

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, COLUMBIA UNIVERSITY]

Preparation of Ethyl D,L-Glycerate-1-C¹⁴ and Glycerol-1-C¹⁴ 1

BY ALBERT P. DOERSCHUK

As part of a study of the reactions of glucose, glucuronic acid, ascorbic acid and lipid substances, we have prepared ethyl D,L-glycerate-1-C¹⁴ and glycerol-1-C¹⁴ by the addition of HC¹⁴N to hydroxyacetaldehyde, followed by hydrolysis, esterification, acetylation and reduction.

Experimental

Ethyl D,L-Glycerate-1-C¹⁴.—Dihydroxymaleic acid,² crystallized first from acetone made slightly acid with hydrochloric acid and then from methanol,³ was dissolved in a minimum of methanol, precipitated as the dihydrate by the slow addition of an equal volume of distilled water, and decarboxylated in pyridine to hydroxyacetaldehyde,⁴ which was washed with acetone and crystallized from anhydrous ethyl ether. A solution of 3.75 millimoles of hydroxyacetaldehyde dissolved in enough 16.7% aqueous sodium bisulfite solution to give 5.0 ml. was added to 10.0 ml. of an aqueous solution of 5.0 millimoles of sodium hydroxide and 2.50 millimoles of sodium cyanide, containing one millicurie of radioactivity as C¹⁴; the mixture was allowed to react for 26 hours at 35.0°. The initial pH was 10.7 and the final pH was 10.0. The course of the cyanide disappearance for this set of conditions and for several other sets of conditions was followed by titrating the residual cyanide in the presence of ammonia and potassium iodide with standard silver nitrate to the point of first turbidity.⁵ Blanks run to determine cyanide disappearance other than by reaction with hydroxyacetaldehyde differed from the cyanohydrin reaction mixture only in having no hydroxyacetaldehyde and in being made from 12.2% sodium bisulfite solution, making the pH value of the blank flasks identical with the pH value of the reaction flasks.⁶ The pH value of the blank remained constant at 10.7. Under the conditions given here, 93.6% of the cyanide disappeared by reaction with hydroxyacetaldehyde in 26 hours and 97.0% disappeared in 45.5 hours. The mixture was then acidified with hydrochloric acid and quickly evaporated *in vacuo* at 45° to a solid which was treated with 10 ml. of a 1:1 mixture of concentrated hydrochloric acid and distilled water in a boiling water-bath for 4 minutes under nitrogen. The mixture was quickly evaporated *in vacuo* at 45° to a solid to which was added 25 ml. of 2.5 N sulfuric acid in absolute ethanol followed by 5 ml. of toluene. After refluxing for 12 hours and removing the water formed as the azeotrope, the sulfuric acid was neutralized with excess barium carbonate and the solution concentrated *in vacuo* at 25° to 5 ml. Forty-five ml. of anhydrous ethyl ether was added slowly and with stirring and the supernatant solution collected after centrifuging. The residue was treated with 5 ml. of absolute ethanol (partial solution resulted) and the precipitation process repeated three times. The supernatant liquids were combined, concentrated *in vacuo* at 25°, placed in a small distilling flask containing 0.30 g. of non-radioactive ethyl D,L-glycerate carrier⁷ and a small quantity of barium carbonate, and distilled at 4 mm. The distillate was treated with 10 ml. of anhydrous ethyl ether and the mixture centrifuged; the clear solution was concentrated, placed in a small distilling flask, and distilled at 4 mm. The colorless liquid product had a saponification equivalent of 134.5⁸; calculated for ethyl D,L-glycerate, 134.1. Three preliminary non-radioactive syntheses gave yields corresponding to incorporation of 30% of the initial radioactivity in the

ester; the one radioactive synthesis so far carried out gave 0.33 g. of product with a specific activity of 1.28×10^6 counts/minute/milligram, corresponding to an incorporation of only 19% of the initial radioactivity. At the end of the radioactive cyanohydrin reaction, a substantial quantity of radioactivity was recovered unreacted.

