n-octyl alcohol.[6] Solutions were prepared by dispersing the required amount of amylose in 1 *N* KOH to yield a concentration of 0.267% following neutralization with 0.5 *N* HCl. Aliquots of the neutralized solutions were diluted to a final concentration of 0.002%. To those solutions which were heated a layer of mineral oil was frequently added to help prevent loss of iodine. Excess iodine was frequently added during the experiments, but with no effect on the wave length of maximum absorption. Spectra were determined in 19 × 105 mm. round cuvettes with the aid of a Model 14 Coleman Universal spectrophotometer.

(6) S. Lansky, M. Kooi and T. J. Schoch, *ibid.*, **71**, 4066 (1949).

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The Chemotherapy of Tuberculosis. III. Thiosemicarbazide Derivatives

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In the course of a program[1,2] on the synthesis of compounds for the chemotherapy of tuberculosis, a number of hitherto unreported thiosemicarbazones, acyl thiosemicarbazides, thiadiazole derivatives, and related classes that have already been reported on by other workers[3-6] have been made.

All of the compounds prepared were inactive in experimental mouse tuberculosis.

Acknowledgment.—We are indebted to Dr. Al Steyermark and his associates for the microanalyses and Drs. R. J. Schnitzer and E. Grunberg for the chemotherapeutic screening.

Experimental

1-(3-Pyridyl)-thiosemicarbazide.—3-Pyridylhydrazine (15 g., 0.137 mole) was refluxed for 6 hours with 12.7 g. of ammonium thiocyanate and 18 ml. of 9 *N* HCl in ethanol in 350 ml. of ethanol. The separated crude product was recovered by filtration and crystallized from aqueous ammonia solution; yield 5 g. (21%), m.p. 223–224°.

Anal. Calcd. for $C_6H_8N_4S$: C, 42.8; H, 4.8. Found: C, 43.0; H, 4.6.

2,3-Dihydro-5-(2′-acetoxy-4′-nitrophenyl)-2-phenylamino-1,3,4-thiadiazole.—Acetyl *p*-nitrosalicylic acid (67.5 g., 0.3 mole) was converted to the acid chloride by 62.4 g. of PCl_5. The acid chloride obtained was treated with 50.1 g. (0.3 mole) of 4-phenyl-3-thiosemicarbazide in nitromethane as a solvent. The reaction solution was heated for 4 hours on the steam-bath. On cooling, 30 g. of the product separated, and further heating and cooling gave 4.3 g. more of the crude product. The combined crude fractions (34.3 g.) were recrystallized from nitrobenzene; yield 23.3 g. (20%), m.p. 316–317°.

Anal. Calcd. for $C_{16}H_{12}O_4N_4S$: C, 53.9; H, 3.4. Found: C, 54.0; H, 3.2.

2,3-Dihydro-5-(2′-hydroxy-4′-aminophenyl)-2-phenylamino-1,3,4-thiadiazole.—2,3-Dihydro-5-(2′-acetoxy-4′-nitrophenyl)-2-phenylamino-1,3,4-thiadiazole (13 g., 0.036 mole) was added in portions to 100 ml. of phenylhydrazine and 150 ml. of anisole. The reduction was heated at reflux for 2 hours and on cooling crystallized a buff-colored product. The recovered product was recrystallized twice from anisole; yield 9.6 g. (92%), m.p. 282–283°.

Anal. Calcd. for $C_{14}H_{12}ON_4S$: C, 59.2; H, 4.2. Found: C, 59.5; H, 4.3.

1-(*p*-Acetamidobenzoyl)-thiosemicarbazide.—*p*-Acetamidobenzoyl chloride (9.5 g., 0.048 mole) was added to 20 ml. of 10% sodium hydroxide solution containing 4.4 g. (0.048 mole) of thiosemicarbazide. The reaction solution became warm and was cooled and stirred for 2 hours. The product was recovered by filtration and washed with water; crude yield 9.1 g. (75%), m.p. 192–194° (dec.). A small sample was recrystallized by solution in ethylene glycol and addition of water to turbidity, m.p. 221° (dec.) of the colorless crystals.

