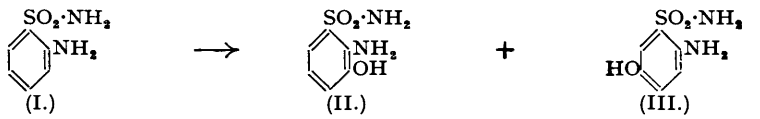
355. *The Synthesis of 2-Amino-3- and -5-hydroxybenzenesulphonamides.*

By D. V. PARKE and R. T. WILLIAMS.

2-Amino-3-hydroxybenzenesulphonamide has been synthesized from *m*-hydroxybenzenesulphonamide by way of 2-nitro-3-hydroxybenzenesulphonamide, and 2-amino-5-hydroxybenzenesulphonamide from *o*-aminobenzenesulphonamide by way of 5-bromo-2-aminobenzenesulphonamide. These compounds have been orientated and they appear to be identical with the two hydroxyorthanilamides which were isolated as metabolites of orthanilamide in the rabbit.

STUBBS AND WILLIAMS (*Biochem. J.*, 1947, **41**, xlix) reported the isolation of two aminohydroxybenzenesulphonamides from the urine of rabbits dosed with orthanilamide (I). These compounds were designated hydroxyorthanilamide-A (m. p. 188°) and -B (m. p. 179—180°) and were believed to be 2-amino-3- (II) and -5-hydroxybenzenesulphonamide (III) respectively, because the aromatic ring is usually hydroxylated *in vivo* in the positions *ortho* and *para* to the



amino-group. An attempt was therefore made to synthesize these compounds through the corresponding nitro-derivatives which theoretically could be produced by the nitration of *m*-hydroxybenzenesulphonamide.

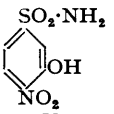
m-Hydroxybenzenesulphonamide was prepared by diazotization of *m*-aminobenzenesulphonamide and nitrated at 0° in concentrated sulphuric acid with 1·14 moles of nitric acid. A complex mixture of nitro-compounds was obtained in excellent yield. This was separated by chromatography, followed by fractional crystallization, into five pure compounds, two of which were mononitro- and three were dinitro-*m*-hydroxybenzenesulphonamides. These pure compounds accounted for some 20% of the starting material, 13% as mononitro- and 7% as dinitro-compounds. The main bulk of the mixture was, of course, sacrificed in the fractionation.

Theoretically *m*-hydroxybenzenesulphonamide could give rise on nitration to the 2-, 4-, and 6-mononitro-derivatives and probably to small amounts of the 2 : 4-, 2 : 6-, and 4 : 6-dinitro-derivatives. In this work we were only interested in the mononitro-compounds; the

dinitro-compounds were isolated incidentally and no attempt was made to determine their structure.

Although we expected to obtain three mononitro-derivatives we only succeeded in isolating two. Whether or not a third occurred in the mixed product could not be proved unequivocally. The least soluble mononitro-compound isolated formed dark yellow plates, m. p. 169—170°, and was readily proved to be 4-nitro-3-hydroxybenzenesulphonamide (IV) by reduction to the known 4-amino-3-hydroxybenzenesulphonamide which had been synthesized by Thorpe and Williams (*Biochem. J.*, 1941, **35**, 61) from benzoxazol-2-one.

The second mononitro-compound isolated formed pale yellow silky needles, m. p. 189—190°, and could be either 2-nitro-3- (V) or -5-hydroxybenzenesulphonamide (VI). On reduction, this nitro-compound yielded an amino-3-hydroxybenzenesulphonamide, m. p. 190°, which was identical (mixed m. p.) with the hydroxyorthanilamide-A isolated from rabbit urine. For reasons given below, this is the 2-amino-3-hydroxy-derivative (II) and therefore the mono-nitro-compound, m. p. 189—190°, is the 2-nitro-3-hydroxy-derivative (V).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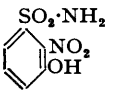
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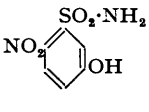
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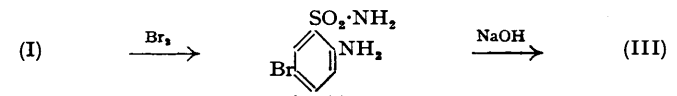
2-Amino-5-hydroxybenzenesulphonamide (III), m. p. 181°, was obtained from 5-bromo-2-aminobenzenesulphonamide (VII), prepared by bromination of orthanilamide in glacial acetic acid. The structure of (VII) was proved by heating it at 180° with 80% sulphuric acid whereby it was converted into *p*-bromoaniline. On fusion of (VII) with sodium hydroxide the hydroxy-


