water. The water-insoluble residue was boiled up with 40 ml. of 0.05 *N* hydrochloric acid and filtered hot. The residue was washed with a little hot water on the filter. The insoluble portion was dissolved in 50 ml. of warm dilute ammonium hydroxide and the cooled solution filtered. The ammoniacal solution was evaporated to dryness and the residue dissolved in 3 l. of boiling water. The filtered solution was cooled overnight at 5°. The white crystalline precipitate was separated and dried in a vacuum over phosphorus pentoxide; yield 1.06 g. *Anal.* Calcd. for $C_5H_4O_2N_4$: N, 36.8. Found: N, 36.6.

The xanthine was identified by its conversion to bromocaffeine (m. p. 208–209°) and ethoxycaffeine (m. p. 140°) by Fischer's methods.[6] Mixed melting points with equal quantities of bromocaffeine and ethoxycaffeine prepared from caffeine[7] showed no depression.

(6) E. Fischer, "Untersuchungen in der Puringruppe," 1882–1906, pp. 462–463.

Summary

1. The structure of the aglycone from the crotonoside is confirmed as 2-oxy-6-aminopurine by its transformation to xanthine.

2. Isoguanine (2-oxy-6-aminopurine) has been obtained in crystalline form and its optical crystallographic properties are recorded.

3. An unusual example of the use of constant boiling hydrochloric acid for deamination of a compound, resistant to the action of nitrous acid, is cited.

(7) Fischer, *ibid.*, pp. 93–95.

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[CONTRIBUTION FROM THE BUREAU OF CHEMISTRY AND SOILS, UNITED STATES DEPARTMENT OF AGRICULTURE]

Some Salts of 2-Oxy-6,8-diaminopurine

BY JOSEPH R. SPIES AND THOMAS H. HARRIS, JR.

The third of the three isomeric diamino-oxypurines was first prepared by Cherbuliez and Bernhard from isoguanine,[1] by reduction of the colored diazonium coupling compound. These authors did not describe characterizing salts of the new purine, presumably due to an insufficient amount of the substance.[2]

This paper describes a method for the preparation of 2-oxy-6,8-diaminopurine, some of its properties and several salts. Table I shows the formulas of the crystalline salts which have been prepared by usual methods. All of these salts exhibit birefringence when viewed with a polarizing microscope. The sulfate of 2-oxy-6,8-diaminopurine, like that of isoguanine, retains one molecule of water of crystallization even on heating in vacuum at 139°. Unlike isoguanine, 2-oxy-6,8-diaminopurine forms salts with both acetic acid and carbon dioxide.

Experimental

Preparation of 2-Oxy-6,8-diaminopurine Sulfate.—Twelve grams of finely pulverized 2,4-dichloroaniline (1.5 mol) was diazotized in 58 ml. of concentrated hydrochloric acid by the slow addition of 5.4 g. of sodium nitrite in 7.5 ml. of water. The suspension was kept at 0–5° and stirred mechanically. The diazonium salt practically all dissolved when crushed ice was added to the acid suspension.

To 10 g. of 2-oxy-6-aminopurine sulfate, dissolved in 400 ml. of 1.2 *N* sodium hydroxide (0–10°), stirred mechanically, was added one-half the cold diazonium solution. Eighteen and one-half milliliters of 19 *N* sodium hydroxide was then added and the remaining diazonium solution was dropped in as before. The cold red solution was stirred for fifteen minutes. Carbon dioxide was passed in until the solution was neutral to phenolphthalein. The red precipitate was filtered off and washed with cold water. This precipitate was dissolved in 600 ml. of 1.5% sodium hydroxide and heated and stirred during the gradual addition of sodium hydrosulfite (approx. 180 g.). The solution, which should be kept alkaline, finally turns from red to yellow. The voluminous precipitate which formed when the cooled solution was saturated with carbon dioxide was filtered off and washed with water. (Acetic acid may be used in place of carbon dioxide.) The slightly yellow precipitate was dissolved in 1300 ml. of boiling 5% sulfuric acid, decolorized with carbon, filtered and allowed to cool to 5°. The sulfate was recrystallized from 1 liter of 5% sulfuric acid. The fine hair-like crystals were washed with water and dried at 110° in a vacuum: yields 2–4 g. Two further recrystallizations from 5% sulfuric acid produced no change in nitrogen content of the sulfate.

The Base.—Purified 2-oxy-6,8-diaminopurine sulfate was dissolved in warm dilute sodium hydroxide and precipitated from the cooled solution with carbon dioxide. The carbonate was dissolved in boiling water (about 1 g./l.) and the hot solution was filtered through a Seitz sterilizing pad to remove a little colloidal carbon. The clear solution was boiled gently for some time and allowed to cool in a flask closed with an Ascarite tube.

