

idly and that adequate numbers of microorganisms are present in all samples to cause biodegradability. The initial inoculum is not critical as long as it is adequate to cause the lauryl sulfate to degrade in two days. Increasing the inoculum beyond this point does not seem to cause an appreciable increase in the rate of degradation and it does not cause a more complete degradation. Final levels of degradation are the same as in the standard river die-away test, with the ac-

celerated test simply compressing the test time to less than one-half.

REFERENCES

1. Hammerton, G., *J. Appl. Chem.* 5, 517 (1955).
2. Sawyer, C. N., R. H. Bogan and J. R. Simpson, *Ind. Eng. Chem.* 48, 2, 236 (1956).
3. The Procter & Gamble Co., personal communication.
4. Standard Methods for Examination of Water and Wastewater, 11th Edition, Part I, Surfactants (Antonie) 1960, p. 246-251.

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Determination of the Glyceride Structure of Fats; Analysis of 14 Animal and Vegetable Fats¹

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Abstract

The glyceride compositions of seven animals and seven vegetable fats have been determined by GLC analysis of the oxidized esterified glycerides as described in an earlier paper in this series. The compositions determined are compared with those calculated from lipase hydrolysis data according to the method of VanderWal. Good agreement was found between the calculated and determined compositions for the majority of the 14 fats. The exceptions were human fat and the more saturated vegetable fats, palm oil and cocoa butter, where some discrepancies occurred.

Introduction

THE GLC ANALYSIS of oxidized esterified glycerides (1) provides a rapid and accurate means of determining the fatty acid distribution in natural fats. Initial investigations on a few fats showed good agreement between the determined glyceride composition and that calculated from lipase hydrolysis data according to the method of VanderWal (2). The present work extends this type of analysis to 14 animal and vegetable fats. The results obtained are compared with those calculated from lipase hydrolysis data.

Experimental

The vegetable oils studied were commercial, refined and bleached samples. The animal fats were cold-extracted with chloroform from fresh adipose tissue. Twenty mg of each fat was oxidized with permanganate-periodate as described earlier (1). The resulting azelao-glycerides were methylated with diazomethane and the oxidized esterified glycerides analysed by GLC. Conditions for GLC analysis and methods of calculation were as previously described (1).

The original fatty acid compositions of the fats were determined by GLC of their methyl esters on an *o*-phthalic-ethylene glycol column. The method of Youngs (3) was used for lipase hydrolysis in which the composition of the liberated fatty acids was determined rather than that of the unhydrolysed mono-glycerides. All results are reported as mole percentages.

Results and Discussion

Tables I and II give the glyceride compositions as determined and as calculated from lipase hydrolysis data. Since the separation of the oxidized esterified

glycerides by GLC is dependent on their effective carbon number, those glycerides giving rise to the same carbon number emerge together and are determined as a group as indicated in the tables. The individual glycerides can be calculated from lipase hydrolysis data and this has been done. Comparisons are then made on the basis of the sum of the calculated glycerides in a group where two or more glycerides have the same carbon number. Since myristic acid is a minor component in the fats investigated, the calculated proportion of myristic-containing glycerides is small. Glycerides containing more than one myristic acid are less than 0.1%.

Tables III and IV give the original fatty acid compositions of the fats, as determined by GLC of their methyl esters, and those calculated from the glyceride compositions obtained. The agreement between these compositions for the individual fats serves as a check on the glyceride analysis. The fatty acid composition in the 1-3 positions is also given in the Tables. These latter figures were used in calculating the glyceride compositions on the basis of VanderWal's theory.

Good agreement between the determined and calculated compositions was found for linseed, corn, olive, cottonseed and soybean oils. For the more saturated fats, cocoa butter and palm oil, the proportion of di-saturated glycerides found was somewhat higher than that calculated, with the remaining glycerides being lower than calculated. In view of the good agreement obtained for the other vegetable fats, this suggests that the actual glyceride distribution for the more saturated vegetable fats may be slightly different than that predicted from lipase hydrolysis data. This however, requires further investigation.