TABLE I

Thiosemicarbazones	Formula	Crystallized from	Color	Yield, %	M.p., °C.	Analyses, % Calcd. Nitrogen	Calcd. Sulfur	Found Nitrogen	Found Sulfur
Streptomycin trihydrochloride[a]	$C_{22}H_{42}O_{11}N_{10}S\cdot 3HCl$	Aq. HCl + EtOH	Colorless	10	205		4.2		4.4
Anisil mono	$C_{17}H_{17}O_3N_3S$	Acetone	Orange	67	227–228		9.4		9.4
Naphthazarin mono	$C_{11}H_9O_3N_3S$	EtOH	Purple-black	27	160 dec.	16.0		15.7	
Acenaphthenequinone mono	$C_{13}H_9ON_3S\cdot H_2O$	Aq. EtOH	Yellow	50	223–224	15.4[b]		15.5	
Acenaphthenone	$C_{13}H_{11}N_3S$	EtOH	Yellow	88	227–228	17.4		17.1	
2-Methyl-3-hydroxy-5-hydroxymethylisonicotinaldehyde	$C_9H_{12}O_2N_4S\cdot{}^1/_2H_2SO_4$	Water	Yellow	63	194 dec.	9.7[c]	16.6	10.1[c]	16.5
2-Pyrryl methyl ketone	$C_7H_{10}N_4S$	EtOH	Colorless	74	184–185		17.5		16.9
D(+)-Mannose[d]	$C_7H_{15}O_5N_3S\cdot C_2H_5OH$	EtOH	Colorless	>90	175–176 dec.		10.5		10.4[e]
L(−)-Mannose[e]	$C_7H_{15}O_5N_3S$	Water	Colorless	>90	173–174 dec.	[f]			
D(+)-Glucose	$C_7H_{15}O_5N_3S$	80% EtOH	Colorless	>90	190[g] dec.		12.7		12.7
D(−)-Arabinose	$C_6H_{13}O_4N_3S$	[h]	Colorless	>90	151–152		14.4		13.9
L(+)-Arabinose	$C_6H_{13}O_4N_3S$	[h]	Colorless	>90	151–152		14.4		14.3
Nicotinaldehyde S-methyl[i]	$C_8H_{10}N_4S$	Aq. EtOH	Yellow	89	110–111		16.5		16.3
4-Acetamidobenzaldehyde-4′-phenyl-	$C_{16}H_{16}ON_4S$	HOAc	Yellow	94	200–201	[j]			

[a] Reported in solution without being isolated, R. Donovick, G. Rake and J. Fried, *J. Biol. Chem.*, **164**, 173 (1946). [b] Calcd.: C, 57.2; H, 4.0. Found: C, 56.5; H, 3.4. [c] Two nitrogens by Kjeldahl. [d] % volatile calcd. 18.2; Found: 18.5. [e] The L(−)-mannose thiosemicarbazone also formed the alcoholate from ethanol similar to the D(+)-isomer and was not analyzed because it had the same melting point as the D(+)-isomer and did not exhibit a mixed m.p. depression with the D-isomer. [f] Calcd.: C, 33.2; H, 5.9. Found: C, 33.1; H, 5.9. [g] Reported m.p. 204°; Neuberg and W. Neiman, *Ber.*, **35**, 2049 (1902). [h] Triturated with hot methanol instead of being recrystallized. [i] Method of E. Hoggarth *J. Chem. Soc.*, 1579 (1950). [j] Calcd.: C, 61.5, H, 5.1. Found: C, 61.3; H, 5.1.

The compounds in Table I were prepared by the reaction of the parent substance with thiosemicarbazide in aqueous ethanol or aqueous ethanol–acetic acid in cases wherein the parent substance was not soluble in aqueous ethanol.

(1) T. S. Gardner, F. A. Smith, E. Wenis and J. Lee, *J. Org. Chem.*, **16**, 1121 (1951).

(2) T. S. Gardner, E. Wenis and F. A. Smith, THIS JOURNAL, **73**, 5455 (1951).

