2-Benzyl-4-chloro-6-dimethylaminomethylphenol.—Using 219 g. (1.0 mole) of 2-benzyl-4-chlorophenol, 190 ml. of aqueous dimethylamine and 80 ml. of formalin, the Mannich reaction was carried out as above. On neutralization of the acid extracts, the Mannich base separated as a solid. It was extracted into benzene (its solubility in ether was very low), and the benzene solution dried by shaking with saturated sodium chloride and filtering through anhydrous sodium sulfate. The benzene was evaporated and the crude product crystallized from methanol, giving 209 g. (76% yield) of 2-benzyl-4-chloro-6-dimethylaminomethylphenol, m.p. 84.5–86.5°. Repeated recrystallization from methanol sharpened the m.p. to 85.5–86.5°.

Anal. Calcd. for $C_{16}H_{18}ClNO$: C, 69.7; H, 6.6. Found: C, 69.7; H, 6.7.

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The Acidity Constant, Solubility Product and Solubility of Dithioöxamide[1]

BY RUTH POWERS YAFFE AND ADOLF F. VOIGT

RECEIVED FEBRUARY 4, 1952

Dithioöxamide, frequently called rubeanic acid, is a well-known complexing agent of the platinum group metals. It is readily soluble in alkaline solutions and forms salts which have been called rubeanates. Although the acidic behavior of dithioöxamide is well known, no determination of its acidity constant has been reported.

The method followed for this determination was the potentiometric titration procedure for acids of limited solubility given by Back and Steenberg.[2]

The potentiometric titrations were made with a Beckman Model G *p*H meter equipped with a #1190-E High *p*H Glass Electrode. A known quantity of dithioöxamide was dissolved in a known excess volume of standard base. This solution was then titrated with a standard acid. At the beginning of the titration the weak acid was in the form of its soluble salt in a slight excess of base. After this excess base was neutralized, a known concentration, C_s, of titrating acid was added and the *p*H measured. This process was repeated with about five additions of acid before flocculation occurred. From each of these measurements a value of pK_a was calculated by means of equation (1)

$$pK_a = pH + \log \left\{ \frac{C_s + C_{OH} - C_H}{C_a + C_H - C_s - C_{OH}} \right\} + \frac{0.5\sqrt{\mu}}{1 + \sqrt{\mu}} \quad (1)$$

where C_a is the total concentration of the weak acid [HA + A^-]. The titration was continued through the stage of flocculation, about five more measurements being obtained before salting out of the dithioöxamide at the reference electrode caused the results to be inconsistent. These data were used in equation (2) to calculate the solubility product.

$$pK_{sp} = pH - \log \{C_s + C_H - C_s - C_{OH}\} + \frac{0.5\sqrt{\mu}}{1 + \sqrt{\mu}} \quad (2)$$

The solubility of the acid is given by the relation

$$S = K_{sp}/K_a \quad (3)$$

It was found that the dithioöxamide behaved as a monobasic acid. Equations (1) and (2) give the thermodynamic constants since they contain an expression for the activity coefficient of the acid ion, and the hydrogen ion activity is determined directly. The values of μ used in the equations were calculated for each point and were approximately 0.01. In each titration, 5 or 6 individual determinations of K_a, K_{sp} and S were obtained. In all, five complete titrations were carried out. The over-all averages of these 25 to 30 determinations and their standard deviations are: for the acidity constant, K_a, $(1.28 \pm 0.04) \times 10^{-11}$, for the solubility product, K_{sp}, $(3.94 \pm 0.12) \times 10^{-14}$ and for the solubility, S, $(3.07 \pm 0.04) \times 10^{-3}$ mole per liter.

In addition to these thermodynamic constants, the non-thermodynamic constants at an ionic strength of 1.0 were desired. Sodium perchlorate, made by dissolving sodium carbonate in perchloric acid, was used as the inert electrolyte. Three complete titrations were made, and the results averaged. The non-thermodynamic constants may be summarized as follows: $k_a = (3.78 \pm 0.04) \times 10^{-11}$; $k_{sp} = (6.27 \pm 0.14) \times 10^{-14}$; $s = (1.66 \pm 0.03) \times 10^{-3}$ mole per liter.

