Specific Distribution of Fatty Acids in the Milk Fat Triglycerides of Goat and Sheep[1]

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ABSTRACT

The triglycerides of the fat globules of sheep and goat milk were isolated and separated into short and long chain lengths by silicic acid column chromatography. The short chain lengths comprised major triglycerides with 34-44 acyl carbon atoms and accounted for nearly 50% of the total milk fat. The long chain lengths contained major triglycerides with 40-54 acyl carbons. Stereospecific analyses of the short chain triglyceride fraction showed that of the 20-23 moles per cent of C_4-C_8 fatty acids present, at least 95% were specifically attached to the glycerol molecule in the position corresponding to carbon 3 of *sn*-glycerol. The distribution of the other fatty acids (C_{10} or greater) did not show such marked specificity for either the 1 or the 2 position. Although individual triglycerides were not identified, the specific placement of the fatty acids could best be accounted for by assuming a common pool of long chain 1,2-diglycerides which served as precursors of the bulk of both short and long chain triglycerides during milk fat synthesis.

INTRODUCTION

Stereospecific analyses of bovine milk fat have shown (1,2) that the butyric acid and other short chain fatty acids, which are specifically derived by de novo synthesis in the mammary gland (3), are largely or exclusively esterified to the 3 position of the glycerol molecule, *sn*-nomenclature used throughout (4). The distributions of the other fatty acids (C_{10} or greater), which are derived from diet and the depot fat, did not exhibit such marked specificity for either the 1 or the 2 position. These data support the hypothesis (5) that the short chain fatty acids are esterified with long chain diglycerides, or are substituted in glycerophosphatide intermediates, in the final step of milk fat biosynthesis. The milk fats of sheep and goat are also rich in butyric and other short-chain acids (6), but their intraglyceride distribution has not been ascertained. A demonstration of comparable stereospecificity in these species would establish the generality of the phenomenon in the ruminants, and might permit extrapolation of data to nonruminant species which also derive their milk fats by mobilization and partial de novo synthesis of fatty acids in the mammary gland.

MATERIALS AND METHODS

The chemical reagents, solvents, chromatographic materials and analytical standards were as described (2). Fresh samples of raw sheep and goat milk were obtained from local farms. The goat milk (450 ml) was collected from one animal, while the sheep milk (360 ml) was pooled from three ewes. The triglycerides were isolated from the milk fat globules by extraction with chloroform-methanol (2:1) as described for cow's milk (7). The various triglyceride preparations were free of contamination with free fatty acids, diglycerides, and any other common lipids by chromatography on columns or thin layers of silicic acid, as described below.

Separation of Short and Long Chain Lengths

Triglycerides of short and long chain length were resolved by chromatography on columns of silicic acid essentially as described by Blank and Privett (8). The fractions obtained from 1.2 g of total milk fat were pooled in two nearly equal portions; the least polar one (0.6-0.7 g) provided the long chain length, the more polar one (0.5-0.6 g) gave the short and medium chain length triglycerides. The latter fraction comprised major triglycerides with 32-46 acyl carbon atoms, the former long chain triglycerides with 40-54 acyl carbons. Smaller quantities of short and long chain triglycerides were resolved by thin layer chromatography (TLC) as previously described (9). This method was also used to resolve the short and long chain diglycerides released from the short chain triglyceride fraction by hydrolysis with pancreatic lipase.

Stereospecific Analysis

The positional distribution of the fatty acids in the short and long chain triglycerides was determined by the method of Brockerhoff as previously described (2). After a brief hydrolysis with pancreatic lipase, the reaction mixture

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TABLE I

Molecular Weight Distribution of Short and Long Chain Triglycerides of Goat and Sheep Milk Fat[a,b]

