expressed in terms of enzyme units and calculated from the expression

$$\text{E.U.} = \frac{T_0 - T}{T}$$

where E.U. represents enzyme units, T_0 the time of the uncatalyzed reaction in sec., and T the time of the catalyzed reaction in sec. All reactions were carried out in a cold room maintained at 5 ± 1°. The inhibitors were preincubated with the enzyme for 10 min. prior to the addition of substrate. This procedure allowed for enzyme–inhibitor equilibrium to take place. The concentration required to inhibit 50% of the enzyme activity was determined graphically. Approximately two units of enzyme activity were utilized in each experiment.

The *in vivo* inhibition of brain carbonic anhydrase was approximated by diluting the tissue throughout a range of 1–10 to 1–200 and determining the enzyme activity of the inhibited tissue relative to the untreated controls.

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The Synthesis of Some Substituted *m*-Benzenedisulfonamides

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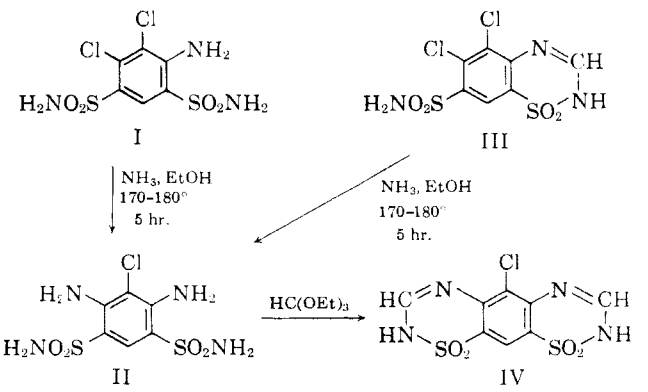
An approach to the synthesis of some substituted *m*-benzenedisulfonamides, not readily available *via* the usual methods for preparing this class of compounds, is described. The diuretic activity of the compounds was evaluated.

Prior to 1957 there was little mention of substituted *m*-benzenedisulfonamides in the organic chemical literature. Lustig and Katscher,[1] in 1927, described a convenient method for the preparation of amino substituted *m*-benzenedisulfonamides by the direct chlorosulfonation of a substituted aniline in the presence of sodium chloride, then treatment of the resulting *m*-disulfonyl chloride with ammonia. Davies and Poole[2] prepared 4,6-dichloro-*m*-benzenedisulfonamide and 2,4,6-trichloro-*m*-benzenedisulfonamide by disulfonation of the corresponding chlorobenzene and transformation of a salt of the disulfonic acid to the disulfonyl chloride with phosphorus pentachloride and thence into the disulfonamide.

With the discovery[3] that substituted *m*-benzenedisulfonamides had pronounced diuretic activity, the literature on such compounds has greatly expanded. The procedure of Lustig and Katscher has been extensively applied to substituted anilines.[3-9] The chlorosulfonation of a substituted 2-aminobenzenesulfonamide is a varient which has been used to advantage.[10] A reaction developed by Meerwein[11] for converting an amino group into a sulfonyl chloride was employed by Petrow[12] in the synthesis of benzenedisulfonamides.

We were interested in synthesizing *m*-benzenedisulfonamides for their evaluation as diuretic agents and also as intermediates for the synthesis of substituted 3,4-dihydro-2H-1,2,4-benzothiadiazine-7-sulfonamide-1,1-dioxides.[13] During the course of this work it became apparent that the methods reviewed in the preceding paragraph were not adequate for the convenient synthesis of certain *m*-benzenedisulfonamides and it is the purpose of this paper to discuss an additional approach which we have found to be very useful in the synthesis of these compounds.

