



Composition and distribution of fatty acids in triglycerides from goat infant formulas with milk fat

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ABSTRACT

Most infant formulas use vegetable oils in place of milk fat to provide an overall fatty acid profile similar to that of breast milk. Vegetable oils have 5 to 20% saturated fatty acids in the *sn*-2 position of triglycerides unless they are modified by interesterification. Interesterification is increasingly used for the fat for infant formulas to raise the level of saturated fatty acids in the *sn*-2 position to 40 to 60%. The objective of this study was to verify an alternative approach to providing the appropriate fatty acid profile, including in the *sn*-2 position, for a goat infant formula. In this method, 55% of total fat was made from goat milk fat and 45% from a mixture of unmodified high oleic sunflower, canola, and sunflower oils in a ratio of 44:30:26. The fatty acid profile was measured by gas-liquid chromatography and the relative percentage of fatty acids in the *sn*-2 position of triglycerides was measured via partial deacylation with Grignard reagent using trimethylsilyl derivatives of monoacylglycerols. Mixing goat milk fat with vegetable oils produced a formula with a profile of essential fatty acids and a ratio of linoleic:α-linolenic fatty acids within the required interval of 5 to 15:1 recommended for infant formula. The proportion of palmitic acid in the *sn*-2 position was 31%.

Key words: fatty acid, infant formula, *sn*-2 position

INTRODUCTION

Fat provides about 40 to 54% of the energy content of formula, but also provides functional fatty acids (Koletzko et al., 2005). It is recommended that infant formula contain 300 to 1,200 mg/100 kcal of linoleic acid (LA; 18:2n-6), a minimum of 50 mg/100 kcal of α-linolenic acid (LNA; 18:3 n-3,) and a LA:LNA ratio of 5 to 15:1 for optimal synthesis of long-chain polyunsaturated fatty acids (PUFA; Koletzko et al.,

2005). Additional recommendations include restricting the sum of myristic acid and lauric acid to less than 20% in consideration of their potential negative effects on serum cholesterol and lipoprotein levels (Koletzko et al., 2005).

Vegetable oils are typically used to provide the essential fatty acids in infant formula (Hansen and Diener, 1997; Berger et al., 2000). When palm olein was used to raise palmitic acid in the fat to 25%, similar to human milk, infants were able to absorb only 90% of fat (Nelson et al., 1996, 1998; Ostrom et al., 2002). This compared with 95% in infants fed formula with only 8 to 9.5% palmitic acid (Nelson et al., 1996, 1998). The reduced fat absorption from formula with palm olein was suggested to be attributable to the high level of palmitic acid and because <15% of this fatty acid was in the *sn*-2 position (Nelson et al., 1996, 1998; Ostrom et al., 2002). Although there is no evidence of a need to have levels of palmitic acid the same as in human milk fat (Nelson et al., 1996), the solution used to overcome the deficiency in fat absorption was to enhance the proportion of this fatty acid in the *sn*-2 position by interesterification of the palm olein (Carnielli et al., 1996; Lien et al., 1997; Lucas et al., 1997; López-López et al., 2001). Using vegetable oils that are modified by interesterification as the sole source of fat has also been suggested for goat infant formulas (Maduko et al., 2007a,b).

We propose an alternative method, which is to use a mixture of approximately 50% milk fat combined with a selection of unmodified vegetable oils. Because 40% of palmitic acid in milk fat is already esterified at the *sn*-2 position (Freeman et al., 1965; Marai et al., 1969), we predict a combination with vegetable oils to supply essential fatty acids should result in a formula with a fatty acid profile recommended for infant formula while still increasing *sn*-2 palmitic acid compared with formula with unmodified vegetable oils. The advantages of this alternative approach are to retain some milk fat globule membrane components that have potential benefits to the infant (Berger et al., 2000; Spitsberg, 2005) and to avoid the need for extensive modification of the ingredients. The objective of this study was to verify

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the fatty acid profile, including the *sn*-2 position, of the fatty acids of a formula made from a mixture of goat milk fat and selected unmodified vegetable oils. A secondary objective was to conduct a preliminary analysis of a limited number of formulas with cow milk.