Carbon No.	Goat					Sheep				
		Short chain		Long chain			Short chain		Long chain	
	Original	Total	Residual[c]	Total	Residual	Original	Total	Residual	Total	Residual
24						Trace	0.2	0.2		
26	0.1	Trace	Trace			0.5	0.5	0.6		
28	0.3	0.2	0.1			0.8	1.3	1.2		
29	Trace					Trace	0.1	0.1		
30	0.7	0.5	0.7			1.2	2.2	2.2	0.1	0.1
31	0.1					Trace	0.2	0.1		
32	1.5	2.0	2.0			2.0	3.9	3.7	0.3	0.3
33	0.1	0.1	0.2			0.2	0.4	0.4		
34	3.9	5.9	6.1			4.0	7.7	7.5	1.0	0.9
35	0.5	1.0	0.7			0.7	1.2	1.1		
36	8.5	16.9	16.3			7.2	15.2	14.6	2.3	2.2
37	0.7	1.2	0.7			1.3	1.9	2.0		
38	12.4	26.9	25.2	Trace	Trace	11.4	23.9	23.6	4.4	4.3
39	Trace			Trace	Trace	0.7				
40	10.4	21.8	21.5	1.0	1.1	11.3	20.3	20.0	5.5	5.5
42	8.1	10.5	11.3	4.5	4.6	6.4	7.2	7.4	5.9	6.0
43	Trace			0.3	0.2					
44	7.4	5.4	6.1	7.5	7.6	5.4	3.7	4.0	6.5	6.6
46	5.9	2.7	3.2	8.3	8.4	5.5	2.4	2.7	7.3	7.5
48	7.6	1.0	1.5	13.4	13.5	7.2	1.8	2.0	10.8	10.6
50	12.3	1.4	1.8	25.1	24.6	10.5	2.2	2.4	16.3	16.4
52	13.1	1.2	1.7	27.3	27.4	13.5	2.1	2.4	22.2	22.3
54	6.2	1.3	0.9	12.1	12.2	9.5	1.5	1.7	15.4	15.6
56	0.2			0.2	0.1	0.4	0.1	0.1	1.0	0.8
58						0.3			0.6	0.6
60						Trace			0.4	0.3

[a]Short and long chain lengths resolved by chromatography on silicic acid columns.

[b]Values are given in mole percentage.

[c]Triglycerides recovered following partial hydrolysis with pancreatic lipase.

was extracted with diethyl ether without acidification. The ether extracts were concentrated and the mono-, di- and triglycerides isolated by TLC. Portions of the mono- and diglycerides were then acetylated by reaction with acetic anhydride-pyridine. The acetates were examined by gas chromatography along with the unhydrolyzed triglycerides and the yield and composition of the products of the lipase hydrolysis assessed by reference to tridecanoin which was added as internal standard. Other aliquots of these fractions were transbutylated and the fatty acid composition determined by gas liquid chromatography (GLC).

The rest of the diglycerides was converted into glycerophosphatidyl phenols by treatment with phenyl dichlorophosphate and the reaction products isolated by TLC. The digestion with phospholipase A was performed as described by Brockerhoff (10), except that less phosphatidyl phenol (50 mg) and only one half as concentrated a buffer was used. The phosphatides derived from 1,2-diglycerides yielded the 2-fatty acid, while those from 2,3-diglycerides having the L-configuration, were not attacked. After incubation for 4 hr at room temperature, the reaction products were isolated by TLC (2).

Gas Liquid Chromatography

GLC analyses of fatty acid butyl esters, monoglyceride diacetates, diglyceride monoacetates and di- and triglycerides were done as previously described (2).

RESULTS AND DISCUSSION

Starting Materials

The overall fatty acid and triglyceride distributions of the goat and sheep milk samples were similar to those described earlier (6). Table I gives the composition of the triglycerides in the short and long chain fractions and compares them to those of the total from which they were derived by chromatography on silicic acid. While the short chain triglycerides (26-42 acyl carbons) have been nearly completely resolved from the long chain length

TABLE II

Fatty Acid Composition of Short and Long Chain Triglycerides of Goat and Sheep Milk Fat[a,b]