The approach in question is dependent upon the activating effect of sulfamoyl groups *ortho* and *para* to a suitable leaving group (halogen or nitro) in the benzene nucleus.[14] Thus reaction of 4-amino-5,6-dichloro-*m*-benzenedisulfonamide (I) with ammonia in ethanol at 170-180° for 5 hr. furnished 5-chloro-4,6-diamino-*m*-benzenedisulfonamide (II) in satisfactory yield. This compound (II) also was obtained under similar reaction conditions from 5,6-dichloro-2H-1,2,4-



(1) O. Lustig and E. Katscher, *Monatsh.*, **48**, 87 (1927).
(2) W. Davies and H. G. Poole, *J. Chem. Soc.*, 1122 (1927).
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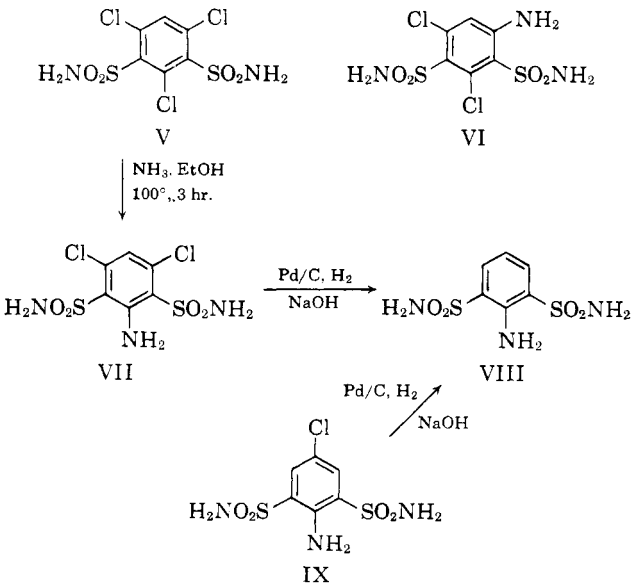
(11) (a) H. Meerwein, E. Büchner, and K. Van Emster, *J. prakt. Chem.*, [2] **152**, 237 (1939); (b) H. Meerwein, G. Dittmar, G. Göllner, K. Hafner, F. Mensch, and O. Steinfort, *Chem. Ber.*, **90**, 841 (1957).
(12) V. Petrow, O. Stephenson, and A. M. Wild, *J. Pharm. and Pharmacol.*, **12**, 705 (1960).
(13) J. G. Topliss, M. H. Sherlock, F. H. Clarke, M. C. Daly, B. Pettersen, J. Lipski, and N. Sperber, *J. Org. Chem.*, **26**, 3842 (1961).
(14) Subsequent to the completion of our work the use of this principle was reported by (a) G. B. Jackman, V. Petrow, O. Stephenson, and A. M. Wild, *J. Pharm. and Pharmacol.*, **12**, 648 (1960), and (b) C. W. Whitehead and J. J. Traverso, *J. Org. Chem.*, **27**, 951 (1962). The particular reactions involved, however, were not the same.

benzothiadiazine-7-sulfonamide-1,1-dioxide (III). The structure of the product was confirmed by its conversion with ethyl orthoformate into 5-chloro-2H,8H-benzo[1,2-e:5,4-e′]bis[1,2,4]thiadiazine-1,1,9,9-tetroxide (IV). It is interesting to note that 4-amino-6-chloro-*m*-benzenedisulfonamide did not react with ammonia under the conditions which converted I into II. The importance of the activating effect of the additional halogen atom in I *ortho* to the leaving group is thereby demonstrated. The possible synthesis of II by the commonly used method would first require the preparation of 2-chloro-*m*-phenylenediamine (not readily available) and the application of the Lustig and Katscher procedure to this compound. However, in this connection it may be noted that in our hands *m*-phenylenediamine gave a very low yield of 4,6-diamino-*m*-benzenedisulfonamide.[15]