MATERIALS AND METHODS

Reagents

All solvents and reagents, unless otherwise specified, were supplied by Scharlau Chemie SA (Barcelona, Spain). Solvents used were of multisolvent grade. Aluminum-backed silica gel thin layer chromatography sheets (20 × 20 cm) were from Merck (Darmstadt, Germany; Nr 1.05554). The following triacylglycerol standards were used: tricaprin, tricaprylin, trilaurin, tricaproin, tripalmitin, trimyristin, tristearin, triolein, tributyrin, tripalmitolein, triarachidin, and tri-11-eicosenoin (all from Supelco, Bellefonte, PA); ethyl magnesium bromide was from Sigma-Aldrich (St. Louis, MO).

Samples

A total of 3 batches of goat whole milk powders and 3 batches of goat infant formulas, manufactured at different times by Dairy Goat Co-operative (NZ) Ltd. (Hamilton, New Zealand), were analyzed. The goat infant formula contained 55% of the total fat from milk fat, with the rest consisting of a mixture of high oleic sunflower (84% oleic acid) and canola and sunflower (30% oleic acid) oils in a ratio of 44:30:26.

Two batches of cow whole milk powder and 2 batches of cow infant formulas were also analyzed. The cow formula contained 25% of total fat from milk fat and a mixture of canola, soybean, high oleic sunflower (84% oleic acid), coconut, and sunflower (30% oleic acid) oils in a ratio of 27:23:20:15:15. The cow whole milk powder was manufactured by Fonterra (Auckland, New Zealand) and the cow formula with milk fat was manufactured by Dairy Goat Co-operative (NZ) Ltd. All powders were dissolved in water to 13 g/100 mL before extraction of the fat for analysis.

Lipid Extraction

The fat in the samples was extracted according to Folch et al. (1957). The lower phase was rotary evaporated and dried in vacuo to a constant weight.

Quantification of Fatty Acids

Fatty acid isopropyl esters were prepared from the extracted fats (Wolff, 1994). Isopropyl esters of fatty acids were used instead of methyl esters to avoid the loss of

otherwise highly volatile ester derivatives of short-chain fatty acids present in ruminant milks. Another advantage of the use of isopropyl esters is that no correction factors are required for quantification of fatty acids in the whole chain length range (Wolff, 1994).

The fatty acid isopropyl esters were measured by capillary gas chromatography on a Hewlett Packard 5890 series II gas chromatograph (Palo Alto, CA) equipped with a flame ionization detector and fused quartz (30 m, 0.32 mm i.d.) BP-20 capillary column (SGE, Ringwood, Victoria, Australia). Helium was used as a carrier gas with a split ratio of 1:50. The oven was operated at 65°C for 3 min; the temperature was then increased to 190°C at a rate of 5°C/min and left for 15 min. Fatty acids were identified by the use of standards and the known equivalent chain length values (Stránský et al., 1997).

The amounts of individual fatty acids were expressed in moles per 100 moles of total fatty acids calculated from the molecular weights of fatty acid isopropyl esters. Total fat was calculated by the sum of individual fatty acids expressed as triglyceride equivalents.

Sn-2 Positional Analysis

Regiospecific analysis of the fats was performed by partial deacylation of the triacylglycerol molecules with Grignard reagent (Takagi and Ando, 1991) as suggested for milk by Bracco (1994).

This consisted of dissolving 20 mg of triacylglycerol in dry diethyl ether (0.6 mL) and adding 66 µL of ethyl magnesium bromide solution (3 M in diethyl ether). The mixture was shaken for 1 min, glacial acetic acid (20 µL) was added, the mixture was shaken, and 0.66 mL of water was added. The mixture was vigorously shaken again and allowed to stand until complete separation of the phases occurred. The upper phase was collected and the water phase was extracted twice with diethyl ether (0.7 mL). All diethyl ether phases were combined and partially evaporated under a stream of argon. The *sn*-2 monoacylglycerol fraction was isolated by thin layer chromatography on boric acid-impregnated thin layer chromatography plates (Thomas et al., 1965). The plates were developed in chloroform:acetone (96:4, vol/vol; Becker et al., 1993). After removal of solvents in the stream of argon, the spots were detected under UV irradiation (254 nm) and sprayed with a primulin solution (5 mg in 100 mL of acetone). The *sn*-1(3) monoacylglycerol zone (retention factor ~0.15) and *sn*-2 monoacylglycerol zone (retention factor ~0.35) were collected separately and each was eluted with diethyl ether (3.5 mL) for further analysis.