Fatty acids	Goat					Sheep				
		Short chain		Long chain			Short chain		Long chain	
	Original	Total	Residual[c]	Total	Residual	Original	Total	Residual	Total	Residual
4:0	5.1	10.2	10.3			4.4	8.0	7.8		
6:0	4.4	8.2	8.5			3.6	7.2	7.2	0.5	0.6
8:0	2.6	4.7	4.8	0.9	0.5	2.4	3.8	3.6	1.3	1.1
10:0	7.8	8.8	9.0	6.4	6.3	5.5	7.5	7.1	4.0	4.6
10:1	0.3	0.3	0.3			0.2	0.3	0.3	0.1	0.1
12:0	3.8	3.9	3.8	2.7	2.3	3.5	4.3	3.9	2.8	2.6
12:1						0.1	Trace	Trace	0.1	Trace
14:0	9.6	10.3	10.0	10.5	9.4	9.8	11.1	10.5	9.6	9.6
14:1	0.2	0.5	0.4	0.5	0.5	0.6	0.5	0.5	0.8	0.9
15:0	2.0	1.5	1.7	2.5	2.3	2.8	2.2	2.7	3.1	2.7
16:0	26.0	24.6	24.9	28.7	28.6	21.2	21.2	21.3	21.5	22.2
16:1	1.8	2.0	1.6	2.3	2.6	1.7	1.4	2.1	2.3	2.2
17:0	0.8	0.7	0.5	1.3	1.4	1.0	0.8	0.6	1.1	1.1
18:0	9.9	6.0	6.6	12.7	13.6	14.0	9.8	10.6	16.8	16.9
18:1	20.6	14.0	14.0	26.4	26.9	21.8	15.1	15.0	28.4	28.5
18:2	2.7	2.0	1.8	3.1	3.7	4.4	3.7	3.4	4.6	3.7
18:3						2.6	2.5	2.7	3.0	2.7
20:1	2.4	2.0	1.8	1.8	1.9	0.4	0.5	0.5	Trace	0.5
20:2	Trace	0.3	Trace	0.2	Trace	Trace	0.1	0.2	Trace	Trace

[a]Short and long chain lengths resolved by chromatography on silicic acid columns.

[b]Values are given in mole percentage.

[c]Triglycerides recovered following partial hydrolysis with pancreatic lipase.

TABLE III

Molecular Weight Distribution and Fatty Acid Composition of the Diglycerides Derived From Short Chain Triglycerides by Lipase Hydrolysis[a,b]

Carbon No.	Diglyceride acetates				Carbon No.	Fatty acids			
	Goat		Sheep			Goat		Sheep	
	Original	Residual[c]	Original	Residual		Original	Residual	Original	Residual
16	0.7	2.1	1.3	3.8	4:0	8.5	15.0	9.5	11.7
18	1.4	3.4	2.6	6.3	6:0	6.9	12.1	6.8	10.6
20	7.9	14.6	7.4	15.4	8:0	4.0	5.7	2.6	5.1
21		1.0	1.7	3.4	10:0	9.6	10.6	6.0	10.0
22	20.8	36.0	14.0	25.6	10:1	0.3	0.4	0.6	0.4
23			0.8	1.3	12:0	4.0	2.8	4.0	4.6
24	21.8	33.2	24.0	40.1	12:1				
25					14:0	13.0	10.7	12.9	10.8
26	7.9	7.8	10.9	3.5	14:1	0.8	1.1	0.8	0.9
27					15:0	1.0	1.5	2.6	2.3
28	5.0	1.5	3.9	0.6	16:0	26.0	16.4	22.0	12.6
29					16:1	2.3	1.2	1.2	1.2
30	4.0	0.4	4.3		17:0	0.7	0.6	0.6	0.8
31					18:0	5.1	3.1	8.8	7.1
32	5.6		4.8		18:1	13.6	13.9	16.3	15.0
33			0.3		18:2	2.0	2.5	3.3	4.3
34	9.9		8.0		18:3			1.6	2.0
35					20:1	1.9	2.1	0.4	0.4
36	10.1		9.1		20:2	0.3	0.3		0.2
37									
38	4.1		6.4						
40	0.8		0.4						

[a]Mixed 1,2- and 2,3-diglycerides released by partial hydrolysis with pancreatic lipase.

[b]Values are given in mole percentage.

[c]2,3-Diglycerides recovered from TLC of mixed 1,2- and 2,3-diglycerides.