(1) Cherbuliez and Bernhard, *Helv. Chim. Acta*, **15**, 471 (1932); Spies, THIS JOURNAL, **61**, 350 (1939).

(2) K. Bernhard, "Recherches sur la Graine de Croton (*Croton tiglium* L.)," Thèse (Genève), 1932, described the sulfate and hydrochloride. The water content of the hydrates does not agree with that reported by us, however.

TABLE I

FORMULAS AND ANALYTICAL DATA OF SALTS OF 2-OXY-6,8-DIAMINOPURINE

	Formula	Carbon % Calcd.	Carbon % Found[c]	Hydrogen % Calcd.	Hydrogen % Found	Nitrogen % Calcd.	Nitrogen % Found	Water % Calcd.	Water % Found
		Hydrated salt							
Sulfate	$B_2 \cdot H_2SO_4 \cdot 3H_2O$[b]	24.8	25.1	4.16	3.98	34.7	35.1		
Hydrochloride	$B \cdot HCl \cdot 1.5H_2O$	26.2	26.5	4.39	4.29	36.6	36.6		
Acetate	$B \cdot C_2H_4O_2 \cdot 3.5H_2O$	29.1	28.8	5.93	6.03	29.1	28.9		
Carbonate[d]	$B_2 \cdot H_2CO_3 \cdot H_2O$	32.0	31.6	3.91	4.47	40.8	40.8		
Picrate[e]	$B \cdot C_6H_3O_7N_3 \cdot 0.5H_2O$	32.7	33.2	2.49	2.54	31.2	31.0		
		Heated salt[a]							
Sulfate	$B_2 \cdot H_2SO_4 \cdot H_2O$ (139°)	26.8	26.5	3.60	3.61	37.5	37.6	7.44	7.47
Hydrochloride	$B \cdot HCl$ (110°)	29.6	30.2	3.49	3.56	41.5	41.3	11.8	11.9
Acetate	$B \cdot C_2H_4O_2$ (80°)					37.2	37.2	21.8	22.0
Carbonate	$B_2 \cdot H_2CO_3$								
Picrate	$B \cdot C_6H_3O_7N_3$ (110°)							2.23	2.39

[a] The hydrated salts were heated *in vacuo* to constant weight at the temperature following the formula. [b] B = ($C_5H_5ON_5$). [c] In all cases, average values are reported where duplicate analyses were obtained. [d] Prepared by allowing a hot solution of the base to cool in a current of carbon dioxide. Crystallizes as needles which slowly lose carbon dioxide on heating *in vacuo*. [e] Crystallizes as orange plates and yellow spheroids.

Ninety-three milligrams of the base so obtained was dissolved in 1300 ml. of boiling water, filtered hot, and the solution boiled gently for about one hour. The solution was then cooled to 5° in a flask closed with an Ascarite tube. The supernatant was decanted and the solid obtained by centrifuging. The amorphous solid was dried *at once* in a vacuum over phosphorus pentoxide and potassium hydroxide. After this preliminary drying, it was heated in a vacuum at 139° for fifteen minutes; yield, 0.81 g. of almost white solid. For analysis, the dried sample must be weighed in a closed system as it gains weight on exposure to air.

Anal. Calcd. for $C_5H_5ON_5$: N, 50.6. Found: N, 50.4.

Optical Properties of the Acetate.—The acetate was prepared by cooling a hot 5% acetic acid solution of the pure base.

Microscopical examination[3] of the crystals of 2-oxy-6,8-diaminopurine acetate hydrate ($C_5H_5ON_5 \cdot CH_3COOH \cdot 3.5H_2O$) showed that they consisted of colorless clear-cut rhombs. In parallel polarized light (crossed nicols), the plates extinguish sharply, showing abnormal interference colors, consisting of peculiar shades of red, blue and purple. The birefringence is strong, $n_\gamma - n_\alpha = 0.239$. Because all the plates extinguish sharply when examined with crossed nicols, interference figures would not be expected in convergent polarized light, indicating that β is more or less perpendicular to their broad faces. The refractive indices are n_α 1.430 (common); n_β indt.; n_γ 1.669 (common) ± 0.002 (immersion method).

(3) The authors are indebted to George L. Keenan of the Food and Drug Administration for the optical crystallographic determinations.

Summary

1. The formulas of several crystalline salts of 2-oxy-6,8-diaminopurine are reported.

2. A method for preparing 2-oxy-6,8-diaminopurine from isoguanine is described.

3. The optical crystallographic properties of 2-oxy-6,8-diaminopurine acetate hydrate are recorded.

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