

experiment during the course of which, at three time intervals, samples were removed for ultimate analysis showed that the residue, except for change in color, had altered only slightly during a 773-hour heating, having changed from C = 59.58 and H = 8.17 to C = 59.32 and H = 7.74. For these reasons it appears quite probable that the main course of the reaction is such as to favor the formation of a polymerization product. Further support for this conclusion may be found in the fact that the observed vapor pressures<sup>1</sup> when measured by a dynamic method<sup>2</sup> were lower than the calculated at any given temperature above that at which this compound begins to decompose. This is to be expected were a polymerization takes place.

The static method<sup>3</sup> of measuring pressure-temperature relationships which was employed is very susceptible to the influence of gaseous decomposition products even though their quantity may be small. An immediate effect of this characteristic is to be found in the higher values which were obtained by this method of measurement. The foregoing observations account, it seems, for the discrepancies which were previously noted in vapor pressure measurements by two different methods of approach.

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### Preparation and Properties of the Normal Barium Salt of *l*-Cystine.<sup>1</sup>

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Several normal metallic salts of cystine have already been described. In 1900, Embden<sup>2</sup> prepared the normal copper salt and employed it in the isolation of the amino acid. A little later, Neuberg and Mayer<sup>3</sup> prepared the corresponding salts of silver, mercury, lead and cadmium as well as of copper. Recently, Toennies and Lavine<sup>4</sup> have succeeded in obtaining the lithium, sodium and potassium salts of cystine in crystalline form.

In the course of a study of the alkaline decomposition of cystine, which will be presented in a separate paper, it was observed while adding crystalline barium hydrate to a hot aqueous suspension of *l*-cystine that most of the cystine went into solution at a certain stage and that immediately thereafter a pale yellow precipitate separated out. By filtering off this pre-

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<sup>2</sup> Embden, *Z. physiol. Chem.*, **32**, 94 (1901).

<sup>3</sup> Neuberg and Mayer, *ibid.*, **44**, 498 (1905).

<sup>4</sup> Toennies and Lavine, *J. Biol. Chem.*, **90**, 203 (1931).

precipitate at once, washing it with a small amount of water, then with alcohol and ether and finally drying at 105°, a product was obtained (Preparation I) which analyzed fairly well for the normal barium salt of cystine.

Since no mention of this substance could be found in the literature and since it is involved in studies dealing with the decomposition of cystine by alkalis, it was thought desirable to prepare the salt in pure form and to determine its more important properties.

**Method of Preparation.**—A very satisfactory method consists in mixing equivalent amounts of aqueous Ba(OH)<sub>2</sub> and cystine in the cold and precipitating the resulting salt with alcohol. More exactly, 16 g. of Ba(OH)<sub>2</sub>·8H<sub>2</sub>O was dissolved in 250 cc. of boiled distilled water and cooled to 0°. Then 12 g. of the *l*-cystine was added and the mixture shaken in a stoppered Erlenmeyer flask until only very little undissolved material remained. The cold mixture was filtered through a Buchner funnel and after washing with distilled water the slight residue was discarded. The combined filtrate and washings, amounting to about 300 cc., were treated with 1600 cc. of 95% ethyl alcohol, which caused abundant precipitation of a white granular material. After thorough shaking, this mixture was placed in a refrigerator at -15° overnight. In the morning, the precipitate was filtered off with suction, washed with 600 cc. of 95% ethyl alcohol, then with ether and finally dried in an air oven at 100°. The yield was 18.1 g., which is 96.5% of the theoretical calculated from the cystine taken.

**Properties.**—The above product (Preparation II) was a perfectly white, finely divided powder which under the microscope showed no clearly defined crystalline form.

**Anal.** Calcd. for C<sub>4</sub>H<sub>10</sub>O<sub>4</sub>N<sub>2</sub>S<sub>2</sub>Ba: N, 7.46; S, 17.07; Ba, 36.57. Found: N (Prepn. I), 7.32, (Prepn. II), 7.31; S (I), 16.20, (II) 16.65; Ba (I), 35.78; (II) 36.42.