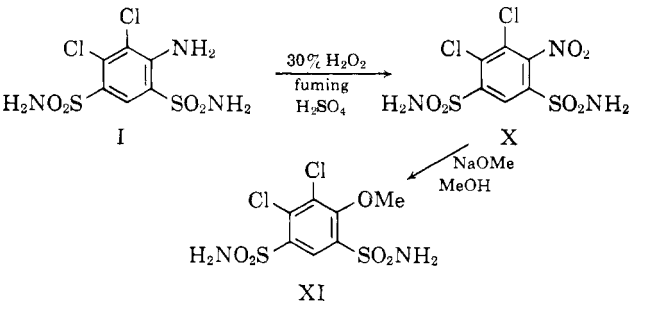
m-Benzenedisulfonamides can be obtained from substituted N-alkylanilines.[4,9] However, such substitution of the amine function in our hands significantly reduced the yield in the chlorosulfonation reaction as compared with that obtained from the parent aniline. Moreover, the synthesis of N-substituted amino-*m*-benzenedisulfonamides by this method is limited to those N-substituents which are unreactive under chlorosulfonation conditions. A convenient method which overcame these difficulties was devised by taking advantage of the activated nature of a chlorine atom in 4,6-dichloro-*m*-benzenedisulfonamide. Thus the latter compound when refluxed with excess of benzylamine in ethanol solution for 16 hr. furnished 4-N-benzylamino-6-chloro-*m*-benzenedisulfonamide in good yield. With methylamine under similar conditions 6-chloro-4-methylamino-*m*-benzenedisulfonamide was obtained.[16]

The displacement of one chlorine atom in 2,4,6-trichloro-*m*-benzenedisulfonamide (V) by an amino group could give rise to two possible products, VI and VII. The nonequivalent chlorine atoms are each activated by the sulfamoyl groups making a prediction



difficult concerning their relative reactivities. The reaction with ammonia in ethanol for 3 hr. at 100° gave, as the only insolable product, a compound whose analytical data were consistent with its formulation as a monoamine VI or VII. Reduction of this product in alkaline solution using a palladium charcoal catalyst yielded an amino-*m*-benzenedisulfonamide identical with that obtained by a similar reduction of 2-amino-5-chloro-*m*-benzenedisulfonamide (IX). The reduction product was, therefore, 2-amino-*m*-benzenedisulfonamide (VIII), thus establishing the structure of the ammonia reaction product as VII. It may be noted that application of the Lustig and Katscher procedure to 3,5-dichloroaniline, a likely path to VI or VII, does not yield any recognizable product.[17]

Oxidation of the amino group in 4-amino-5,6-dichloro-*m*-benzenedisulfonamide (I) with 30% hydrogen peroxide and fuming sulfuric acid afforded a good yield of 4,5-dichloro-6-nitro-*m*-benzenedisulfonamide (X). This compound is interesting in that all five substituents on the benzene ring could be activated in some degree toward attack by nucleophilic reagents. The action on the compound of a large excess of sodium methoxide in methanol solution at reflux temperature for a period of 16 hr. was investigated. The main product of the reaction was a compound of empirical formula $C_7H_8Cl_2N_2O_4S_2$ whose infrared spectrum gave no evidence of change in the *meta*-disulfonamide groups. It was apparent from these considerations that the nitro group had been displaced by methoxide and that the product in question was 4,5-dichloro-6-methoxy-*m*-benzenedisulfonamide (XI). An attempt to prepare compound



XI by application of the Lustig and Katscher procedure to 2,3-dichloroanisole afforded a monosulfamoyl derivative of 2,3-dichlorophenol.

Pharmacological Evaluation.—The compounds were assayed for diuretic activity[18] in the rat (oral route) by a previously described procedure[19] with the exception that the urine collection period was 6 hr. instead of 4 hr. The standard compound was chlorothiazide and the activity of the compounds tested was determined for urine, sodium, potassium and chloride excretion. Compound 2 was included for comparative purposes.[20] The results are presented in Table I.

Compound 7 was significantly more potent, and compound 6 significantly less potent, than chlorothiazide in terms of sodium and urine excretion. The other

(15) Lustig and Katscher, ref. 1, report m.p. 187° for the compound obtained by this procedure. The structure of our product, m.p. 258–259°, was established by an independent synthesis involving the reduction of 4-amino-6-nitro-*m*-benzenedisulfonamide.

(16) Also, when ammonia was used in ethanol at 150–160° for 4 hr., the product was 4-amino-6-chloro-*m*-benzenedisulfonamide.

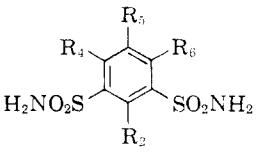
(17) Short and Biermacher (ref. 6) using chlorosulfonic acid alone, were also unable to obtain a disulfonamide from 3,5-dichloroaniline. However, under milder conditions they obtained a monosulfonamide.

