Sulfuric Acid Treatment of Phendimetrazine Diastereoisomers.—Samples (50 mg.) of *d-threo*- (I) and *l-erythro*-3,4-dimethyl-2-phenylmorpholine (II) were dissolved in concentrated H_2SO_4 (0.5 ml. each) and left standing at room temperature. After 15 and 64 hr., respectively, the samples were diluted with water to make a 10% solution, and the optical rotation was determined (Table I). The 64-hr. samples (in triplicate) were rendered alkaline, the free bases were extracted with ether, dried (Na_2SO_4), and treated with HCl, and the mixture was evaporated to dryness under reduced pressure. The residue[19] was dissolved in water to make a final concentration of 1×10^{-3} *M*, and the effect on guinea pig liver MAO was determined *in vitro*.[17] The inhibition was 50 ± 4%. Compared to the values listed in Table III, such a degree of inhibition corresponds to a mixture consisting of about 80% of the *threo* diastereoisomer.

Acknowledgment.—We wish to thank Dr. Frank A. L. Anet for the n.m.r. spectra and their interpretation and Jean Dubuc and his group, respectively, for the MAO assays.

(19) Thin layer chromatography [methanol–benzene, 1:10; sprayed with concentrated H_2SO_4 followed by Draggendorf's reagent as modified by Munier and Macheboeuf (*cf.* K. Randerath, "Dünnschichtchromatographie," Verlag Chemie, Weinheim, 1962, p. 128)] indicated a mixture consisting of the unchanged *erythro* (R_f 0.25) and the newly formed *threo* (R_f 0.33) isomers.

The Synthesis of Thyromimetic Substances and Potential Inhibitors of Thyroxine[1]

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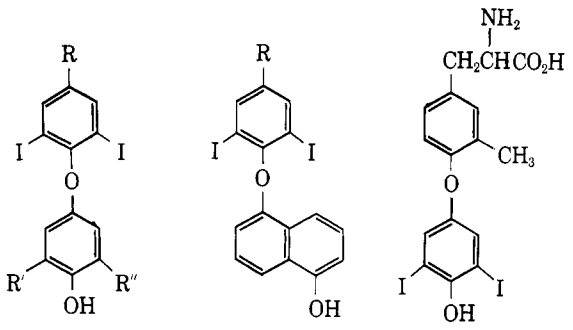
The syntheses of eight novel analogs of thyroxine are reported. Advantages of using the monobenzoate esters of requisite hydroquinone intermediates in the Glaxo method are described. Preliminary testing data for 3,5-diiodo-3′-methyl-L-thyronine and the corresponding *t*-butyl compound are noted. R_f values for the thyroxine analogs are recorded.

In a previous communication,[3] the basis for our approach to the synthesis of thyroxine analogs and potential inhibitors of thyroxine was summarized. The syntheses of eight novel analogs, Ia–d, IIa–c, and III are now reported; the synthetic scheme for Id was briefly communicated previously.[3]

In view of current theories of thyroxine action and results of testing, the choice of compounds was made on the basis of the postulated relation between activity and presumed ability to form a quinoid structure,[4] the easy oxidation of *t*-butylhydroquinone, the tendency of highly hindered phenols to be converted to cyclohexadienone derivatives, and the thyroxine-like activities of methyl analogs in the tadpole.[3,5] For further references, the monographs of Pitt-Rivers and Tata[6] and the papers of Lissitzky and Bouchilloux[7] are also of direct interest in connection with biochemical activities of substances related to those of the present study.



I II DL-III

Ia, R = L-$CH_2CH(NH_2)COOH$; R′ = CH_3; R″ = H
b, R = L-$CH_2CH(NH_2)COOH$; R′ = *t*-C_4H_9; R″ = H
c, R = CH_2CH_2COOH; R′ = *t*-C_4H_9; R″ = H
d, R = CH_2CH_2COOH; R′ = R″ = *t*-C_4H_9

IIa, R = CH_2COOH
b, R = CH_2CH_2COOH
c, R = L-$CH_2CH(NH_2)COOH$

Pittman, Shida, and Barker[8] found that Ia (the 3′-methyl analog of triiodothyronine) was 44% as active as L-triiodothyronine and twice as active as L-thyroxine in basal metabolic tests in thyroidectomized rats. The testing of two of the analogs prepared in the present study was carried out with Mr. Roy G. Robinson, but only in tadpoles. Using the method for detecting the relative rates of the induced metamorphosis of *Rana catesbeiana* tadpoles developed by Bruice, Winzler, and Kharasch,[5d] it was found that the 3′-methyl analog (Ia), as well as the 3′-*t*-butyl analog (Ib), had activities equivalent to that of L-thyroxine. Thus, the biological effects of these 3′-alkyl analogs as thyromimetic agents appears to be substantiated in these preliminary screenings and is of particular interest in view of earlier considerations on the activities of alkyl-substituted thyronines.[5d,8] It is of special interest to note that a single alkyl group in the prime ring (in place of iodine) exerts a definite positive effect on activity, since it is known

(1) This study was supported in part by Grant A-703 from the National Institutes of Health and in part by supporting grants from the Travenol Laboratories, Skokie, Ill., and the Upjohn Company, Kalamazoo, Mich. An independent synthesis and a rat antigoiter assay of L-3,5-diiodo-4-(3-*t*-butyl-4-hydroxyphenoxy)phenylalanine (Ib) is reported by E. C. Jorgensen and J. A. W. Reid, *J. Med. Chem.*, **8**, 533 (1965).

(2) To whom inquiries should be addressed.

(3) N. Kharasch and N. N. Saha, *Science*, **127**, 756 (1958).

(4) C. Niemann and C. E. Redemann, *J. Am. Chem. Soc.*, **63**, 1549 (1941); C. Niemann and J. F. Mead, *ibid.*, **63**, 2685 (1941); C. Niemann, *Fortsch. Chem. org. Naturstoffe*, **7**, 167 (1950).

