Determination of Glyceride Composition of Several Solid and Liquid Fats

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Abstract

Cocoabutter, Sumatra palm oil, lard, groundnut oil, soybean oil and cottonseed oil have been analyzed by separating the triglycerides according to their degree of unsaturation by means of thin-layer chromatography on silica impregnated with silver nitrate. Of each triglyceride fraction obtained in this way, the fatty acid composition—overall and at the 2-position—has been determined. Moreover, the triglyceride composition of the fractions of cocoabutter, Sumatra palm oil and lard has been determined by means of gas-liquid chromatography. The results confirm the correctness of Vander Wal's theory on the distribution of fatty acids in the triglycerides of vegetable natural fats.

Introduction

THE TRIGLYCERIDE COMPOSITION of natural fats has been studied by different methods (1). Fractional crystallization (2), counter current distribution (3–5) and various oxidation techniques (6–9) are valuable methods. Determination of the fatty acid composition—overall and at the 2-position—by the lipase splitting technique (10,11) offers the possibility to calculate the triglyceride composition (12) of natural fats taking into account the assumptions of Vander Wal (13,14). The separation of triglycerides according to their mol wt by means of GLC (15,16) is very useful in combination with other separation techniques (17).

The quantitative analysis of triglycerides by separating 60–100 mg of sample according to their degree of unsaturation by means of column (18) and especially by TLC on silver nitrate impregnated silica (19, 20) is a very accurate and rapid method. The triglyceride fractions obtained are large enough for the fatty acid as well as the triglyceride composition to be determined by GLC. It is also possible to determine the fatty acid composition at the 2-position of the triglyceride fractions by a micro lipase splitting technique (21). By means of the analytical results thus obtained the triglyceride composition can be accurately calculated.

Experimental

The triglycerides are separated from the more polar substances by column chromatography over silica (22). An amt of 60–80 mg of triglyceride mixture is separated into fractions by TLC on silica impregnated with silver nitrate (19). The glycerides are extracted from the adsorbent and the amt in each fraction determined by means of a glycerol determination with periodic acid after saponification (20). The fatty acids are converted into methyl esters with diazomethane and analyzed gas chromatographically.

The triglyceride fractions obtained from a second separation are used for enzymatic hydrolysis (21) and for the triglyceride analysis by means of temp-programmed GLC. The instrument used for the triglyceride analysis is a F & M Model 609 with a hydrogen flame ionization detector. Analyses of the fully hardened glycerides (unsaturated ones degrade strongly) are carried out on a silver column (50 x 0.3 cm), packed with 10% (w/w) silicone rubbergum S.E. 30 (ex General Electric) on Diatoport S, 80–100 mesh (ex F & M Scientific Corp.). The packing was conditioned by heating it up to 350C for 18 hr. The triglycerides were brought on to the column which was subsequently connected to the F & M apparatus and heated to 200C. Then the programmed heating was switched on (rate 1.8C/min). Nitrogen was used as mobile phase (flow-rate 70–100 ml/min).

Analysis of a mixture of pure synthetic triglycerides (trilaurin, trimyristin, tripalmitin, tristearin) showed that the standard deviation of the mean for each component was less than 0.4% ($\Phi = 5$). The degradation of the tripalmitin and tristearin was constant for one column (ca. 5 and 15%, respectively). The results were corrected accordingly.

Calculation and Results

In the separation of triglycerides on silica layers (measuring 20 x 40 cm) impregnated with silver nitrate, triglycerides having the same number of double bonds, as for instance, OOO and SOL, OOL and SLL, also are separated. Glycerides with the same number of double bonds were nevertheless combined, because from the total fatty acid composition the glyceride composition of the fractions can be calculated without difficulty. The fatty acid composition—overall and at the 2-position—of the fats and their fractions are presented in Tables I to VI.

For the calculation of the glyceride composition of the fractions from their fatty acid compositions, the fatty acids are divided into groups. In addition, the following symbols are introduced:

S	saturated fatty acids	
M	12:0–14:0	main component myristic acid
P	15:0–17:0	main component palmitic acid
St	18:0–20:0	main component stearic acid
O	14:1–22:1	main component oleic acid
L	18:2	linoleic acid
Le	18:3	linolenic acid

From the data in the tables the triglyceride composition of the fractions was calculated (21), making the following assumptions:

1) Each fraction exclusively contains triglycerides having the same number of double bonds.

2) The saturated acyl groups: St, P and M found at the 1- and 3-positions, are statistically distributed over these positions.

Considering that each fraction contains only glycerides having the same number of double bonds and that the acyl groups occupying the 2-position are known, the distribution of the unsaturated fatty acids over the 1- and 3-positions in the fractions is fully established.

The overall fatty acid composition of the fractions found by analysis, has been made to conform to the first assumption by applying minor corrections to the values for the unsaturated acids. The percentages of the saturated fatty acids were changed proportionally. In no case did these corrections exceed 1.5%.