The isolated *sn*-2 monoacylglycerols were derivatized using trimethylsilyl by the method according to Liu

Table 1. Energy and fat content of samples¹

Item	Cow milk	Goat milk	Cow IF	Goat IF
Energy (kcal/100 mL)	52	55	68	68
Total fat (g/100 mL)	3.0	3.5	3.4	3.3
Milk fat (% of total fat)	100	100	25	55
Energy from fat (% of total energy)	52	57	45	44

¹Cow IF = infant formula with cow milk, cow milk fat, and vegetable oils (n = 2); Goat IF = infant formula with goat milk, goat milk fat, and vegetable oils (n = 3). Both manufactured by Dairy Goat Co-operative (NZ) Ltd. (Hamilton, New Zealand).

and Kinderlerer (1999) and quantified using a Hewlett Packard 5890 series II gas chromatograph equipped with a flame ionization detector and fused quartz (30 m, 0.32 mm i.d.) BP-5 capillary column (SGE). Helium was used as a carrier gas with a split ratio of 1:50. The oven temperature started at 120°C; the temperature was then increased at a rate of 4°C/min up to 290°C and left for 10 min.

The relative proportions of each fatty acid in the *sn*-2 position were calculated as $(M/T \times 3) \times 100\%$, where M is the mol% of fatty acid in 2-monacylglycerol and T is mol% of fatty acid in triglycerides (Bracco, 1994; López-López et al., 2002; Straarup et al., 2006).

RESULTS

The energy and fat contents of each sample are given in Table 1. All formulas contained approximately 45% energy from fat. The fatty acid composition of the triacylglycerols isolated from the samples is presented in Table 2. The infant formulas had some short-chain fatty acids (C4:0 and C6:0), reflecting the use of ruminant milk fats (Table 2). Goat milk and goat infant formulas with milk fat tended to have more medium-chain fatty acids (C8:0 and C10:0) compared with cow milk or cow infant formula with milk fat. Palmitic acid was slightly lower in the goat and cow infant formulas compared with values of mature breast milk, but total saturated fatty acids levels were similar (Table 2). Goat infant formula contained slightly less monounsaturated fatty acids and PUFA compared with reference values for breast milk. The LA concentration of the goat and cow infant formula equated to 675 and 930 mg/100 kcal, respectively, and the LNA concentrations were 58 and 95 mg/100 kcal, respectively. This equated to a LA:LNA ratio of 12 for the goat infant formula and 10 for the cow infant formula. Lauric (C12:0) plus myristic (C14:0) acids for goat and cow infant formulas were 11 and 15% of total fat, respectively (Table 2).

The fatty acid composition of the *sn*-2 monoacylglycerols obtained by the Grignard reaction is presented in Table 3. The main differences in the fatty acid profiles of the 2-monoacylglycerols from goat and cow milks

were the approximately 3-fold higher number of medium-chain fatty acids and half the amount of palmitic acids in goat milk compared with cow milk. There were negligible short-chain 2-monoacylglycerols in the milks. The infant formulas had more oleic acid than palmitic acid in the *sn*-2 position.

Table 4 shows the relative percentage of the individual fatty acids that are present in the *sn*-2 position of triglycerides. Short-chain fatty acids (C4:0 and C6:0) from cow or goat milk were preferentially located in the *sn*-1(3) position, whereas other fatty acids did not show such marked specificity. Palmitic acid in cow and goat milk was equally distributed between *sn*-2 and *sn*-1(3).