(5) (a) See, *e.g.*, H. S. Blanchard, *J. Org. Chem.*, **25**, 264 (1960); E. Muller, *et al.*, *Chem. Ber.*, **92**, 2278 (1959), and references therein; (b) C. Hansch and T. Fujita [*J. Am. Chem. Soc.*, **86**, 1621 (1964)] have also recently commented on the problems of correlating biological activities with structures in the thyroxine series; (c) *cf.* also T. C. Bruice, N. Kharasch, and R. J. Winzler, *Arch. Biochem. Biophys.*, **62**, 305 (1956); (d) T. C. Bruice, R. J. Winzler, and N. Kharasch *J. Biol. Chem.*, **210**, 1 (1954).

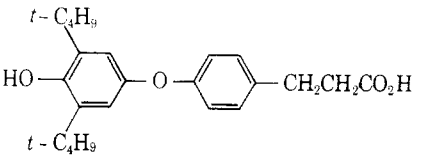
(6) R. Pitt-Rivers and J. R. Tata, "The Thyroid Hormones," Pergamon Press, London, 1959.

(7) S. Lissitzky and S. Bouchilloux, *Bull. soc. chim. biol.*, **39**, 1215 (1957), and earlier articles.

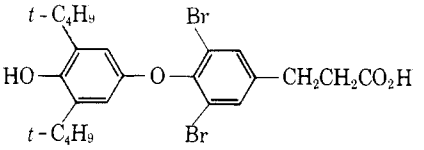
(8) C. S. Pittman, H. Shida, and S. B. Barker, *Endocrinology*, **68**, 248 (1961); *cf.* also, S. B. Barker, *Federation Proc.*, **21**, 635 (1962); A. Wahlborg, C. Bright, and E. Frieden, *Endocrinology*, **75**, 561 (1964).

that the 3′,5′-dihydro compound (L-3,5-diiodothyronine) has a lower activitity than does thyroxine.[5c]

The studies of Cahnmann and co-workers,[9] concerning the nonenzymatic coupling of two diiodotyrosine derivatives, admirably illustrate the formation of cyclohexadienone precursors of thyroxine and its analogs and provide a unique route to substances related to those of the present study. Compounds IV and V, so prepared by Matsuura and Cahnmann,[9b] were tested



IV



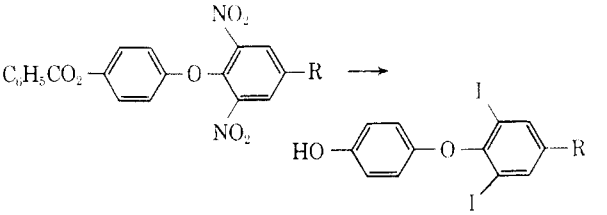
V

by them, since these substances were of special interest, in view of positive activities reported earlier[5c,d] for 3,5-dimethyl-3′,5′-diiodothyronine, and the considerations of Kharasch and Saha[3] on the possible effects of compounds like Id as antagonists of thyroxine. Cahnmann and Matsuura found that IV and V were both devoid of activity in tadpole metamorphosis tests, and IV was also inactive in the oxygen consumption assay in the rat. Furthermore, both IV and V showed no effect as thyroxine antagonists in the rat. The conclusion of Matsuura and Cahnmann[9b] that the presence of two bulky substituents, as *t*-butyl, *ortho* to the phenolic hydroxyl group precludes both thyroxine activity, as well as activity as a thyroxine antagonist, thus seems definitive. However, since IV lacks iodine atoms in the 3,5-positions, the presence of which is known to be important for thyroxine activity, and since it is known that bromo analogs are of considerably lower activities than are the iodo compounds,[5c] the desirability for further testing of selected analogs is still indicated.

Compounds of types I and II were synthesized by modifications of the Glaxo method.[10] Compound Ia was prepared by using 3-methyl-4-benzoyloxyphenol for condensation to the diphenyl ether. The latter was obtained by a sequence which involved benzylation of toluhydroquinone, followed by benzoylation and removal of the benzyl group by hydrogenation to yield the desired protected toluhydroquinone derivative, m.p. 144–145°, which gave a melting point depression on admixture with the alternate isomer.

The benzoate group proved to be a satisfactory blocking group for later steps in the syntheses. These included coupling to the diphenyl ether with the appropriately blocked 3,5-dinitrotyrosine, reduction of the nitro groups, diazotization of the amino groups, and conversion to the 3,5-diiodo compounds. After these conversions, the blocking groups were readily removed by refluxing with mixtures of hydrochloric and acetic acids.

The use of the benzoate function as a protecting group in the synthesis of thyroxine compounds has been previously considered. Dickson[11] reported the ethyl ester of N-acetyl-4-(4-benzoyloxyphenoxy)-3,5-diiodophenylalanine, but to our knowledge details of the hydrolysis to 3,5-diiodothyronine have not been recorded. The success of the Glaxo synthesis requires purification of intermediates at appropriate stages. To test the effectiveness of the benzoate blocking group, we investigated the comparative synthesis of VIa–d with the benzoates and by conventional methods. In our experience we found that the use of the benzoates was advantageous. Particularly, for example, the blocked di-



VIa, R = COOH
b, R = CH_2COOH
c, R = CH_2CH_2COOH
d, R = L-$CH_2CH(NH_2)COOH$

nitrodiphenyl ether in the case of the acetic acid analog (VIb) is an especially difficult intermediate to purify because of its relatively low melting point (76°) and high solubility if the methoxy derivative[12] is used in place of the benzoate. The latter, however, melts at 146–147° and is easily purified by crystallization. The mild conditions for removing the benzoate blocking group is another advantage, especially with more sensitive compounds, where the use of hydrobromic or hydriodic acid may lead to undesired side reactions.