As an example, the glyceride composition of the fraction having three double bonds is calculated.

TABLE I

Fatty Acid Compositions (Mole %) of Cocoa Butter and Its Triglyceride Fractions

Fatty acids	0 Double bond		1 Double bond		2 Double bonds		3 Double bonds		Cocoa butter	
	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position
12:0										
14:0	1.2	0.9	1.2	0.6	0.3	0.4	1.3	1.0	0.7	0.3
16:0	38.6	65.1	26.8	2.0	21.2	4.3	16.0	10.7	25.2	2.4
18:0	58.7	34.0	38.7	2.4	27.0	2.1	15.1	8.8	35.5	1.6
20:0	1.5									
18:1			33.3	95.0	36.3	53.5	41.0	48.7	35.2	89.0
18:2					15.2	39.7	20.8	24.3	3.2	6.4
18:3							5.8	6.5	0.2	0.3

TABLE II

Fatty Acid Compositions (Mole %) of Sumatra Palm Oil and Its Triglyceride Fractions

Fatty acids	0 Double bond		1 Double bond		2 Double bonds		3 Double bonds		4 Double bonds		Sumatra palm oil	
	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position
12:0	0.9	trace	trace	0.3	0.1	0.7	0.2	0.9	trace	0.8	0.1	0.1
14:0	3.6	4.3	1.8	1.0	1.0	0.8	0.6	0.6	0.6	0.8	1.2	0.8
16:0	89.5	91.1	61.2	17.1	38.2	9.0	21.0	4.1	16.2	4.6	46.8	18.2
18:0	5.6	4.6	3.5	1.3	4.8	1.0	2.2	0.6	2.1	0.8	3.8	1.3
20:0	0.4		0.2		0.3		0.1	trace	1.0	trace	0.2	
16:1			trace	1.2	0.5	1.1	1.3	1.0	1.0	1.5		0.6
18:1			33.3	79.1	44.0	61.4	50.0	51.7	29.3	24.5	37.6	59.5
20:1							0.5	trace	0.5		0.3	
18:2					11.1	26.0	24.1	41.1	45.3	63.0	10.0	19.5
18:3									0.4	4.0		

TABLE III

Fatty Acid Compositions (Mole %) of Lard and Its Triglyceride Fractions

Fatty acids	0 Double bond		1 Double bond		2 Double bonds		3 Double bonds		4 Double bonds		Lard	
	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position
12:0	1.2	4.0	0.5	0.2	0.2	0.6	trace	1.2	trace	trace	0.3	0.2
14:0	4.4	11.0	2.4	4.0	1.8	4.5	0.9	1.5	0.6	1.5	1.7	5.7
15:0	0.4	0.8	trace	0.2	0.2	0.3		0.4	trace	1.0	0.1	trace
16:0	44.1	67.6	37.7	71.2	25.8	58.2	11.5	32.0	7.1	17.2	26.2	52.0
17:0	1.0	1.4	0.4	0.9	0.6	0.5	trace	0.6	0.4	1.5	0.5	0.8
18:0	48.0	14.9	25.3	9.0	9.2	4.0	3.1	5.7	1.6	2.0	13.5	5.6
20:0	0.9	0.3	0.4							trace	0.2	trace
14:1				trace	trace					1.0	0.2	trace
15:1					trace				trace	2.5	trace	
16:1			1.0	trace	4.4	6.0	7.0	11.5	6.5	4.5	4.0	8.3
17:1					0.3	0.6	0.5	1.0	0.7	2.0	0.3	0.7
18:1			31.8	14.5	52.8	23.1	60.4	42.1	43.4	24.2	42.9	23.0
20:1			0.5	trace	0.2	0.7	1.1		0.7	trace	0.8	
18:2					4.5	1.5	15.5	4.0	35.0	38.6	9.0	3.7
18:3									4.0	4.0	0.3	