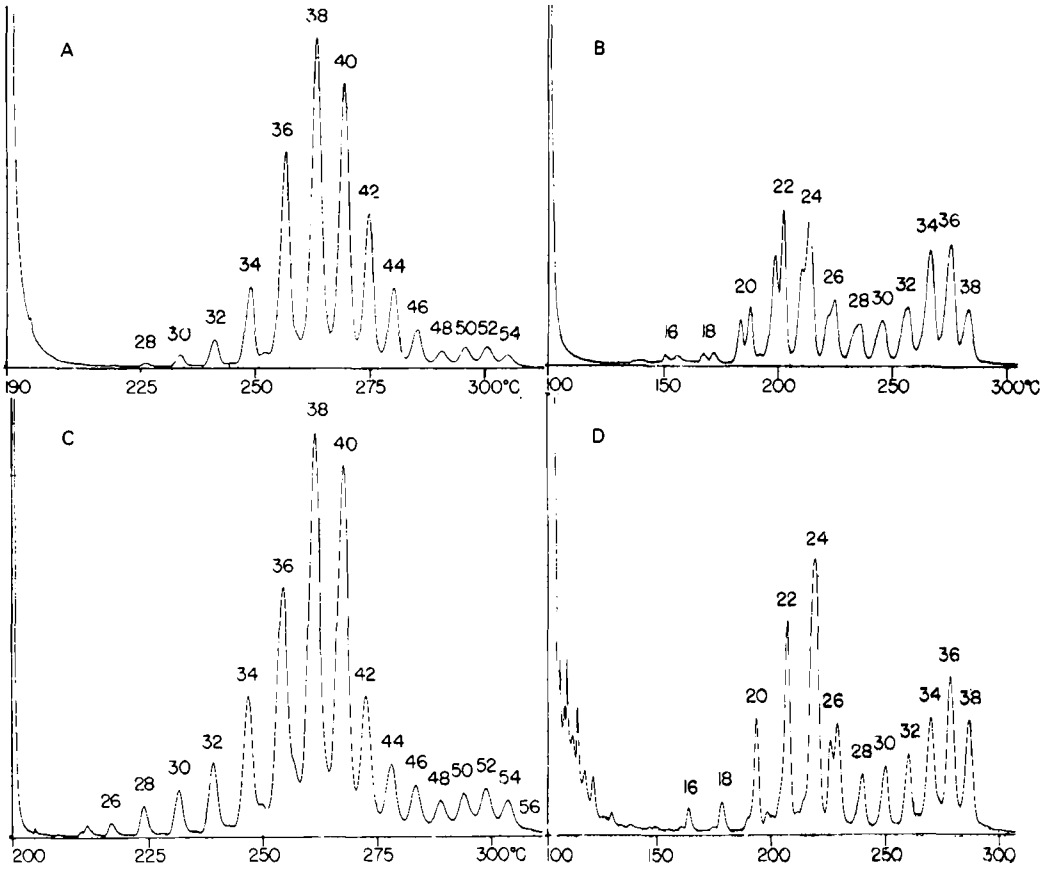

FIG. 1. GLC separation of short chain triglycerides and the derived diglyceride acetates. The peaks are identified by the total number of acyl carbon atoms. Beckman GC-4 gas chromatograph; columns 3% (w/w) JXR (dimethylpoly-siloxane gum) on Gas Chrom Q (100-120 mesh); temperature program as shown. A, short chain triglycerides of goat milk fat; B, diglyceride acetates of A; C, short chain triglycerides of sheep milk fat; D, diglyceride acetates of C.

glycerides (44-54 acyl carbons) of the goat milk, the removal has been less effective for the sheep milk glycerides. Furthermore, in both cases the short chain fraction still contains significant amount of long chain triglycerides (C_{44}-C_{54}). This contamination, however, was not sufficient to impare the reliability of the subsequent stereospecific analysis. The short chain fractions of the goat and sheep milk triglycerides made up 52 and 40 moles per cent respectively, of the total milk fat triglycerides. A proportional summation of the distributions of the short and long chain glycerides gave the reconstituted total distributions which differed little from those of the corresponding original milk fats.

Pancreatic Lipase Hydrolysis

Table I also gives the composition of the triglycerides remaining after the limited exposure of the short and long chain triglycerides to pancreatic lipase. On the basis of the molecular weight distribution there is little indication of a selective hydrolysis of any triglyceride types. This is confirmed by the fatty acid composition given in Table II, which shows close qualitative and quantitative correspondence between the original and the residual glyceride mixtures.

The diglycerides from the short chain fraction were isolated in 15-20% yield. The distribution of their molecular weights and fatty acids is given in Table III. Since the C_4-C_8 acids comprised 20-23 moles per cent of the total fatty acids, almost all of the diglycerides of carbon number 24 or less (about 50% of total) must consist of one short and one long chain acid. The simultaneous appearance in the enzyme digest of short chain acids and diglycerides containing the same short chain acids was a further indication that the pancreatic lipase did