In the preparations of IIa–c, the use of 1,5-dihydroxynaphthalene monobenzoate also proved effective. However, we were unable to prepare the required ester ether of 2,6-di-*t*-butylhydroquinone, and in this case used the hydroquinone directly, with attendant side reactions, for the coupling to the diphenyl ether.

Attempted conversions of Ia to 3′-methyl-3,5,5′-triiodothyronine by iodination of Ia were not successful, although it might be anticipated that this could be readily achieved.[13]

Compound III was obtained *via* VII as shown below.

Assays of Ia and Ib.—Using the procedure of Bruice, *et al.*,[5d] and with 30 selected *Rana catesbeiana* tadpoles in the test groups, in the groups subjected to standard L-thyroxine, and in the control groups, Ia was found to be equally as active as L-thyroxine, both tested at 10^{-6} *M* concentrations. The per cent activity of Ia was 100.8 and of Ib was 99.3 (10^{-6} *M* for Ib and L-thyroxine

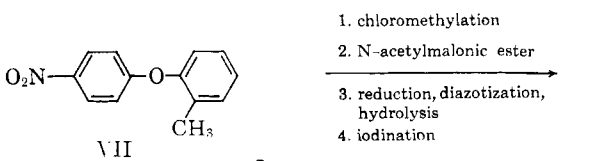
(9) (a) H. J. Cahnmann, and T. Matsuura, *J. Am. Chem. Soc.*, **82**, 2050 (1960); (b) T. Matsuura and H. J. Cahnmann, *ibid.*, **82**, 2055 (1960); (c) A. Hofer and H. J. Cahnmann, *J. Med. Chem.*, **7**, 326 (1964); *cf.* also A. Nishinaga and T. Matsuura, *J. Org. Chem.*, **29**, 1812 (1964).

(10) (a) J. H. Barnes, R. C. Cookson, G. T. Dickson, J. Elks, and V. D. Poole, *J. Chem. Soc.*, 1448 (1953), and previous articles; (b) *cf.* especially, J. R. Chambers, G. T. Dickson, J. Elks and B. A. Hems, *ibid.*, 3424 (1949).

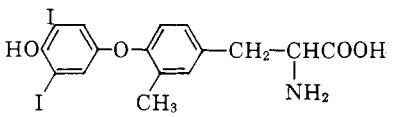
(11) G. T. Dickson, British Patent 671,070 (April 30, 1952); *Chem. Abstr.*, **47**, 7543 (1953); Glaxo Laboratories Ltd., German Patent 896,946 (Nov. 16, 1953); G. T. Dickson, U. S. Patent 2,680,762 (June 8, 1954).

(12) J. H. Wilkinson, *Biochem. J.*, **63**, 803 (1956); J. H. Wilkinson, British Patent 805,761 (Dec. 10, 1958); *Chem. Abstr.*, **53**, 8074 (1959).

(13) Compare reported difficulties in the iodination of L-thyronine: P. Block and G. Powell, *J. Am. Chem. Soc.*, **64**, 1070 (1942); *cf.* C. L. Gemmill, *Arch. Biochem. Biophys.*, **63**, 177 (1956); and of 3,5-diiodo-2′,5′-dimethylthyronine: N. Zenker and E. C. Jorgensen, *J. Am. Chem. Soc.*, **81**, 4643 (1959).



VII



DL-III

also). In a similar series of tests, but using Ia (5×10^{-7} *M*) admixed with L-thyroxine (also 5×10^{-7} *M*) *vs.* L-thyroxine at 10^{-6} *M*, the activity was 87%. With Ib, similarly, 94% activity was detected.

Experimental[14]

Hydroquinone Monobenzoate.—To 27.5 g. (0.25 mole) of hydroquinone and 20.6 g. (0.26 mole) of pyridine in 250 ml. of carbon tetrachloride was added dropwise during 1 hr., with rapid stirring, 34 g. (0.24 mole) of benzoyl chloride in 75 ml. of carbon tetrachloride. The mixture was acidified with 100 ml. of 1.2 *N* HCl, and solid products were collected and washed with water. The precipitate contained hydroquinone mono- and dibenzoate. The dibenzoate was sparingly soluble in hot ethanol and was separated using 200 ml. of boiling alcohol; crude yield 16 g. The hot filtrate was diluted with 1 vol. of water and cooled. The yield of hydroquinone monobenzoate was 24 g. (50%), m.p. 162–163°.[15]

5-Benzoyloxy-1-naphthol.—A solution of 16.1 g. (0.10 mole) of 1,5-naphthalenediol and 9.8 g. (0.26 mole) of dry pyridine in 600 ml. of ether was stirred, chilled in ice, and treated with 15.2 g. (0.24 mole) of benzoyl chloride in 1-ml. portions during 30 min. The mixture was kept at room temperature for 1 hr. and filtered. The filtrate was evaporated to dryness and the residue was triturated with dilute HCl. The dibenzoate was separated from the monobenzoate as above, giving 10.6 g. (37%) of product, m.p. 165–166° after recrystallization from dilute ethanol.

Anal. Calcd. for $C_{17}H_{12}O_3$: C, 77.25; H, 4.57. Found: C, 77.51; H, 4.65.

4-Benzoyloxyphenyl Benzyl Ether.—This was prepared in 79% yield by benzoylating 10 g. of hydroquinone monobenzyl ether by the Schotten–Baumann technique; and in 89% yield, using pyridine–benzoyl chloride, in benzene. The samples were crystallized from 95% ethanol and melted at 135–136°.

Anal. Calcd. for $C_{20}H_{16}O_3$: C, 78.93; H, 5.17. Found: C, 78.75; H, 5.17.