Glycerol-1-C¹⁴.—One gram of acetic anhydride and 0.6 g. of pyridine were added to 0.090 g. of ethyl D,L-glycerate-1-C¹⁴ containing 7.54×10^7 counts/minute as C¹⁴ and the mixture was refluxed for 3 hours. Then 0.30 g. of non-radioactive acetylated ethyl D,L-glycerate carrier was added. The pyridine and the acetic anhydride were removed *in vacuo*. The residue was twice distilled over silver carbonate at 4 mm., dissolved in 2 ml. of anhydrous ethyl ether, and added slowly through a dropping funnel to a solution of 0.20 g. of commercial lithium aluminum hydride⁹ in 40 ml. of anhydrous ethyl ether contained in a 125-ml. flask equipped with dropping funnel, reflux condenser and magnetic stirrer. The mixture was allowed to stand for half an hour with stirring; then the excess lithium aluminum hydride was neutralized by the cautious addition of 2 ml. of distilled water. The complex was hydrolyzed by slowly adding 5.2 ml. of cold, aqueous 25% sulfuric acid, resulting in a clear, colorless, two-phase system. All but approximately 5 ml. of the ethyl ether was removed at room temperature under nitrogen. The slow addition of 13.2 ml. of cold 10% potassium hydroxide in methanol resulted in a white precipitate and a homogeneous supernatant liquid. The precipitate was centrifuged out and washed twice with isopropyl alcohol. The solutions were combined and made faintly acid to litmus with 25% aqueous sulfuric acid; 3 g. of barium carbonate was added, followed by 10 g. of anhydrous magnesium sulfate added slowly with stirring and cooling. The mixture was allowed to stand 1 hour with occasional stirring. The solids were centrifuged out and washed with isopropyl alcohol and the solvent removed *in vacuo* at 25° from the combined solutions. The residue was taken up in 7 ml. of isopropyl alcohol, made alkaline to litmus with 10% potassium hydroxide in methanol, centrifuged and refluxed on a water-bath for 30 minutes. The mixture was made faintly acid to litmus with 25% aqueous sulfuric acid and barium carbonate was added. The solids were centrifuged out, the solvent was removed *in vacuo* at 25°, the residue was dissolved in 5 ml. of distilled water containing 0.20 g. of non-radioactive glycerol carrier and the solution was washed three times each with chloroform, ligroin (b.p. range of 70 to 90°), and ethyl ether, discarding the washings. The aqueous solution was transferred to a small distilling flask and made slightly alkaline with a few grains of potassium carbonate; the solvent was removed and the residue distilled at 4 mm. The distillate was dissolved in methanol, treated 15 minutes with "Norite A" decolorizing charcoal and transferred to a small distilling flask containing a few ml. of benzene. Most of the solvent was boiled off at atmospheric pressure, residual water coming over as the ternary azeotrope. The residue was distilled at 4 mm., yielding 0.27 g. of product with a specific activity of 1.77×10^6 counts/minute/milligram which corresponds to an incorporation of 63.4% of the initial radioactivity. The product was a colorless, odorless, viscous liquid,⁸ not reacting with alkali under the conditions of the saponification equivalent determination,⁸ and possessing a refractive index within 0.002 of that of authentic glycerol.⁸

Anal. Calcd. for C₃H₈O₃: C, 39.1; H, 8.76. Found⁸: C, 39.4, 39.2; H, 8.72, 8.59.

The product was benzoylated^{8,10} to give a solid which

(1) This work was supported in part by a grant from the Nutrition Foundation, Inc.

(2) H. O. L. Fischer and L. Feldmann, *Ber.*, **62B**, 854 (1929).

(3) The crystals lost methanol of crystallization and became amorphous shortly after exposure to air at room temperature.

(4) H. O. L. Fischer and C. Taube, *Ber.*, **60B**, 1704 (1927).

(5) W. E. Miltzer, *Arch. Biochem.*, **9**, 91 (1946).

(6) With different sodium bisulfite preparations, slightly different solution concentrations were required to attain the given pH values.

(7) The non-radioactive ethyl D,L-glycerate carrier was prepared and purified in the manner described above from D,L-glyceric acid made by the oxidation of glycerol ("Beilstein," Fourth Aufl., III, p. 395).

(8) This observation was made on the product of a preliminary, larger scale, non-radioactive synthesis in which no carrier was added.

(9) Amend Drug and Chemical Co., Inc., New York.

(10) E. H. Huntress and S. P. Mulliken, "Identification of Pure Organic Compounds," John Wiley and Sons, Inc., New York, N. Y., 1941, p. 479.

after crystallization from ligroin melted at 72°; the melting point of glycerol tribenzoate crystallized from ligroin is 72°.¹¹ No depression was observed on mixed melting point with a sample of authentic glycerol tribenzoate.⁸

Benzoylation of a sample of the radioactive glycerol and

(11) A. Fairbourne and G. E. Foster, *J. Chem. Soc.*, **127**, 2759 (1925).

purification of the resulting glycerol tribenzoate by crystallization from ligroin gave a product whose radioactivity per millimole differed by 3% from that of the glycerol; this error lies well within the limits of precision of the radioactivity determination.

All samples were counted as barium carbonate on paper mountings.

NEW YORK 27, N. Y.

RECEIVED AUGUST 21, 1950

[RESEARCH LABORATORIES, AMERICAN HOME FOODS, INC.]

