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Aliphatic Beta Monoglycerides[1]

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The preparation and identification of aromatic and fatty acid beta monoglycerides has been established by several investigators. Helferich and Sieber[3] were the first to prepare a true beta aromatic ester of glycerol. They prepared the β-monobenzoate and β-mono-(*p*-nitrobenzoate) of glycerol by acid hydrolysis of the α,α′-ditrityl ether derivatives. Jackson and King[4] and Verkade and associates[5] on application of this method to the preparation of beta monoglycerides of the aliphatic acids found that a shift of the acyl group from the beta to the alpha position occurred. Because of this tendency for aliphatic groups to shift from the beta to alpha position under conditions of acid hydrolysis, the method obviously is not adaptable to the preparation of aliphatic beta monoglycerides.

Bergmann and Carter,[6] employing α,α′-benzylidene glycerol, prepared by the method of Hibbert and Carter,[7] reduced the corresponding beta esters of this intermediate catalytically to obtain beta monoacetin, beta monopalmitin, and the beta monobenzoate of glycerol. King and associates[8,9] verified the method by preparing the glycerol beta monoesters of capric, lauric, myristic, palmitic and stearic acids, and proved their structures by conversion of the beta esters to symmetrical triglycerides; the latter were identical with those prepared by other independent methods.

Daubert,[10] using the method suggested by Verkade and associates,[11] succeeded in preparing beta monopalmitin by catalytic detritylation of the esterified α,α′-ditrityl ether of glycerol. Two methods, now well established, are thus available for the preparation of fatty acid beta monoglycerides.

Since all beta monoglycerides of C_2 to C_{18} fatty acids have been prepared and identified, except beta monocaproin and beta monocaprylin, the purpose of the present investigation was to obtain the experimental data for these two beta esters and their benzylidene intermediates, and also to correlate the melting point data of the complete series.

Experimental

Preparation of β-Caproyl-α,α′-benzylidene Glycerol.—α,α′-Benzylidene glycerol (33.5 g.) was dissolved in dry pyridine (30 ml.) and the solution cooled in an ice-bath. To this solution there was added slowly caproyl chloride (25 g.). During the addition of the fatty acid chloride the temperature of the mixture was maintained at approximately 20° by cooling in an ice-bath. After the mixture was allowed to stand four hours at room temperature, ice water (200 ml.) was added and the product separated as a white, semi-solid mass. After washing several times with 200-ml. portions of ice water to remove the pyridine, the mass was dissolved in warm petroleum ether (250 ml.) and the solution dried over anhydrous calcium sulfate. On cooling the filtered solution for twelve hours at 5°, the fine, needle-like crystals which separated were filtered with suction and purified by recrystallization several times from petroleum ether; yield 50.2 g. (97%), m. p. 34.1°; molecular weight, 278 ± 2 (calcd., 278.34). Molecular weight determinations of all compounds were made by the ebullioscopic method of Menzies and Wright,[12] as modified by Hanson and Bowman.[13] Benzene was used as the solvent in all determinations.

Anal. Calcd. for $C_{16}H_{22}O_4$: C, 69.04; H, 7.97. Found: C, 69.35, 69.50; H, 7.90, 8.03.

Preparation of β-Caprylyl-α,α′-benzylidene Glycerol.—Caprylyl chloride (22.5 g.) was added slowly to a cooled solution of α,α′-benzylidene glycerol (25 g.) in dry pyridine (30 ml.). The mixture was allowed to stand at room temperature for twenty-four hours. Ice water (200 ml.) was added and the oily liquid which separated was washed several times with 200-ml. portions of ice water until the odor of pyridine was no longer perceptible. The residue was dissolved in ethyl ether (250 ml.) and dried over anhydrous calcium sulfate. After the ether was removed under reduced pressure, the residue on cooling to 5° solidified to a white, crystalline mass. The crystalline product was dissolved in warm petroleum ether (250 ml.) and placed in a cold chest at −20°. The fine, white crystals which separated were filtered with suction and recrystallized several times from petroleum ether; yield 35.6 g. (84%), m. p. 35.0°; molecular weight, 306 ± 2 (calcd., 306.39).