(18) We are indebted to Dr. R. M. Taylor of the Department of General Pharmacology, Biological Research Division, Schering Corporation, for these results.

(19) R. M. Taylor and J. G. Topliss, *J. Med. Pharm. Chem.*, **5**, 312 (1962).

(20) Electrolyte excretion data for this compound were reported by F. J. Lund and W. Kobinger, *Acta Pharm. Tox., Kbh.*, **16**, 297 (1960).

TABLE I



Compound	R_3	R_4	R_5	R_6	Urine volume	Na	Cl	K
1		Chlorothiazide			1.0	1.0	1.0	1.0
2	H	NH_2	H	Cl	1.6[a]	1.5[a]	0.9[a]	3.3
3	H	$NHCH_3$	H	Cl	1.5	2.1[a]	0.1[b]	8.1[a]
4	H	$NHCH_2C_6H_5$	H	Cl	2.3[a]	2.3[a]	0.8	6.3[a]
5	H	NH_2	Cl	NH_2	1.7[a]	1.8[a]	0.3[b]	[c]
6	NH_2	Cl	H	Cl	0.2[b]	0.1[b]	0.05	3.1[a]
7	H	Cl	Cl	NO_2	2.4[b]	1.9[b]	0.2[a]	17.8[a]
8	H	Cl	Cl	OCH_3	2.8[b]	1.4	0.02[a]	16.6[a]

[a] A rough estimate of potency. There is evidence of non-parallelism. [b] Significant difference between standard and test drug at p. 0.05. [c] Assay invalid.

compounds appeared to show a slightly higher potency than chlorothiazide with respect to these parameters. However, only compound 4 approached the activity of chlorothiazide with regard to chloride excretion and resembled compound 2 in this respect. There was a marked tendency for the compounds to exhibit a much greater effect on potassium excretion than with chlorothiazide.

The diuretic activity of chlorothiazide and the *m*-benzenedisulfonamides may be considered to result from an intrinsic saluretic effect, in addition to an inhibitory action of the compound on carbonic anhydrase. With chlorothiazide the desirable saluretic effect is much the larger component. In the case of the *m*-benzenedisulfonamides, compounds 2 and 4 appear to have a moderate saluretic effect[21] but probably derive a higher proportion of their total urine and sodium excreting effects from the inhibition of carbonic anhydrase than does chlorothiazide. The low chloride and high potassium excreting properties of compounds 3, 5, 7 and 8 indicate that the carbonic anhydrase inhibition mechanism probably plays a more important role in their diuretic activity.

Experimental[22]

5-Chloro-4,6-diamino-*m*-benzenedisulfonamide (II).—(a) A mixture of 4-amino-5,6-dichloro-*m*-benzenedisulfonamide[6] (9.45 g.), ethanol (250 ml.) and ammonia (50 g.) was heated in an autoclave for 5 hr. at 170–180°. The solvent and excess of ammonia were evaporated and water was added to the residual material. The crude product was collected by filtration, washed with water and air dried affording a discolored solid (6.05 g.). Recrystallization from methanol (charcoal) yielded the desired product (4.3 g.), m.p. 255–256°; λ_{max} 239 mμ (ε 57,500); 262 mμ (ε 12,900); 300 mμ (ε 2,100) (broad).

Anal. Calcd. for $C_6H_9ClN_4O_4S_2$: Cl, 11.79; N, 18.63. Found: Cl, 12.03; N, 18.20.

(b) 5,6-Dichloro-2H-1,2,4-benzothiadiazine-7-sulfonamide-1,1-dioxide[6] (5 g.) was heated with ammonia (10 g.) and ethanol (100 ml.) in an autoclave at 175° for 5 hr. The solvent and excess of ammonia were evaporated and dilute hydrochloric acid was added to the residue. The crude product was collected by filtration, washed with water and air dried; yield 3.25 g., m.p. 249–250°. Recrystallization from 95% ethanol (charcoal) raised the melting point to be 255–256°. The product was shown to be the same as that obtained in (a) (mixture m.p. and infrared spectrum).