3-Methyl-4-benzoyloxyphenyl Benzyl Ether.—To 41 g. (1.02 moles) of NaOH in 100 ml. of water was added rapidly 124 g. (1 mole) of toluhydroquinone and 127 g. (1 mole) of benzyl chloride, in 600 ml. of 95% ethanol. The resulting solution was steam heated for 3 hr. while solvent escaped. The mixture was cooled and neutralized with 1 l. of 5% aqueous NaOH. The dibenzyl ether was separated by extraction with benzene, the aqueous layer was acidified with 5 *N* HCl, and the crude monobenzyl ether was extracted with benzene. The benzene layer was washed with water and solvents were distilled under reduced pressure. The residue was dissolved in a solution of 26.7 g. (0.66 mole) of NaOH in *ca.* 500 ml. of water at 0° and, with stirring in an ice bath, 94 g. (0.66 mole) of benzoyl chloride was added in 10-g. portions. Sodium acetate was added as required to maintain alkalinity. After 2 hr. the precipitate was collected and washed with water and 95% ethanol. The product was dissolved in benzene and the solution was distilled until water and alcohol were removed. The solution was cooled, partially decolorized with alumina, filtered, and evaporated to dryness. The residue was crystallized from 95% ethanol; yield 60 g. (19%), m.p. 71–72°, and raised to 81–82° by further recrystallization.

Anal. Calcd. for $C_{21}H_{18}O_3$: C, 79.22; H, 5.70. Found: C, 78.85; H, 5.82.

(14) All melting points are uncorrected.

(15) T. Kehrmann, M. Sandoz, and R. Monnier, *Helv. Chim. Acta*, **4**, 943 (1921).

The product was also obtained by benzoylation of the sodium sulfate–dried benzene solution of the crude monobenzyl ether with equimolar amounts of pyridine and benzoyl chloride. The mixture was washed with water, dilute HCl, and water, and concentrated *in vacuo*. The residue was crystallized from 95% ethanol; yield 67 g. (21%), m.p. 75–77°.

4-Benzoyloxy-3-*t*-butylphenyl Benzyl Ether.—This product was prepared as above, using pyridine-benzoyl chloride. The desired monobenzyl ether of *t*-butylhydroquinone was insoluble in dilute alkali and was extracted with benzene directly from the alkaline solution. The crude sample thus contained the dibenzyl ether formed, but this did not interfere with isolation of the desired product. After recrystallization from absolute ethanol the melting point was 78–79°, yield 33% (1 *M* run).

Anal. Calcd. for $C_{24}H_{24}O_3$: C, 79.99; H, 6.69. Found: C, 80.12; H, 6.81.

4-Hydroxy-3,5-di-*t*-butylphenyl Benzyl Ether.—This was obtained by benzylating 2.0 g. of 2,6-di-*t*-butylhydroquinone, as above, and recrystallizing from ligroin (60–70°); m.p. 88–89°, yield 60%.

Anal. Calcd. for $C_{21}H_{28}O_2$: C, 80.73; H, 9.03. Found: C, 81.08; H, 9.40.

The ether did not undergo benzoylation or methylation normally at the hindered hydroxyl group.

Hydroquinone Monobenzoate by Hydrogenolysis.—A suspension of 5.5 g. of 4-benzoyloxyphenyl benzyl ether in 150 ml. of 95% ethanol was shaken in the presence of palladium on charcoal at 2.81 kg./cm.^{2} (40 p.s.i.) for 6 hr. The organic material dissolved slowly. After separation of catalyst, the filtrate was concentrated and diluted with water; yield 3.5 g., m.p. and m.m.p. 161–162° with an authentic sample.

3-Methyl-4-benzoyloxyphenol.—This phenol was prepared by hydrogenolysis of the corresponding benzyl ether (53 g. in 500 ml. of absolute ethanol) for 24 hr. and crystallization from benzene; yield (in two crops) 33 g., 87%, m.p. 142–144°; analytical sample, m.p. 145–146°. A mixture melting point with product prepared by direct benzoylation of toluhydroquinone[16] (m.p. 110–111°) was 101–108°.

Anal. Calcd. for $C_{14}H_{12}O_3$: C, 73.67; H, 5.30. Found: C, 73.37; H, 5.35.

3-*t*-Butyl-4-benzoyloxyphenol.—This was obtained similarly as the methyl analog from benzene; 88% yield, m.p. 148–149°.

Anal. Calcd. for $C_{17}H_{18}O_3$: C, 75.53; H, 6.71. Found: C, 75.42; H, 6.91.

2-*t*-Butyl-4-benzoyloxyphenol.—This was prepared by benzoylation of 10 g. of *t*-butylhydroquinone, using equimolar amounts of benzoyl chloride and sodium acetate by the Schotten–Baumann method; yield 48%, m.p. 112–113° after recrystallization from benzene.

Anal. Calcd. for $C_{17}H_{18}O_3$: C, 75.53; H, 6.71. Found: C, 75.54; H, 6.61.

A mixture melting point with the 3-*t*-butyl isomer was 102–109°.

2,6-Di-*t*-butylhydroquinone.—2,6-Di-*t*-butylphenol was nitrosated, in 68% yield, to give 2,6-di-*t*-butyl-4-nitrosophenol,[17] m.p. 211–212°. The latter was hydrogenated at 1.4 kg./cm.^{2} (20 p.s.i.) over Raney nickel, and 2,6-di-*t*-butyl-4-aminophenol was isolated as the bisulfate, m.p. 151.0–151.5°, in 98% yield. Oxidation of this in concentrated HCl at 55–60°, with ferric chloride, gave 81% of 2,6-di-*t*-butylbenzoquinone,[18] m.p. 68°. The latter was reduced to the hydroquinone, m.p. 99–101° (97% yield), using zinc dust and acetic acid.

