

Further work is planned involving the adaptation of the method to the study of the formation of hydrogen sulfide by bacteria in various prepared culture media. It should be possible to secure some interesting quantitative data on this subject.

### Summary

A method for the estimation of hydrogen sulfide in food products which comprises the following features has been developed.

1. Removal of sulfide sulfur from the material by a current of carbon dioxide acting on an aqueous acidified suspension of the product.

2. Absorption of the evolved hydrogen sulfide in dilute zinc acetate solution.

3. Evaluation of the sulfide content of the absorbing solution, by a comparison with standards of the "methylene blue" color produced by interaction of the hydrogen sulfide with *p*-amino-dimethylaniline, hydrochloric acid and ferric chloride, according to the well-known Lauth's reaction.

Analyses of beef, pork and fish showed that hydrogen sulfide was formed progressively during the putrefaction of these products.

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## THE UTILIZATION OF PARA-TOLUIDINE IN THE SYNTHESIS OF AROMATIC ESTERS OF THE NOVOCAINE TYPE<sup>1</sup>

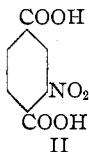
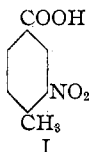
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This paper is a report of an investigation in the field of anesthetics, and contains data contributing to our knowledge of the relationship between chemical constitution and pharmacological action. The research was developed around the aromatic base *p*-toluidine, and by application of new syntheses and improvement in the technique of known reactions several new combinations have been prepared which promise to be of immediate physiological and pharmacological interest. The derivatives of major interest are some new esters of aromatic acids prepared from diethyl-amino-ethyl alcohol,  $(C_2H_5)_2N.CH_2CH_2.OH$ .

The two aromatic acids selected as the basis of our work were 2-nitro-4-methylbenzoic acid I and nitroterephthalic acid II.



<sup>1</sup> Constructed from part of a dissertation presented by Magnus A. Soderman to the Faculty of the Graduate School of Yale University, June, 1922, in candidacy for the degree of Doctor of Philosophy.



methyl-2-nitro-4-aminobenzene. Later, Banse<sup>2</sup> investigated the action of fuming nitric acid on *p*-toluyl nitrile and obtained, by using a large excess of acid, the same nitrile described by Noyes. The yield obtained is not stated. Borsche and Böcker<sup>5</sup> obtained later the same compound by nitration of *p*-toluyl nitrile with ordinary concd. nitric acid, but they give no directions regarding the proportions used and make no statement about yields of nitration product. A description of a single experiment will serve to illustrate the method used by us for the preparation of this compound.

Twelve hundred g. of concd. sulfuric acid and 384 g. of concd. nitric acid were thoroughly mixed and cooled to 20°. The molecular proportion of nitric acid was, therefore, 1.5 for each molecule of nitrile. To this acid mixture 300 g. of *p*-toluyl nitrile was added, during efficient stirring, at such a rate that the temperature of the solution did not rise above 40°. When a little more than one-half of the nitrile had been added, crystals of the nitro compound began to appear, and after the final addition of the nitrile the reaction mixture became a thick, yellow paste. After standing for 30 minutes, the mixture was poured into 6000 cc. of cold water and the precipitate separated by filtration and washed free from acids with water. The weight of the crude nitro compound was 405 g., or 97%.

Although but slightly volatile with steam, this nitro compound can be purified easily by this means giving an excellent yield of pure, colorless nitro compound, melting sharply at 107°. Ten g. of the crude product when subjected to this method of purification gave 9.3 g. of pure material or a yield of 93%.

*Anal.* Calcd. for C<sub>8</sub>H<sub>6</sub>O<sub>2</sub>N<sub>2</sub>: N, 17.28. Found: 17.39.

This nitrile can also be purified by distillation under diminished pressure and boils at 171° (12 mm.).