TABLE IV

Fatty Acid Compositions (Mole %) of Groundnut Oil and Its Triglyceride Fractions

Fatty acids	1 Double bond		2 Double bonds		3 Double bonds		4 Double bonds		5 Double bonds		6 Double bonds		Groundnut oil	
	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position
12:0	0.7	0.9	0.3	0.4	0.1								0.3	0.1
14:0	1.8	1.7	0.4	0.5	0.3	0.1	0.2	1.3	trace	trace			0.1	0.1
16:0	39.4	15.9	26.5	5.4	16.2	2.7	6.6	1.9	1.2	1.0	0.5		12.3	1.8
18:0	11.6	3.0	7.1	1.0	3.6	0.5	1.6	0.3					2.5	0.2
20:0	3.4	0.6	2.7	0.4	1.8		0.8	0.1	trace				0.5	0.6
22:0	6.9	1.1	6.2	1.2	2.6	0.3	3.1	0.3	trace		trace		2.5	
24:0	2.9	0.5	2.9	0.4	1.3	0.1	1.6	0.2					1.0	
16:1	trace	0.5	trace	1.1		0.5	0.5	0.7					trace	trace
18:1	32.3	75.7	37.9	54.6	44.2	39.5	37.0	26.7	32.4	10.6	3.5		37.7	31.4
20:1	0.4	0.1	1.2	0.2	2.3		1.0	0.1					0.7	trace
22:1	0.6		2.0	0.3	1.7		0.4						1.0	0.1
18:2			12.8	34.5	25.9	56.3	47.2	68.4	64.9	85.4	93.0		41.1	65.3
18:3									1.5	3.0	3.0		0.3	0.4

TABLE V

Fatty Acid Compositions (Mole %) of Soybean Oil and Its Triglyceride Fractions

Fatty acids	1 Double bond		2 Double bonds		3 Double bonds		4 Double bonds		5 Double bonds		6 and more double bonds		Soybean oil	
	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position
12:0	0.3	0.1	0.1	trace	0.2	trace	trace	trace	trace	trace			trace	
14:0	0.9	2.0	0.3	2.5	0.3	0.1	0.1	0.2	0.2	0.2	0.2	0.1	0.1	trace
15:0	0.6	0.5	0.1	0.5			trace	trace			trace	trace	trace	trace
16:0	46.8	13.5	38.3	8.0	20.7	2.7	15.9	2.5	1.2	1.0	3.3	0.1	10.5	1.4
17:0	0.9	0.3	0.3	0.6	0.4		trace	trace	trace				trace	trace
18:0	15.7	4.5	11.0	2.5	6.5	0.8	4.3	0.8	0.4	0.8	0.9	0.4	3.2	0.3
20:0	1.5		0.2	trace	1.0	0.5	0.5	0.2	trace	0.3			0.2	trace
16:1	2.8	3.4	trace	2.8	trace	0.7	1.0	2.1	0.6	1.0	0.4	1.0	trace	0.3
18:1	30.5	75.7	31.9	41.0	41.9	50.0	24.5	25.9	31.5	24.7	7.0	10.9	22.3	24.8
20:1	trace	trace	0.8		0.2	trace	0.2	0.2	0.8	0.3	trace	0.2	0.9	trace
18:2			17.0	42.1	28.5	45.2	52.9	67.3	62.1	66.7	72.2	75.9	54.5	67.4
18:3					0.3		0.6	0.8	3.2	5.0	16.0	10.5	8.3	5.8

TABLE VI

Fatty Acid Compositions (Mole %) of Cottonseed Oil and Its Triglyceride Fractions

Fatty acids	0 Double bond		1 Double bond		2 Double bonds		3 Double bonds		4 Double bonds		5 Double bonds		6 Double bonds		Cottonseed oil	
	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position	Overall	2-Position
12:0	3.5															
14:0	10.5		2.5		1.3	0.5	0.5	0.3	0.8	0.4	trace	trace	trace		0.8	
16:0	75.5		56.5	14.8	50.2	3.5	27.5	2.7	23.7	2.8	1.0	1.5	1.6		27.3	3.0
18:0	10.5		6.7	0.9	5.0	0.6	4.0	0.4	2.3	0.5	0.2	0.4	0.1	2.5	2.0	0.1
20:0			1.0	trace	0.8										0.3	
16:1			1.5	4.0	0.3	0.8	0.9	0.5	trace	0.9	0.7	2.1	trace		0.8	0.9
18:1			31.8	80.3	18.4	25.9	35.1	52.5	12.8	13.0	31.2	47.2	2.5	3.5	18.3	31.7
20:1			trace						0.3							
18:2					24.0	68.7	32.0	43.6	60.1	82.4	65.9	48.8	93.3	94.0	50.5	64.3
18:3											1.0		2.5		trace	

OOO + SOL = %O at 2-position
SLO = %L at 2-position
OSL + SSLe = %S at 2-position
SLeS = %Le at 2-position

Since 3 × %Le total = SSLe + SLeS, SSLe and subsequently OSL can be calculated. 3 × %L total = SOL + SLO + OSL; since OSL and SLO are known, SOL can now be calculated, so that also OOO is known.

With the help of assumption 2, the percentages of StOL, POL and MOL can be calculated from the percentage of SOL and the ratio St : P : M at the 1,3-positions. From the ratio St : P : M at the 2-position and the percentage of OSL, the percentages of OStL, OPL and OML are calculated. In this way exact calculation of the fractions having 0,1,2 and 3 double bonds is possible (Table VII, columns "Calculated").