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(1) Contribution No. 171 from the Institute for Atomic Research and the Department of Chemistry, Iowa State College, Ames, Iowa. Work performed in the Ames Laboratory of the Atomic Energy Commission.

(2) E. Back and B. Steenberg, *Acta Chem. Scand.*, **4**, 810 (1950).

NEW COMPOUNDS

Derivatives of *p*-Aminosalicylic Acid[1,2]

Two derivatives of *p*-aminosalicylic acid have been prepared.

Methyl N-(2-Hydroxyethyl)-*p*-aminosalicylate.—To a vigorously stirred solution of 33.4 g. (0.2 mole) of methyl *p*-aminosalicylate (prepared from commercial *p*-aminosalicylic acid[3]) and 700 ml. of anhydrous ether in a one-liter flask surrounded by an ice-bath were added simultaneously over a period of 30 minutes 61 ml. (0.1 mole) of a 10% solution of ethylene oxide in anhydrous ether and 40 ml. of a 2% solution of boron trifluoride etherate in anhydrous ether. After two hours of additional stirring, the ice-bath was removed and stirring continued for 30 minutes. The mixture was then heated at reflux temperature for one hour, transferred to a separatory funnel and extracted quickly with three 150-ml. portions of 10% sodium hydroxide solution. The extracts were combined, treated with carbon dioxide, and filtered at intervals to obtain the precipitate in several fractions. These were dried in a vacuum desiccator, dissolved in anhydrous ether, filtered from any carbonates, and the ether evaporated. Each fraction was a mixture of methyl *p*-aminosalicylate (I) and methyl N-(2-hydroxyethyl)-*p*-aminosalicylate (II), the latter being present in increasing proportions in successive fractions. By fractional sublimation at 0.02–0.05 mm. pressure 24.1 g. of I and 2.9 g. (37%) of II were obtained. From a mixture of benzene and petroleum ether II was obtained as a white crystalline solid, m.p. 90°.

Anal. Calcd. for $C_{10}H_{13}NO_4$: N, 6.63. Found: N, 6.53.

N-(2-Hydroxyethyl)-*p*-aminosalicylic Acid.—In a pear-shaped 25-ml. flask a mixture of 0.75 g. (0.0036 mole) of methyl N-(2-hydroxyethyl)-*p*-aminosalicylate and 4.27 ml. (0.0106 mole) of 10% sodium hydroxide solution were re-

(1) From the M. S. thesis of E. Kenneth Brakebill, June, 1951.

(2) This work was supported in part by a Research Corporation Grant-in-Aid.

(3) J. J. Schaefer and Leonard Doub, THIS JOURNAL, **71**, 3564 (1949).

fluxed for 1.5 hours, cooled, filtered, and made acid to congo red with 15% hydrochloric acid. The N-(2-hydroxyethyl)-*p*-aminosalicylic acid was filtered and dried in a vacuum desiccator; yield 0.14 g. (25%); m.p. 126°.

Anal. Calcd. for $C_9H_{11}NO_4$: N, 7.11. Found: N, 6.89.

The filtrate was made alkaline and saturated with carbon dioxide yielding 0.11 g. of methyl N-(2-hydroxyethyl)-*p*-aminosalicylate.

Hydrolysis of methyl *p*-aminosalicylate under identical conditions gave a 61% yield of *p*-aminosalicylic acid.

DEPARTMENT OF CHEMISTRY
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RECEIVED FEBRUARY 4, 1952

Vanadium Monoboride

A vanadium boride of the composition VB was prepared by simultaneous reduction of V_2O_5 and B_2O_3 with carbon. The reaction was carried out under a protective atmosphere of hydrogen by heating the well mixed and pelleted ingredients in a graphite crucible at 3000°F.

Anal. V, 77.6; B, 16.7; C, 0.07. V/(V + B) ratio found 17.7; calcd., 17.5.

Crystal Structure.—Using X-ray diffraction, the compound was found to be isomorphous with CrB[1]. The X-ray techniques employed were the same as those used by J. T. Norton and co-workers[2].

The VB structure is orthorhombic with four molecules per unit cell. The lattice constants were calculated to be for *a* 3.10 Å., *b* 8.17 Å., *c* 2.98 Å., the calculated density is 5.44 g./cc.