**Hydrolysis of 1-Methyl-2-nitro-4-cyanobenzene to 2-Nitro-*p*-toluic Acid.**—This acid was first obtained by Noad<sup>6</sup> by the action of nitric acid on cymol. It has also been prepared by the nitration of *p*-toluic acid and various cymol derivatives.<sup>7</sup> The yields obtained are not stated and the melting points given vary from 188° to 190°. Noyes<sup>3</sup> prepared the toluic acid by hydrolysis of its nitrile, heating with concd. hydrochloric acid under pressure, but here again no yields are recorded. We have used sulfuric acid as the hydrolytic agent, and investigated the efficiency of the method, using various dilutions of acid. When 10 g. of the 2-nitro-*p*-toluyl nitrile and 75 g. of an acid mixture (d., 1.52) consisting of equal volumes of concd. sulfuric acid and water are heated in an oil-bath at 180° the nitrile dissolves quickly and within one-half an hour the toluic acid, m. p. 186–187°, is produced in a quantitative yield. In experiments in which the volume of water was one-third and two-thirds that of the concd. acid, the time of hydrolysis was lessened somewhat. The molecular proportions of sulfuric acid and nitrile were also considered. In the experiment described above,

<sup>5</sup> Borsche and Böcker, *Ber.*, **36**, 4359 (1903).

<sup>6</sup> Noad, *Ann.*, **63**, 297 (1847).

<sup>7</sup> Fittica and Ramsay, *Ann.*, **168**, 250 (1878). Van Scherpenzeel, *Chem. Centr.*, **72**, II, 290 (1901).

the weight of sulfuric acid used corresponds to 8.5 molecular equivalents of sulfuric acid per equivalent of nitrile. It was found that even when the sulfuric acid used (d., 1.52) contained as small a quantity of sulfuric acid as 1.5 molecular equivalents of sulfuric acid to each equivalent of nitrile the hydrolysis was complete in one-half an hour and the yield of acid melting at 185–188° was practically quantitative.

**2-Nitro-*p*-toluylchloride**,  $C_6H_4(CH_3)(NO_2)(COCl)_2(1,2,4)$ .—This acid chloride is easily prepared by the action of phosphorus pentachloride on the acid described above, using the proportion of 50 g. of acid and 62 g. of pentachloride. After the mixture had been heated for two hours to complete the reaction the phosphorus oxychloride was removed at 120° under diminished pressure and the acid chloride purified by distillation in a partial vacuum. The following boiling points were observed: 165° at 18 mm.; 185° at 36 mm.; 183° at 32 mm.; and 181° at 30 mm. The yield was 52 g., or 95%. This acid chloride is a light yellow oil possessing a characteristic odor.

*Anal.* Calcd. for  $C_8H_8O_2NCl$ : Cl, 17.7. Found: 17.5.

**Nitroterephthalic Acid**,  $C_6H_3(NO_2)(COOH)_2(2,1,4)$ .—Noyes<sup>8</sup> attempted to prepare this acid by oxidation of nitro-*p*-xylene with potassium permanganate but without success. When, however, he employed potassium ferricyanide in alkaline solution as an oxidizing reagent the acid was obtained melting at 266–268°. The only other method described for the preparation of the acid is that recommended by Burkhardt,<sup>8</sup> and modified by later workers, namely, the nitration of terephthalic acid with a mixture of fuming nitric acid and concd. sulfuric acid. We have prepared the acid by oxidation of 2-nitro-*p*-toluic acid with potassium permanganate according to the procedure described by Johnson and Bigelow.<sup>9</sup>

Fifty g. of the toluic acid was dissolved, in a 10-liter enameled pail, with 64 g. of sodium hydroxide in 3150 cc. of water. Heat was then applied until the temperature of the solution registered 90–100°, when 98 g. of potassium permanganate (or 10% more than the calculated amount) was added in five portions and after addition of the last portion the mixture was boiled until complete decolorization of the permanganate was effected. The manganese dioxide was then separated by filtration, washed with hot water and the solution concentrated to a volume of one liter. On acidification of the alkaline solution with sulfuric acid the terephthalic acid separated in a yield of 56 g., or 96%. It was purified by crystallization from hot water; m. p., 268°.