DISCUSSION

The use of goat milk fat and unmodified vegetable oils produces an infant formula with a fatty acid profile that is similar, but not identical, to reference values for breast milk. The total levels of PUFA, which are derived from the vegetable oils in formulas, exhibit the greatest difference between the formulas and breast milk. However, PUFA levels in breast milk are heavily influenced by the mother's diet and vary considerably among women and among different regions (Jensen, 1999; Koletzko et al., 2001). The formulas produced in this study also do not have added docosahexaenoic acid or arachidonic acids, which are sometimes added to formulas for term infants (Simmer et al., 2008). Importantly, the present study shows that goat milk fat and unmodified vegetable oils provides essential fatty acids and a LA:LNA ratio within the range of 5 to 15:1 and less than 20% of total fat as lauric and myristic fatty acids as recommended for infant formulas (Koletzko et al., 2005). Thus, although the fatty acid profile of the formulas does not exactly match that of breast milk, they were still within the normal range reported in the literature (Jensen, 1999; Koletzko et al., 2001; López-López et al., 2002).

The key analysis in the present study was the regio-specific distribution of individual fatty acids within the triglyceride. Several techniques for regiospecific analysis of fatty acids have been used. Monoacylglycerols were

Table 2. Fatty acid profile (mol/100 mol of total fatty acids) of cow, goat, and human milks and infant formulas^{1,2}

Fatty acid ¹	Cow milk		Goat milk	Cow IF		Goat IF	Breast milk ⁴
	1	2		1	2		
C4:0	8.4	8.2	5.7 ± 0.2	1.9	1.2	3.1 ± 1.0	
C6:0	4.5	4.0	4.6 ± 0.2	1.4	0.8	2.5 ± 0.9	
C8:0	2.2	2.1	4.4 ± 0.2	2.3	2.3	2.0 ± 0.6	0.14 ± 0.04
C10:0	4.3	4.5	12.0 ± 0.2	2.5	2.0	7.3 ± 1.6	1.2 ± 0.4
C12:0	4.3	3.6	6.0 ± 2.0	9.3	9.6	4.2 ± 0.6	5.5 ± 1.1
C14:0	13	12	10.7 ± 1.2	6.4	5.2	7.0 ± 1.1	6.0 ± 1.4
C15:0	1.4	1.4	0.8 ± 0.1	0.4	0.2	0.6 ± 0.1	0.23 ± 0.07
C16:0	31	32	22 ± 1	14	12	17 ± 3	21 ± 2
C17:0	0.8	0.9	0.6 ± 0.1	0.2	0.2	0.4 ± 0.1	0.35 ± 0.06
C18:0	9.6	9.4	10.0 ± 1.7	5.0	4.5	6.3 ± 1.4	7.4 ± 1.2
C18:1 n-9	14	15	17.1 ± 0.5	33	36	31 ± 9	42 ± 3
C18:2 n-6	0.6	0.8	1.4 ± 0.1	19	20	14 ± 3	18 ± 5
C18:3 n-3	0.7	0.7	0.7 ± 0.2	1.9	2.5	1.2 ± 0.1	0.8 ± 0.3
C20:0	0.1	0.1	0.2 ± 0.1	0.3	0.3	0.3 ± 0.1	0.20 ± 0.05
SFA	79	78	77 ± 1	44	38	51 ± 9	42 ± 4
MUFA	17	18	19 ± 1	35	38	33 ± 9	42 ± 4
PUFA	1.3	1.5	2.1 ± 0.3	21	23	15 ± 3	20 ± 4

¹Results for goat milk and goat infant formula are mean ± SD of 3 batches of powders. Individual values are presented for 2 batches of cow milk and cow infant formula.

²Cow IF = infant formula with cow milk, cow milk fat, and vegetable oils (n = 2); Goat IF = infant formula with goat milk, goat milk fat, and vegetable oils (n = 3). Both manufactured by Dairy Goat Co-operative (NZ) Ltd. (Hamilton, New Zealand).

³SFA = saturated fatty acids; MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids.

