umn chromatography (and/or GLP chromatography) to give the ratio of EFA-linoleic acid to saturated fatty acids.

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Isomerization of Mono Ethenoid Acids During Hydrogenation[1]

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Abstract

Methyl petroselinate, methyl oleate, and methyl erucate were hydrogenated under conditions used in industry for selective hydrogenation. The resulting products were separated into saturated esters and *trans*- and *cis*-unsaturated esters on a silver nitrate impregnated silicic acid column. The positional isomers in the total hydrogenated samples and the *cis* and *trans*-fractions were determined by oxidation with permanganate-periodate and GLC analysis of the resulting dicarboxylic fragments. Positional isomers were found in both *trans* and *cis*-fractions with equal shifting of the bond toward and away from the carboxyl group, regardless of whether the bond was originally in the 6,9, or 13 position. The ratio of *trans* to *cis*-form in the positional isomers in all cases was higher than the reported equilibrium proportions of 2:1.

Introduction

ISOMERIZATION of mono ethenoid acids during partial hydrogenation has been studied by several investigators. From partially hydrogenated ethyl oleate, Moore (9) isolated both positional and geometric isomers, while Hilditch and Vidyarthi (7), working with methyl oleate, obtained *trans*-isomers with bonds in the 8 and 10 positions. Boelhouwer et al. (4) and Knegtel et al. (8) showed the formation of large amounts of positional isomers during the hydrogenation of unsaturated fatty acids. Allen and Kiess (3) demonstrated that bond shifting toward and away from the carboxyl group was equal during hydrogenation of oleic acid. By separating the *trans* from the *cis*-acids by acetone crystallization and determining the positional isomers present in the purified *trans*-fractions, these authors concluded that the positional isomers were composed of an equilibrium mixture of *cis* and *trans*-isomers. Feuge and Cousins (6) studied the influence of temperature, rate of hydrogen dispersion, type of catalyst, and catalyst concentration on the bond shifts during hydrogenation of methyl oleate. The amount of *trans*-isomers formed was not proportional to either the degree of hydrogenation or the extent of migration of the double bonds. Effects of operating variable during partial hydrogenation of methyl oleate were also studied by Albright and Wisniak (1). They found that the ratio of *trans* to *cis*-isomers approached 2, and that the rates of hydrogenation for *cis, trans* and positional isomers were the same. This latter point was also noted in a recent paper by Allen (2) on the hydrogenation of methyl *cis* 6-, *cis* 9-, and *cis* 12-octadecenoates. Scholfield et al. (11), studying the partial hydrogenation of methyl linolenate, isolated the monoenes formed and fractionated these into the *cis* and *trans*-isomers by countercurrent distribution between methanolic silver nitrate and petroleum ether. In the *cis*-fraction, the major part of the double bonds remained in the original positions, while in the *trans*-fraction the bonds were widely scattered. In the monoenes with double bonds in other than the original positions 13-25% were *cis*.

The present paper describes the application of a silver nitrate impregnated silicic acid column, as described by de Vries (5), to the quantitative separation of saturated esters and *trans* and *cis*-isomers formed during the partial hydrogenation of methyl petroselinate, methyl oleate, and methyl erucate under selective conditions. Positional isomers in the hydrogenated esters and their *cis* and *trans*-fractions

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were determined by oxidation by the permanganate-periodate method of von Rudloff (10) and GLC analysis of the resulting dicarboxylic acids.

Experimental

Materials

Methyl oleate. Olive oil was saponified with alcoholic KOH and the mixed fatty acids were esterified with methanol. The esters were fractionally distilled in a Podbielniak column and methyl oleate obtained from the C_{18} fraction by five low temp recrystallizations from acetone. The final product as determined by GLC on a polyester column was: 98.5% oleate, 1% linoleate, 0.5% stearic. Iodine value (I.V.) was 85.2.

Methyl erucate. Prepared as above from the C_{22} fraction from rapeseed oil. Composition determined as above was: 99.0% erucate; 1% behenic. I.V. was 70.1.

Methyl petroselinate. Oil from coriander seeds was hydrolysed with alcoholic KOH and the fatty acids liberated from the soaps. The petroselinic acid was obtained by two crystallizations from 90% ethanol at −20C. The acid was esterified with methanol and GLC of the methyl ester as such and, after oxidation, showed 97.5% methyl petroselinate, 0.5% methyl oleate and 2% methyl palmitate. I.V. was 83.5.

Hydrogenation

Hydrogenations were carried out in a small scale hydrogenator constructed in this laboratory. The unit was similar to the Parr shaking hydrogenator except that the glass hydrogenation tube was replaced by an electrically heated aluminum block. This block was drilled to take three stainless steel tubes so that three samples could be run simultaneously. Five g of the esters and the required amount of commercial Rufert nickel catalyst were put in the tubes, the tubes placed in the preheated block, and the unit sealed. After evacuation, hydrogen was admitted and the hydrogenation carried out at 190C and 20 psig with shaking for 30 min. After hydrogenation, the samples were filtered at 70C on a bed of celite contained in a filter paper. *Trans*-isomers were determined by IR analysis using a Perkin Elmer Model 21 double beam instrument and calculating the *trans*-content by the AOCS tentative method.