5-Chloro-2H,8H-Benzo[1,2-e:5,4-e′]bis[1,2,4]thiadiazine-1,1,-9,9-tetraoxide (IV).—5-Chloro-4,6-diamino-*m*-benzenedisulfonamide (7.1 g.) was dissolved in a hot mixture of ethyl orthoformate (25 ml.) and 2-methoxyethanol (75 ml.) and the solution refluxed for 12 hr. Solid separated out during the reflux period. The cooled reaction mixture was filtered and the solid product washed with methanol and air dried; yield 5.3 g., m.p. > 360°. Recrystallization was effected from a mixture of dimethylformamide–water (charcoal) affording a white crystalline, highly insoluble solid, m.p. > 360°. No N–H absorptions corresponding to an aromatic amine were present in the infrared spectrum.

Anal. Calcd. for $C_5H_3ClN_4S_2O_4$: Cl, 11.06; S, 20.00. Found: Cl, 10.78; S, 20.42.

4,6-Diamino-*m*-benzenedisulfonamide.—4-Amino-6-nitro-*m*-benzenedisulfonamide (40.4 g.) was dissolved in 80% ethanol (840 ml.), iron filings (40.4 g.) were added and the mixture was brought to reflux with stirring. To the stirred refluxing reaction mixture 10% hydrochloric acid (405 ml.) was added, dropwise, and refluxing then continued for 3 hr. The hot mixture was filtered and the insoluble material extracted with boiling water. The filtrate and the aqueous extract were combined, concentrated and then cooled, affording crude product (25.3 g.), m.p. 240–242°. Recrystallization from water (charcoal) gave 16.5 g. of pure product, m.p. 258–259° (reported[1] m.p. 187°; 261–262°).[20]

Anal. Calcd. for $C_6H_8ClN_5O_4S_2$: Cl, 12.41; N, 14.71; S, 22.44. Found: Cl, 12.22; N, 14.71; S, 22.54.

The same product, m.p. 258–259°, was obtained in very low yield from *m*-phenylenediamine hydrochloride by the procedure of Lustig and Katscher.[1]

4-N-Benzylamino-6-chloro-*m*-benzenedisulfonamide.—A solution of 4,6-dichloro-*m*-benzenedisulfonamide[2] (1.0 g.), benzylamine (1.77 g.) and ethanol (20 ml.) was refluxed for 16.5 hr. The solvent was evaporated on the steam-bath, the viscous residue triturated with 10% hydrochloric acid (*ca.* 20 ml.) and the crude product collected by filtration, washed with water and air dried. Recrystallization from aqueous methanol (charcoal) gave 0.99 g. of product m.p. 212–213°. Further recrystallization from the same solvent yielded white crystals, m.p. 213–214° (reported,[13b] m.p. 200°). λ_{max} 225 mμ (ε 37,000); 270 mμ (ε 23,400); 320 mμ (ε 5,500).

Anal. Calcd. for $C_{13}H_{14}ClN_3O_4S_2$: Cl, 9.44; N, 11.18. Found: Cl, 9.39; N, 11.35.

4-Chloro-6-N-methylamino-*m*-benzenedisulfonamide.—A mixture of 4,6-dichloro-*m*-benzenedisulfonamide[2] (5.0 g.), methylamine (21 g.) and 95% ethanol (200 ml.) was refluxed for 16 hr. The reaction mixture was concentrated almost to dryness and 10% hydrochloric acid added. The crude product was collected, washed with water and air dried; (4.33 g.) m.p. 251°. Recrystallization from 50% ethanol gave 4.06 g., m.p. 254° (reported,[9] m.p. 248–249°); λ_{max} 227 mμ (ε 35,600); 270 mμ (ε 19,800); 320 mμ (ε 4,600).

Anal. Calcd. for $C_7H_9ClN_3S_2O_4$: Cl, 11.82; N, 14.02. Found: Cl, 11.69; N, 13.95.

4-Amino-6-chloro-*m*-benzenedisulfonamide.—A mixture of 4,6-dichloro-*m*-benzenedisulfonamide (5 g.), ammonia (20 g.) and ethanol (100 ml.) was heated in an autoclave for 4 hr. at 150–160°. The solvent was evaporated, water added to the residue and the crude product collected by filtration, washed with water and air dried (2.8 g.). Crystallization from methanol (charcoal) gave the pure product; (1.4 g.), m.p. 260–261°, identical with a sample prepared from *m*-chloroaniline[3] (mixture m.p. and infrared spectrum).