A saturated solution of the salt was prepared by shaking 4 g. with 50 cc. of distilled water, allowing it to stand overnight at 25°, and filtering to remove excess salt. The saturated solution was very faintly yellow in color but seemed to be perfectly stable under the conditions of observation. Forty cc. of this solution was diluted to 200 cc. with water and aliquots were taken for nitrogen, sulfur and barium analyses to determine solubility, as indicated. This new salt of cystine then is water soluble at 25° to the extent of 6.15 g. per 100 cc. of saturated solution.

#### SOLUBILITY OF THE NORMAL BARIUM SALT OF *l*-CYSTINE IN WATER AT 25°

Basis of calculation.....	Nitrogen content	Sulfur content	Barium content	
Barium salt in 100 cc. of satsd. soln.....	6.14	6.17	6.14	Av. 6.15

When placed in a 1-decimeter tube, the saturated solution gave a saccharimeter reading of -11.0° at 25° and on dilution to five times its volume with water a proportionate value of -2.2°. Calculated on the basis of concentration of the barium salt, this gives a specific rotation of -61.9°; or calculated for cystine contained in the salt, of -96.9°. The latter agrees quite well with similar figures reported by Toennies and Lavine<sup>4</sup> for the lithium, sodium and potassium salts of cystine. Free cystine was isolated from some of the salt and 0.2 g. of this dissolved in 25 cc. of normal hydrochloric acid at 25°, when placed in a 1-decimeter tube, gave a reading of -4.6° saccharimeter units. The corresponding specific rotation of -199.1° as compared with -204.2° for the original cystine indicates that racemization had not occurred to any considerable extent during the formation of the barium salt. Hence, it is safe to conclude that Preparation II consisted almost entirely of the barium salt of *l*-cystine.

If the saturated aqueous solution of the substance is diluted sufficiently with water,

hydrolysis takes place and a portion of the cystine precipitates out in the form of hexagonal plates. That hydrolysis had not taken place in preparing the saturated solution is clearly shown by the very close agreement between solubilities calculated from contents of nitrogen and sulfur on the one hand and barium on the other.

When the dry salt was heated in a melting point tube it showed no signs of decomposition or fusion up to about 250°; then it began to brown very gradually but without sintering.

When a portion of the salt equivalent to 2.5 g. of cystine was refluxed with 50 cc. of distilled water for twelve hours, nitrogen and sulfur analyses made after the removal of free hydrogen sulfide and ammonia indicated that decomposition to the extent of approximately 50% had taken place.

In its property of being water soluble, the barium salt of *l*-cystine resembles the alkali salts and differs from the heavy metal salts so far described. This characteristic as well as its ease of preparation and the ease of determination of its metallic constituent should make the barium salt useful in further studies on the mechanism of the alkaline decomposition of cystine. It might also find use in the preparation of derivatives of cystine, particularly in cases where the reaction requires anhydrous conditions.

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## The Structure of the Bromination Product of Ortho-Nitrotoluene

BY DAVID L. YABROFF

Wachendorff<sup>1</sup> obtained a dibromo product upon the bromination of *o*-nitrotoluene which he called a dibromonitrotoluene. He made the observation, however, that this product was soluble in alkali. Greiff<sup>2</sup> showed that the compound was a dibromo-anthranilic acid and suggested that it might be the *p*-*m*-dibromo-anthranilic acid obtained by Hübner<sup>3</sup> upon the nitration and reduction of *p*-*m*-dibromobenzoic acid since both of the products melted at 225°.

If an intramolecular oxidation and reduction of the nitrotoluene occurs before the bromination, we should expect 3,5-dibromo-2-aminobenzoic acid as the final product. If bromination occurs before the oxidation-reduction process, we should expect 4,6-dibromo-2-aminobenzoic acid to be formed. These two compounds may be readily distinguished by allowing them to react for twenty-four hours in a dilute alcoholic solution with bromine water in the presence of a mineral acid. Under these conditions<sup>4,5</sup> all positions ortho or para to the amino group which are not already substituted are brominated, and at the same time bromine is substituted for the carboxyl group. This may be represented as

<sup>1</sup> Wachendorff, *Ann.*, **185**, 259 (1877).

<sup>2</sup> Greiff, *Ber.*, **13**, 288 (1880).

<sup>3</sup> Hübner, *ibid.*, **10**, 1706 (1877).

<sup>4</sup> Francis and Hill, *THIS JOURNAL*, **46**, 2498 (1924).

<sup>5</sup> Flürscheim and Holmes, *J. Chem. Soc.*, 448 (1928).