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Isolation of Lignoceric Acid from Acorns

by

Charles Pratt

Introduction

Lignoceric acid belongs to a group of organic compounds popularly known as "fatty acids." The name fatty acid is due to the occurrence of higher homologs such as palmitic, the C_{16} member, and stearic, the C_{18} member, acids in natural facts in the form of glycerides (1). It is generally the acids with an even number of carbon atoms which are found in normal fats. The fats with very small numbers of carbon atoms or with unsaturated chains tend to be liquid and are called oils while those with larger numbers of carbons and saturated chains are solids.

Lignoceric acid has twenty-four carbon atoms and like all acids in this group with 10 carbons or more, is a solid. Its formula is: $CH_3(CH_2)_{22}COOH$. It is not as widely distributed in nature as palmitic acid, stearic acid, and oleic acid. Detection of its presence in acorns was somewhat a surprise inasmuch as no published accounts of its occurrence in acorns seem to exist.

Experimental

One kg of acorns from the common pine oak trees were crushed with a Waring Blendor and the mean was extracted with ether in a soxhlet extractor. When the liquid coming over was no longer colored, the extraction was assumed to be completed. The solvent (ether) was removed by evaporation under the hood, and the saponification number of the oil was determined. To 1.50 g of oil was added 25 ml of alcoholic KOH, .2501N, and the solution was refluxed for one hour. The solution was allowed to cool and the excess KOH was titrated with a .200N hydrochloric acid.

Saponification Number:

ml of KOH = 20
Normality of KOH = 0.2501
m.e. of KOH = 5.0020

ml of acid = 5
Normality of acid = .2000
m.e. of acid = 1.0

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*The investigator is indebted to John Lang, a chemistry student at Savannah State College, for his cooperation and assistance.

$$\text{m.e. of KOH neutralized by oil} = \text{m.e. base} - \text{m.e. HCl}$$

$$5.002 - 1.000 = 4.002$$

$$\text{grams of KOH hydrolyzing the oil} = 0.224$$

$$\text{Wt. of oil} = 1.5 \text{ g}$$

$$\text{S. N.} = \frac{.224\text{g KOH}}{1.5\text{g oil}} \times \frac{X\text{g KOH}}{1\text{g oil}}$$

$$X = .49 \text{ g or } 149 \text{ mg KOH}$$

$$\text{S.N.} = 149$$

Once the saponification number was determined, it was used as a guide to hydrolysis.

One hundred grams of acorn oil were mixed with 500 ml of water and 16 g (a slight excess) of solid KOH was added to this mixture. After refluxing, for one hour, the flask was cooled and its contents neutralized with 6N hydrochloric acid. Upon cooling, a solid separated and formed a top layer which was removed and its water pressed from it with filter paper. The fatty acid mixture was placed on the Fisher Zone Refiner and the zone which appeared in largest quantity, about 80 per cent, was removed and further analyzed.

Analysis

Calculated	*Found*
Carbon = 78.26	78.01
Hydrogen = 13.04	12.99
M.p. 81-82°C(2)	82.1°C

Summary

The oil of acorns was extracted and its Saponification number was determined. This information was used as a guide in the hydrolysis of larger quantities of oil.

More than one fatty acid was found to be in the acorn oil, but one acid composed about 80 per cent of the fatty acid mixture. It was purified by use of the Fisher Zone Refiner, and its element composition and melting point indicate that it is lignoceric acid.

References

1. Whitmore, F. C., "Organic Chemistry", Van Nostrand Company, Inc., pp. 286-7, 1937.
2. Lange, N. A., "Handbook of Chemistry", Handbook Publishers, Sandusky, Ohio, 8th Ed., p. 574, 1952.

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Synthetic Preparation of Apiose From Dihydroxy Acetone

by

Charles Pratt*

Abstract

A five-carbon sugar of unusual interest is apiose. This sugar differs from the normal pentoses in that it possesses a branched chain. The sugar has been isolated from parsley and its structure determined (1). In 1962 it was isolated by paper chromatography in the Savannah State College laboratories (2).

This paper deals with the synthesis of apiose by consecutive cyanide additions beginning with dihydroxy acetone.

Experimental

Twenty-two and one half grams [.25 mol.] of dihydroxy acetone were added to 0.25 mol of sodium bisulfite dissolved in 100 ml of water, and the mixture was refluxed for about 30 minutes until all materials were in solution. The solution was then evaporated to dryness under reduced pressure.

To the dried product, one-fourth mole of sodium cyanide and 400 ml of water were added, and the mixture was refluxed for eight hours.

After refluxing, the cyano-compound formed was oxidized by refluxing for 3 hours with 150 ml of concentrated hydrochloric acid. The reactions assumed to take place are shown in figure 1.

The 2-hydroxymethyl-2, 3-dihydroxy propionic acid was dialyzed and the resulting ion-free solution was concentrated under reduced pressure to approximately 200 ml. To this solution were added lumps of sodium amalgam. The solution was constantly stirred with a magnetic stirrer. As the Na-Hg decomposed the solution tended to become basic [pH was measured constantly by zeromatic pH meter] and therefore, hydrochloric acid was added from time to time to maintain a pH of 2.5-3.0.