4-Hydroxy-3,5-dinitrophenyl Esters.—*p*-Hydroxybenzoic acid, *p*-hydroxyphenylacetic acid, and N-acetyltyrosine were nitrated at −10 to −20° and esterified as follows. To 100 g. of the 3,5-dinitro-4-hydroxyphenyl acid, in 250 ml. of absolute ethanol, was cautiously added 40 ml. of acetyl chloride. The warm solution was refluxed 1 hr. (omitted in case of tyrosine derivative) and the volume was reduced to one-third by distillation *in vacuo*. The solution was chilled and the precipitate was collected. The product was washed with a little cold ethanol and dried. Yields of the compounds thus obtained were: ethyl 3,5-dinitro-4-hydroxybenzoate,[19] m.p. 84–85°, 70%; ethyl 3,5-dinitro-4-hydroxyphenylacetate,[10] m.p. 72–73°, 90%; and the ethyl ester of N-

(16) A. Jacob, F. K. Sutcliffe, and A. R. Todd, *J. Chem. Soc.*, 327 (1940).

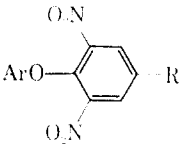
(17) S. J. Metro, *J. Am. Chem. Soc.*, **77**, 2901 (1955).

(18) W. R. Vaughan and G. K. Finch, *J. Org. Chem.*, **21**, 1201 (1956); A. F. Bickel and E. C. Kooyman, *J. Chem. Soc.*, 3211 (1953).

(19) H. Salkowski, *Ann.*, **163**, 44 (1872).

TABLE I

4-(BENZOYLOXYARYLOXY)-3,5-DINITROPHENYL ESTERS



Ar

A = 4-Benzoyloxyphenyl
B = 4-Benzoyloxy-3-methylphenyl
C = 4-Benzoyloxy-3-*t*-butylphenyl
D = 5-Benzoyloxy-1-naphthyl
E = 4-Hydroxy-3-*t*-butylphenyl
F = 4-Hydroxy-3,5-di-*t*-butylphenyl

R

G = COOEt
H = CH_2COOEt
I = $CH{=}C(COOEt)_2$
J = L-$CH_2CH(NHAc)COOEt$
K = CH_2CH_2COOEt
L = $CH_2CH(COOEt)_2$

Ar	R	Yield, %	M.p., °C.	Formula	Carbon, % Calcd.	Carbon, % Found	Hydrogen, % Calcd.	Hydrogen, % Found
A	G	50	158–160	$C_{22}H_{16}N_2O_9$	58.41	58.58	3.57	3.55
A	H	45	146–147	$C_{23}H_{18}N_2O_9$	[a]			
A	I	73	152–153	$C_{27}H_{22}N_2O_{11}$	58.91	58.55	4.03	4.08[b]
A	J	72	159–160	$C_{26}H_{23}N_3O_{10}$	[c]	[d]		
B	J	70	153–154	$C_{27}H_{25}N_3O_{10}$	58.80	58.70	4.57	4.76[e]
C	J	60	103–104	$C_{30}H_{31}N_3O_{10}$	[f]	[g]		
D	H	50	184–185	$C_{27}H_{20}N_2O_9$	62.80	62.73	3.90	4.08
D	I	78	147–148	$C_{31}H_{24}N_2O_{11}$	62.00	62.04	4.03	4.20
D	J	69	167–168	$C_{30}H_{25}N_3O_{10}$	61.32	61.72	4.29	4.29
E	I	23	141–142	$C_{24}H_{26}N_2O_{10}$	57.36	57.44	5.22	5.29
F	K	47	94–95	$C_{25}H_{32}N_2O_8$	[h]			

[a] *Anal.* Calcd.: N, 6.09. Found: N, 6.26. [b] *Anal.* Calcd.: N, 5.09. Found: N, 5.28. [c] *Cf.* ref. 20. [d] $[\alpha]^{30}D$ −33° (*c* 1, acetone); lit.[20] $[\alpha]^{20}D$ −12° (dioxane). Our sample had a similar low rotation in this solvent. [e] $[\alpha]^{27}D$ −3.34° (*c* 1, acetone). [f] *Anal.* Calcd.: N, 7.08. Found: N, 6.95. [g] $[\alpha]^{27}D$ −35.8° (*c* 1, acetone). [h] *Cf.* ref. 3.

acetyl-3,5-dinitro-L-tyrosine,[8b] m.p. 118–120°, 90%. Ethyl 3-(4-hydroxy-3,5-dinitrophenyl)propionate was prepared as previously described.[20]

Propionic Acid Analogs.—Ethyl *p*-hydroxybenzalmalonate[21] was prepared and nitrated in 73% yield. The side-chain double bond was hydrogenated simultaneously with the nitro groups.

Condensation of the benzoyloxyphenols with 3,5-dinitro-4-hydroxyphenyl acids were carried out in general by the method of Clayton, Green, and Hems.[20] The reaction mixtures were refluxed for 15–45 min. instead of 1 hr., and methanesulfonyl chloride was sometimes substituted for *p*-toluenesulfonyl chloride. In the preparation of Ic and Id, the appropriate esters were condensed with unprotected *t*-butylhydroquinone and 2,6-di-*t*-butylhydroquinone. The yields were low in these cases, because of formation of corresponding benzoquinones. Yields and data are in Table I.

The conversion of 4-(benzoyloxyaryloxy)-3,5-dinitro esters to diiodo esters was as follows. A suspension of 15 g. of the dinitro compound and 5 g. of 5% palladium on charcoal, in 150 ml. of acetic acid, was hydrogenated 3 hr. initially at 3.16 kg./cm.^{2} (45 p.s.i.). Raney nickel (15 g. in 15 ml. of acetic acid) was added cautiously and hydrogenation continued for 10 hr. A solution containing nitrosylsulfuric acid was prepared by adding 4.5 g. of finely powdered sodium nitrite to H_2SO_4 cooled to 0°. The temperature was raised slowly, with good stirring, to 45–50°, giving a clear solution. The solution was cooled to 5° and 84 ml. of acetic acid was added while the temperature was held below 10°. The filtered pale green diamine solution was added dropwise with stirring at 0–4°. The resulting orange solution was stirred 30 min. more and poured in a thin stream during 3–4 min. into a well-stirred mixture containing 25 g. of iodine, 39 g. of sodium iodide, 560 ml. of water, 5 g. of urea, and 165 ml. of chloroform. The mixture was stirred 16 hr., and the chloroform layer was separated and extracted with 2 *M* sodium bisulfate solution and water. Chloroform was removed by distillation *in vacuo* and 50 ml. of ethyl alcohol was added to the residue. The flask was scratched and let stand overnight. The crystals were collected and recrystallized from *ca.* 85% ethanol. The product was collected, washed with water, and dried; yields and other data are in Table II.