⁴Values for breast milk are mean ± SD from López-López et al. (2002).

used in this study because this results in more accurate data than use of diacylglycerols (Turón et al., 2003). Monoacylglycerols may be produced from triglycerides by either pancreatic lipase or by Grignard deacylation

(Bracco, 1994). Pancreatic lipase gives variable selectivity for fatty acid chain length, particularly short-chain fatty acids that are present in ruminant milks (Ha and Lindsay, 1993). In contrast, no significant selectivity

Table 3. Fatty acid profile (mol/100 mol of total fatty acids) in the sn-2 position of triacylglycerols from cow, goat, and human milks and infant formulas^{1,2}

Fatty acid	Cow milk		Goat milk	Cow IF		Goat IF	Breast milk ³
	1	2		1	2		
C4:0	ND ⁴	ND	ND	ND	ND	ND	
C6:0	0.9	0.8	0.9 ± 0.3	0.1	0.1	0.2 ± 0.1	
C8:0	1.7	1.7	3.8 ± 0.1	0.7	0.4	1.0 ± 0.7	ND
C10:0	4.2	4.0	14.2 ± 0.3	2.1	1.4	7.0 ± 2.3	0.28 ± 0.08
C12:0	5.9	6.3	9 ± 3	16	16	6 ± 1.1	4.2 ± 0.7
C14:0	21.7	20.8	19 ± 2	8.5	5.8	13 ± 2	9.6 ± 1.6
C15:0	2.0	2.0	1.0 ± 0.2	0.6	0.4	0.8 ± 0.2	0.5 ± 0.1
C16:0	40.8	35.8	23 ± 2	12.4	8.5	16 ± 3	57 ± 5
C17:0	0.7	0.8	0.6 ± 0.1	0.2	0.2	0.4 ± 0.1	0.4 ± 0.1
C18:0	4.9	4.5	4.7 ± 3.8	0.2	0.2	0.4 ± 0.1	1.4 ± 0.3
C18:1 n-9	10.2	11.2	14 ± 1	32	36	30 ± 9	16 ± 3
C18:2 n-6	1.2	1.1	2.2 ± 0.6	21	24	16 ± 4	13 ± 3
C18:3 n-3	0.6	0.5	0.5 ± 0.2	1.8	2.7	1.1 ± 0.1	0.7 ± 0.1
C20:0	ND	ND	0.30 ± 0.04	0.2	0.2	0.3 ± 0.1	0.13 ± 0.04

¹Results for goat milk and goat infant formula are mean ± SD of 3 batches of powders. Individual values are presented for 2 batches of cow milk and cow infant formula.

²Cow IF = infant formula with cow milk, cow milk fat, and vegetable oils (n = 2); Goat IF = infant formula with goat milk, goat milk fat, and vegetable oils (n = 3). Both were manufactured by Dairy Goat Co-operative (NZ) Ltd. (Hamilton, New Zealand).

³Values for breast milk are mean ± SD from López-López et al. (2002).

⁴ND = not detected.

Table 4. Relative percentage of each fatty acid in *sn*-2 position^{1,2}

Fatty acid	Cow milk		Goat milk	Cow IF		Goat IF	Breast milk ³
	1	2		1	2		
C4:0	ND ⁴	ND	ND	ND	ND	ND	
C6:0	7	7	6 ± 2	2.4	4.2	2.7 ± 0.6	
C8:0	26	27	29 ± 1	10	6	13 ± 5	ND
C10:0	33	30	39 ± 1	28	23	32 ± 3	8 ± 3
C12:0	46	58	52 ± 2	58	56	52 ± 3	25 ± 4
C14:0	56	58	61 ± 2	44	37	63 ± 1	53 ± 6
C15:0	48	48	43 ± 1	50	66	46 ± 2	78 ± 9
C16:0	44	37	35 ± 1	29	24	31 ± 1	88 ± 7
C17:0	29	30	33 ± 6	33	33	33 ± 1	40 ± 5
C18:0	17	16	14 ± 11	1.3	1.5	1.8 ± 0.1	9 ± 2
C18:1 n-9	24	25	28 ± 1	32	34	32 ± 1	12 ± 2
C18:2 n-6	67	46	52 ± 9	38	38	37 ± 2	22 ± 3
C18:3 n-3	29	24	23 ± 3	32	36	31 ± 5	25 ± 5
C20:0	ND	ND	67 ± 28	22	22	28 ± 6	21 ± 9

