Synthesis of Metabolic Products of Benzoxazoles[1]

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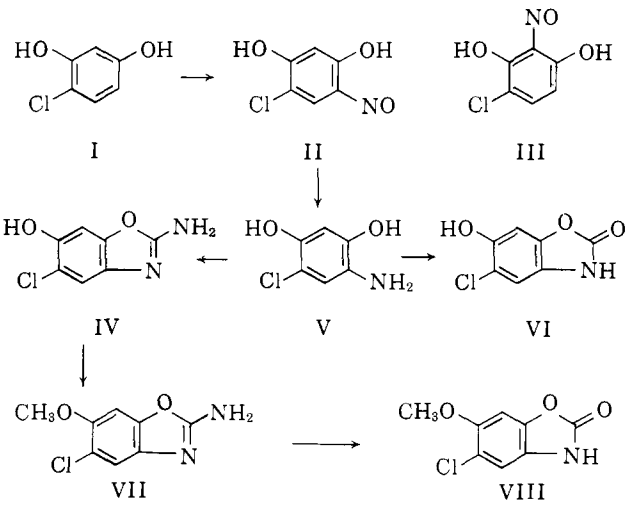
The synthesis is reported of 2-amino-5-chloro-6-hydroxybenzoxazole and 5-chloro-6-hydroxy-2-benzoxazolinone, metabolic products of 2-amino-5-chlorobenzoxazole and 5-chloro-2-benzoxazolinone, respectively. These metabolites, as well as the corresponding 6-methoxy derivatives, have little, if any, of the skeletal muscle relaxant activity of the parent compounds.

Conney, Trousof, and Burns[2] reported that 2-amino-5-chlorobenzoxazole[3] and 5-chloro-2-benzoxazolinone[4] are metabolized in man primarily to hydroxylated compounds in which the ring system remains intact. Earlier, Gressly and Nencki[5] reported that 2-benzoxazolinone is metabolized in the dog to a hydroxylated compound, Bray, Clowes, and Thorpe[6] stated that 2-benzoxazolinone is metabolized in the rabbit to 6-hydroxy-2-benzoxazolinone, and assigned the position of the hydroxyl group on the basis of qualitative tests on the sulfuric acid hydrolysis product of the metabolite.



In view of the fact that electrophilic substitution reactions take place in the 6-position of 2-aminobenzoxazoles and 2-benzoxazolinones,[7-9] it seemed most likely that biological hydroxylation does, in fact, occur in the 6-position. Accordingly, the 6-hydroxy derivatives of 2-amino-5-chlorobenzoxazole and 5-chloro-2-benzoxazolinone were prepared for comparison with the metabolites isolated by Conney, Trousof, and Burns.

Compounds IV and VI were prepared by treating 4-amino-6-chlororesorcinol (V) with cyanogen bromide and with phosgene, respectively. Compound V was obtained by catalytic hydrogenation of 4-chloro-6-nitrosoresorcinol (II), which resulted from treatment of 4-chlororesorcinol (I) with butyl nitrite in ethanolic sodium ethoxide. Fabré[10] reported the isolation of a nitrosochlororesorcinol from treatment of 4-chlororesorcinol with amyl nitrite and assigned structure III to the product, although he presented no evidence to support his conclusion.

The expected product of a substitution reaction of a resorcinol is the 4-derivative. Moreover, Henrich[11] showed that nitrosation of resorcinol gives 4-nitrosoresorcinol, and McLamore[12] established that the nitrosation product of 4-cyclohexylresorcinol is 4-cyclohexyl-6-nitrosoresorcinol.

The nitrosochlororesorcinol which we obtained was shown to have structure II by examination of its nuclear magnetic resonance spectrum in dimethyl sulfoxide and in dioxane. The spectrum showed two *single* peaks of equal intensity at 2.25 and 4.11 τ in dimethyl sulfoxide and at 1.70 and 3.67 τ in dioxane consistent with structure II. These peaks were assigned to the aromatic protons on the basis of failure to shift when acid was added. In alternative structure III, the peaks of the *ortho* hydrogen atoms should show considerable splitting.

The amino compound V was not isolated due to its extreme instability to light and air even in acid solution. The reaction with cyanogen bromide to give IV and with phosgene to give VI was carried out under nitrogen, but even under these conditions, considerable color developed and the best yields were about 50 to 60%.

Comparison of compound IV with the isolated metabolic product of 2-amino-5-chlorobenzoxazole[2] by melting point, mixture melting point, and ultraviolet and infrared spectra showed that they were identical. Similarly, compound VI was shown to be identical with the isolated metabolic product of 5-chloro-2-benzoxazolinone.