2-Amino-*m*-benzenedisulfonamide (VIII).—2-Amino-5-chloro-*m*-benzenedisulfonamide[7] (1.0 g.) was dissolved in 5% sodium hydroxide solution (100 ml.) and reduction effected at room temperature and 40 p.s.i. in the presence of 5% palladium-on-carbon (0.2 g.) over a period of 16 hr. The catalyst was filtered off, the filtrate neutralized with hydrochloric acid, concentrated and chilled overnight. The solid which separated was collected and recrystallized from acetone–water, m.p. 212–213° (reported[9] m.p. 206–207°); λ_{max} 212 mμ (ε 32,500); 250 mμ (ε 7,700); 319 mμ (ε 5,300).

Anal. Calcd. for $C_6H_9N_3O_4S_2$: N, 16.72; S, 25.51. Found: N, 16.80; S, 25.76.

2-Amino-4,6-dichloro-*m*-benzenedisulfonamide (VII).—A mixture of 2,4,6-trichloro-*m*-benzenedisulfonamide[2] (10 g.), ammonia (30 g.) and ethanol (300 ml.) was heated in an autoclave for 3 hr. at 100°. The reaction mixture was concentrated almost to dry-

(21) This has been reported for compound 2 by J. M. Sprague, *Ann. N. Y. Acad. Sci.*, **71**, 328 (1958).

(22) Ultraviolet absorption spectra were measured in methanol solution and infrared absorption spectra as Nujol mulls unless otherwise stated. Melting points were determined on a Thomas–Hoover capillary melting point apparatus.

ness and dilute hydrochloric acid added. The crude solid product was collected by filtration, washed with water and recrystallized from methanol–chloroform (charcoal) giving white crystals (2.0 g.), m.p. 248–250°. Further recrystallization from 50% ethanol gave the compound, m.p. 255°; λ_{max} 229 mμ (ε 23,500); 236 mμ (ε 23,000); 257 mμ (ε 5,400); 263 mμ (ε 4,600); 318 mμ (ε 7,200).

Anal. Calcd. for $C_6H_7Cl_2N_3O_4S_2$: Cl, 22.16; N, 13.13. Found: Cl, 21.96; N, 12.99.

Catalytic reduction of the compound in alkaline solution under conditions similar to those employed for the reduction of 2-amino-5-chloro-*m*-benzenedisulfonamide yielded 2-amino-*m*-benzenedisulfonamide as shown by comparison with the sample previously obtained.

4,5-Dichloro-6-nitro-*m*-benzenedisulfonamide (X).—4-Amino-5,6-dichloro-*m*-benzenedisulfonamide (1.0 g.) was dissolved in sulfuric acid (5 ml.) and the solution added to a mixture of 20% fuming sulfuric acid (20 ml.) and 30% hydrogen peroxide solution (10 ml.), cooled in an ice-bath. After 2 hr. at ice-bath temperature and 4 hr. at room temperature, the reaction mixture was diluted with water (200 ml.) and then allowed to stand overnight. The crude product was collected by filtration, washed with water and air dried; yield, 0.58 g. m.p. 237–242°. Recrystallization was effected from dilute ethanol yielding the pure product, m.p. 248–249°; λ_{max} 214 mμ (ε 41,400); plateau 286–290 mμ (ε 1,900); 295 mμ (ε 2,200); λ_{max} 6.50 μ (s).

Anal. Calcd. for $C_6H_5Cl_2N_3O_4S_2$: Cl, 20.25; N, 12.00. Found: Cl, 20.56; N, 11.96.

4,5-Dichloro-6-methoxy-*m*-benzenedisulfonamide (XI).—A mixture of 4,5-dichloro-6-nitro-*m*-benzenedisulfonamide (1.0 g.), sodium methoxide (2.0 g.) and methanol (20 ml.) was refluxed for 16 hr. The cooled reaction mixture was diluted with water, acidified with dilute hydrochloric acid, and the crude product collected by filtration, washed with water and air dried. Recrystallization was effected from water yielding product of m.p. 240–242°; λ_{max} 222 mμ (ε 17,500); sh. 245 mμ (ε 8,600); 269 mμ (ε 4,400); 299 mμ (ε 3,200).