(VII.)

derivative (III), m. p. 181°, was obtained in poor yield. The structure of (III) follows from that of (VII). Hence by elimination the structure of the amino-derivative (II) and its parent nitro-derivative (V) is also proved. 2-Amino-5-hydroxybenzenesulphonamide appeared to be identical in all its reactions with the hydroxyorthanilamide-B of Stubbs and Williams (*loc. cit.*), but comparison by mixed melting point was not possible as none of the metabolite was available.

Experimental.

m-*Hydroxybenzenesulphonamide.*—Metanilamide (5 g.; m. p. 140°), dissolved in a mixture of sulphuric acid (20 c.c.) and water (40 c.c.), was diazotised at 0° with sodium nitrite (2 g.) in water (20 ml.) and then heated on the water-bath until the evolution of nitrogen ceased. After the mixture had been kept overnight at 0°, the crystals of m-*hydroxybenzenesulphonamide* were collected. A further quantity of the compound was obtained by exhaustively extracting the filtrate with ether (total yield, 4 g.). On recrystallization from water (charcoal), the sulphonamide was obtained as large, faintly yellow, thick, hexagonal plates, m. p. 165—166° (Found: C, 41·5; H, 4·05; N, 8·5; S, 19·0. $C_6H_7O_3NS$ requires C, 41·6; H, 4·1; N, 8·1; S, 18·5%). A strong aqueous solution gives a purple colour with dilute ferric chloride solution. Light absorption: $\lambda_{max.}$ 285—286 mμ.; $\varepsilon_{max.} = 2700$.

Nitration of m-*Hydroxybenzenesulphonamide.*—To the sulphonamide (10 g.), dissolved in concentrated sulphuric acid (65 c.c.) and cooled to 0°, was slowly added a mixture of nitric acid (4·2 c.c.; *d* 1·42) and concentrated sulphuric acid (4·5 c.c.), the temperature being kept below 5°. After an hour at room temperature, the mixture was poured on ice. The resulting yellow solution was extracted with ether which, when evaporated, left a yellow crystalline mass (11 g.). Repeated fractional crystallization of this product from water yielded only one pure compound—4-*nitro*-3-*hydroxybenzenesulphonamide*. This formed long dark yellow plates, m. p. 170° (Found: C, 33·3; H, 2·6; N, 12·9; S, 14·4. $C_6H_6O_5N_2S$ requires C, 33·0; H, 2·8; N, 12·9; S, 14·7%). It was slightly soluble in cold water but readily soluble in hot. It gives a deep orange colour with aqueous sodium hydroxide and a slight reddish-violet colour with aqueous ferric chloride.

The nitro-compound (0·4 g.) in water (10 c.c.) was heated with iron powder (1 g.) and dilute acetic acid (1 c.c.) until the yellow colour disappeared (*ca.* 10 minutes). The mixture was filtered and the filtrate continuously extracted with ether for 24 hours. The ether was evaporated off and the brown residue dissolved in a little ethanol and treated with charcoal. Carbon tetrachloride was then added to the filtered alcoholic solution until incipient precipitation occurred. The solution was warmed until the precipitate redissolved; on cooling, long pale brown plates, m. p. 164°, separated and were identified as 4-amino-3-hydroxybenzenesulphonamide by a mixed m. p. (164°) and colour reactions (cf. Thorpe and Williams, *loc. cit.*; Williams, *Biochem. J.*, 1946, **40**, 219; 1947, **41**, 1).

Chromatography of the Nitration Product.—The product of the nitration of *m*-hydroxybenzenesulphonamide was obviously complex. The crude nitration product obtained by ether-extraction as

above was dissolved in water and the solution poured on a column (1 × 40 cm.) of activated alumina (B.D.H.). The chromatogram was developed with water at pH 6. Two principal yellow bands developed, the upper consisting of dinitro- and the lower of mononitro-compounds.

