was present in the reaction of α-methyl glucoside and ketene, acetylation progressed nearly to completion.

Pyridine is not a satisfactory solvent for the glucose–ketene reaction. Wollenberg's compound, $C_{13}H_{11}O_3N$, formed by reaction of ketene and pyridine, is not formed by reaction of pyridine with either acetylketene or dehydroacetic acid.

EVANSTON, ILLINOIS RECEIVED NOVEMBER 25, 1938

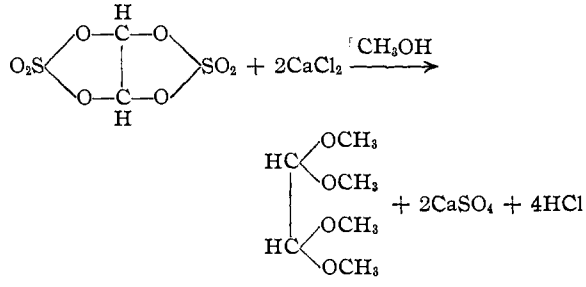
[CONTRIBUTION FROM THE RESEARCH LABORATORY OF ORGANIC CHEMISTRY, MASSACHUSETTS INSTITUTE OF TECHNOLOGY, NO. 179]

The Preparation and Estimation of Glyoxal Tetramethyl Acetal

BY D. H. GRANGAARD AND C. B. PURVES

Although the tetraethyl acetal of glyoxal has been synthesized in various ways,[1-3] the tetramethyl acetal apparently has never been isolated in a pure condition.[2] We required a specimen of the latter for reference purposes[4] and to obtain it we adapted the synthesis used by Baker and Field[3] for the former.

Acetylene tetrachloride and 65% oleum readily gave the white, insoluble sulfate of glyoxal in about 30% yield.[5] A reaction occurred when this was heated in absolute methyl alcohol containing calcium chloride and the result was a nearly quantitative formation[6] of glyoxal tetramethyl acetal.



This method was also valid for other acetals in the series.

Glyoxal tetramethyl acetal was a mobile, colorless liquid, b. p. 159°, with the interesting solubility and volatility characteristics described in the experimental portion. The acetal was easily and quantitatively hydrolyzed to glyoxal by hot, dilute mineral acid and was therefore estimated readily as glyoxal *bis*-2,4-dinitrophenylhydrazone.[7] Smaller amounts could be determined colorimetrically, also as glyoxal, by a method involving Benedict's uric acid reagent (arsenophosphotungstic acid),[8] provided certain precautions were taken. The pure acetal served as an excellent source for standard glyoxal solutions, since it was easy to prepare and its aqueous solutions were stable for months.

Experimental

Glyoxal Tetramethyl Acetal.—The glyoxal sulfate was used without further purification. One mole (30.8 g.) was dissolved in 800 cc. of cold absolute methanol containing two moles (31.4 g.) of anhydrous calcium chloride. After boiling for three hours under a reflux condenser, the solution was set aside for about twelve hours to allow the very finely divided precipitate of calcium sulfate to settle. The decanted mother liquor, together with the methanol washings from the precipitate, was made slightly alkaline with sodium methylate solution and then diluted with an equal volume of water. When this aqueous alcoholic system was fractionally distilled through an efficient column, the methyl alcohol volatilized without carrying over any of the product, which was recovered in the first 750 cc. of the aqueous fraction. Sodium chloride (225 g.) was dissolved in this fraction prior to a twelve-hour extraction with ether in an efficient continuous extractor. The ethereal extract was dried over anhydrous magnesium sulfate, the solvent evaporated, and the slightly yellow liquid residue (16.7 g. or 79%,[9] $n^{20}D$ 1.3998) distilled under diminished pressure. The pure acetal boiled at 98–100° (110 mm.).

Anal. Calcd. for $C_2H_2(OCH_3)_4$: C, 47.97; H, 9.39; OCH_3, 82.61; mol. wt., 150.1; *MR*, 36.48. Found: C, 48.04, 47.96; H, 9.44, 9.33; OCH_3, 82.08, 81.98; mol. wt. (dioxane), 145, 146; *MR*, 36.72.

(1) Pinner, *Ber.*, **5**, 151 (1872).

(2) Harris and Temme, *ibid.*, **40**, 171 (1907). From sodium ethoxide and the diethyl acetal of dichloracetaldehyde. The two corresponding methyl derivatives gave a product so volatile that it could not be separated from the excess methyl alcohol.

(3) Baker and Field, *J. Chem. Soc.*, 86 (1932).

(4) We have been studying the isolation of glyoxal, as the tetramethyl acetal, from various oxy-starches and oxycelluloses for the past eighteen months. In the meantime, Davidson [*J. Text. Inst.*, **29**, T 215–218 (1938)] has independently developed our exact line of thought in this connection.

(5) Ruggli and Henzi, *Helv. Chim. Acta*, **12**, 362 (1929); M. A. Perkins, U. S. Patent 1,999,995 (1935); British Patent 447,135 (1936).

(6) *Cf.* the 13% yield of the tetraethyl acetal obtained by Baker and Field[3] without the calcium chloride. These authors preferred the structural formula used here for glyoxal sulfate.

(7) (a) Glasstone and Hickling, *J. Chem. Soc.*, 824 (1936); (b) Neuberg and Simon, *Biochem. Z.*, **256**, 485–491 (1932).

(8) Ariyama, *J. Biol. Chem.*, **77**, 359 (1928).