Anal. Calcd. for $C_7H_8Cl_2N_2O_2S_2$: Cl, 21.15. Found: Cl, 21.08.

Monosulfonamide of 2,3-Dichlorophenol.—2,3-Dichloroanisole was chlorosulfonated and the resulting product treated with ammonia according to the procedure of Lustig and Katscher.[1] The product obtained had m.p. 252–253°; λ_{max} 241 mμ (ε 8,600); 272 mμ (ε 3,200); sh. 279 mμ (ε 2,800); 291 mμ (ε 2,500).

Anal. Calcd. for $C_6H_5Cl_2NO_3S$: N, 5.79; Cl, 29.29. Found: N, 5.90; Cl, 29.08.

The same product was obtained when 2,3-dichlorophenol was used instead of 2,3-dichloroanisole.

Acknowledgments.—The authors are indebted to Mr. R. Wayne for the infrared spectral data, Mr. E. Connor for the microanalytical results, and Mr. W. Boraczek for the preparation of certain of the compounds described.

The Synthesis and Pharmacological Action of Tremetone

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Tremetone (I), the principal levorotatory ketonic constituent of "tremetol," the crude toxin of *Eupatorium urticaefolium*, has been synthesized. The synthetic sequence involved: coumarilic acid—(NaHg)→ hydrocoumarilic acid —(EtOH, H^+)→ ethyl hydrocoumarilate —(MeMgBr)→ 2-(2,3-dihydro-2-benzofuryl)-2-propanol —(Ac_2O, $SnCl_4$)→ 2-(2,3-dihydro-5-acetyl-2-benzofuryl)-2-propyl acetate —(pyrolysis)→ I. Optical resolution was accomplished at the hydrocoumarilic acid stage, and the synthetic enantiomer (−)-I proved identical in all respects with natural levorotatory tremetone. Preliminary toxicity data for tremetone and tremetol are discussed, as applied to goldfish, several species of insects, mice, rabbits, sheep and chickens. Crude tremetol proved toxic to chickens, whereas tremetone was harmless, suggesting that the ketone may not be the active toxin in tremetol responsible for "trembles" in cattle and "milk sickness" in humans.

White snakeroot (*Eupatorium urticaefolium*) is a toxic weed which grows extensively in damp areas of the central United States. Its ingestion by cattle causes the veterinary disease "trembles,"[3,4] while human consumption of dairy products from infected cattle engenders the fatal malady known as "milk sickness,"[5] an incurable illness which periodically ravished early pioneer communities of the central west. In 1929 J. F. Couch isolated from white snakeroot a straw-colored oil, "tremetol," which produced the symptoms of trembles in sheep and which was considered to be the homogeneous active toxin of this weed.[3,6] No subsequent confirmation of Couch's observations has been reported, and indeed this challenging problem has lain dormant until only recently, when we became interested in its pursuit.[7]

Application of column chromatography to the crude tremetol described by Couch afforded six individual components: a sesquiterpene hydrocarbon ($C_{15}H_{24}$), two steroids ($C_{27}H_{46}O$ and $C_{30}H_{48}O$) and three ketones, namely, tremetone ($C_{13}H_{14}O_2$), dehydrotremetone ($C_{13}H_{12}O_2$) and hydroxytremetone ($C_{13}H_{14}O_3$).[8] These ketones proved toxic to goldfish (*vide infra*) and produced a red color test with sulfuric acid, a test which Couch found characteristic of trembles-producing fractions.[3,6] Accordingly, one or more of these ketones was suspected of being the active toxin of white snakeroot, and their structural investigations were therefore undertaken.

Degradative reactions and catalytic hydrogenation revealed that tremetone, the principal ketone con-

(1) National Science Foundation Faculty Fellow, 1961–1962.

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(3) J. F. Couch, *J. Agr. Res.*, **35**, 547 (1927); see this article for an extensive review of the earlier literature.

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