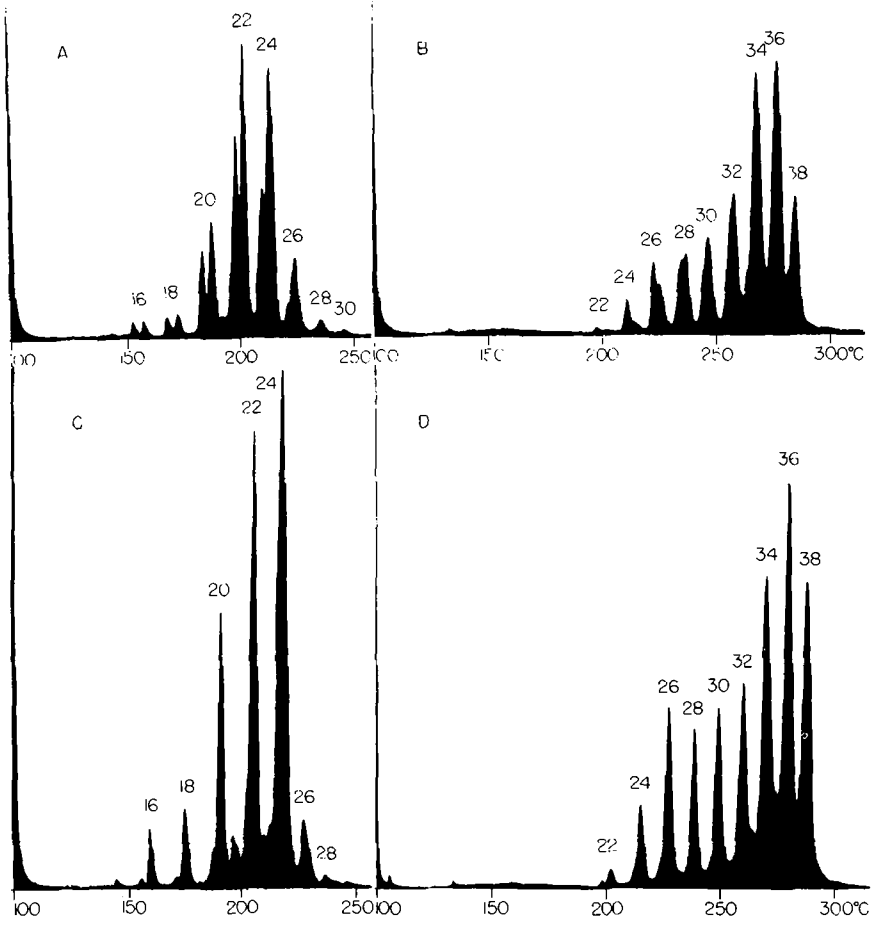

FIG. 2. GLC separation of the short and long chain diglyceride acetates of short chain triglycerides. Peak identification and instrumentation as in Figure 1. A and B, respectively, short and long chain diglyceride acetates of goat milk fat; C and D, respectively, short and long chain diglyceride acetates of sheep milk fat.

not discriminate between short and long chain fatty acids in the 1 and 3 positions of the short chain triglycerides. Figure 1 shows the GLC elution patterns of the original short chain triglycerides and of the diglycerides derived from them by pancreatic lipase hydrolysis. The diglyceride mixture shows the characteristic two hump distribution seen for the original milk fat of these species. These diglycerides can be resolved on the basis of chain length by means of TLC on silica gel. Figure 2 shows the GLC elution patterns recorded for the two diglyceride subgroups of the short chain triglycerides of goat and sheep milk fat. The molecular weight and fatty acid distribution of the short chain diglycerides approximated closely the composition of the residual lysophosphatidyl phenols discussed below. The long chain length diglycerides resembled those of the phosphatidyl phenols which were attacked by phospholipase A. These observations suggested that the TLC resolution of the diglycerides effectively divided them into the 1,2- and the 2,3-diglyceride types. Only the 2,3-diglycerides contained the short chain fatty acids.

Table IV gives the molecular weight and fatty acid distributions of the total diglycerides derived from the long chain triglycerides and of the residual phosphatidyl phenols (2,3-diglycerides) recovered following degradation of the mixed phosphatidyl phenols with phospholipase A. In both the goat and the sheep the glycerides have comparable distributions of molecular weights which are due to similarities in composition and distribution of the fatty acids. Furthermore, pancreatic lipase appears to

TABLE IV

Molecular Weight Distribution and Fatty Acid Composition of the Diglycerides Derived From Long Chain Triglycerides of Goat and Sheep Milk[ab]