The lower band was run off the column with water. The yellow solution was acidified with dilute hydrochloric acid, saturated with sodium chloride, and extracted with ether. Evaporation of the ether left a yellow crystalline solid which on fractional crystallisation from water yielded two compounds. The less soluble was 4-nitro-3-hydroxybenzenesulphonamide (m. p. 169—170°) described above. The more soluble 2-*nitro*-3-*hydroxybenzenesulphonamide* formed pale yellow, silky needles, m. p. 189—190° (Found : C, 33·0; H, 2·5; N, 12·9; S, 15·1. $C_6H_6O_5N_2S$ requires C, 33·0; H, 2·8; N, 12·9; S, 14·7%), readily soluble in water and giving a slight reddish-violet colour with aqueous ferric chloride and an orange colour with aqueous sodium hydroxide. The yields from 11 g. of nitration mixture were 4-nitro- 0·7 g. and 2-nitro- 0·58 g.

The 2-nitro-compound (0·3 g.) was reduced by heating it in water (8 c.c.) for a few minutes with iron powder (0·75 g.) and dilute acetic acid (0·7 ml.) Next morning, the solution was filtered and extracted continuously with ether. The ethereal extract was evaporated and the residue recrystallized from ethanol–carbon tetrachloride. Thus 2-*amino*-3-*hydroxybenzenesulphonamide*, m. p. 190°, was obtained (Found : C, 38·2; H, 4·25; N, 14·8; S, 16·6. $C_6H_8O_3N_2S$ requires C, 38·3; H, 4·3; N, 14·9; S, 17·0%) (see following paper for absorption spectrum). This compound gives a green colour with ferric chloride under certain conditions.

On diazotisation it gives an intense yellow colour, and coupling with dimethyl-α-naphthylamine takes place very slowly to give a red dye. It does not depress the m. p. (188°) of hydroxyorthanilamide-A (Found : C, 38·5; H, 4·7; N, 14·5; S, 16·7%) described by Stubbs and Williams (*loc. cit.*) from the urine of rabbits fed with orthanilamide and is identical with it in all respects.

The upper chromatogram band, on further development with water, was resolved into three bands, the lowest being pale lemon-coloured, the middle orange-yellow, and the uppermost mid-yellow. These bands were separated by cutting the column and extraction with water. Three *dinitro*-m-*hydroxybenzenesulphonamides* whose properties are given in the table were thus obtained.

Dinitro-m-*hydroxybenzenesulphonamides.*

No.	Location in upper band.	Description.	M. p.	Comparative solubility in water.	Yield, g.	Found, % : C.	H.	N.	S.
1	Middle	Dark yellow clusters	208—209°	Least soluble	0·08	27·8	1·95	15·9	12·25
2	Top	Short dark yellow needles	235	—	0·45	28·1	1·8	15·3	11·9
3	Bottom	Pale yellow short needles	248	Most soluble	0·18	25·7	2·55	15·1	—
				$C_6H_5O_7N_3S$ requires		27·4	1·9	16·0	12·2
				$C_6H_5O_7N_3S,H_2O$ requires		25·6	2·5	14·9	—

Reduction of the Nitration Product.—The separation of the components of the product of nitration of *m*-hydroxybenzenesulphonamide was extremely tedious and the final yields of the required amino-compounds small. Better yields, however, were obtained when the crude nitration product was first reduced and the amino-compounds separated.

A solution of the nitration product (20 g.) in hot water (250 c.c.) was boiled with iron powder (50 g.) and dilute acetic acid (5 ml.). A few crystals of sodium dithionite were added to the hot solution to reduce the black oxidation products of the aminophenolsulphonamides, and the solution was filtered hot and then continuously extracted with ether for 12 hours. Evaporation of the ether left a mixture of aminophenolsulphonamides, which was fractionated by crystallization from water after clarification with charcoal. During the crystallizations the solutions were kept in stoppered flasks and a few crystals of sodium dithionite were added. 2-Amino-3-hydroxybenzenesulphonamide (4 g.) separated first, as a mixture of red and yellow crystals (m. p. 190°) which were identical except in colour. From the mother-liquors, pale reddish-brown plates of 4-amino-3-hydroxybenzenesulphonamide (m. p. 164°; 0·1 g.) were obtained.