Column Chromatography

The partially hydrogenated samples were separated into saturated, *trans*, and *cis*-fractions on a silver nitrate impregnated silicic acid column as described by de Vries (5). One hundred g Mallinckrodt silicic acid (100 mesh, chromatographic grade) and 200 ml of aqueous solution silver nitrate (500 g/liter) were heated in a water bath for 10 min with stirring. After cooling, the silicic acid was filtered off and dried at 120C for 16 hr. Two parts of the silver nitrate containing silicic acid were ground with one part of celite, and this material was used in all subsequent column separations. Fifteen g of this material was slurried with Skellysolve F and packed in an 18-mm diam column to give a column length of 15 cm. The column was covered with black paper during operation. Elution of the samples was followed by collecting 5 ml fractions, evaporating, and weighing. Preliminary experiments with a mixture (60 mg) containing equal proportions of methyl stearate, methyl elaidate, and methyl oleate gave clear cut separations by eluting respectively with 15,30, and 45% benzene in Skellysolve F. Figure 1 shows a typical separation for partially hydrogenated methyl petroselinate.

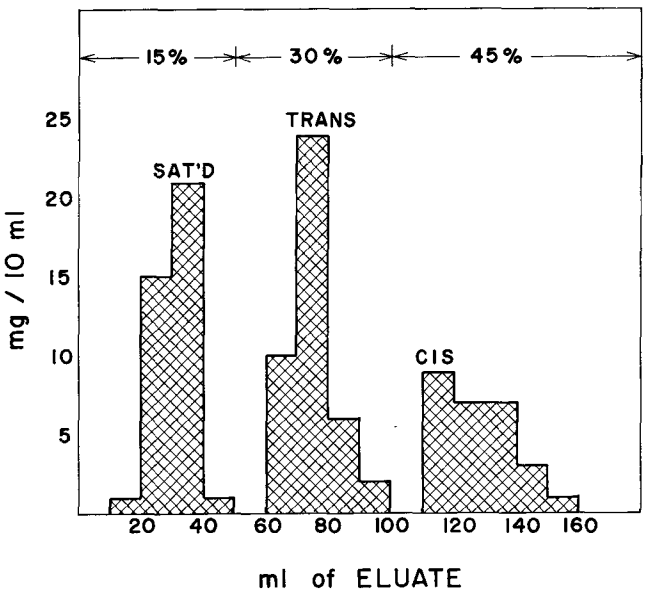

FIG. 1. Column chromatography of 110 mg hydrogenated methyl petroselinate. Adsorbent 15 g $AgNO_3$/silicic acid/celite. Eluant benzene/Skellysolve F (15/85 to 45/55).

Oxidation

The original esters, partially hydrogenated esters, and the *trans* and *cis*-fractions from the partially hydrogenated esters, were oxidized with permanganate-periodate as described by von Rudloff (10). In a typical oxidation, 7 ml of oxidant solution (21 g sodium meta periodate and 25 ml 0.1M potassium permanganate made up to 1 litre with water) was taken in a 50-ml Erlenmeyer flask. To this was added 9 mg potassium carbonate in 1.5 ml of water, 12 ml of water and 8.5 ml tertiary butanol. After the addition of 25 mg of the esters in 5 ml tertiary butanol, the mixture was shaken on a wrist action shaker for 3 hr at room temp. In the case of the erucate esters shaking time was increased to 6 hr. After this time, ethylene was bubbled in to destroy excess oxidant and the tertiary butanol was removed on a water bath with a stream of air. The remaining solution was acidified with 6-7 drops concentrated HCl and extracted 3 times with 25-ml portions of chloroform. The chloroform solution was evaporated at room temp in a fume hood, the remaining mono- and dicarboxlyic acid fragments esterified with diazomethane, and analysed by GLC.

Gas Chromatography

An isothermal gas chromatographic unit with thermal conductivity detectors, constructed in the laboratory, was used for the analysis of original methyl oleate, methyl erucate, and methyl petroselinate. The column was an 8 ft, 3/16 in. diam copper tube packed with phthalic-ethylene glycol polyester on chromosorb W, 1:6 by weight. An F and M 500-temp programmed unit with thermal conductivity detector was used for analysis of the oxidation products. For the oxidation products of methyl oleate and methyl erucate the 2 ft × ¼ in. stainless steel column, with a silicone coated packing supplied with the F and M unit, was used. Programming was from 100–250C at 5.6C/min with an 80 ml/min helium flow rate. With the oxidized products of methyl petroselinate overlapping of some of the mono- and dicarboxylic esters occurred on the silicone column and it was necessary to use a 6 ft × ¼ in. column containing 3% apiezon L on chromosorb W. In the case of the oxidized methyl petroselinate it was also necessary to measure both the