For the fractions containing linolenic acid, and having four, five or six double bonds, exact calculation of the triglyceride composition is not possible for lack of sufficient information. This problem was solved by attributing a certain value to one minor component. As an example, the calculation is given of the triglyceride composition of the soybean oil fraction having 5 double bonds.

$$\text{LSLe} = 2.3\%\ \text{S at 2-position} \quad [a]$$
$$\text{LOL} + \text{OOLe} = 26.0\%\ \text{O at 2-position} \quad [b]$$
$$\text{OLL} + \text{SLLe} = 66.7\%\ \text{L at 2-position} \quad [c]$$
$$\text{OLeO} + \text{SLeL} = 5.0\%\ \text{Le at 2-position} \quad [d]$$
$$\text{SLLe} + \text{LSLe} + \text{SLeL} = 3 \times 1.8\%\ (\text{S total}) \quad [e]$$
$$(\text{LOL} + \text{OLL}) + 2(\text{OOLe} + \text{OLeO}) = 3 \times 32.9\%\ (\text{O total}) \quad [f]$$
$$2(\text{LOL} + \text{OLL}) + (\text{SLLe} + \text{LSLe} + \text{SLeL}) = 3 \times 62.1\%\ (\text{L total}) \quad [g]$$
$$(\text{OOLe} + \text{OLeO}) + (\text{SLLe} + \text{LSLe} + \text{SLeL}) = 3 \times 3.2\%\ (\text{Le total}) \quad [h]$$

Of these eight equations, six are independent. As there are seven unknowns, no exact solution is possible. Substracting the sum of [a] and [d] from equation [h] we find: OOLe + SLLe = 2.3. To enable calculation of all the components, a value of 1.2 was attributed to SLLe. In this way only a minor uncertainty is introduced into the triglyceride composition of soybean oil.

However, the triglyceride composition of the most unsaturated fraction of soybean oil cannot be calculated in this manner, because it contains triglycerides with six, seven and eight double bonds. Although it has been possible to separate this fraction again into three groups on silica impregnated with a small amt of silver nitrate (6 g $AgNO_3$ on 140 g silica) no reproducible results have been obtained till now. The fatty acid compositions—overall and at the 2-position—of this fraction of soybean oil are in agreement with those which can be deduced from the triglycerides with six and more double bonds found by Coleman's method (Table IX).

The triglyceride composition of each fat was calculated from the percentages and the triglyceride composition of the fractions (Tables VIII and IX). In these tables the triglyceride compositions calculated according to Coleman's method are also given. For the liquid oils the saturated fatty acids are incorporated in the group saturated fatty acids S.

For fractions having three or more double bonds and containing only few triglycerides with two saturated acyl groups, the triglyceride composition according to total number of carbon atoms in the acyl groups can be calculated directly, without having recourse to assumption 2, from their total fatty acid composition. Using the techniques described here, it is, for example, impossible to establish the distribution of the saturated fatty acids in a mixture of SOL and SLO.

For fractions having 0,1 and 2 double bonds, representing the major components in solid fats, assumption 2 is used in the calculation of the triglyceride composition of the fractions. In this case, gas-chromatographic analysis of the triglycerides supplies fresh data, which enable the correctness of assumption 2

TABLE VII

Triglyceride Compositions (Mole %) of the Fractions of Solid Fats

	Cocoa butter											
0 Double bond[a]	Calculated[b]	GLC[c]	1 Double bond	Calculated	GLC	2 Double bonds	Calculated	GLC	3 Double bonds	Calculated	GLC
46 PPM	0.4		48 MOP	1.2		48		1.3			
48 PMSt	0.3		48 PMO	0.2		50 PLP	7.3		50 MOL	1.1	
48 MPSt	1.3	8.1	50 MOSt	1.7		50 PPL	1.6	11.6	50 MLO	1.0	6.2
48 PPP	4.2		50 POP	15.3	17.2	52 POO	22.9		50 PLeP	1.6	
48 PStM	0.2		50 StMO	0.4		52 PLSt	19.3		50 PPLe	2.8	
50 StMSt	0.4		50 PPO	0.8		52 PStL	0.8	41.1	52 POL	13.8	
50 PPSt	24.2	27.0	52 POSt	44.5		52 StPL	2.1		52 PLO	11.7	
50 MStSt	0.7		52 StPO	1.2	49.0	54 StOO	30.4		52 OPL	5.0	33.9
50 PStP	2.2		52 PStO	1.0		54 StLSt	12.8	42.5	52 PLeSt	3.0	
52 StPSt	35.0	42.7	54 StOSt	32.3	33.8	54 StStL	1.0		52 StPLe	2.7	
52 PStSt	12.6		54 StStO	1.4		56		3.5	52 PStLe	2.3	
54 StStSt	18.3	22.2							54 OOO	20.2	
									54 StOL	13.6	
									54 StLO	11.6	55.6
									54 OStL	4.1	
									54 StLeSt	1.5	
									54 StStLe	2.3	
									56		4.3
Remaining ones	0.2					Remaining ones	1.8		Remaining ones	1.7	

to be checked. As liquid fats contain only a small percentage of glycerides having 0,1 and 2 double bonds, only the triglyceride fractions of cocoabutter, Sumatra palm oil and lard were analyzed with temp-programmed gas chromatography (Table VII). The fractions of lard with three and four double bonds have not been analyzed in this way, because they contain mono-unsaturated acyl groups with different chain-lengths.