2-*Amino*-5-*hydroxybenzenesulphonamide.*—5-*Bromo*-2-*aminobenzenesulphonamide.* To orthanilamide (5·2 g.) in glacial acetic acid (75 c.c.), bromine (0·9 c.c., 2·8 g.) in acetic acid (15 c.c.) was gradually added during 5—10 minutes with gentle warming and vigorous shaking. After 15 minutes the solution was poured into water (250 c.c.), whereby the *bromo-orthanilamide* was precipitated; it was filtered off after several hours (4·2 g.). Repeated recrystallization from 25% aqueous ethanol gave the pure compound as almost white glistening elongated plates, m. p. 179° (Found : C, 28·5; H, 2·8; N, 10·5; Br, 33·0. $C_6H_7O_2N_2SBr$ requires C, 28·7; H, 2·8; N, 11·2; Br, 31·8%) (see Parke and Williams, *loc. cit.*, for absorption spectrum).

Conversion of 5-*bromo*-2-*aminobenzenesulphonamide into* p-*bromoaniline.* The foregoing bromo-compound (1 g.) in 80% sulphuric acid (10 ml.) was heated at 180° for 10 minutes. The pale pink solution gradually became violet, this colour gradually becoming more intense. After cooling, the solution was poured into water (50 c.c.) and neutralized with 40% aqueous sodium hydroxide, with cooling. When the solution was made slightly acid with dilute sulphuric acid, unchanged bromo-orthanilamide was precipitated; this was removed by filtration. The filtrate was made slightly alkaline with dilute sodium hydroxide solution, bromoaniline being precipitated. This was extracted with ether, and the extract dried (Na_2SO_4). Evaporation of the ether left *p*-bromoaniline as a violet solid which was dissolved in 2N-hydrochloric acid (5 c.c.) and water (20 ml.) by heat. After crystallization at 0°, *p*-bromoaniline hydrochloride separated as pale violet needles (220 mg., 28%). The free base was regenerated and after recrystallization from 20% aqueous ethanol formed nearly colourless clusters of needles (Found : Br, 47·0. Calc. for C_6H_6NBr : Br, 46·5%), m. p. 63—64° not depressed on admixture

with authentic *p*-bromoaniline (m. p. 64°). The free base (0·1 g.) was acetylated in the usual way and, after recrystallization of the product from 25% aqueous ethanol, *p*-bromoacetanilide (55 mg.), m. p. and mixed m. p. 168°, was obtained (Found: Br, 37·8. Calc. for C_8H_8ONBr: Br, 37·3%).

Conversion of 5-bromo-2-aminobenzenesulphonamide into 2-amino-5-hydroxybenzenesulphonamide. Sodium hydroxide (25 g.) and water (10 c.c.) were heated together in a silica dish and to the melt was added the 5-bromo-compound (5 g.) and sodium dithionite (1 g.). After being heated for a few minutes, the dark violet melt was allowed to cool and then dissolved in water. The solution was acidified with concentrated hydrochloric acid with cooling and then made neutral with saturated aqueous sodium carbonate. The solution (250 c.c.) was continuously extracted with peroxide-free ether for four periods of 8 hours each. Each ethereal extract was evaporated to dryness, and the residues were dissolved in a little hot water with the addition of little sodium dithionite, treated with charcoal, and filtered. The first extract gave only tars; the other three gave only tars on cooling, but, on concentration of the mother-liquors from these tars, 160 mg. (4·3%) of crude 2-*amino*-5-*hydroxybenzenesulphonamide*, m. p. 179—180°, were obtained. Recrystallization from water eventually gave the pure compound, m. p. 180—181°, as violet-brown long plates (Found: C, 38·7; H, 4·3; S, 17·4. $C_6H_8O_3N_2S$ requires C, 38·3; H, 4·3; S, 17·0%). Light absorption: Maxima at 211, 255, and 338 mμ.; $\varepsilon_{max.}$ = 18,100, 7,800, and 3,200 respectively). It was readily soluble in hot water and moderately soluble in cold. With ferric chloride it gave an intense violet-red colour, soluble in ether. On addition of a drop of dilute aqueous ammonia to its aqueous solution, an intense violet colour gradually appeared; this colour is not given by 2- or 4-amino-3-hydroxybenzenesulphonamide, but is given by the metabolite, hydroxyorthanilamide-B. The same colour is also produced very slowly when an aqueous or ethanolic solution is exposed to air. Diazotization followed by coupling with dimethyl-α-naphthylamine gives an intense violet colour, as does that of *p*-aminophenol.

Department of Biochemistry,
St. Mary's Hospital Medical School, London, W.2.

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