Anal. Calcd. for $C_{18}H_{22}O_4$: C, 70.56; H, 8.55. Found: C, 70.23, 70.14; H, 8.57, 8.61.

(1) The authors are indebted to Swift and Company and to the Buhl Foundation for grants in support of this investigation.

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(3) Helferich and Sieber, *Z. physiol. Chem.*, **175**, 311 (1928).

(4) Jackson and King, THIS JOURNAL, **55**, 678 (1933).

(5) Verkade and Meerburg, *Rec. trav. chim.*, **54**, 716 (1935).

(6) Bergmann and Carter, *Z. physiol. Chem.*, **191**, 211 (1930).

(7) Hibbert and Carter, THIS JOURNAL, **51**, 1601 (1929)

(8) Stimmel and King, *ibid.*, **56**, 1724 (1934).

(9) Daubert and King, *ibid.*, **60**, 3003 (1938).

(10) Daubert, *ibid.*, **62**, 1713 (1940).

(11) Verkade, van der Lee, de Quant and Zuydewijn, *Proc. Acad. Sci., Amsterdam*, **40**, 580 (1937).

(12) Menzies and Wright, THIS JOURNAL, **43**, 2312 (1921).

(13) Hanson and Bowman, *Ind. Eng. Chem., Anal. Ed.*, **11**, 440 (1939).

Preparation of β-Monocaproin.—β-Caproyl-α,α′-benzylidene glycerol (8.5 g.) was dissolved in absolute alcohol (100 ml.) to which was added palladium black (0.5 g.). The mixture was transferred to the hydrogenation bottle of a Burgess–Parr apparatus and the air evacuated. The reduction, which was complete in two hours, was carried out at room temperature and 36 lb. hydrogen pressure. The catalyst was removed by filtration and the filtrate reduced to a volume of 10 ml. Attempts to obtain well-defined crystals from alcohol and various combinations of mixed solvents were not successful. Therefore, the product after removal of solvent, was placed in a vacuum desiccator for three weeks. On cooling to −20° the beta monocaproin crystallized. The crystalline mass was washed repeatedly at −20° with pre-cooled petroleum ether and then dried in a vacuum desiccator. At room temperature the crystalline mass liquefied. The beta monocaproin was placed in a small glass tube in which was inserted a thermometer and then it was held at −20° for twenty-four hours. In the determination of the melting point, a slow temperature rise was controlled in a dry-ice–acetone bath until the product melted; m. p. −8 to −10°, molecular weight, 189 ± 1 (calcd. 190.24); refractive index at 40°, 1.4462.

Anal. Calcd. for $C_9H_{18}O_4$: C, 56.62; H, 9.54. Found: C, 56.45, 56.51; H, 9.34, 9.28.

For further identification the beta monocaproin was used to prepare β-caproyl-α,α′-distearin, m. p. 47.0° (Robinson, Roche and King,[14] 47.2°).

Preparation of β-Monocaprylin.—β-Caprylyl-α,α′-benzylidene glycerol (6 g.) was dissolved in absolute alcohol (50 ml.) and palladium black (1 g.) was added. The hydrogenation was carried out as described for beta monocaproin. After removal of the catalyst, the alcohol was removed *in vacuo*. The product was crystallized from a 1:1 mixture of ether and petroleum ether; m. p. 29.8°, molecular weight 217 ± 1 (calcd., 218.29); refractive index at 40°, 1.4473.

Anal. Calcd. for $C_{11}H_{22}O_4$: C, 60.52; H, 10.16. Found: C, 60.24, 60.34; H, 10.09, 10.00.

For further identification the beta monocaprylin was used to prepare β-caprylyl-α,α′-distearin, m. p. 51.5° (Robinson, Roche and King,[14] 51.8°).