Discussion

The method by which 60–80 mg of triglycerides can be separated into fractions according to their number of double bonds on silver nitrate/silica plates (20 x 40 cm) followed by extraction of the glycerides from the adsorbent, offers the possibility of obtaining an almost complete triglyceride analysis. The fractions are large enough to determine the amt of each fraction by means of a simple and reliable titration method of the glycerol formed by saponification of the glycerides.

Moreover, the fatty acid composition—overall and that at the 2-position—can be determined; the latter by means of a micro lipase splitting technique. From these data the percentages of the groups SSS, SSO and SOS; SOO, OSO, SSL and SLS; OOO, SOL, SLO, OSL, SSLe and SLeS, etc. can be calculated. The results obtained in this way agree reasonably to those obtained by Coleman's method of calculation (Tables VIII, IX). This only proves that the fatty acids—saturated (taken as one group), oleic, linoleic and linolenic—at the 1- and 3-position are randomly distributed over these positions.

In this stage nothing is known about the different saturated fatty acids at the 1- and 3-position. A gas-chromatographic analysis of the triglyceride fractions will only yield fresh data if the triglycerides contain at least two saturated acyl groups, for in this case the triglycerides are separated according to their mol wt, and the most frequently occurring unsaturated acyl groups have 18 C-atoms (except in the case of two lard fractions). For this reason, the gas-chromatographic analysis of triglycerides was only applied to the fractions of the solid fats. In Table VII the triglyceride compositions of the fractions of cocoabutter, Sumatra palm oil and lard—assuming a random distribution of the saturated fatty acids at the 1- and 3-position—have been compared with the gas-chromatographic analyses. It appeared that for the first two fats there is a good agreement between the calculated values and those determined experimentally. Lard is an exception; in the fraction with

TABLE VIII

Triglyceride Compositions (Mole %) of Solid Fats

Cocoa butter

0 Double bond	A[a]	B[b]	1 Double bond	A	B	2 Double bonds	A	B	3 and more double bonds	A	B
PMSt		0.1	MOP	0.9	0.6	POO	3.7	5.4	OOO	0.5	0.6
StMSt		0.1	MOSt	1.3	0.8	StOO	4.9	7.7	POL	0.4	1.1
PPP	0.1	0.3	POP	12.0	11.9	PLP	1.2	0.8	StOL	0.4	1.5
PPSt	0.5	0.9	POSt	34.8	34.2	PLSt	3.1	2.5	PLO	0.3	0.4
StPSt	0.8	0.7	StOSt	25.2	24.5	StLSt	2.0	1.8	StLO	0.3	0.6
PStP		0.2	PMO	0.2					SLeS		0.2
PStSt	0.3	0.6	StMO	0.3					Remaining ones	0.7	0.1
StStSt	0.4	0.4	PPO	0.6	0.1				4 Double bonds	0.9	0.8
			StPO	0.9	0.2						
			PStO	0.8	0.1						
			StStO	1.1	0.1						
Remaining ones	0.1	0.2	Remaining ones	0.2	0.2	Remaining ones	1.1	0.3			
	2.2	3.5		78.3	72.7		16.0	18.5		3.5	5.3

Sumatra palm oil

0 Double bond[a]	Calculated[b]	GLC[c]	1 Double bond	Calculated	GLC	2 Double bonds	Calculated	GLC	3 Double bonds	Calculated	GLC	4 Double bonds	Calculated	GLC
44 MMP	0.3	0.7	48 MOP	3.4	4.1	48 MLP	0.7	1.3	48		1.5	50		2.6
46 PMP	3.1	7.5	48 PMO	1.2		50 MOO	1.0		50 MOL	0.3		52 PLL	37.4	
46 MPP	7.6		50 POP	68.3	82.9	50 PPL	4.8	25.8	50 MLO	0.6	2.6	52 LPL	2.0	
48 PMSt	0.5		50 PPO	15.7		50 PLP	19.5		50 OML	1.5		52 POLe	3.3	44.4
48 PPP	71.5	75.1	52 POSt	8.2		52 POO	54.1		52 POL	22.3		52 PLeO	3.3	
48 MPSt	0.3		52 StPO	0.9	13.0	52 OPO	3.3	66.7	52 PLO	36.6	61.3	52 OPLe	2.6	
50 PPSt	10.8	15.1	52 PStO	1.2		52 PLSt	5.3		52 OPL	4.1		54 OOL	22.0	
50 PStP	3.3		54 StOSt	0.2		54 StOO	7.4	6.2	54 OOO	27.7		54 OLO	18.2	50.3
52 StPSt	0.4	1.6							54 StOL	2.4	34.6	54 StLL	7.2	
52 PStSt	0.5								54 StLO	3.9		56		2.7
									54 OStL	0.6				
Remaining ones	1.7		Remaining ones	0.9		Remaining ones	3.9					Remaining ones	4.0	