Carbon No.	Diglyceride acetates				Carbon No.	Fatty acids			
	Goat		Sheep			Goat		Sheep	
	Original	Residual[c]	Original	Residual		Original	Residual	Original	Residual
20			1.2						
22			3.1						
24	0.2		5.9		4:0				
26	2.4		4.5		6:0			0.3	0.6
28	5.1	Not	4.6	Not	8:0	0.4	0.7	1.1	1.6
29			0.2		10:0	4.7	8.7	4.1	5.9
30	6.6	determined	6.5	determined	10:1				0.1
31			0.2		12:0	2.5	3.0	2.9	3.7
32	11.1		10.3		12:1			0.3	0.2
33			0.5		14:0	13.6	12.2	10.5	12.0
34	27.2		19.1		14:1	0.4	0.7	0.7	1.1
35					15:0	2.6	2.6	3.3	3.2
36	31.4		23.7		16:0	34.1	20.4	25.1	13.6
37					16:1	2.0	2.3	2.1	2.4
38	15.0		19.7		17:0	1.0	1.6	1.3	0.8
39					18:0	12.0	10.0	18.8	14.1
40	1.0		0.4		18:1	23.3	30.5	24.0	30.7
42					18:2	1.6	4.3	3.6	5.8
44					18:3			1.4	4.0
46					20:1	1.6	2.3	0.5	0.2
48					20:2	0.2	0.7	Trace	Trace

[a]Mixed 1,2- and 2,3-diglycerides released by partial hydrolysis with pancreatic lipase.

[b]Values are given in mole percentage.

[c]Residual phosphatidyl phenols containing 2,3-diglycerides.

have released nearly the same fatty acids from the long chain glycerides of both species, which would indicate that the two milk fats also possess comparable positional placement of the acids in these glycerides.

Positional Distribution of Fatty Acids

Table V presents the fatty acid compositions of the individual positions in the glyceride molecule of the short chain triglycerides. The fatty acids in position 1 were derived from the data for the lysophosphatidyl phenols. No C_4-C_6 acids were found in this location. The fatty acids in position 2 were determined by analysis of the monoglycerides liberated by pancreatic lipase and by analysis of the free fatty acids released by phospholipase A. Although the two determinations showed slight differences in the proportions of some of the medium chain length fatty acids, there were no signs of any short chain fatty acids. Furthermore, acetylation of the 2-monoglycerides, followed by GLC, gave molecular weight distributions which agreed closely with the fatty acid proportions, once again with no indication of any butyric or caproic acid residues in the 2 position.

The fatty acids in position 3 were obtained by summation and by subtraction as described in the footnotes to Table V. The two methods gave similar estimates for the major acids but the relatively large error (4-7%) precluded quantitative conclusions about the minor components. The C_4-C_{10} acids comprised approximately 75% and 60% of the total acids in the short chain triglycerides of goat and sheep milk, respectively. The remainder consisted of a variety of medium and long chain fatty acids which were due to the contamination of the short chain fraction by long chain triglycerides during the silicic acid column chromatography. The proportion of the long chain fatty acids in this fraction was consistent with the finding of only 20-23 moles per cent of short chain fatty acids in the original short chain triglyceride fraction. This distribution was also in agreement with the fatty acid composition of the 1,2- and 2,3-diglycerides recovered from the TLC of the total diglyceride fraction.

Table VI gives the fatty acid composition of the three positions of the glycerol molecule for the long chain triglycerides of goat and sheep milk. The most obvious characteristic of the acid distribution is the preferential association of myristic acid with the 2 position. Palmitic acid was distributed nearly equally between the 1 and 2 positions in the goat milk fat, while in

TABLE V

Positional Distribution of Fatty Acids in Short Chain Triglycerides of Goat and Sheep Milk Fat[a,b]