(3) R. Behnisch, F. Mietzsch and H. Schmidt, *Angew. Chem.*, **60**, 113 (1948).

(4) E. Hoggarth, A. R. Martin, N. E. Storey and E. H. P. Young, *Brit. J. Pharmacol.*, **4**, 248 (1949).

(5) B. Croshaw and L. Dickinson, *ibid.*, **5**, 178 (1950).

(6) J. Bernstein, H. L. Yale, K. Losee, M. Holsing, J. Martins and W. A. Lott, THIS JOURNAL, **73**, 906 (1951).

Anal. Calcd. for $C_{10}H_{12}O_2N_4S$: N, 11.1 (2 N by Kjeldahl). Found: N, 10.7.

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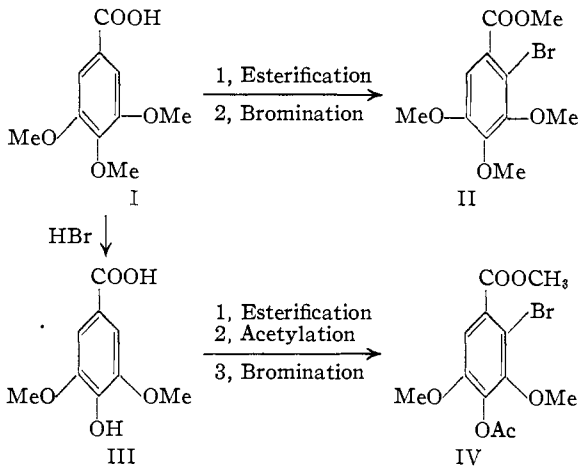
Polymethoxybromobenzenes[1]

BY E. C. HORNING[2] AND JOHN A. PARKER[3]

Investigations of the structure and physiological activity of compounds related to colchicine, which have been underway for some time, have made it necessary to develop a satisfactory means of obtaining monobromo derivatives of a variety of polymethoxybenzenes. The usual methods of direct bromination in inert solvents generally give small amounts of the desired products, but demethylation and polybromination occur as well, and as a result indirect or special methods have usually been employed to provide better yields of the compounds. A method which we have found satisfactory involves a ferric chloride-catalyzed bromination in acetic anhydride, with pyridinium bromide perbromide as the source of bromine. Although no new reagent or new solvent is employed, we have found no record of previous use of this method.



The bromination of methyl 3,4,5-trimethoxybenzoate by this procedure proceeds smoothly and gives good yields (85%) of product (II). This bromination has been reported several times, but with divergent results. The method reported here leads to a low-melting (34–36°) monobromo ester, correctly described by Feist and Dschu,[4] although if the reaction mixture is heated an unidentified product, m.p. 88–89°, perhaps identical with the material prepared in another way and also described as methyl 2-bromo-3,4,5-trimethoxybenzoate,[5] may also be obtained. A related ester, methyl 2-bromo-3,5-dimethoxy-4-acetoxybenzoate, was prepared by the same bromination method. The generality of the procedure was examined through the bromination of veratrole and 1,2,3-trimethoxybenzene, but has not been extended beyond these examples.

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(2) National Heart Institute, Bethesda, Maryland.

(3) National Institutes of Health Predoctoral Fellow, 1950–1951.

(4) K. Feist and G. Dschu, "Festchrift A. Tschirch," Leipzig, 1926, p. 29; *Chem. Zentr.*, **98**, II, 58 (1927).

(5) M. T. Bogert and E. Plaut, THIS JOURNAL, **37**, 2723 (1915).

Experimental

Methyl 3,4,5-Trimethoxybenzoate.—A 100-g. quantity of 3,4,5-trimethoxybenzoic acid was suspended in 500 ml. of dry methanol, and hydrogen chloride was bubbled rapidly through the mixture without temperature control until the methanol was saturated and the acid dissolved. The mixture was chilled (24 hours) and the crystalline product was removed by filtration, washed with chilled methanol, and dried to yield 101.5 g. (94%) of the ester, m.p. 83–84° (reported[6] m.p. 83–84°).