Lard

0 Double bond[a]	Calculated[b]	GLC[c]	1 Double bond	Calculated	GLC	2 Double bonds	Calculated	GLC	3 Double bonds	Calculated	GLC	4 Double bonds	Calculated	GLC
46 PMP	1.6	1.6	48 PMO	1.5	3.1	48 PML	0.4	0.7	OML	2.7		LML	1.0	
48 PMSt	6.5	16.2	48 MPO	2.8		48 MPL	0.2		POL	0.3		OMLe	0.5	
48 PPP	7.7		50 POP	2.0		50 MOO	0.5		PLO	1.1		LPL	13.3	
50 StMSt	6.5		50 StMO	2.5	22.5	50 OMO	4.2		OPL	33.0		PLL	0.7	
50 PPSt	30.6	52.0	50 PPO	26.5		50 StML	0.5	9.5	OOO	53.5		POLe	0.2	
50 PStP	1.7		52 POSt	6.3		50 PPL	4.7		StOL	0.8		PLeO	2.0	
52 StPSt	30.2	28.9	52 StPO	43.0	69.1	50 PLP	0.3		StLO	2.9		OPLe	6.4	
52 PStSt	6.6		52 PStO	3.3		52 POO	13.8		OStL	5.7		OOL	33.8	
54 StStSt	6.5	1.3	54 StOSt	5.1	5.3	52 OPO	48.6	69.7				OLO	37.2	
			54 StStO	5.4		52 StPL	5.5					LStL	1.3	
						52 PLSt	0.7					StLL	0.7	
						54 StOO	16.1					StOLe	0.2	
						54 OStO	3.3	19.1				StLeO	2.0	
						54 StStL	0.4					OStLe	0.7	
						54 StLSt	0.4							
						56		1.0						
Remaining ones	2.1		Remaining ones	1.6		Remaining ones	0.4							

[a] Figure before the triglyceride is the sum of the number of C-atoms of the acyl groups.
[b] Calculated from fatty acid compositions of the fractions.
[c] Separation according to the number of C-atoms.

0 double bonds there is a preference for triglycerides with 50 C-atoms and in the fraction with one double bond for triglycerides with 52 C-atoms. This is not surprising, however, since in lard palmitic and shorter fatty acids are preferably at the 2-position; being in contrast with stearic acid (Table III). There is therefore a clear difference between the saturated fatty acids with different chain lengths.

From the analytical results in may be concluded that cocoabutter and Sumatra palm oil completely follow the distribution pattern of Vander Wal and Coleman. In the case of the vegetable oils—groundnut, soybean and cottonseed oil—this could only be demonstrated for the saturated fatty acids taken as one group, S, and for the individual, unsaturated acids.

In lard, the saturated fatty acids taken as one group, S and the individual, unsaturated fatty acids are likewise distributed over the triglycerides in accordance with Vander Wal and Coleman's theory. The individual, saturated fatty acids at the 1,3-positions, however, are not randomly distributed over these positions.

TABLE IX
Triglyceride Compositions (Mole %) of Liquid Fats

Groundnut oil

1 Double bond	A[a]	B[b]	2 Double bonds	A	B	3 Double bonds	A	B	4 Double bonds	A	B	5 Double bonds	A	B	6 Double bonds	A	B
SOS	2.3	2.4	SOO	8.7	7.5	OOO	6.2	5.9	OOL	7.8	7.9	OLL	17.8	16.4	LLL	3.9	5.5
SSO	0.7	0.4	OSO	0.8	0.5	SOL	5.0	5.0	OLO	8.7	12.3	LOL	2.2	2.6	OLLe }		
			SSL	0.6	0.3	SLO	15.7	15.5	SLL	10.7	10.3	SLeL }			LOLe }	0.4	0.4
			SLS	5.3	4.9	OSL	1.0	1.0	LSL	1.2	0.6	LSLe }	0.7	0.3	OLeL }		
									SLeO		0.1	SLLe }					
												OLeO }	0.2	0.2			
												OOLe }					
															Remaining ones	0.1	
	3.0	2.8		15.4	13.2		27.9	27.4		28.4	31.2		20.9	19.5		4.4	5.9