Methoxy compound VII was prepared by methylation of IV with dimethyl sulfate and sodium hydroxide. Hydrolysis of VII by refluxing a solution in dilute hydrochloric acid gave VIII.

Behavioral testing in mice by the method of Goodsell, *et al.*,[13] of compounds IV, VI, VII, and VIII administered orally showed very little, if any, of the skeletal muscle relaxant activity of the parent compounds.

(1) Presented at the Fourth Delaware Valley Regional Meeting of the American Chemical Society, Philadelphia, Penna., January 25, 1962.

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(4) Chlorzoxazone, Paraflex®.

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Experimental[14]

4-Chloro-6-nitrosoresorcinol.—4-Chlororesorcinol (14.4 g., 0.1 mole) was dissolved in a solution of sodium ethoxide prepared from 2.3 g. (0.1 g. atom) of sodium and 100 ml. of anhydrous ethanol. The resulting solution was cooled to 0° and treated with a solution of 10.3 g. (0.1 mole) of butyl nitrite in 10 ml. of anhydrous ethanol. The dark reaction mixture was stirred for 3 hr. at 0° then poured into 300 ml. of water. Neutralization of the dark solution with hydrochloric acid gave a yellow precipitate which was recrystallized from aqueous ethanol to give 12 g. (69%) of yellow crystals which slowly decomposed without melting upon heating; $\lambda_{max}^{CH_3OH}$ 308 mμ (ε 18,000); λ_{max}^{Nujol} 2.85, 2.98, 3.16, 6.20, 6.41 μ.

Anal. Calcd. for $C_6H_4ClNO_3 \cdot H_2O$: C, 37.7; H, 3.2; N, 7.3; H_2O, 9.4. Found: C, 37.8; H, 3.3; N, 7.0; H_2O, 9.2.

2-Amino-5-chloro-6-hydroxybenzoxazole.—A solution of cyanogen bromide in methanol was prepared by slowly adding 17.6 g. (0.11 mole) of bromine to 5.4 g. (0.11 mole) of sodium cyanide in 200 ml. of methanol. The air in the flask was displaced with nitrogen and a solution of 4-amino-6-chlororesorcinol [prepared by shaking at room temperature a solution of 17.3 g. (0.1 mole) of 4-chloro-6-nitrosoresorcinol in 200 ml. of methanol with hydrogen under a pressure of 2.8 kg./cm.^{2} and 2 g. of 10% palladium-charcoal catalyst until 0.2 mole of hydrogen was absorbed] was added rapidly with stirring. The reaction mixture was heated quickly to reflux and allowed to cool to room temperature. The solution was neutralized with sodium bicarbonate solution, and most of the methanol was removed by distillation under reduced pressure. The black precipitate was purified by several recrystallizations from methanol after treatment with Norit A to give 6.2 g. (33.7% yield) of colorless crystals, m.p. 215–217° dec.; $\lambda_{max}^{CH_3OH}$ 242 and 302 mμ (ε 11,000 and 8,1000); $\lambda_{max}^{0.1\ N\ HCl}$ 235 and 292 mμ (ε 9,500 and 8,600); $\lambda_{max}^{0.1\ N\ NaOH}$ 315 and 323 mμ (ε 19,500 and 19,600); λ_{max}^{Nujol} 2.98, 3.01, 5.99, 6.11, 6.37, 6.71 μ.

Anal. Calcd. for $C_7H_5ClN_2O_2$: C, 45.5; H, 2.7; Cl, 19.2; N, 15.2. Found: C, 45.7; H, 2.6; Cl, 19.2; N, 15.1.

5-Chloro-6-hydroxy-2-benzoxazolinone (VI). 1. From 4-Amino-6-chlororesorcinol.—A solution of 4-amino-6-chlororesorcinol was prepared as described above, using ethyl acetate in place of methanol. This solution was transferred under nitrogen to a flask containing 18 g. (0.22 mole) of sodium acetate in 100 ml. of ethyl acetate. The mixture was stirred and treated rapidly with a solution of 9.8 g. (0.10 mole) of phosgene in 20 ml. of ethyl acetate. The reaction mixture was heated to reflux and then allowed to cool to room temperature. Water was added, the organic layer was separated, and the aqueous layer was extracted with ether. The combined organic solution was washed with dilute sodium bicarbonate solution, then with dilute hydrochloric acid and finally with water. The solution was dried over anhydrous magnesium sulfate and evaporated under reduced pressure to give 15 g. of crude red product which was purified by several recrystallizations from methanol after treatment with Darco to give colorless crystals (6.0 g., 32.3%), m.p. 245–247° $\lambda_{max}^{CH_3OH}$ 232 and 300 mμ (ε 5,700 and 7,300); $\lambda_{max}^{0.1\ N\ HCl}$ 229 sh and 293 mμ (ε 5,100 and 6,000); $\lambda_{max}^{0.1\ N\ NaOH}$ 238 and 321 mμ (ε 11,100 and 7,200); λ_{max}^{Nujol} 3.06, 5.59, 5.68, 6.07, 6.67, 6.78 μ.