The blocked diiodo compounds were hydrolyzed, and in the cases of the malonic acid derivatives decarboxylated, in a HCl–acetic acid mixture. The samples were mixed with 5 parts of concentrated HCl and treated with enough acetic acid (2–5 parts) to dissolve hot. The solutions were refluxed 10–16 hr. and concentrated *in vacuo* to 0.5 vol., cooled, and diluted with concentrated HCl, if necessary, to precipitate the product. The alanine analogs separated as the hydrochlorides. When desired, these compounds were converted to the free amino acids by neutralizing with sodium acetate solution, in methanol. Data are in Table III.

***p*-Nitrophenyl *o*-Tolyl Ether.**—This was prepared by an adaptation of the method in "Organic Syntheses"[22] for *p*-nitrophenyl phenyl ether. A mixture of 32.4 g. (0.30 mole) of *o*-cresol and 15.2 g. (0.27 mole) of KOH was heated at 160° for 30 min. After cooling to 100–110°, the mixture was treated with 0.1 g. of activated copper metal[22] and 7.8 g. (0.05 mole) of *p*-chloronitrobenzene. The stirred mixture was refluxed at 138–145° for 10 min. (exothermic reaction) and cooled, 7.8 g. (0.05 mole) more of *p*-chloronitrobenzene was added, and the mixture finally was heated at 138–145° for 90 min. The resulting dark red mixture was cooled, poured onto an ice–aqueous NaOH mixture and extracted with chloroform. The chloroform layer was washed well with water and distilled. The desired ether distilled at 174–175° (5 mm.),[23] $n^{24}D$ 1.6053, yield 16 g. (70%).

4-(Chloromethyl)-2-methylphenyl *p*-Nitrophenyl Ether.—This was prepared by adapting the method of Southwick, Foltz, and McIntyre[24] for chloromethylation of *p*-nitrophenyl phenyl ether. A solution of 160 g. (0.70 mole) of *p*-nitrophenyl *o*-tolyl ether, 45 g. of paraformaldehyde, 82 ml. of sirupy phosphoric acid, 210 ml. of concentrated HCl, and 400 ml. of glacial acetic acid was steam heated for 48 hr., with stirring. More paraformaldehyde (30 g.) and 150 ml. of concentrated acid was added, and heating continued 18 hr. The orange-yellow mixture separated into two layers; the lower, oily layer was poured onto ice. The oil solidified to a granular product which was separated and washed well with water, then 5% Na_2CO_3 solution, and finally with water. After drying *in vacuo*, the pale yellow product

(20) J. C. Clayton, G. F. H. Green, and B. A. Hems, *J. Chem. Soc.*, 2473 (1951).

(21) R. I. Meltzer, D. M. Lustgarten, and A. Fischman, *J. Org. Chem.*, **22**, 1577 (1957).

(22) R. Q. Brewster and T. Groening, "Organic Syntheses," Coll. Vol. II, John Wiley and Sons, Inc., New York, N. Y., 1943, p. 445.

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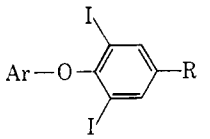
(24) P. L. Southwick, G. E. Foltz, and W. E. McIntyre, *ibid.*, **75**, 5877 (1953).

TABLE II

4-(BENZOYLOXYARYLOXY)-3,5-DIIODOPHENYL ESTERS



Compd.[a] Ar	R	Yield, %	M.p., °C.	Formula	Carbon, % Calcd.	Carbon, % Found	Hydrogen, % Calcd.	Hydrogen, % Found	Iodine, % Calcd.	Iodine, % Found
A	G	60	138–139	$C_{22}H_{16}I_2O_5$	43.02	42.10	2.63	2.71	41.33	41.44
A	H	50	102–103	$C_{23}H_{18}I_2O_5$	43.97	44.42	2.89	3.05	40.41	39.95
A	L	60	129–129	$C_{27}H_{24}I_2O_7$	45.40	45.23	3.38	3.50	35.54	35.47
A	J	77	147–148	$C_{26}H_{23}I_2NO_6$	*b*					
B	J	45	170–171	$C_{27}H_{25}I_2NO_6$	45.46	45.20	3.53	3.71		
C	J	55	159–160	$C_{30}H_{31}I_2NO_6$	47.70	47.42	4.14	3.80[c]		
D	H	62	183–184	$C_{27}H_{20}I_2O_5$	47.81	47.81	2.97	3.14	37.42	37.60
D	L	65	138–139	$C_{31}H_{26}I_2O_7$	48.71	48.88	3.43	3.36		
D	J	55	164–165	$C_{30}H_{25}I_2NO_6$	48.08	48.20	3.36	3.51	33.87	33.69
E	L	70	73–74	$C_{25}H_{28}I_2O_6$	43.26	43.19	4.24	4.66		
F	K	15	120–121	$C_{24}H_{32}I_2O_4$	*d*					

[a] See Table I for groups represented by letters for Ar and R. [b] Ref. 11. [c] $[\alpha]^{27}D$ 2°. [d] Ref. 3.

