_2NO_4$: C, 40.36; H, 3.90; I, 42.66. Found: C, 39.99; H, 4.12; I, 42.38.

The following conditions were tried in unsuccessful attempts to hydrolyze *and* demethylate the amino acid (IVb). (a) A solution of IVb (250 mg.), red phosphorus (0.35 g.), glacial acetic acid (8.0 ml.), and 48% aqueous HBr (2 ml.) was heated under reflux for 3.25 hr.[18]; this gave mainly the O-methyl ether (Vb).

(12) (a) K. Tomita, H. A. Lardy, D. Johnson, and A. Kent, *J. Biol. Chem.*, **236**, 2981 (1961); (b) B. Blank, C. M. Greenberg, and J. F. Kerwin, *J. Med. Chem.*, **7**, 53 (1964).

(13) E. C. Jorgensen, *Proc. Staff Meetings Mayo Clinic*, **39**, 560 (1964).

(14) Melting points were determined with a Thomas–Hoover capillary melting point apparatus and are corrected. Microanalyses were performed by the Microanalytical Department, University of California, Berkeley Calif. Optical rotations were obtained in a 0.5-dm. tube with a Rudolph photoelectric polarimeter. Paper chromatography was carried out on Whatman No. 1 paper which had been washed with 2 *N* NH_4OH. N.m.r. spectra were obtained at a field strength of 60 Mc. on samples in deuteriochloroform solution on a Varian A-60 instrument using tetramethylsilane (δ 0) as internal standard. Infrared spectra were obtained with a Beckman IR-5 instrument.

(15) We are grateful to Dr. Benjamin Blank for providing additional experimental details of this reaction.

(16) J. R. Chalmers, G. T. Dickson, J. Elks, and B. A. Hems, *J. Chem. Soc.*, 3424 (1949).

(17) P. A. Lehman and E. C. Jorgensen, *Tetrahedron*, **21**, 363 (1965).

(18) Conditions used for hydrolyzing related compounds containing labile iodine atoms on the "inner ring."[19]

(19) E. C. Jorgensen and J. A. W. Reid, *J. Med. Chem.*, **7**, 701 (1964).

(b) A solution of IVb (250 mg.), glacial acetic acid (10 ml.), water (4 ml.), and concentrated HCl (4.0 ml.) was heated under reflux for 4 hr. To this was added 47% aqueous HI (5.0 ml.), and heating under reflux was continued for 30 min. Only 3,5-diiodothyronine could be isolated from the reaction mixture as deduced from mixture melting point and a comparison of infrared spectra.

N-Acetyl-3-[4-(3-*t*-butyl-4-hydroxyphenoxy)-3,5-dinitrophenyl]-L-alanine Ethyl Ester (III).—The method is similar to that described by Meltzer, *et al.*[8] A solution of N-acetyl-3,5-dinitro-L-tyrosine ethyl ester (8.55 g., 0.025 mole) in dry pyridine (40 ml.) was stirred and heated on a steam bath. Freshly distilled methanesulfonyl chloride (3.15 g., 0.027 mole) was then added; a vigorous reaction occurred. After 2 min., nitrogen was passed into the system for 5 min. before 2-*t*-butyl-1,4-hydroquinone[9] (6.2 g., 0.037 mole) was added in one portion. Heating was continued for a further 15 min. in an atmosphere of nitrogen before the reaction mixture was poured onto crushed ice (100 g.). The mixture was then extracted twice with benzene (100 ml.), the benzene extracts were combined and washed consecutively with 100-ml. portions of water, aqueous 2 *N* HCl, water, aqueous 2 *N* NaOH, and finally water. Evaporation of the benzene gave a dark brown oily residue (2.6 g.) which was dissolved in the minimum quantity of benzene and chromatographed on acid-washed alumina (125 g.). Elution with ethyl acetate gave a fraction (1.4 g., 11%) which solidified on addition of petroleum ether (b.p. 30–60°), m.p. 105–120°. Crystallization from benzene–ethyl acetate gave the diphenyl ether (III) as yellow crystals, m.p. 124–126°, $[\alpha]^{25}D$ +33° (*c* 1, $CHCl_3$), containing 1 molecule of benzene of recrystallization. The n.m.r. spectrum showed the characteristic pattern of the dinitrothyronine structure[17] (*t*-BuMe, δ 1.33), also a singlet at δ 7.37 (6 protons) assigned to the benzene of crystallization. The benzene was retained even following recrystallization from aqueous methanol.

Anal. Calcd. for $C_{23}H_{27}N_3O_9 \cdot C_6H_6$: C, 61.37; H, 5.87. Found: C, 61.16; H, 5.88.

Considerable quantities of black tars were obtained, but these were not investigated.

N-Acetyl-3-[4-(3-*t*-butyl-4-hydroxyphenoxy)-3,5-diiodophenyl]-L-alanine Ethyl Ester (IVa).—A solution of III (2.8 g., 4.1 mmoles) in glacial acetic acid (80 ml.) was hydrogenated, bisdiazotized, and converted to the 3,5-diiodo ether as described by Jorgensen and Kaul[20] to give a dark brown residue (2.78 g.) which was dissolved in the minimum quantity of $CHCl_3$ and chromatographed on acid-washed alumina (100 g.). Elution with $CHCl_3$ gave the crude diiodo ether (IVa) (2.6 g., 86%) as a viscous oil. Crystallization from aqueous methanol gave crystals, m.p. 107–109°, $[\alpha]^{25}D$ +44° (*c* 1, $CHCl_3$). The n.m.r. spectrum confirmed the structure[17] (*t*-BuMe, δ 1.37).