	Goat					Sheep				
	One	Two		Three		One	Two		Three	
Fatty acids	LPP	MG	FFA	Method A[c]	Method B[d]	LPP	MG	FFA	Method A	Method B
4:0				30.6	30.0				24.0	23.4
6:0				24.6	24.2				21.6	21.2
8:0	2.3	2.4	2.0	9.4	9.0	0.9	2.7	2.2	7.8	7.5
10:0	5.0	11.1	10.0	10.3	10.1	2.3	8.2	7.5	12.0	11.8
10:1		0.3	0.3	0.6	0.5		0.2		0.7	0.6
12:0	6.2	5.7	5.7	0.2	0.1	3.3	6.6	6.0	3.0	2.6
14:0	9.5	21.4	22.4	0.0	0.0	11.4	19.2	18.3	2.7	2.4
14:1	Trace	0.8	0.8	0.7	1.4		1.1	1.0	0.4	0.7
15:0	1.5	2.8	3.2	0.2	0.2	2.2	2.7	3.5	1.7	1.9
16:0	41.3	29.2	29.8	3.3	3.6	38.4	21.8	20.3	3.4	3.4
16:1	4.0	2.0	1.9	0.0	0.4	2.1	1.9	2.4	0.2	0.5
17:0	1.6	0.5	0.3	0.0	0.7	1.4	0.6	0.6	0.4	1.0
18:0	11.9	5.2	5.8	0.9	1.0	14.8	10.7	11.5	3.9	3.5
18:1	14.3	14.3	14.1	13.4	13.5	16.4	17.3	17.5	11.6	12.7
18:2	0.8	2.6	2.7	2.6	2.4	2.9	4.5	5.3	3.7	4.1
18:3						3.9	2.0	3.8	1.6	2.0
20:1	1.2	1.4	1.0	3.4	2.8	Trace	0.5	0.3	1.0	0.3
20:2	0.4	0.3		0.2	0.3		Trace		0.3	0.4

[a]Position relative to *sn*-glycerol 3-phosphate; LPP, L-lysophosphatidyl phenol produced from L-phosphatidyl phenols by phospholipase A; MG, 2-monoglycerides produced from original triglycerides by pancreatic lipase; FFA, free fatty acids produced from the L-phosphatidyl phenols by phospholipase A.

[b]Values are given in mole percentage.

[c]Values obtained by subtracting the sum of the acids of monoglycerides and lysophosphatidyl phenols from the original fatty acids.

[d]Values obtained by subtracting the monoglyceride acids from the acids of residual phosphatidyl phenols.

the sheep milk this acid was preferentially placed in the 1 position. Neither milk contained much palmitic acid in the 3 position, which in both species was occupied to a large extent by oleic acid. In both fats the stearic acid was preferred for the 1 position. These distributions are nearly identical to those reported earlier (10) for the long chain glycerides of cow's milk fat, except for oleic acid, which in the bovine milk samples was found in a somewhat higher concentration in position 1 than in the other two positions.

Table VII gives the fatty acid composition of the 1,2-diglycerides of the short and long chain triglycerides of goat and sheep milk. Although not matching in numerical values, the proportions of the acids in the corresponding diglyceride moieties are closely similar. These findings coincide with those made previously regarding the fatty acid composition of the 1,2-diglyceride moieties of the short and long chain triglycerides of bovine milk fat. As in the case of the bovine milk fats (11), it may be suggested that the 1,2-diglycerides are derived from a common pool during the resynthesis of the plasma triglycerides in the mammary gland. The peculiar milk fat triglyceride pattern could then be produced by a nearly random association of these diglycerides with the remaining C_4-C_{18} fatty acids.

The characteristic distribution of the fatty acids in the 1,2-diglycerides of the milk fats could arise either from any mono- and diglycerides derived by partial hydrolysis of blood triglycerides. The latter possibility, however, has been ruled out in the goat and cow by the results of pancreatic lipase hydrolyses of the milk fat and the blood triglycerides (12,13).

Mechanism of Biosynthesis

The extensive analyses of the milk triglycerides presented here and in previous publications (2,9,11) have allowed the exclusion of complete randomization or any simple modification of it as a system for milk fat triglyceride biosynthesis. The recognition of a common pool of 1,2-diglycerides in both short and long chain triglycerides of the ruminant milks is in agreement with the results obtained by Kinsella and

TABLE VI

Positional Distribution of Fatty Acids in Long Chain Triglycerides of Goat and Sheep Milk Fat[a,b]