TABLE III

THYROXINE ANALOGS

Compd.	Yield, %[a]	M.p., °C.[b]	Formula	Carbon, % Calcd.	Carbon, % Found	Hydrogen, % Calcd.	Hydrogen, % Found
VIa	87	256–258[c]	$C_{13}H_8I_2O_4$				
VIb	94	215–216[d]	$C_{14}H_{10}I_2O_4$				
VIc	92	244–245[e]	$C_{15}H_{12}I_2O_4$				
VId	85	254–255[f]	$C_{15}H_{13}I_2NO_4$				
Ia	82	225–227[g]	$C_{16}H_{15}I_2NO_4$	35.64	35.74	2.81	3.04[h]
Ib·HCl	67	195–197	$C_{19}H_{22}ClI_2NO_4$	36.93	37.08	3.59	3.55[i]
Ic	79	127–129[j]	$C_{19}H_{20}I_2O_4$	40.30	40.62	3.57	3.90[k]
Id	57	197–198	$C_{23}H_{28}I_2O_4$	*l*			
IIa	80	215–217	$C_{18}H_{12}I_2O_4$	39.58	39.69	2.22	2.61
IIb	84	186–188	$C_{19}H_{14}I_2O_4$	40.74	40.74	2.52	2.94
IIc	73	214–217[m]	$C_{19}H_{15}I_2NO_4$	39.67	39.61	2.63	2.97

[a] Yield on the removal of the blocking groups. [b] Samples melted with decomposition. [c] C. R. Harington and G. Barger [*Biochem. J.*, **21**, 169 (1927)] report m.p. 252–254°. [d] C. R. Harington and R. Pitt-Rivers [*ibid.*, **50**, 438 (1952)] report m.p. 214–216.5°. [e] Lit.[20] m.p. 250°. [f] Lit.[10b] m.p. 255°. [g] 3,5-Diiodo-3′-methylthyronine hydrochloride melts at 248° dec. [h] *Anal.* Calcd.: I, 47.08. Found: I, 47.70. [i] *Anal.* Calcd.: N, 2.27. Found: N, 2.09. [j] Recrystallized from dilute acetic acid. [k] *Anal.* Calcd.: I, 44.83. Found: I, 45.45. [l] *Cf.* ref. 3. [m] 4-(5-Hydroxy-1-naphthyloxy)-3,5-diiodo-L-phenylalanine hydrochloride melts at 235° dec.

melted at 44–45° (189 g., 98% yield). It was converted to the thiuronium salt by heating with an equimolar amount of thiourea, in alcohol, for 3 hr.; m.p. 215–216°, yield 95%.

Anal. Calcd. for $C_{15}H_{16}ClN_3O_3S$: C, 50.91; H, 4.56; S, 9.06. Found: C, 50.92; H, 4.90; S, 9.00.

Diethyl 4-(4-Nitrophenoxy)-3-methylbenzylacetamidomalonate.—To 3.22 g. (0.14 g.-atom) of sodium in 330 ml. of absolute ethanol, protected from atmospheric moisture, was added 30.45 g. (0.14 mole) of diethyl acetamidomalonate. The mixture was refluxed 90 min., 36.2 g. (0.13 mole) of 4-(chloromethyl)-2-methylphenyl *p*-nitrophenyl ether was added, the mixture was refrigerated, and the product was collected and washed well with water, yielding pale yellow needles (52 g., 85%), m.p. 133–134°.

Anal. Calcd. for $C_{23}H_{26}N_2O_8$: C, 60.25; H, 5.72; N, 6.11. Found: C, 60.35; H, 5.56; N, 6.30.

3-Methyl-DL-thyronine.—A solution of 50 g. (0.109 mole) of ethyl 4-(4-nitrophenoxy)-3-methylbenzylacetamidomalonate in 50 ml. of absolute methanol was hydrogenated over 1.5 g. of 5% palladium on charcoal for 2 hr. at 2.11 kg./cm.^{2} (30 p.s.i.). After removing the catalyst, the filtrate was treated with dry HCl. Ethyl 4-(4-aminophenoxy)-3-methylbenzylacetamidomalonate hydrochloride separated as a paste. This was collected, washed with ether, and dried *in vacuo;* yield 45 g. (88%), m.p. 124–125°. The hydrochloride (4 g.) was dissolved in 160 ml. of warm 50% H_2SO_4 solution and cooled below 10°. With continuous stirring, an aqueous solution containing 0.8 g. of sodium nitrite in 8 ml. of water was added. Then 0.5 g. of urea was added and the mixture was refrigerated for 3 hr.

To a refluxing solution of 224 ml. of concentrated H_2SO_4 in 300 ml. of water, the diazonium salt solution was added dropwise with vigorous stirring during 45 min. Water (150 ml.) was added to the resulting golden yellow solution, which was then refluxed for 4.5 hr. The material was allowed to cool, concentrated to 0.5 vol. by distillation *in vacuo*, cooled by adding ice, and neutralized (pH 5–6) with concentrated NH_4OH. After refrigeration overnight, the tan flaky product was collected, 1.91 g. (83% yield), m.p. 230–232° dec. The material was purified by dissolving in dilute H_2SO_4 and precipitation with NH_4OH, m.p. 249–250°. This was converted to the hydrochloride (hydrate) by adding concentrated HCl to the acetic acid solution.

Anal. Calcd. for $C_{16}H_{17}NO_4 \cdot HCl \cdot H_2O$: C, 56.22; H, 5.90. Found: C, 55.92; H, 5.91.

3-Methyl-3′,5′-diiodo-DL-thyronine.—To 0.30 g. (0.00104 mole) of 3-methyl-DL-thyronine in 10 ml. of 40% aqueous ethylamine was added dropwise 2.1 ml. of a solution of 4.82 g. (0.038 g.-atom) of iodine in 10 ml. of saturated KI solution. The resulting solution was shaken mechanically for 45 min., cooled, and neutralized with 15% HCl to pH 4–5. The mixture was refrigerated 2 days, the pale tan product was collected, washed well with distilled water, and dried; yield 0.23 g. (43%). To purify, the sample was dissolved in methanol containing 2 drops of concentrated HCl, charcoaled, and poured into a solution of 1.7 g. of sodium acetate in 40 ml. of water. The mixture was distilled *in vacuo* until a flocculent precipitate appeared. The product was separated as before; m.p. 202–203° dec.

