The Preparation of Alkyl Esters from Highly Unsaturated Triglycerides[1]

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Abstract

The alcoholysis reaction has been applied to the preparation of highly unsaturated alkyl esters from menhaden oil. This reaction proceeded very rapidly, and nearly quantitative yields were obtained with virtually no loss in double-bond structure. The formation of esters was studied, using straight- and branched-chain alcohols having 1–6 carbon atoms. The reactions were monitored by the technique of thin-layer chromatography (TLC). Maximum conversion of straight-chain esters was found to be a linear function with respect to the number of carbon atoms in the alcohol. Reaction time varied from 2 min for methanol to 60 min for n-hexanol. Branched-chain alcohols reacted more slowly than did the corresponding straight-chain compounds.

This reaction was found to be applicable to laboratory and large scale preparations of highly unsaturated alkyl esters.

Introduction

A NUMBER of papers (1–7) have described methods for the preparation of methyl esters from long-chain fatty acids or triglycerides. In general, these methods use anhydrous methanol containing either an acidic or basic catalyst and involve reaction times varying from 0.5–2 hr at reflux temperature. The methods using boron-trifluoride-methanol reagent or diazomethane are rapid and give excellent yields. However, the difficulty in handling and the hazards involved have discouraged their use, especially in the preparation of large quantities of methyl esters.

Since it was our desire to prepare various alkyl esters from highly unsaturated fish oil triglycerides, the alcoholysis reaction appeared to be the most promising. This reaction is simple, gives excellent yields, and would be expected to be non-destructive to the polyenoic structures if conducted under mild conditions. The objective of the research reported here was to apply this reaction to the preparation of various alkyl esters from fish oils.

Experimental

Materials and Equipment. Menhaden oil triglycerides [iodine value (I.V.) 184.5; sap. eq. 290.5; 1.09% conjugated diene, and a trace of conjugated triene], free of unsaponifiable matter, were obtained by molecular distillation of crude menhaden oil. The alcohols used were purified and dried by conventional methods wherever necessary.

As reported previously (8), thin-layers (250–275 μ) of silicic acid on glass plates (20 x 20 cm) were prepared according to the method of Stahl (9).

Preparation of esters. Into a reaction flask equipped with mechanical stirrer, condenser, and gas-bubbling tube, were placed 50.0 g (0.057 mole) of menhaden oil triglycerides and 7.0 g (0.218 mole) of anhydrous methanol. The reaction vessel was placed in a constant temperature bath at 60.0 ± 0.1C and vigorously stirred under an atmosphere of purified (99.99%) nitrogen. When equilibrium was attained, a solution of 0.15 g (0.0065 mole) of sodium in 4.0 g (0.125 mole) of methanol was added to the reaction mixture. Samples of the reaction mixture were withdrawn at regular intervals, and the reaction was quenched by immediately dropping the sample into cold water. Water insoluble compounds were extracted with petroleum ether (bp 30–60C), and the extract was dried over anhydrous sodium sulfate. After removal of the solvent, the extent of conversion of triglycerides to methyl esters was determined by TLC.

Ethyl, n-propyl, i-propyl, n-butyl, i-butyl, and n-hexyl esters were prepared under the same conditions, except in the cases of the water insoluble alcohols that were removed from the product by distillation under vacuum. Figures 1 and 2 are typical examples of TLC analysis of the reaction products. Yields of about 99% were obtained in all cases. The time required to convert menhaden oil triglycerides to the various alkyl esters is shown in Figure 3. There was no reaction when t-butyl alcohol was used.

Methyl, ethyl, and n-butyl esters have been prepared in quantities up to 5 gal with no change in the mole ratios of the reactants. The large quantities of alkyl esters were prepared from crude menhaden oil rather than from the purified menhaden oil that was used in the initial study of the reaction. The reaction times were the same as indicated in Figure 3. The hot reaction-mixture was washed with water until the washings were neutral. Any emulsions encountered were effectively broken with 20% saline solution. The product was dried over anhydrous sodium

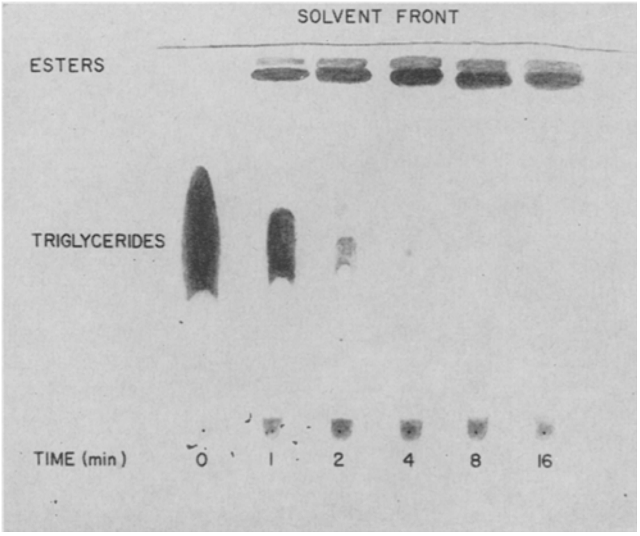

FIG. 1. Thin-layer silicic acid adsorption chromatogram of ethyl esters.
Solvent system: 90:10 petroleum ether (bp 30–60C)—ethyl ether.
Development time: 30 min.
Indicator: Iodine vapors.

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[1] Presented at the AOCS meeting, St. Louis, Mo., 1961.