Methyl 2-Bromo-3,4,5-trimethoxybenzoate (II).—A few crystals (approximately 200 mg.) of ferric chloride (hydrated) were dissolved in a solution of 22.6 g. (0.10 mole) of methyl 3,4,5-trimethoxybenzoate in 100 ml. of acetic anhydride. A total of 35.5 g. of pyridinium bromide perbromide was added in small portions, with stirring or shaking,[7] and the resulting solution was heated at 50–55° for five minutes to ensure completeness of reaction. The mixture was added slowly with good stirring to 500 ml. of hot (60–70°) water in order to hydrolyze the solvent and, after cooling, the mixture was extracted with 1:1 ether–ethyl acetate. The combined organic extracts were washed with 5% hydrochloric acid, 5% sodium hydroxide solution, water and 2% aqueous acetic acid. The organic material was dried (magnesium sulfate), the solvents were removed, and the crude yield of 32.5 g. of product was distilled under reduced pressure to provide 2.0 g. of fore-run (b.p. to 160° (2 mm.)) and 28.5 g. of product, b.p. 160–161° (2 mm.). The colorless ester was recrystallized from ether–pentane to give 25.8 g. (85%) of methyl 2-bromo-3,4,5-trimethoxybenzoate, m.p. 34–36°. The correct structure was assigned to this compound by Feist and Dschu,[4] who reported m.p. 33°.

When the reaction mixture was heated at 90–100° for periods of 30 minutes or longer, the reaction product was a colorless crystalline material, m.p. 89–90°, of unidentified structure, which was perhaps similar to the products usually obtained by bromination of the ester with bromine in acetic anhydride at room temperature.[5]

3,5-Dimethoxy-4-hydroxybenzoic Acid (Syringic Acid).—A mixture of 50.0 g. of 3,4,5-trimethoxybenzoic acid, 200 ml. of acetic acid and 40 ml. of concentrated hydrobromic acid was heated under reflux for three hours. After dilution with 800 ml. of water, followed by chilling (24 hours), a 38.6 g. (83%) yield of syringic acid, m.p. 202–203° (reported m.p. 204°), was obtained.

Methyl 3,5-Dimethoxy-4-acetoxybenzoate.—A solution of 30.0 g. (0.15 mole) of 3,5-dimethoxy-4-hydroxybenzoic acid in 100 ml. of dry methanol was saturated with hydrogen chloride. After standing for 24 hours, the solvent was evaporated, leaving a residue of 31.2 g. of crude ester. A 30.0 g. quantity of this material was dissolved in 120 ml. of acetic anhydride, and the mixture was heated under reflux for 30 minutes. The solution was added to 500 ml. of warm (50–60°) water, and the crystalline product was separated after chilling. The yield was 24.5 g. (64%) of colorless ester, m.p. 128–129°. This material has been prepared previously.[5]

Anal. Calcd. for $C_{12}H_{14}O_6$: C, 56.69; H, 5.55. Found: C, 56.62; H, 5.46.

Methyl 2-Bromo-3,5-dimethoxy-4-acetoxybenzoate (IV).—The bromination procedure described previously was carried out with 20.0 g. (0.074 mole) of methyl 3,5-dimethoxy-4-acetoxybenzoate and 28.0 g. of pyridinium bromide perbromide in 125 ml. of acetic anhydride. The crude product was crystallized from ether–pentane to yield 16.0 g. (62%) of colorless ester, m.p. 75–76°.

Anal. Calcd. for $C_{12}H_{13}O_6B$: C, 43.26; H, 3.93. Found: C, 43.53; H, 4.19.

1,2,3-Trimethoxy-4-bromobenzene.—To a stirred solution of 67.2 g. (0.40 mole) of 1,2,3-trimethoxybenzene in 400 ml. of acetic anhydride, containing a few crystals of ferric chloride, was added, in small portions, 144 g. of pyridinium bromide perbromide. The solution was heated for

(6) F. W. Semmler, *Ber.*, **41**, 1774 (1908).

(7) It is advisable to follow the precautions mentioned by Bogert,[5] although we have noted no difficulties.