Anal. Calcd. for $C_{16}H_{15}I_2NO_4$: C, 35.64; H, 2.81; I, 47.08; N, 2.60. Found: C, 36.06; H, 3 04; I, 46.94; N, 2.15.

5-[4-(4-Nitrophenoxy)-3-methylbenzal]hydantoin.—4-(Chloromethyl)-2-methylphenyl *p*-nitrophenyl ether was converted to the hexaminium salt (m.p. 205–206° dec.).[25] The salt was

(25) S. J. Angyal, *Org. Reactions*, **8**, 205 (1954).

decomposed to the aldehyde (m.p. 94–95°) in 50% acetic acid. A mixture of 0.7 g. of the aldehyde, 0.7 g. of fused sodium acetate, 0.27 g. of hydantoin, 5 ml. of acetic acid, and 3 drops of acetic anhydride was refluxed 2 hr. The mixture was charcoaled, cooled, treated with 2.0 ml. of water, and refrigerated. The precipitated solid was separated washed with water, and oven-dried. The straw yellow product (0.83 g., 90% yield) was recrystallized from absolute ethanol, m.p. 264–265°.

Anal. Calcd. for $C_{17}H_{13}N_3O_5$: C, 60.17; H, 3.86; N, 12.39. Found: C, 59.80; H, 4.08; N, 12.15.

Paper Chromatography.—The thyronines were chromatographed in *t*-amyl alcohol saturated with 2 *N* NH_4OH.[26] All samples except III gave one spot. R_f values observed for the substituted thyronines were: 3′-methyl-3,5-diiodo-, 0.63; 3′-*t*-butyl-3′,5′-diiodo-, 0.77; 3-methyl-, 0.55; 3-methyl-3′,5′-diiodo-, 0.35 (compared to 3,5-diiodothyronine, 0.58[25]). A second minor spot in the sample of III was obtained at R_f 0.48. This may be due to the presence of a small amount of 3-methyl-3′-iodothyronine. The naphthalene derivatives were similarly chromatographed and spotted with ninhydrin or 1-nitroso-2-naphthol. R_f values obtained were: 4-(5-hydroxy-1-naphthyloxy)-3,5-diiodo-L-phenylalanine, 0.52; the acetic acid analog, 0.62; and the propionic acid analog, 0.56.

(26) G. I. Gleason, *J. Biol. Chem.*, **213**, 837 (1955).

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Hypocholesterolemic Agents. Thyroalkanols

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A series of thyroalkanols was prepared and tested for hypocholesterolemic activity. The thyroalkanols, prepared by diborane reduction of the corresponding thyroalkanoic acids, showed comparable potency to the thyroalkanoic acids and less toxicity as exemplified by their effect on weight gain of treated and control rats.

The importance of thyroxine as a "metabolic regulator," and in particular the role of thyroxine in cholesterol metabolism, stimulated our interest in the chemical modification of the thyroxine side chain with the objective of effecting a split between hypocholesterolemic activity and calorigenic activity. The interesting hypocholesterolemic activity recently reported[1] for thyroalkanoic acids and the possibility that changes in the polarity of the side chain might be of importance in the absorption, distribution, metabolism, and thus the over-all activity of a thyroxine analog prompted the preparation and evaluation of a series of thyroalkanols as hypocholesterolemic agents (Table I).

The synthesis of triiodothyroethanol has been reported by Tomita and Lardy[2] who coupled an appropriately substituted phenylethanol derivative with *p*-methoxyphenol to afford a diphenyl ether bearing an ethanol side chain. Subsequent reactions yielded triiodothyroethanol. The general method of synthesis of thyroalkanols developed in this work depends upon the diborane reduction of the corresponding thyroalkanoic acids. It is of interest that diborane reduction was selective and did not adversely affect the iodinated diphenyl ether intermediates usually attacked by many other reducing agents.[2,3]

The synthesis of **8**, the thyroalkanoic acid precursor of the thyroethanol **1**, was accomplished by the Borrows[4] modification of the general method of Ullmann and Nadai.[5] Methyl 3,5-dinitro-4-hydroxyphenylacetate (**25**) was condensed with 3,5-dimethyl-4-methoxyphenol (**24**) in the presence of *p*-toluenesulfonyl chloride to afford the diphenyl ether **26** (Scheme I). Reduction, diazotization, and iodination yielded **7** which was treated with hydrogen iodide to yield **8**. The same type of synthesis in the 4′-deoxy series yielded the thyroalkanoic acid analog **10**.

Biological Methods.—Hypercholesterolemia was induced in male Sprague–Dawley rats (fasted weight about 220 g.) by using a high cholesterol diet[6] containing 10% coconut oil as fat source and 18% casein supplemented by 0.2% methionine as a protein source. Test compounds suspended in 0.25% methylcellulose at concentrations adjusted to 1 ml. of vehicle/100 g. of body weight were administered orally to groups of 10 rats. Control groups received vehicle only. The rats, weighed three times per week, were fed *ad libitum* until 17 hr. before the end of the experiment (14 days). Blood was drawn from the aorta after treatment with cyclopal.[7] Food consumption and weights were recorded at the end of each experiment.

Ferric chloride–sulfuric acid reagent was used for the determination of total sterols according to Zak, *et al.*,[8] and samples were analyzed by means of an Auto-Analyzer.[9]

Discussion

A summary of the hypocholesterolemic activity of a group of thyroalkanols and thyroalkanoic acids is

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(9) Technicon Controls Inc., Chauncey, N. Y.