Sumatra palm oil

0 Double bond	A[a]	B[b]	1 Double bond	A	B	2 Double bonds	A	B	3 Double bonds	A	B	4 Double bonds	A	B
PMP	0.3	0.3	MOP	1.3	1.1	POO	18.9	19.1	OOO	3.2	4.2	OOL	1.5	2.2
MPP	0.6	0.3	POP	25.9	21.8	StOO	2.6	1.7	POL	2.6	4.8	OLO	1.3	1.5
PPP	6.1	6.6	POSt	3.1	3.9	OPO	1.2	1.3	PLO	4.3	6.2	PLL	2.6	1.5
PPSt	0.9	1.2	PPO	6.0	5.8	PPL	1.7	1.5	StLO	0.5	0.6	StLL	0.5	0.1
PStP	0.3	0.5	StPO	0.3	0.5	PLP	6.8	7.1	OPL	0.5	0.6	SOLe }		
			PStO	0.5	0.4	PLSt	1.9	1.2				SLeO }	0.8	
												OSLe }		
Remaining ones	0.3	0.1	Remaining ones	0.8	0.7	Remaining ones	1.9	1.2	Remaining ones	0.6	0.7	Remaining ones	0.2	1.3
	8.5	9.0		37.9	34.2		35.0	33.1		11.7	17.1		6.9	6.6

Lard

0 Double bond	A[a]	B[b]	1 Double bond	A	B	2 Double bonds	A	B	3 Double bonds	A	B	4 and more double bonds	A	B
PMP	0.1	0.1	POP	0.6	0.6	POO	5.2	5.0	OOO	11.7	10.2	OOL	1.4	4.2
PMSt	0.4	0.3	POSt	1.9	1.6	StOO	6.1	6.4	PLO	0.2	0.6	OLO	1.5	1.2
StMSt	0.4	0.2	StOSt	1.5	1.0	OMO	1.6	1.9	StLO	0.6	0.7	LPL	0.5	0.7
PPP	0.5	0.1	PMO	0.4	0.9	OPO	18.4	16.8	OML	0.6	0.8	LStL	0.1	0.1
PPSt	2.0	2.6	StMO	0.7	1.2	OStO	1.2	1.8	OPL	7.2	7.0	OPLe	0.3	0.2
StPSt	2.0	1.7	MPO	0.8	....	PPL	1.8	1.7	OStL	1.2	0.7	OLL	....	0.5
PStP	0.1	....	PPO	7.9	8.2	StPL	2.1	2.2						
PStSt	0.4	0.3	StPO	12.8	10.6									
StStSt	0.4	0.2	PStO	0.9	0.9									
			StStO	1.6	1.1									
Remaining ones	0.3	0.1	Remaining ones	0.6	0.4	Remaining ones	1.5	1.4	Remaining ones	0.3	2.6	Remaining ones	0.2	1.2
	6.6	5.6		29.7	26.5		37.9	37.2		21.8	22.6		4.0	8.1

[a] "A" Calculated from the fatty acid compositions of the fractions.
[b] "B" Calculation according to Coleman.

Soybean oil

1 Double bond	A[a]	B[b]	2 Double bonds	A	B	3 Double bonds	A	B	4 Double bonds	A	B	5 Double bonds	A	B	6 and more double bonds	A	B
SOS	1.1	1.0	SOO	2.9	2.3	OOO	1.5	1.2	OOL	6.7	5.4	OLL	13.5	14.4	LLL		15.7
SSO	0.3	0.2	OSO	0.4	0.1	SOL	4.1	4.9	OLO	2.5	3.3	LOL	5.1	5.8	OLLe		2.9
			SSL	0.6	0.3	SLO	5.0	6.0	SLL	13.9	13.0	OOLe	0.2	1.0	OLeL		1.2
			SLS	2.8	2.7	OSL	0.4	0.4	LSL	0.9	0.4	OLeO	0.6	0.3	LOLe		2.3
						SSLe	0.1	0.1	SOLe	0.2	1.0	SLLe	0.3	2.6	SLeLe		0.2
						SLeS		0.2	SLeO	0.2	0.5	SLeL	0.4	1.1	OLeLe		0.3
									OSLe		0.1	LSLe	0.5	0.2	LeOLe		0.2
															LLLe		6.2
															LLeL		1.3
															LLeLe		0.6
															LeLLe		0.5
															LeLeLe		0.1
	1.4	1.2		6.7	5.4		11.1	12.8		24.4	23.7		20.6	25.4		35.8	31.5

Cottonseed oil

0 and 1 double bond	A[a]	B[b]	2 Double bonds	A	B	3 Double bonds	A	B	4 Double bonds	A	B	5 Double bonds	A	B	6 Double bonds	A	B
SSS	0.5	0.6	SOO	4.8	3.5	OOO	0.8	0.5	OOL	4.1	3.5	OLL	6.4	6.9	LLL	13.0	12.2
SOS	4.5	6.3	OSO	0.3	0.1	SOL	9.4	12.5	OLO	1.6	1.0	LOL	6.5	6.2			
SSO	0.8	0.4	SSL	0.6	1.2	SLO	8.4	7.0	SLL	22.5	24.7						
			SLS	12.4	12.5	OSL	0.6	0.3	LSL	1.1	0.6						
												Remaining ones	0.4		Remaining ones	1.3	
	5.8	7.3		18.1	17.3		19.2	20.3		29.3	29.8		13.3	13.1		14.3	12.2

[a] "A" Calculated from the fatty acid compositions of the fractions.
[b] "B" Calculation according to Coleman.