The mercury which was formed by the decomposition of Na-Hg was removed, and the remaining solution was neutralized and dialyzed to remove excess salts. Following dialysis, the 4-carbon alde-

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*The investigator is indebted to Ellen Polite and Andrew Zeigler, chemistry students, at Savannah State College, for their cooperation and assistance.

hyde was subjected to the same cyanide addition steps as was the dihydroxy acetone. Apiose was the final result. This is shown in figure 2.

Fehlings Solution gave a positive result when used to test a small portion of the final product. Another portion was spotted with Whatman No. 1 chromatography paper and its R_f value was compared with that of authentic apiose which had been isolated from parsley [2].

Summary

The branched 5-carbon sugar, apiose, was prepared by cyanide addition to 1,3-dihydroxy acetone; hydrolysis to the acid of the resulting cyano compound; reduction of the acid to a 4-carbon aldehyde, and the cyanide addition repeated. The results were interpreted by paper chromatography, and by standard methods of sugar identification.

Acknowledgments

This project was sponsored in part by grants-in-aid from the Research Corporation and the National Science Foundation through an Undergraduate Research Participation Program.

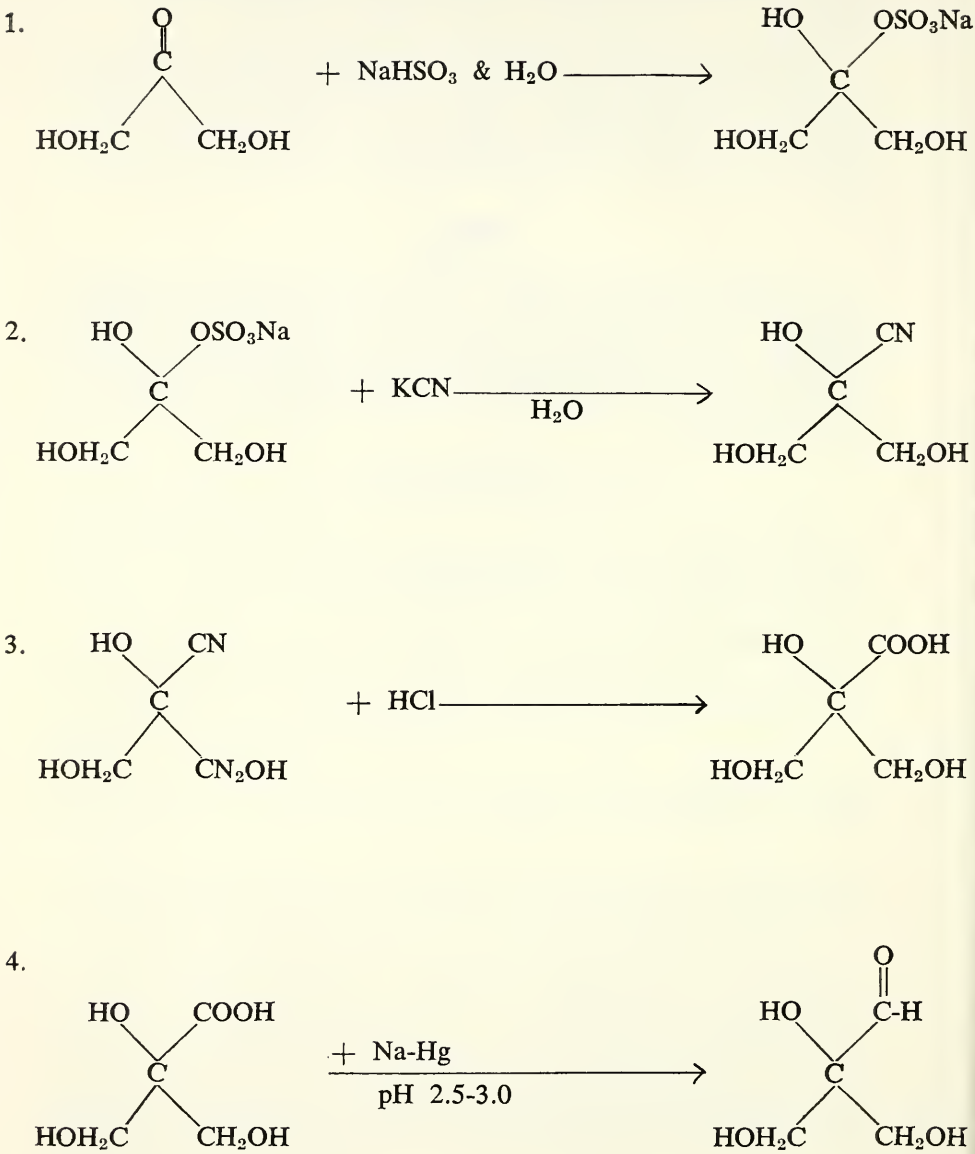
1.
O
C
HOH2C
CH2OH
+ NaHSO3 & H2O
HO
OSO3Na
C
HOH2C
CH2OH
2.
HO
OSO3Na
C
HOH2C
CH2OH
+ KCN
H2O
HO
CN
C
HOH2C
CH2OH
3.
HO
CN
C
HOH2C
CN2OH
+ HCl
HO
COOH
C
HOH2C
CH2OH
4.
HO
COOH
C
HOH2C
CH2OH
+ Na-Hg
pH 2.5-3.0
O
HO
C-H
C
HOH2C
CH2OH

Figure 1.