TABLE I
Isomerization of Methyl Erucate

	I.V.	Sat. mole %	Composition of dicarboxylic esters mole %							
			10	11	12	13	14	15	16	17
Non-hydrogenated	70.1	1.0	....	....	0.5	98.5	.....	....	....	....
Hydrogenated, 0.4% catalyst	46.6	32.6	1.4	4.6	12.7	28.8	13.0	4.8	1.4	0.7
Trans 39.4%			0.8	3.1	10.2	10.6	10.4	3.2	1.0	0.1
Cis 28.0%			....	0.7	2.5	20.0	2.4	1.9	0.5	....
Hydrogenated, 0.2% catalyst	59.9	12.7	....	2.5	12.0	60.5	10.5	1.8	....	....
Trans 33.0%			....	1.1	9.1	11.4	9.8	1.3	0.3	....
Cis 54.3%			....	....	2.4	49.5	2.4	....	....	....

TABLE II
Isomerization of Methyl Oleate

	I.V.	Sat. mole %	Composition of dicarboxylic esters mole %					
			7	8	9	10	11	12
Non-hydrogenated	85.2	0.5	....	0.5	99.0	.....	....	....
Hydrogenated, 0.4% catalyst	64.4	23.0	3.9	11.4	45.4	11.2	4.0	1.1
Trans 37.3%			2.3	9.4	9.9	11.1	2.7	1.7
Cis 39.7%			0.8	1.7	34.2	1.2	1.5	0.3
Hydrogenated, 0.2% catalyst	76.0	8.5	....	7.5	72.0	8.7	3.3	....
Trans 24.8%			....	7.3	6.9	9.1	1.2	0.3
Cis 66.7%			....	....	66.0	....	0.7	....

TABLE III
Isomerization of Methyl Petroselinate

	I.V.	Sat. mole %	Composition of dicarboxylic esters mole %				
			4	5	6	7	8
Non-hydrogenated	83.5	2.0	....	0.5	97.5	....	....
Hydrogenated, 0.075% catalyst	55.5	35.6	2.5	12.2	35.0	12.2	2.5
Trans 40.4%			2.2	10.3	14.0	11.3	2.6
Cis 24.0%			....	1.4	21.2	1.4	....

mono- and dicarboxylic fragments and to see that they agreed, as dimethyl adipate and shorter dicarboxylic esters are appreciably volatile and a portion may be lost on standing at room temp.

In all cases, areas under the peaks were measured by triangulation and compositions were converted to mole %.

Results and Discussion

Analytical data on the isomerization of the monoethenoid esters show in Tables I–III. For methyl oleate and methyl erucate, 0.2 and 0.4% Rufert nickel catalyst flakes containing 20% nickel gave satisfactory partial hydrogenations. With methyl petroselinate, 0.2% catalyst completely hydrogenated the sample under the conditions used. Satisfactory partial hydrogenation was obtained with 0.075% catalyst. The difference in catalyst concentration required was probably because the methyl petroselinate was prepared from oil freshly extracted from the seed, whereas the methyl oleate and erucate had been prepared a year before use. In the latter case small amounts of peroxides formed on standing, and probably resulted in destruction of a portion of the catalyst.

For calculations, the *trans*-content was taken from the IR analysis, the saturated esters from GLC analysis, and the *cis* obtained by difference. These results agreed with the weights of the fractions obtained from the silver nitrate-silicic acid column, within 2 unit %, and within 1 unit with the I.V. The proportions of positional isomers in the partially hydrogenated samples and their *cis* and *trans*-fractions are given as the mole % of dicarboxylic as determined by GLC after oxidation The sum of the individual positional isomers in the fractions agreed well with those in the total sample.

The migration of the double bond during hydrogenation takes place equally in both directions from the initial position. This is in agreement with the observation of Allen and Kiess (3) and Knegtel et al. (8). The present results also show that this migration is independent of the location of the double bond along the carbon chain. The same pattern of migration was observed in the three esters, methyl petroselinate, methyl oleate, and methyl erucate, where the double bond is situated in the 6,9 and 13 positions, respectively. Allen and Kiess (3) reported a 2:1 proportion for the *trans* and *cis*-forms of individual positional isomers. In the present study this ratio varied 4:1–7:1, which agrees with the findings of Scholfield et al. (11). This would indicate that during the migration of the double bonds, equilibrium proportions of the *trans* and the *cis*-forms are not obtained under the conditions used or that the *cis* is more readily hydrogenated than the *trans*.

The present method for determining the geometric and positional isomers in fatty acids appears highly satisfactory. The silver nitrate treated silicic acid column gives a sharp separation of the saturated *trans* and *cis*-esters. The oxidation with permanganate-periodate and GLC analysis of the resulting fragments provided a convenient determination of bond positions. The method should prove equally useful in studying the hydrogenation of the polyunsaturated acids.

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