Agreement between the calculated and determined glyceride compositions for the animal fats was generally good with the exception of human fat. In the latter case considerably more monosaturated glycerides were found than would be expected from lipase hydrolysis data, with a corresponding drop in the proportions of the fully unsaturated glycerides and more saturated glycerides. A similar pattern was found for two other samples of human fat. Since humans undoubtedly receive a much higher proportion of dietary fat than the other animals tested, this discrepancy may represent the effect of combined endogenous and exogenous fats.

In general it appears that glyceride composition calculated on the basis of lipase hydrolysis data provides a good estimate for the majority of natural

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TABLE I
 Glyceride Composition of Vegetable Fats

Carbon No.	36	38	40	42	44	46	48	50	52	54
Possible glycerides	U ₃	MU ₂	PU ₂ M ₂ U	SU ₂ MPU M ₃	P ₂ U MSU M ₂ P	PSU MP ₂ M ₂ S	SU ₂ P ₃ MPS	P ₂ S MS ₂	S ₂ P	S ₃
Palm oil										
Found	5.9	0.9	30.5	7.6	35.6	8.8	9.1	1.3	0.3	...
Calc.	9.4	1.1	29.9	6.1	30.1	9.1	9.8	3.8	0.5	...
Linseed oil										
Found	73.0	...	17.3	8.0	1.0	0.6
Calc.	69.6	...	18.9	8.4	1.5	1.3	0.3
Corn oil										
Found	60.5	...	30.0	4.3	4.0	1.1	0.2
Calc.	60.2	...	31.1	3.8	4.4	0.3	0.3	0.1
Cocoa butter										
Found	0.8	...	6.2	8.8	19.1	39.9	22.7	1.4	1.1	...
Calc.	1.6	...	9.7	11.3	15.1	35.3	21.3	2.2	2.5	1.0
Olive oil										
Found	59.6	...	29.8	5.8	3.8	1.0
Calc.	59.9	...	28.7	6.0	3.7	1.4	0.3
Cottonseed oil										
Found	27.1	2.0	42.4	6.5	18.0	3.0	1.0
Calc.	26.6	1.5	43.6	5.8	18.8	3.0	0.7
Soybean oil										
Found	55.5	...	28.0	9.2	4.8	2.3	0.2
Calc.	56.5	...	27.2	10.0	3.3	2.4	0.6

U—unsaturated acids; M—myristic acid; P—palmitic acid and S—stearic acid.

 TABLE II
 Glyceride Composition of Animal Fats

Carbon No.	36	38	40	42	44	46	48	50	52	54
Possible glycerides	U ₃	MU ₂	PU ₂ M ₂ U	SU ₂ MPU M ₃	P ₂ U MSU M ₂ P	PSU MP ₂ M ₂ S	SU ₂ P ₃ MPS	P ₂ S MS ₂	S ₂ P	S ₃
Human										
Found	21.8	3.6	45.7	13.6	12.6	2.7
Calc.	32.0	3.4	34.5	10.1	11.9	5.4	1.8	0.7	0.1	...
Dog										
Found	20.3	4.2	24.8	20.7	14.1	10.2	4.4	0.9	0.4	...
Calc.	22.8	4.4	25.8	17.1	11.7	10.8	4.2	2.0	0.8	...
Ground squirrel										
Found	55.0	3.0	33.1	4.2	4.2	0.5
Calc.	55.8	2.0	31.6	4.0	5.2	1.1	0.3
Chicken										
Found	31.6	1.5	36.7	10.6	12.9	4.7	1.6	0.4
Calc.	31.7	1.6	34.9	11.1	11.9	6.3	1.7	0.7	0.1	...
Pig										
Found	14.5	2.3	38.0	14.3	6.7	18.3	1.9	1.8	2.2	...
Calc.	13.6	2.0	37.1	12.7	7.7	19.7	2.4	2.2	2.4	0.2
Rabbit										
Found	27.6	3.6	36.4	13.5	12.0	4.3	2.0	0.6
Calc.	31.1	4.3	34.2	9.5	13.4	4.9	2.0	0.6
Guinea pig										
Found	31.1	...	34.3	14.0	13.2	5.0	2.0	0.4
Calc.	33.0	...	35.9	9.2	13.7	6.0	1.3	0.9

U—unsaturated acids; M—myristic acid; P—palmitic acid and S—stearic acid.