**Nitroterephthaloyl Chloride**,  $C_6H_3(NO_2)(COCl)_2(2,1,4)$ .—One hundred g. of dry nitroterephthalic acid was mixed with 202 g. of phosphorus pentachloride when almost immediately reaction set in with evolution of hydrogen chloride and almost complete liquefaction. After the liquid had been heated at 100° to complete the reaction it was distilled under diminished pressure, yielding 110 g. of this acid chloride as a light yellow oil; b. p., 174° (8 mm.).

*Anal.* Calcd. for  $C_8H_5O_4NCl_2$ : Cl, 28.6. Found: 28.7.

**Preparation of Diethylamino-ethyl Alcohol** ( $C_2H_5$ )<sub>2</sub>NCH<sub>2</sub>CH<sub>2</sub>OH.—All of the amino alcohol used in this research was prepared according to the Ladenburg<sup>10</sup> method by heating diethylamine with ethylene chloro-

<sup>8</sup> Burkhardt, *Ber.*, **10**, 145 (1877).

<sup>9</sup> Johnson and Bigelow, *THIS JOURNAL*, **41**, 1559 (1919).

<sup>10</sup> Ladenburg, *Ber.*, **14**, 1878 (1881).

hydrin. A series of experiments was performed to determine the best conditions for obtaining a good yield of this reagent by a practical laboratory method. The reaction was carried out in a cylindrical, iron pipe autoclave. The method of operating was to mix the diethylamine, chlorohydrin and the condensing reagent with enough water to dissolve it in the autoclave, and then heat the mixture to the desired temperature, by submerging the autoclave in an oil-bath. After the reaction was complete and the autoclave had cooled, the reaction mixture was extracted with ether, the latter dried over anhydrous sodium sulfate and then distilled. A large fraction boiling between 159° and 163° was always easily obtained. The boiling point of the amino alcohol is given in the literature as 160–161° at ordinary pressure.

As a result of many experiments, in which we varied the temperature of reaction, time of heating and the catalyst used for accelerating condensation, we concluded that sodium hydroxide in water serves as the most efficient condensing agent; that heating for six hours at 110–120°, or three hours at 160°, is the most favorable temperature condition and finally that by using an excess of diethylamine the yield of amino alcohol is greatly increased. The results of a series of three experiments are recorded in Table I.

TABLE I  
EFFECT OF VARIATION IN THE PROPORTION OF AMINE

Expt.	Time Hours	Temp. °C.	Condensing agent NaOH	Chloro- hydrin G.	Diethyl- amine G.	Equiv.	Yield of amino alcohol	
							G.	%
1	6	110–120	10 g. in H <sub>2</sub> O	17	22.6	1.5	17	70
2	6	110–120	10 g. in H <sub>2</sub> O	17	30.1	2	19	77
3	3	160	15 g.	25.5	45.2	2	30	83

**Diethylamino-ethyl 2-Nitro-*p*-toluate**, C<sub>6</sub>H<sub>3</sub>(CH<sub>3</sub>)(NO<sub>2</sub>)(COOCH<sub>2</sub>CH<sub>2</sub>N(C<sub>2</sub>H<sub>5</sub>)<sub>2</sub>)<sub>2</sub>—(1, 2, 4).—This ester is obtained in the form of its hydrochloric acid salt by allowing diethylamino-ethyl alcohol to interact with 2-nitrotoluy chloride in anhydrous ether at ordinary temperature. The yield was practically quantitative. The salt was purified for analysis by crystallization from 95% alcohol and melted at 152–153° with decomposition.

*Anal.* Calcd. for C<sub>14</sub>H<sub>21</sub>O<sub>4</sub>N<sub>2</sub>Cl: Cl, 11.20. Found: 11.08.

This salt crystallizes as needles and is very soluble in cold water, hot alcohol and acetone, but is insoluble in ether, benzene and toluene. When the salt is treated with alkali the ester separates as an oil.