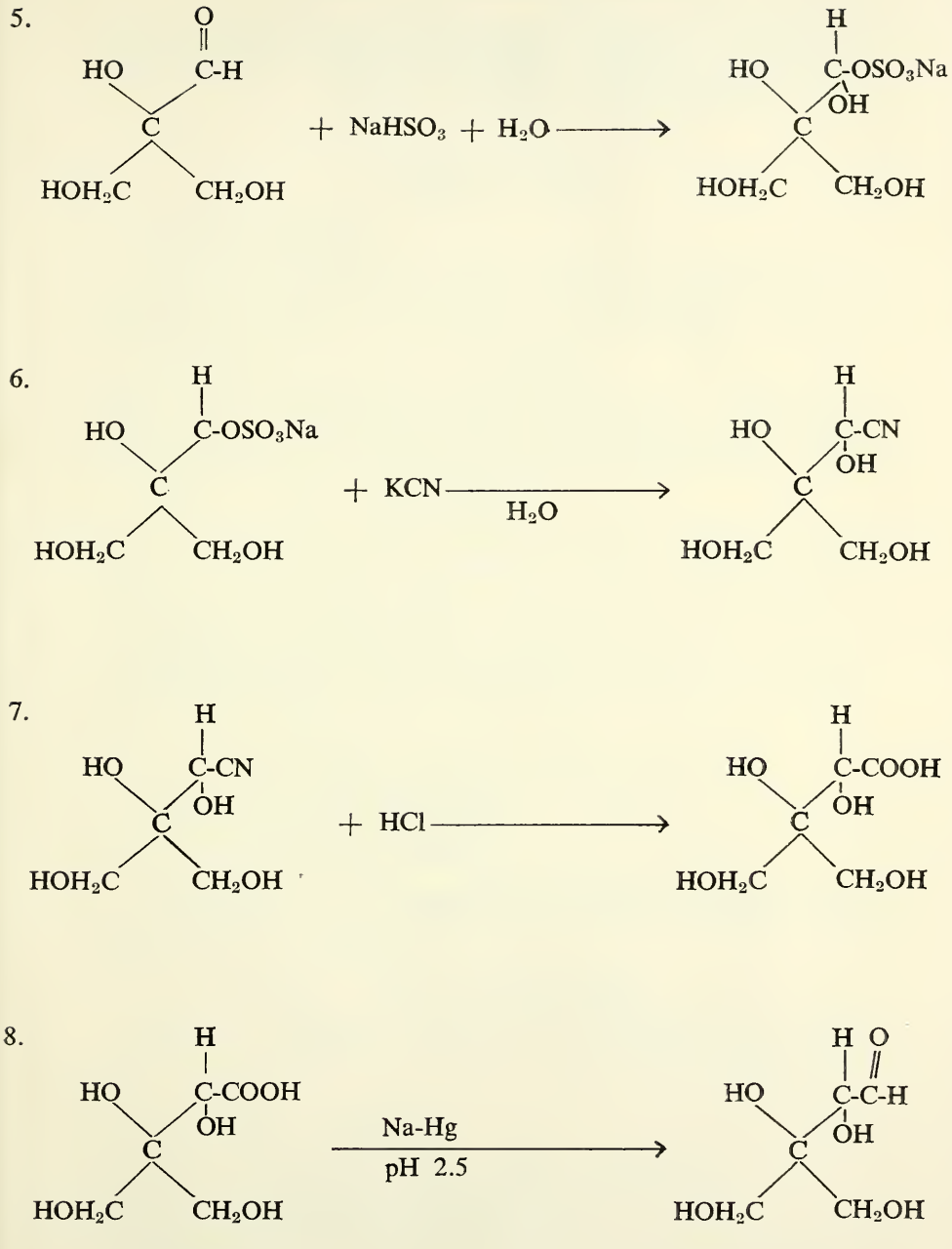
5.
HO
C-H
O
C
HOH2C
CH2OH
+ NaHSO3 + H2O
H
HO
C-OSO3Na
OH
C
HOH2C
CH2OH
6.
H
HO
C-OSO3Na
C
HOH2C
CH2OH
+ KCN
H2O
H
HO
C-CN
OH
C
HOH2C
CH2OH
7.
H
HO
C-CN
OH
C
HOH2C
CH2OH
+ HCl
H
HO
C-COOH
OH
C
HOH2C
CH2OH
8.
H
HO
C-COOH
OH
C
HOH2C
CH2OH
Na-Hg
pH 2.5
H O
HO
C-C-H
OH
C
HOH2C
CH2OH

Figure 2.

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2. Pratt, Charles, "Isolation of Apiose from Parsley", Faculty Research Bulletin. Savannah State College, *16,* 37, [1962].