(9) Some of the product was lost in the dry-ice traps.

Glyoxal tetramethyl acetal was a mobile, water-clear liquid with the following physical constants: b. p. 158–159° (754 mm.); $n^{20}D$ 1.4010; $n^{25}D$ 1.3985; d^{25}_{25} 0.987$_6$. It was readily soluble in the common organic solvents including petroleum ether, and was not easily separated from aqueous or alcoholic solutions by distillation. This separation was possible when ether was the solvent. Ether was also capable of extracting the acetal from water although the partition coefficient was heavily in favor of the latter. The tetramethyl acetal did not reduce hot Fehling's solution. It gave the Ariyama[8] color test for glyoxal only after previous hydrolysis with dilute mineral acid.

Estimation of Glyoxal Tetramethyl Acetal.—The stock solution used was prepared by dissolving 0.3913 g. in 1 liter of distilled water. It was kept in a stoppered flask at 0°.

(a) **With 2,4-Dinitrophenylhydrazine.**[7]—Aliquots (50 cc.) of the stock solution were heated with an equal volume of 2 *N* hydrochloric acid for fifteen minutes on the steam-bath. The glyoxal formed in the hydrolysis was weighed as the *bis*-hydrazone[10] of the above reagent. Calcd. for 19.57 mg. of tetramethyl acetal or 7.56 mg. of glyoxal, 54.5 mg. Found, 53.4, 53.6 and 54.0, 54.7 mg. six months later. The acetal thus gave the quantitative amount of glyoxal.

(b) **By Ariyama's Method.**—The above stock solution was diluted (1:20) to contain 1.965 mg. of the acetal in 100 cc. and 5-cc. aliquots of this were hydrolyzed as in (a) for varying times. Quick cooling was followed by the successive addition of 2 cc. of Benedict's uric acid reagent,[11] 1 cc. of *N* sodium cyanide and 25 cc. of *N* sodium carbonate. After standing for five minutes, the mixtures were diluted to 100 cc. and the depths of the blue colors compared. The hydrolysis was thus found to be 48, 93, 100 and 100% complete after 2.5, 5, 10 and 60 minutes of heating. No detectable hydrolysis of the acetal occurred during ten minutes when the acid was cold nor in six months when distilled water was the solvent. In carrying out an estimation of glyoxal or of the tetramethyl acetal, 5-cc. samples of the unknown and the standard were hydrolyzed as above for fifteen minutes, the other reagents added, and after suitable dilution the final color comparison was made in an Ernst Leitz Wetzler No. 461 colorimeter. With our instrument, the comparison was accurate to within 5% provided the concentration of the unknown lay within the limits indicated in Table I for the standards used. Standards whose concentrations lay outside the 0.05–0.75 mg./100 cc. range were quite unsuitable for comparative purposes.

(10) The *bis*-2,4-dinitrophenylhydrazone had the correct m. p. of 318° but accurate analyses were only occasional, owing to the combustion difficulties reported by others.[7a]

(11) Benedict, *J. Biol. Chem.*, **51**, 189 (1922).

TABLE I

PERMISSIBLE DEVIATIONS OF STANDARD AND UNKNOWN SOLUTIONS

Concn. of standard Mg. glyoxal/100 cc.	Concn. of unknown Mg. glyoxal/100 cc. Minimum	Maximum
0.05	0.04	0.06
.25	.13	.48
.40	.20	.76
.60	.30	.76
.75	.37	.76

Summary

1. Glyoxal sulfate, readily prepared from acetylene tetrachloride and oleum, gave a nearly quantitative yield of glyoxal tetramethyl acetal when heated with calcium chloride dissolved in absolute methyl alcohol. The new acetal had the following physical constants: b. p. 159° (754 mm.); 98–100° (100 mm.); $n^{20}D$ 1.4010; $n^{25}D$ 1.3985; d^{25}_{25} 0.987$_6$.

2. The tetramethyl acetal gave on acid hydrolysis a quantitative yield of glyoxal and could be estimated by methods used for the latter.

3. The acetal was an accessible, stable and satisfactory standard in the colorimetric determination of glyoxal.

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[A COMMUNICATION FROM THE LABORATORY OF ORGANIC CHEMISTRY OF THE UNIVERSITY OF WISCONSIN]

Oxidation of Methyne and Methylene Groups by Ozone

BY JOHN R. DURLAND[1] AND HOMER ADKINS

Recently an attempt was made to determine with ozone the position of unsaturation in a hydrophenanthrene containing one double bond.[2] These experiments were made upon the basis of the naïve assumption that ozone was a rather specific reagent for alkene linkages. The chief product of the ozonization of the unsaturated hydrophenanthrene was an unsaturated ketone, produced by the oxidation of a methylene group, the alkene linkage remaining intact. It therefore seemed advisable to ozonize representative saturated cyclic hydrocarbons. As a result of these experiments there have been obtained saturated alcohols, ketones and acids, and even more surprisingly, unsaturated ketones and hydrocarbons. The yields, in several cases, have been from 20 to 35% of the theoretical.

Among the saturated compounds, cyclohexane

(1) At various times during the progress of this work Mr. Durland has been a teaching assistant of the University, a research assistant supported by the Wisconsin Alumni Research Foundation and a Fellow supported by the Monsanto Chemical Company.

(2) Durland and Adkins, THIS JOURNAL, **60**, 1501 (1938).