**Diethylamino-ethyl 2-Nitroterephthalate**, C<sub>6</sub>H<sub>3</sub>(NO<sub>2</sub>)(COOCH<sub>2</sub>CH<sub>2</sub>N(C<sub>2</sub>H<sub>5</sub>)<sub>2</sub>)<sub>2</sub>—(2, 1, 4).—This ester was prepared from diethylamino-ethyl alcohol and the acid chloride of nitroterephthalic acid according to the procedure described in the preceding experiment. The yield of the hydrochloride of the ester was practically quantitative. When purified by crystallization from 95% alcohol the substance melted at 199°.

*Anal.* Calcd. for C<sub>20</sub>H<sub>33</sub>O<sub>6</sub>N<sub>3</sub>Cl<sub>2</sub>: Cl, 14.71. Found: 14.53.

The salt crystallizes from alcohol in the form of needles. It is very soluble in acetone and cold water, but extremely insoluble in ether and benzene. When the salt is decomposed by alkali the free ester separates as an oil.

**Diethyl Nitroterephthalate**,  $C_6H_3(NO_2)(COOC_2H_5)_2(2,1,4)$ .—This was prepared by interreaction of ethyl alcohol with the acid chloride of nitroterephthalic acid. The ester crystallizes from dil. alcohol in the form of needles; m. p.,  $51^\circ$ . It is soluble in benzene, alcohol, ether and acetone, but insoluble in water.

*Anal.* Calcd. for  $C_{12}H_{13}O_6N$ : N, 5.24. Found: 5.10.

**Diethylamino-ethyl 2-Amino-*p*-toluate**,  $C_6H_3(CH_3)(NH_2)(COOCH_2CH_2N(C_2H_5)_2)(1, 2, 4)$ .—This ester was obtained in the form of its hydrochloride by reduction of the *p*-nitrotoluate described above with tin and hydrochloric acid. Fifty g. of the nitro ester hydrochloride, 375 cc. of water and 28 g. of tin were used in each experiment and the reduction brought about with 300 cc. of concd. hydrochloric acid at  $35^\circ$ . After the reduction was complete the tin was precipitated in the usual manner as tin sulfide, the concentrated filtrate made alkaline and the amino ester extracted with ether after making alkaline. After the liquid was dried over anhydrous sodium sulfate and the ether evaporated, we obtained 32 g. of ester as an oil; yield, 72%. The hydrochloride was prepared by bubbling dry hydrogen chloride through an ether or benzene solution of the ester. The product was then purified by crystallization from 95% alcohol and melted at  $208^\circ$  with decomposition.

*Anal.* Calcd. for  $C_{14}H_{24}O_2N_2Cl_2$ : Cl, 21.95. Found: 21.95.

This compound crystallizes from absolute alcohol as cubes. It is very soluble in water, moderately soluble in warm alcohol and acetone and insoluble in ether and benzene. It is not hygroscopic.

**Diethylamino-ethyl Amino-terephthalate**,  $C_6H_3(NH_2)(COOCH_2CH_2N(C_2H_5)_2)(2, 1, 4)$ .—Using 50 g. of nitroterephthalate ester the reduction was conducted according to the procedure described for the previous experiment. After removing the tin in the usual manner and extracting the ester from an alkaline solution with ether we obtained 32 g. of the crude amino ester; yield, 83%. The hydrochloride was prepared by saturating a benzene solution of the ester with dry hydrogen chloride. The product was purified by crystallization from absolute alcohol and separated as needles, melting at  $213^\circ$  with decomposition. The dihydrochloride is very soluble in water, warm alcohol and acetone, and insoluble in ether and benzene. Dilute aqueous solutions of this salt give a very beautiful blue fluorescence.

*Anal.* Calcd. for  $C_{20}H_{33}O_4N_3Cl_2$ : Cl, 15.69. Found: 15.40.

The pharmacological study of these new diethylamino-ethyl esters described above is in progress.

### Summary

1. Improved methods for the preparation of 2-nitro-*p*-toluic acid and nitroterephthalic acid have been developed. The starting point for the synthesis of both acids is *p*-toluidine.

2. The diethylamino-ethyl alcohol esters of these two acids have been prepared and reduced to their corresponding amino compounds. The latter compounds are physiologically active and exhibit the characteristic properties of local anesthetics of the novocaine type.

3. The results of a pharmacological study of these combinations will be reported later.

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