2-Substituted Tetrahydropyranyl Sulfides

BY FRANK KIPNIS¹ AND JOHN ORNFELT

2-Substituted tetrahydropyranyl sulfides have been prepared by the interaction of dihydropyran with mercaptans in the presence of catalytic quantities of hydrogen chloride.

In 1934, Paul² reported that it was possible to prepare tetrahydropyran ethers by the interaction of dihydropyran with alkanols in the presence of catalytic amounts of hydrogen chloride. This work was extended by Woods and Kramer³ who

Experimental

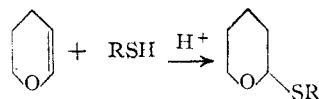
Directions are given for the preparation of ethyl 2-tetrahydropyranyl sulfide. Other sulfides may be prepared by similar methods. The properties of the new materials are listed in Table I.

TABLE I

R	B.p.		Yield, %	Formula	Analyses, ^a %					
	°C.	Mm.			Calculated			Found		
Methyl	47-48	5	57.8	C ₆ H ₁₂ OS	54.50	9.14		54.72	9.27	
Ethyl	42	2	40.5	C ₇ H ₁₄ OS	57.49	9.65	21.92	57.57	9.99	21.87
<i>n</i> -Propyl	55-58	3	36.7	C ₈ H ₁₆ OS	59.95	10.06	20.00	60.09	9.73	20.75
Isopropyl	48	2.5	66.7	C ₈ H ₁₆ OS	59.95	10.06	20.00	59.40	10.23	20.30
<i>n</i> -Hexyl	84-86	2.5	25.0	C ₁₁ H ₂₂ OS	65.29	10.96	15.84	65.37	10.48	15.52
Benzyl	86-87	2	42.3	C ₁₂ H ₁₆ OS	69.19	7.74	15.39	69.59	8.06	15.43

^a Analyses by Oakwold Laboratories, Alexandria, Virginia.

synthesized a series of these ethers by a similar method. It was of interest to us to attempt the preparation of the corresponding sulfides by substituting a mercaptan for the alcohol. These syntheses were realized, and it was found possible to obtain a series of sulfides in comparatively good yields.



The new sulfides are liquids with pleasant, minty aromas, and are quite stable to alkali, but are easily decomposed, even by very dilute acids, to the initial mercaptan and 5-hydroxypentenal.⁴ From a study of the molecular structure of these new compounds, it may be noted that these tetrahydropyranyl sulfides are, in reality, semimercaptals, which are notoriously unstable to acids. An additional analogy may be pointed out here: the sulfides may be considered to be thioglycosides of desoxysugars, and thus are the primitive members of the pyranose form of the carbohydrate series.

(1) Oxford Products, Inc., Cleveland, Ohio.

(2) R. Paul, *Bull. soc. chim.*, (5) **1**, 971 (1934).

(3) G. F. Woods and D. N. Kramer, *This Journal*, **69**, 2246 (1917).

(4) G. F. Woods and H. Sanders, *ibid.*, **68**, 2183 (1916).

Ethyl 2-Tetrahydropyranyl Sulfide.—To a 300-ml. pressure bottle chilled in an ice-bath was added 15.5 g. (0.25 mole) of ethyl mercaptan, 21 g. (0.25 mole) of dihydropyran⁵ and 5 mg. of hydrogen chloride in 1 ml. of anhydrous ether. The bottle was stoppered immediately, shaken thoroughly and the contents allowed to warm to room temperature, after which the temperature was maintained at 90° for 3 hours. At the end of this time, the bottle was chilled and opened, the contents washed into a separatory funnel with 50 ml. of ether and then shaken with 5 ml. of saturated potassium carbonate solution and separated. The organic layer was dried over calcium sulfate, filtered and the volatiles stripped under slightly reduced pressure. The residue was fractionated through a 20-cm. Vigreux column to give a 40.5% yield of a colorless oil boiling at 42° (2 mm.).

Treatment with ammoniacal sodium nitroprusside gave no coloration, indicating absence of unreacted mercaptan, as well as stability of the sulfide to base. Treatment of the sulfides with sodium nitrite-hydrochloric acid caused the development of a red color⁶ indicative of easy hydrolysis to the initial mercaptan, while an immediate pink color was given with fuchsin-sulfur dioxide reagent, confirming hydrolysis to the aldehyde.

MORRIS PLAINS, N. J.

RECEIVED SEPTEMBER 29, 1950

(5) Supplied through the courtesy of E. I. du Pont de Nemours and Company, Wilmington, Delaware. The commercial product was dried with anhydrous potassium carbonate and distilled through a 30-cm. Widmer column.

(6) H. Rheinboldt, *Ber.*, **60**, 181 (1927).