Anal. Calcd. for $C_7H_4ClNO_3$: C, 45.4; H, 2.2; N, 7.5. Found: C, 45.1; H, 2.2; N, 7.3.

2. From 2-Amino-5-chloro-6-hydroxybenzoxazole.—A solution of 1.4 g. of 2-amino-5-chloro-6-hydroxybenzoxazole (IV) in 50 ml. of 2 *N* hydrochloric acid was refluxed for 5 hr. The solid was collected by filtration, washed with water and purified by recrystallization from a mixture of acetone and benzene to give 0.6 g. of product which was shown to be identical with the material obtained from the phosgene method by melting point, mixture melting point and ultraviolet and infrared spectra.

2-Amino-5-chloro-6-methoxybenzoxazole (VII).—A suspension of 5.0 g. (0.027 mole) of 2-amino-5-chloro-6-hydroxybenzoxazole (IV) in 25 ml. of water was cooled to 0–5° and treated with a solution of 1.2 g. (0.03 mole) of sodium hydroxide in 25 ml. of water to give a dark blue solution to which was added slowly 4.0 g. (0.03 mole) of dimethyl sulfate. The ice bath was removed and stirring was continued until the mixture was neutral to litmus. The precipitate was collected, washed with water and purified by crystallization from a mixture of acetone and benzene to give 3.0 g. (56.0% yield) of colorless crystals which decomposed from 195–215°; $\lambda_{max}^{CH_3OH}$ 245 and 302 mμ (ε 11,900 and 8,100); λ_{max}^{Nujol} 2.89, 5.87, 6.36, 6.79 μ.

Anal. Calcd. for $C_8H_7ClN_2O_2$: C, 48.4; H, 3.6; N, 14.1. Found: C, 48.2; H, 3.6; N, 14.1.

5-Chloro-6-methoxy-2-benzoxazolinone (VIII).—A solution of 1.0 g. of 2-amino-5-chloro-6-methoxybenzoxazole (VII) in 25 ml. of 2 *N* hydrochloric acid was refluxed for 4 hr. The precipitate was collected and purified by three recrystallizations from methanol to give 0.25 g. (26.0%) m.p. 225–227°; $\lambda_{max}^{CH_3OH}$ 235 and 298 mμ (ε 6,600 and 6,860); λ_{max}^{Nujol} 3.11, 5.30, 5.60, 6.10, 6.18, 6.70 μ.

Anal. Calcd. for $C_8H_6ClNO_3$: C, 48.1; H, 3.0; N, 7.0. Found: C, 48.0; H, 3.1; N, 7.0.

Acknowledgment.—We wish to thank Mrs. Mary C. Christie for determining the ultraviolet and infrared spectra and for certain of the nitrogen analyses.

(14) Melting points are corrected and were determined on a Thomas-Hoover capillary apparatus. Infrared spectra were determined on a Perkin Elmer Model 21 spectrophotometer and ultraviolet spectra on a Cary Model 14 spectrophotometer.

Thio Derivatives of 2,3-Dihydro-4H-1,3-benzoxazin-4-one. Synthesis and Pharmacological Properties

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A series of 2,3-dihydro-4H-1,3-benzoxazin-4-one-2-thio derivatives has been prepared either by condensation of salicylamide with S-substituted 3-thiopropionaldehyde or thioacetaldehyde acetals or by treating 2-(2-chloroethyl)-2,3-dihydro-4H-1,3-benzoxazin-4-one (chlorthenoxazine) with alkali salts of mercaptans. The pharmacological screening of the most active members of the series, 2,3-dihydro-2-[2-(methylthio)ethyl]-4H-1,3-benzoxazin-4-one (1) and 2,3-dihydro-2-[2-(phenylsulfinyl)ethyl]-4H-1,3-benzoxazin-4-one (8), has shown that they may be classified as interesting antiinflammatory and antipyretic agents of very low toxicity.

2-Substituted 2,3-dihydro-4H-1,3-benzoxazin-4-ones have not often been tested for pharmacological properties and the results of those tests have proved of little interest. Kaufmann[2] found no pharmacological activity for 2,3-dihydro-2-(trichloromethyl)-4H-1,3-benzoxazin-4-one, while Horrom,[3] who synthetized a series of mono- and disubstituted benzoxazinones of this class, reported slight analgesic activity for some of these com-

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