Discussion

It has been observed previously by Stimmel and King[8] that the melting points of the esterified α,α′-benzylidene glycerol derivatives of saturated fatty acids (C_{10} to C_{18}) progressively increased as the length of the carbon chain of the fatty acid in the beta position increased. The melting point (99–100°) of the acetyl compound reported by Bergmann and Carter[6] was verified by Daubert,[10] who interpreted the high melting point as inconsistent with analogous members of the series. This interesting melting point relationship of acetyl compounds was observed by McElroy and King[15] in mixed triglycerides containing one mole of acetic acid. The melting points were relatively high compared with higher members of the series. Since other series based on homologous aliphatic acids exhibit a minimum melting point in compounds containing an aliphatic chain having 6 to 10 carbon atoms, the acetyl and butyryl compounds of this series of benzylidene derivatives lie on the low-carbon branch of the melting point curve and are in agreement with other members which lie on the high-carbon branch. The melting points found for the C_6 and C_8 benzylidene compounds in this investigation (Fig. 1) thus lie on the high-carbon branch of the melting point curve, with the melting point at a minimum in a compound of four carbon atoms. The melting point of the β-caproyl-α,α′-benzylidene glycerol was somewhat higher than expected, but its position on the curve is clearly established.

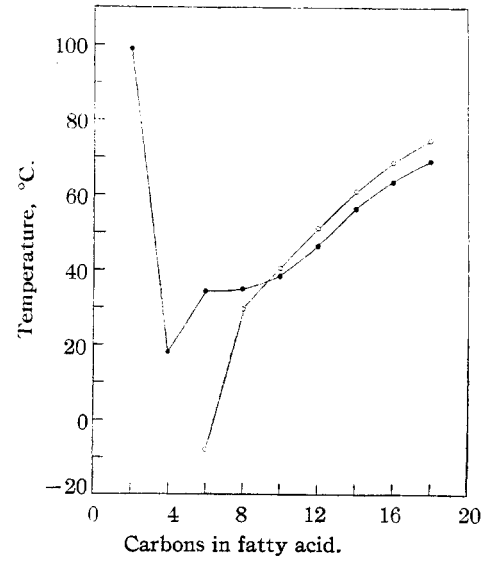

Fig. 1.—Relation between melting point and length of fatty acid carbon chain in β-monoglycerides and their corresponding benzylidene intermediates: O, β-monoglycerides; ●, benzylidene intermediates.

Stimmel and King[8] further observed that the melting points of the beta monoglycerides of C_{10} to C_{18} saturated fatty acids were higher than the corresponding benzylidene intermediates, with the exception of beta monomyristin. Recent investigations in our Laboratory on the preparation of beta monomyristin and beta monocaprin gave verification to the reported melting points of the esters. However, the melting points of β-myristyl-α,α′-benzylidene glycerol and β-capryl-α,α′-

(14) Robinson, Roche and King, THIS JOURNAL, **54**, 705 (1932).
(15) McElroy and King, *ibid.*, **56**, 1191 (1934).

benzylidene glycerol were found to be 56.2 and 38.6°, respectively (Stimmel and King,[8] 62 and 32.5°). Thus, the melting points of the C_{10} to C_{18} saturated fatty acid beta monoglycerides are all higher than the corresponding benzylidene intermediates. In this, and previous, investigations the reverse relationship was evident for lower members (C_2 to C_8) of the series.

Summary

The new benzylidene glycerol intermediates, β-caproyl-α,α′-benzylidene glycerol and β-caprylyl-α,α′-benzylidene glycerol have been prepared, and subsequently reduced to the two new monoesters, β-monocaproin and β-monocaprylin, respectively.

Melting points of the new beta monoglycerides and their benzylidene intermediates increased with increasing length of the carbon chain of the fatty acids consistent with higher members of the aliphatic series.