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Preparation of Malonaldehyde Acetals by Ozonolysis of Polyunsaturated Fatty Esters[1]

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Abstract

Malonaldehyde acetals were prepared in more than a 70% yield by ozonolysis of the methyl esters of linseed oil, safflower oil and linoleic acid, and by ozonolysis of linseed oil alone. Malonaldehyde tetramethyl acetal could not be separated readily from caproaldehyde dimethyl acetal by fractional distillation. However, conversion of the methyl acetals to propylene glycol acetals resulted in sufficient spread in boiling points for their effective separation by distillation.

Introduction

IN OUR LABORATORIES, studies have been made on the reaction of ozone with unsaturated fatty acids and on the chemistry of the products, as part of a program to increase the utilization of linseed and soybean oils.

It has previously been reported (5) that ozonolysis of methyl oleate in methanol at 0C, followed by reduction, usually by zinc and acetic acid, gives the products expected by cleavage of the 9,10 double bond, pelargonaldehyde and methyl azelaaldehydate, in high yields. Methyl azelaaldehydate is, of course, also formed by the ozonolysis of all fatty acids in which the first site of unsaturation is at C_9. It is, therefore, a readily accessible product from the ozonolysis of oils rich in these unsaturated acids, such as linseed and soybean oils. Methyl azelaaldehydate is a potentially important monomer intermediate, and preliminary studies on polymers from pentaerytheritol acetal have already been reported (7).

Since methyl azelaaldehydate is produced only from the first 9 carbon atoms of the fatty acid molecule, an investigation was started to recover useful reactive products from the other half of the molecule. Ozonolysis of methyl linoleate would be expected to give 1 mole of caproaldehyde, 1 mole of malonaldehyde, and 1 mole of methyl azelaaldehydate. Similarly, methyl linolenate should give 1 mole of propionaldehyde, 2 moles of malonaldehyde, and 1 mole of methyl azelaaldehydate. Accordingly, malonaldehyde should be a major product of the ozonolysis of oils rich in these acids. As a simple β-dialdehyde it should be a reactive and potentially useful product.

Malonaldehyde has been prepared previously as its tetraalkyl acetal by the acid-catalyzed condensation of an allyl-alkyl ether with a trialkylorthoformate (3), a method limited by the high cost of the orthoformate ester, by the ozonolysis of 1,4-dibutoxybut-3-ene (2), and by the ozonolysis of the esters of fatty acids (1). Ozonolysis of methyl linoleate in methanol at 0C, followed by reduction with hydrogen and a 3% palladium-on-barium-sulfate catalyst reportedly (1) gave a mixture of aldehydes. Treating this mixture with a catalytic amt of H_2SO_4 in methanol gave the methyl acetals. These were separated by distillation in good yields; however, the products were not characterized, and no mention was made of impurities. Attempts were made to repeat this work.

Results

The methyl esters of linseed oil fatty acids were ozonized and the product was reduced. Reduction was slow if a 3% palladium-barium sulfate catalyst was used. Lindlar catalyst (4) gave better yields more rapidly. In Figure 1 are shown analyses by gas-liquid chromatography (GLC) of the acetals produced from both catalysts in the reduction of ozonolysis products of linseed oil esters. As clearly seen, the amt of malonaldehyde formed relative to the other products is high when Lindlar catalyst is used.

If the ozonolysis product was reduced with zinc and acetic acid, a more complex mixture of products was obtained, and of this mixture, malonaldehyde was only a minor constituent.

The aldehydes in methanol were converted to their methyl acetals by standing overnight with a catalytic amt of H_2SO_4, after which time the acid was removed by stirring the product for 2 hr with a large excess of sodium carbonate. This step was necessary to remove every trace of acid; otherwise failure to do so always led to much polymerization during distillation of the acetals. Fractionation of the acetals was not as straightforward as one would be led to believe from reference 1, and complete fractionation could not be achieved even when a spinning band distillation column was used with a reflux ratio of 120:1. Malonaldehyde tetramethyl acetal fractions were always contaminated with 5–10% of caproaldehyde dimethylacetal, and methyl azelaaldehydate fractions always contained ca. 10% dimethyl azelate (6).

Other alcohols were tried as common ozonization solvents and acetalating agents without much success.

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