Anal. Calcd. for $C_{23}H_{27}I_2NO_5$: C, 42.42; H, 4.18. Found: C, 42.08; H, 4.37.

3-[4-(3-*t*-Butyl-4-hydroxyphenoxy)-3,5-diiodophenyl]-L-alanine (Va).—A solution of IVa (1.0 g., 1.5 mmoles) in glacial acetic acid (75 ml.), concentrated HCl (36 ml.), and water (36 ml.) was heated under reflux for 6 hr. under nitrogen. After 3 hr. of reflux, a portion (30 ml.) of concentrated HCl was added. After 6 hr., distillation decreased the volume of the reaction mixture by two-thirds, and the pH was adjusted to 4.5 with sodium acetate. Addition of water (40 ml.) precipitated amino acid as a brown solid (0.33 g., 37%), m.p. 226–228° dec. Two isoelectric precipitations from the alkaline side gave a tan-colored precipitate (0.22 g.), m.p. 229–230° dec., $[\alpha]^{25}D$ +14° (*c* 1, 1:1 ethanol–aqueous *N* HCl), R_f 0.90 (solvent system: isoamyl alcohol–2*N* NH_4OH). In the same solvent system, 3,5-diiodothyronine had R_f 0.67.

Anal. Calcd. for $C_{19}H_{21}I_2NO_4 \cdot 0.5H_2O$: C, 38.68; H, 3.76; I, 43.02. Found: C, 38.82; H, 3.78; I, 42.70.

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(20) E. C. Jorgensen and P. N. Kaul, *J. Am. Pharm. Assoc., Sci. Ed.*, **48**, 653 (1959).

17-Chloroethynylated Steroids. III. The Synthesis of 17α-Chloroethynyl-5,7,9-estratrien-17β-ol-3-one

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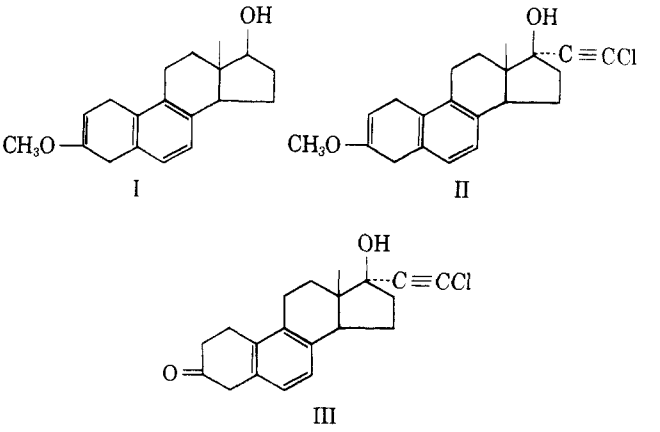
Examination of the structure–activity relationship among the pituitary gonadotrophin-inhibiting and progestational chloroethynylestrenones[1] suggested a rough correlation of activity with increasing planarity of the B-ring. It was, therefore, of interest to prepare the B-ring aromatic derivative III in order to test this correlation.



Birch reduction[2] of 3-methoxy-1,3,5,7,9-estrapentaen-17β-ol[3] afforded 3-methoxy-2,5,7,9-estratetraen-17β-ol (I). Oppenauer oxidation[4] of I led to the corresponding C-17 ketone, which after chloroethynylation[5] to yield II, afforded the required 17α-chloroethynyl-5,7,9-estratrien-17β-ol-3-one (III) after acid hydrolysis.

Compounds II and III were tested in the Merck Institute for Therapeutic Research.[6] Results are summarized in Table I.

Experimental[7]

3-Methoxy-2,5,7,9-estratetraen-17β-ol (I).—A solution consisting of 1.347 g. of 3-methoxy-1,3,5,7,9-estrapentaen-17β-ol[3] (m.p. 150–151°), 20 ml. of tetrahydrofuran,[8] and 20 ml. of *t*-butyl alcohol[8] was added with stirring to 50 ml. of liquid ammonia.[8] A total of 440 mg. of sodium was then added in four approximately equal portions over a period of 5 min. The re-

(1) J. H. Fried, T. S. Bry, A. E. Oberster, R. E. Beyler, T. B. Windholz, J. Hannah, L. H. Sarett, and S. L. Steelman, *J. Am. Chem. Soc.*, **83**, 4663 (1961).

(2) H. L. Dryden, Jr., G. M. Webber, R. R. Burtner, and J. A. Cella, *J. Org. Chem.*, **26**, 3237 (1961).

(3) W. E. Bachmann and A. S. Dreiding, *J. Am. Chem. Soc.*, **72**, 1323 (1950).

(4) C. Djerassi, *Org. Reactions*, **6**, 207 (1951).

(5) H. G. Viehe, *Chem. Ber.*, **92**, 1950 (1959).

(6) We are indebted to Dr. S. L. Steelman for carrying out this determination.

(7) Melting points were taken on a micro hot stage and are corrected. Rotations were determined in benzene at a concentration of 7 mg./ml. at 24–26°. We are indebted to A. Kalowski for the ultraviolet spectra and to R. Boos and his associates for microanalysis herein reported.

(8) The tetrahydrofuran was freshly distilled from $LiAlH_4$. The *t*-butyl alcohol and the ammonia were distilled from sodium.