¹Results were calculated as $(M/T \times 3) \times 100\%$, where M is the mol/100 mol of fatty acid in 2-monacylglycerol and T is mol/100 mol of fatty acid in triglycerides. Results for goat milk and goat infant formula are mean ± SD of 3 batches of powders. Individual values are presented for 2 batches of cow milk and cow infant formula.

²Cow IF = infant formula with cow milk, cow milk fat, and vegetable oils (n = 2); Goat IF = infant formula with goat milk, goat milk fat, and vegetable oils (n = 3). Both were manufactured by Dairy Goat Co-operative (NZ) Ltd. (Hamilton, New Zealand).

³Values for breast milk are mean ± SD from López-López et al. (2002).

⁴ND = not detected.

was observed with respect to chain length or level of saturation in the Grignard deacylation of triglycerides (Turon et al., 2003).

The distribution of the fatty acids within the triglycerides of cow and goat milk fat from the present study are in good agreement with previous studies of goat and cow milk (Freeman et al., 1965; Marai et al., 1969). Thus, the differences in the analytical techniques did not seem to affect the outcome of the analysis.

The relative percentage of palmitic acid in the *sn*-2 position in the formulas with milk fat was greater than 7 of the 11 commercial infant formulas measured by López-López et al. (2002) and all 8 of the infant formulas measured by Straarup et al. (2006). The percentage of palmitic acid in the *sn*-2 position of these formulas ranged from 7 to 28%.

The stereospecificity of palmitic acid in formula determines its digestion and absorption (Bracco, 1994). Absorption of palmitic acid increases in direct proportion to the percentage of palmitic acid in the *sn*-2 position between 10 and 75% (Lien et al., 1997). Similarly, absorption of fat by infants fed formula containing 24% palmitic acid, but with only 13% residing in the *sn*-2 position, was 90% compared with 93 to 98% from formula with the same level of palmitic acid, but with 39 to 66% residing at the *sn*-2 position (Carnielli et al., 1996). However, fat absorption is also dependent on the amount of palmitic acid. Infants fed formula containing 8 to 9.5% palmitic acid absorbed 95% of the

fat despite having only 12% of palmitic acid esterified to *sn*-2 (Nelson et al., 1996, 1998). This level of absorption is similar to the absorption efficiency of breast milk fat (Fomon et al., 1970), which has 25% palmitic acid with more than 60% at *sn*-2 (López-López et al., 2002; Straarup et al., 2006). In a trial with preterm infants, fat absorption was 92 and 89% from formula with fat containing 15 or 24% palmitic acid and 8 and 28% esterified to the *sn*-2 position, respectively (Lucas et al., 1997). Combined, these studies suggest there is no advantage to having more palmitic acid esterified to the *sn*-2 position until the level of palmitic acid in fat is increased to around 24%.

In conclusion, this analysis confirms that it is possible to make infant formula with a mixture of milk fat and vegetable oils to achieve a fatty acid profile recommended for infants (Koletzko et al., 2005). Further, at 17% palmitic acid and with 31% of this fatty acid in the *sn*-2 position, the formula with goat milk fat and vegetable oils is comparable to the formulas considered to have superior fat absorption in infants (Carnielli et al., 1996; Nelson et al., 1996; Lien et al., 1997; Lucas et al., 1997; Nelson et al., 1998; López-López et al., 2001). An infant feeding study has also demonstrated that growth rates of infants fed the formula are similar to that of breast-fed infants (Grant et al., 2005). Therefore, there is no reason to indicate that further increasing the proportion of palmitic acid in the *sn*-2 position by interesterification of the fat ingredients would be

required, thereby avoiding another processing step. The other advantage of this alternative approach is that the formulas contain potential functional components that are supplied by the milk fat globule membrane, including phospholipids, gangliosides, and glycoproteins (Berger et al., 2000; Spitsberg, 2005).