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The Chemistry of Allergens. VIII. Isolation and Properties of an Active Protein-polysaccharidic Fraction, CB-1A, from Castor Beans[1a,b]

BY JOSEPH R. SPIES AND E. J. COULSON

Isolation of the protein-polysaccharidic fraction, CS-1A[2a,b] which contained the principal allergens of cottonseed[1,3] and chemical characterization of the active constituents of CS-1A have been described in previous papers.[4] Similar studies on other substances are limited because suitably hypersensitive human subjects are not always available during the time required to develop isolation methods. Multiple allergenic sensitivity to cottonseed, nuts, and other oil-bearing seeds has long been recognized to form a distinct clinical grouping.[5] Therefore, it was assumed that the allergenic components of some of these other oil seeds might be *chemically* similar enough to CS-1A to permit their isolation by the procedure developed for obtaining CS-1A from cottonseed.

Castor beans, among several oil seeds subjected to the CS-1A procedure,[6] yielded 1.8% of a non-toxic allergenic protein-polysaccharidic fraction, CB-1A. The present paper describes the isolation, chemical and physiological properties and chemical composition of CB-1A.

Alilaire,[7] who first described sensitivity to the castor-oil plant, believed the allergen to be identical with the powerful toxalbumin, ricin. Ratner and Gruehl,[8] however, showed that castor beans contained an anaphylactogenic agent in addition to ricin. Barnard,[9] without giving experimental details, prepared non-toxic, allergenic extracts from castor beans. According to Barnard the allergen was water soluble, alcohol precipitable, heat stable and non-dialyzable. Grabar and Koutseff[10] separated a non-toxic allergenic fraction from castor beans which they called "ricin-allergene." Their preparation was water soluble, heat stable and dialyzable.

Experimental

Isolation of Allergenic Fraction, CB-1A.—Good quality shelled castor beans, *Ricinus communis*, U. S. Department of Agriculture Type 4, were kindly supplied for this investigation by Dr. Donald M. Crooks of the Bureau of

(1) (a) Presented before the Division of Biological Chemistry at the 105th meeting of the American Chemical Society held at Detroit, Michigan, April, 1943. Not copyrighted.

(1) (b) For Article VII of this series entitled "The Nature of the Unidentified Allergens of Cottonseed" see Spies, Chambers, Bernton and Stevens, *J. Allergy*, **14**, 7 (1942).

(2) (a) Spies, Bernton and Stevens, *ibid.*, **10**, 113 (1939); (b) Spies, Coulson, Bernton and Stevens, THIS JOURNAL, **62**, 1420 (1940).

(3) Bernton, Spies and Stevens, *J. Allergy*, **13**, 289 (1942). This article and reference 1 contain conclusive evidence that CS-1A (and therefore subfractions obtained from CS-1A) was immunologically distinct from other allergens present in cottonseed.

(4) See Spies and Umberger, THIS JOURNAL, **64**, 1889 (1942), for the previous paper published in THIS JOURNAL.

(5) Coca, Walzer and Thommen, "Asthma and Hay Fever in Theory and Practice," Charles C. Thomas, Baltimore, Md., 1931, p. 394.

(6) Kapok seeds, black mustard seeds, flaxseed, croton beans, soybeans and pecan nuts also have been subjected to the CS-1A procedure. Yields of 0.9, 0.7, 0.07, 0.02, 0.02, and 0.00%, respectively, were obtained. The products contained nitrogen and carbohydrate in varying proportions and some possessed chemical properties similar to CS-1A. Results of these studies will be described when allergenic or antigenic properties have been determined.

(7) Alilaire, *Ann. Inst. Pasteur*, **28**, 605 (1914).

(8) Ratner and Gruehl, *Am. J. Hygiene*, **10**, 236 (1929); these authors review the literature on castor bean allergy in *J. Allergy*, **2**, 1 (1930).

(9) Barnard, *J. Allergy*, **1**, 473 (1930).

(10) Grabar and Koutseff, *Compt. rend. soc. biol., Paris*, **117**, 700-703 (1934).