Electrical Resistivity.—65.5 microhm-cm. for a hot pressed piece of 65% of the theoretical density. The resistivity of a dense specimen would probably be between 35 and 40 microhm-cm., making VB a metallic conductor.

Work done under Contract with the Office of Naval Research.

(1) S. J. Sindeband, *Transactions of A.I.M.E.*, 185, 198 (1949).

(2) J. T. Norton, H. Blumenthal and S. J. Sindeband, *ibid.*, **185**, 749 (1949).

AMERICAN ELECTRO METAL CORPORATION
YONKERS, NEW YORK H. BLUMENTHAL

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COMMUNICATIONS TO THE EDITOR

THE NATURE OF THE ACTIVE METHYL DONOR FORMED ENZYMATICALLY FROM L-METHIONINE AND ADENOSINETRIPHOSPHATE[1,2]

Sir:

The participation of ATP in the enzymatic transmethylation reaction in which methionine is the methyl donor is well established.[3-5] As has been shown earlier, the role of ATP in such reactions is related to the activation of methionine,[6] as described by equation 1.

L-Methionine + ATP ⟶
Active methionine + orthophosphate (1)

The enzyme catalyzing this reaction has been partially purified, using rabbit liver as its source. The most significant property of active methionine is its ability to function as a methyl donor, even in the absence of ATP. Originally it had been assumed that the role of ATP in the activation reaction was to serve as a source of phosphate bond energy. However, the elucidation of the chemical nature of active methionine which is described below suggests that, regardless of the intermediate steps involved, ATP functions in the activation process in a novel and unexpected way as a donor of its adenosine moiety.

Active methionine, prepared enzymatically, has been purified from the deproteinized reaction mixture by (a) removal of Mg^{++} as Mg pyrophosphate at *p*H 7.0; (b) precipitation of organic and inorganic phosphates with barium and 80% ethanol, *p*H 7.8; (c) paper chromatography with 80% ethanol–5% acetic acid. The location of active methionine on the paper has been greatly facilitated by the observation that when methionine-S^{35} was used for the enzymatic reaction, the intermediate was labeled with S^{35}. Methionine-2-C^{14} also yielded labeled active methionine. After elution from the paper, active methionine exhibited an ultraviolet absorption spectrum nearly identical to that of adenylic acid. On the assumption that the extinction coefficient of active methionine is equal to that of adenylic acid it was found that preparations of active methionine, obtained as above, contained, for each mole of adenine, the equivalent of 0.78 mole of pentose[7] and 0.8 mole of labile methyl groups.[8] Three fragments have been recognized after hydrolysis in 0.5 *N* HCl at 100° for 2 hours. Adenine has been identified conclusively as one of them by chromatography on paper, by ion exchange chromatography on Dowex 1[9] and by oxidation of 2,8-dihydroxyadenine with xanthine oxidase.[10] An amino acid which when chromatographed on paper with different solvents appears to be identical with homoserine is another one of the products of hydrolysis. The nature of a third fragment has not been ascertained as yet; it is a sulfur-

(1) This investigation was aided by grants from the Williams Waterman Fund for the Combat of Dietary Diseases of the Research Corporation of New York and from the American Cancer Society (recommended by the Committee on Growth of the National Research Council).

(2) Adenosinetriphosphate = ATP.

(3) H. Borsook and J. W. Dubnoff, *J. Biol. Chem.*, **171**, 363 (1947).

(4) G. L. Cantoni, *ibid.*, **189**, 203 (1951).

(5) S. Cohen, *ibid.*, **193**, 851 (1951).

(6) G. L. Cantoni, *ibid.*, **189**, 745 (1951); and in "Phosphorus Metabolism," Vol. I, Johns Hopkins Press, Baltimore, Md., 1951, p. 641.

(7) Determined by the Bial-orcinol reaction with heating for 45 minutes at 100°.

(8) The latter were determined enzymatically by guanidoacetate methylpherase, an enzyme catalyzing the reaction

(2) Active methionine + guanidoacetate ⟶ creatine + X

P. J. Vignos, Jr., and G. L. Cantoni, to be published.

(9) W. E. Cohn, *Science*, **109**, 377 (1950).

(10) H. Klenow, *Biochem. J.*, **50**, 404 (1952).