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REFERENCES

- Berger, A., M. Fleith, and G. Crozier. 2000. Nutritional implications of replacing bovine milk fat with vegetable oil in infant formulas. *J. Pediatr. Gastroenterol. Nutr.* 30:115-130.
- Becker, C. C., A. Rosenquist, and G. Holmer. 1993. Regiospecific analysis of triacylglycerols using allyl magnesium bromide. *Lipids* 28:147-149.
- Bracco, U. 1994. Effect of triglyceride structure on fat absorption. *Am. J. Clin. Nutr.* 60(Suppl):1002S-1009S.
- Carnielli, V. P., I. H. Luijendijk, J. B. Van Goudoever, E. J. Sulkers, A. A. Boerlage, H. J. Degenhart, and P. J. Sauer. 1996. Structural position and amount of palmitic acid in infant formulas: Effects on fat, fatty acid, and mineral balance. *J. Pediatr. Gastroenterol. Nutr.* 23:553-560.
- Folch, J., M. Lees, and G. H. Sloane Stanley. 1957. A simple method for the isolation and purification of total lipides from animal tissues. *J. Biol. Chem.* 226:497-509.
- Fomon, S. J., E. E. Ziegler, L. N. Thomas, R. L. Jensen, and L. J. Filer Jr. 1970. Excretion of fat by normal full-term infants fed various milks and formulas. *Am. J. Clin. Nutr.* 23:1299-1313.
- Freeman, C. P., E. L. Jack, and L. M. Smith. 1965. Intramolecular fatty acid distribution in the milk fat triglycerides of several species. *J. Dairy Sci.* 48:853-858.
- Grant, C., B. Rotherham, S. Sharpe, R. Scragg, J. Thompson, J. Andrews, C. Wall, J. Murphy, and D. Lowry. 2005. Randomized, double-blind comparison of growth in infants receiving goat milk formula versus cow milk infant formula. *J. Paediatr. Child Health* 41:564-568.
- Ha, J. K., and R. C. Lindsay. 1993. Release of volatile branched-chain and other fatty-acids from ruminant milk fats by various lipases. *J. Dairy Sci.* 76:677-690.
- Hansen, J. W., and U. Diener. 1997. Challenges of matching human milk fatty acid patterns technically and functionally. *Eur. J. Med. Res.* 2:74-78.
- Jensen, R. G. 1999. Lipids in human milk. *Lipids* 34:1243-1271.
- Koletzko, B., S. Baker, G. Cleghorn, U. F. Neto, S. Gopalan, O. Hernell, Q. S. Hock, P. Jirapinyo, B. Lonnerdal, P. Pencharz, H. Pzyrembel, J. Ramirez-Mayans, R. Shamir, D. Turck, Y. Yamashiro, and D. Zong-Yi. 2005. Global standard for the composition of infant formula: Recommendations of an ESPGHAN coordinated international expert group. *J. Pediatr. Gastroenterol. Nutr.* 41:584-599.
- Koletzko, B., M. Rodriguez-Palmero, H. Demmelmair, N. Fidler, R. Jensen, and T. Sauerwald. 2001. Physiological aspects of human milk lipids. *Early Hum. Dev.* 65(Suppl.):S3-S18.
- Lien, E. L., F. G. Boyle, R. Yuhas, R. M. Tomarelli, and P. Quinlan. 1997. The effect of triglyceride positional distribution on fatty acid absorption in rats. *J. Pediatr. Gastroenterol. Nutr.* 25:167-174.
- Liu, Q. T., and J. L. Kinderlerer. 1999. Preparative thin-layer chromatographic separation and subsequent gas chromatographic-mass spectrometric analysis of monoacylglycerols derived from butter oil by fungal degradation. *J. Chromatogr. A* 855:617-624.
- López-López, A., A. I. Castellote-Bargalló, C. Campoy-Folgozo, M. Rivero-Urgel, R. Tormo-Carnicé, D. Infante-Pina, and M. C. López-Sabater. 2001. The influence of dietary palmitic acid triacylglyceride position on the fatty acid, calcium and magnesium contents of at term newborn faeces. *Early Hum. Dev.* 65(Suppl.):S83-S94.