	Goat					Sheep				
	One	Two		Three		One	Two		Three	
Fatty acids	LPP	FFA	MG	Method A[c]	Method B[d]	LPP	FFA	MG	Method A	Method B
4:0										
6:0									1.5	1.2
8:0	1.2	0.2	0.3	1.2	1.1	0.5	1.0	1.4	2.0	1.8
10:0	2.0	2.5	3.6	13.6	13.8	0.7	2.6	2.7	8.6	9.1
10:1		Trace	Trace				Trace	Trace	0.3	0.2
12:0	2.2	2.9	3.8	2.1	2.2	1.2	3.4	3.1	4.1	4.3
14:0	7.5	18.9	19.5	4.5	4.9	5.5	15.4	16.3	7.0	7.7
14:1		0.9	0.8	0.7	0.6	0.4	0.6	0.7	1.3	1.5
15:0	2.2	2.7	2.7	2.6	2.5	3.0	4.3	5.3	1.0	1.1
16:0	45.4	38.4	37.6	3.1	3.2	37.7	26.0	25.5	1.3	1.7
16:1	2.1	2.7	2.0	2.8	2.6	2.2	2.0	2.4	2.3	2.4
17:0	1.0	1.0	0.5	2.4	2.7	1.9	1.2	1.1	0.3	0.5
18:0	17.9	8.0	7.1	13.1	12.9	22.8	14.4	14.3	13.3	13.9
18:1	17.5	17.6	17.6	44.1	43.4	20.7	21.4	21.8	42.7	39.6
18:2	Trace	2.3	2.5	6.8	6.1	2.6	3.2	3.9	7.3	7.7
18:3						0.8	2.7	1.5	6.7	6.5
20:1	0.6	1.7	2.0	2.8	2.6	Trace	0.3	Trace	Trace	0.4
20:2	0.4	0.2	Trace	0.2	1.4	Trace	1.2	Trace	Trace	Trace

[a]Position relative to *sn*-glycerol 3-phosphate; LPP, L-lysophosphatidyl phenol produced from L-phosphatidyl phenols by phospholipase A; FFA, free fatty acids produced from the L-phosphatidyl phenols by phospholipase A; MG, 2-monoglycerides produced from the original triglycerides by pancreatic lipase.

[b]Values are given in mole percentage.

[c]Values obtained by subtracting the sum of the acids of monoglycerides and lysophosphatidyl phenols from the original fatty acids.

[d]Values obtained by subtracting the monoglyceride acids from the acids of residual phosphatidyl phenols.

TABLE VII

Distribution of Major Fatty Acids in the 1,2-Diglyceride Moieties of the Short and Long Chain Triglycerides of Goat and Sheep Milk[a,b]

	Goat				Sheep			
	Short chain		Long chain		Short chain[d]		Long chain	
Fatty acids	Pos. 1	Pos. 2[c]	Pos. 1	Pos. 2	Pos. 1	Pos. 2	Pos. 1	Pos. 2
8:0	2.3	2.2	1.2	0.3	0.9	2.4	0.5	1.2
10:0	5.0	10.5	2.0	3.0	2.3	7.8	0.7	2.6
12:0	6.2	5.6	2.2	3.4	3.3	6.3	1.2	3.2
14:0	9.5	22.0	7.5	19.2	11.4	18.8	5.5	15.8
16:0	41.3	29.5	45.4	38.0	38.4	21.0	37.7	25.8
16:1	4.0	2.0	2.1	2.4	2.1	2.2	2.1	2.2
17:0	1.6	0.4	1.0	0.8	1.4	0.6	1.9	1.1
18:0	11.9	5.5	17.9	7.5	14.8	11.0	22.8	14.3
18:1	14.3	14.2	17.5	17.6	16.4	17.4	20.7	21.6
18:2	0.8	2.6	Trace	2.4	2.9	5.0	2.6	3.5
20:1	1.2	1.2	0.6	1.8	Trace	0.4	Trace	0.2
20:2	0.4	0.3	0.4	0.1		0.1		0.6

[a]Position relative to *sn*-glycerol 3-phosphate.

[b]Values are given in mole percentage.

[c]Average values computed from independent estimates of the fatty acid composition of the monoglycerides and free fatty acids as explained in Table VI.

[d]The short chain triglycerides of sheep milk contained 3.9 and 3.0 moles per cent of 18:3 in positions 1 and 2, respectively.

McCarthy (14-16) with dispersed bovine mammary cells. Using 2-C^{14}-acetate and 3-C^{14}-glycerol, these workers observed specific activity-time curves which suggested that the milk fat triglycerides were produced by the acylation of 1,2-diglycerides with endogenously synthesized fatty acids.

The mechanism of biosynthesis of the 1,2-diglycerides is less obvious and possibly depends upon as yet unrecognized enzymic specificities. The present data reveal a continuous shift of fatty acids from position 3 to positions 2 and 1 as the chain length increases from C_6 to C_{14} and as the C_{18} acids become progressively more saturated. The extent to which this peculiarity may reflect the structural requirements of the triglyceride end products or the enzyme systems involved in their assembly may become more apparent when the molecular species of the milk fat glycerides are identified.

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