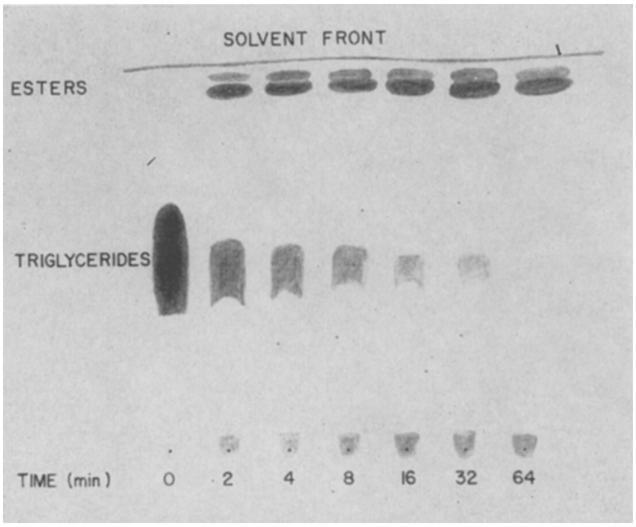

FIG. 2. Thin-layer silicic acid adsorption chromatogram of n-hexyl esters.
Solvent system: 90:10 petroleum ether (bp 30–60C)—ethyl ether.
Development time: 30 min.
Indicator: Iodine vapors.

sulfate and was then filtered. The percent triglycerides converted to alkyl esters was essentially identical to that observed in the initial studies. Total recovery of the product was possible through solvent extraction of the drying agent.

Five gallons of ethyl esters prepared by this method from crude menhaden oil (I.V. 174.5, sap. eq. 294.3) were fractionated into 4 approximately equal fractions in a centrifugal-type molecular still (Model CMS-14, Consolidated Electrodynamics Corp.) A similar 5 gal of ethyl esters were distilled with no fractionation. The distillation data and analysis are shown in Table I. The difference in I.V. between the distilled esters and the calculated theoretical value can be accounted for in the small forerun and pot residue from the distillation.

Discussion

The alcoholysis reaction was carried out at 60C to keep the possible destruction of the polyene structures of the fatty acid moieties to a minimum. This temperature also results in homogeneous reaction mixtures, which simplifies the reaction. It was also found that anhydrous conditions were essential for maximum yields.

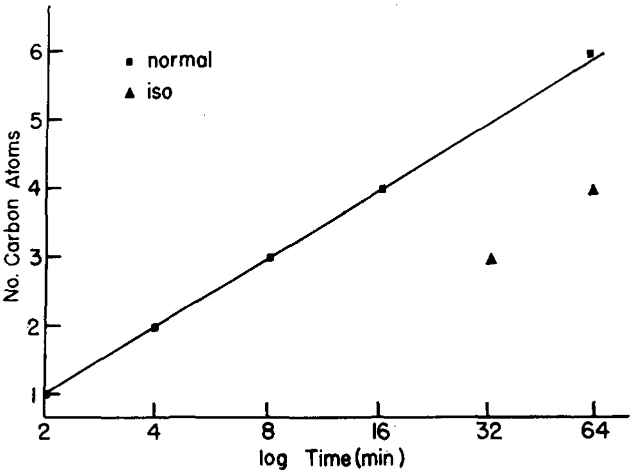

FIG. 3. Effect of chain-length and structure of alcohols on the rate of alcoholysis of menhaden oil.

TABLE I

Molecular Distillation of Menhaden Oil Ethyl Esters

Fraction	Temperature C[a]	Volume %	I.V. (Hanus)	Gardner Number
1	68–69	25	78	<1
2	69–70	25	109	<1
3	70–78	25	170	1
4	78–83	25	284	2
Σ	68–83	100	160	....
Unfractionated esters	67–80	100	159	2

Theoretical I.V. calculated from original oil: 166.
[a] 16–17 μ pressure.

As can be noted in Figures 1 and 2, the technique of TLC provides a rapid and a simple method of determining the time required to convert triglycerides to alkyl esters. When the observed reaction times are plotted versus the chain length of the respective alcohols (Fig. 3), it is seen that an increase of one methylene group in the alcohol doubles the reaction time and that the effect of a branch chain is equivalent to an increase of two methylene groups.

In addition to observing the disappearance of triglycerides and the appearance of alkyl esters, the presence of side products in the reaction can also be detected by TLC. When the same samples were chromatographed by use of a more polar solvent system (10), the spots at the origin in Figures 1 and 2 were resolved into two distinct spots. By comparison with authentic compounds, these spots were found to be free fatty acids and monoglycerides. They were estimated on the thin-layer plates to be present in the mixture to the extent of about 0.5% each at the completion of the reaction. This was subsequently confirmed by standard chemical methods. It was interesting to note that no diglycerides were found even when excess amounts of the reaction mixtures were analyzed by TLC. Only trace amounts (<0.1%) of polymeric material were observed.

Under the conditions of the reaction, it was anticipated that there would be no cis-trans isomerization and little or no effect on the percent conjugation of the polyenoic structures. Infrared analysis of the menhaden oil triglycerides and the alkyl esters derived from them showed no detectable trans isomers (10.3 μ) in any instance. Ultraviolet analysis indicated only a slight increase in the percent conjugation over the starting triglycerides. The greatest increase in conjugation was in the case of the n-hexyl esters which contained 1.42% conjugated diene and a trace of conjugated triene. Spectral analysis of the alkyl esters prepared in large quantities also showed negligible destruction of the unsaturated linkages. It was also observed that no further destruction occurred during the molecular distillation of the esters.

ACKNOWLEDGMENT

Molecular distillation of the alkyl esters by N. K. Allen.

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