- López-López, A., M. C. López-Sabater, C. Campoy-Folgozo, M. Rivero-Urgel, and A. I. Castellote-Bargalló. 2002. Fatty acid and sn-2 fatty acid composition in human milk from Granada (Spain) and in infant formulas. *Eur. J. Clin. Nutr.* 56:1242-1254.
- Lucas, A., P. Quinlan, S. Abrams, S. Ryan, S. Meah, and P. J. Lucas. 1997. Randomised controlled trial of a synthetic triglyceride milk formula for preterm infants. *Arch. Dis. Child.* 77:F178-F184.
- Maduko, C. O., C. C. Akoh, and Y. W. Park. 2007a. Enzymatic interesterification of tripalmitin with vegetable oil blends for formulation of caprine milk infant formula analogs. *J. Dairy Sci.* 90:594-601.
- Maduko, C. O., C. C. Akoh, and Y. W. Park. 2007b. Enzymatic production of infant milk fat analogs containing palmitic acid: Optimization of reactions by response surface methodology. *J. Dairy Sci.* 90:2147-2154.
- Marai, L., W. C. Breckenridge, and A. Kuksis. 1969. Specific distribution of fatty acids in the milk fat triglycerides of goat and sheep. *Lipids* 4:562-570.
- Nelson, S. E., R. R. Rogers, J. A. Franz, and E. E. Ziegler. 1996. Palm olein in infant formula: Absorption of fat and minerals by normal infants. *Am. J. Clin. Nutr.* 64:291-296.
- Nelson, S. E., J. A. Frantz, and E. E. Ziegler. 1998. Absorption of fat and calcium by infants fed a milk-based formula containing palm olein. *J. Am. Coll. Nutr.* 17:327-332.
- Ostrom, K. M., M. W. Borschel, J. E. Westcott, K. S. Richardson, and N. F. Krebs. 2002. Lower calcium absorption in infants fed casein hydrolysate and soy protein based infant formulas containing palm olein versus formulas without palm olein. *J. Am. Coll. Nutr.* 21:564-569.
- Simmer, K., S. K. Patole, and S. C. Rao. 2008. Longchain polyunsaturated fatty acid supplementation in infants born at term. *Cochrane Database Syst. Rev.* 23:CD000376. doi:10.1002/14651858.CD000376.pub2
- Spitsberg, V. L. 2005. Invited review: Bovine milk fat globule membrane as a potential nutraceutical. *J. Dairy Sci.* 88:2289-2294.
- Straarup, E. M., L. Lauritzen, J. Faerk, C.-E. Høy, and K. F. Michaelsen. 2006. The stereospecific triacylglycerol structures and fatty acid profiles of human milk and infant formulas. *J. Pediatr. Gastroenterol. Nutr.* 42:293-299.
- Stránský, K., T. Jursik, and A. Vitek. 1997. Standard equivalent chain length values of monoenoic and polyenoic (methylene interrupted) fatty acids. *J. High Resolut. Chromatogr.* 20:143-158.
- Takagi, T., and Y. Ando. 1991. Stereospecific analysis of triacyl-sn-glycerols by chiral HPLC. *Lipids* 26:542-547.
- Thomas, A. E., J. E. Scharoun, and H. Ralston. 1965. Quantitative estimation of isomeric monoglycerides by thin-layer chromatography. *J. Am. Oil Chem. Soc.* 42:789-792.
- Turon, F., B. Bonnot, Y. Caro, M. Pina, and J. Graille. 2003. Acyl migration incidence on accuracy of a triacylglycerol regioanalysis—A theoretical evaluation. *Chem. Phys. Lipids* 125:41-48.
- Wolff, R. L. 1994. Contribution of trans-18:1 acids from dairy fat to European diets. *J. Am. Oil Chem. Soc.* 71:277-283.