 TABLE III
 Fatty Acid Composition of Vegetable Fats

	M	P	S	U
Palm oil				
Original	1.7	45.5	6.3	46.5
Calc.	1.7	46.7	5.7	45.9
1-3 positions	2.2	51.9	6.5	39.4
Linseed oil				
Original	...	7.8	3.4	88.8
Calc.	...	6.8	2.9	90.3
1-3 positions	...	10.2	5.1	84.7
Corn oil				
Original	...	13.5	1.8	84.7
Calc.	...	13.1	1.9	85.0
1-3 positions	...	17.7	2.5	79.8
Cocoa butter				
Original	...	28.1	32.9	38.3
Calc.	...	29.1	32.8	38.1
1-3 positions	...	40.1	46.8	13.1
Olive oil				
Original	...	12.6	2.6	84.8
Calc.	...	12.7	2.3	85.0
1-3 positions	...	19.0	4.1	76.9
Cottonseed oil				
Original	1.0	29.8	2.4	66.8
Calc.	1.1	28.5	2.8	67.6
1-3 positions	1.2	42.9	2.4	53.5
Soybean oil				
Original	...	12.2	4.4	83.4
Calc.	...	13.3	4.0	82.7
1-3 positions	...	18.1	6.6	74.7

U—unsaturated acids; M—myristic acid; P—palmitic acid and S—stearic acid.

 TABLE IV
 Fatty Acid Composition of Animal Fats

	M	P	S	U
Human				
Original	2.4	23.4	5.2	69.0
Calc.	2.0	25.3	4.6	68.1
1-3 positions	2.4	28.5	6.8	62.3
Dog				
Original	4.2	22.8	11.7	61.0
Calc.	3.8	24.0	11.3	60.9
1-3 positions	2.7	22.9	16.5	58.2
Ground squirrel				
Original	1.0	14.8	1.5	82.7
Calc.	1.0	14.2	1.5	83.3
1-3 positions	0.9	20.1	2.0	77.0
Chicken				
Original	1.1	23.6	6.2	69.1
Calc.	1.0	24.0	5.1	70.1
1-3 positions	1.1	28.9	9.0	61.0
Pig				
Original	1.5	26.0	12.5	60.0
Calc.	1.0	25.8	13.8	59.4
1-3 positions	0.7	8.2	18.9	72.2
Rabbit				
Original	3.2	24.6	5.3	66.9
Calc.	3.5	24.0	5.5	67.0
1-3 positions	1.8	27.4	7.8	63.0
Guinea pig				
Original	...	24.0	5.9	70.1
Calc.	...	23.6	6.9	69.5
1-3 positions	...	27.3	8.5	64.2

U—unsaturated acids; M—myristic acid; P—palmitic acid and S—stearic acid.

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REFERENCES

1. Youngs, C. G., and M. R. Subbaram, *JAACS* 41, 218 (1964).
2. VanderWal, R. J., *Ibid.* 37, 18 (1960).
3. Youngs, C. G., *Ibid.* 38, 62 (1961).
4. Subbaram, M. R., and C. G. Youngs, *Ibid.*, in press.

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fats, particularly for those of commercial interest. Only the individual saturated acids have been considered here, the unsaturated acids being considered as a group. That the distribution of individual unsaturated acids may also be calculated in this way is indicated by a previous publication (4) where the distribution of each individual acid was determined for two fats.