Prostaglandins, Leukotrienes and Essential Fatty Acids 89 (2013) 139–143

Contents lists available at ScienceDirect

Prostaglandins, Leukotrienes and Essential Fatty Acids

journal homepage: www.elsevier.com/locate/plefa

Review

Review of *sn*-2 palmitate oil implications for infant health

Fabiana Bar-Yoseph *, Yael Lifshitz, Tzafra Cohen

Enzymotec Ltd., Sagi 2000 Industrial Park, Kfar Baruch, Israel

ARTICLE INFO

Keywords:
Sn-2 palmitate
Human milk
Structured triglyceride

ABSTRACT

Human milk provides the optimal balanced nutrition for the growing infant in the first months after birth. The human mammary gland has evolved with unusual pathways, resulting in a specific positioning of fatty acids at the outer *sn*-1 and *sn*-3, and center *sn*-2 of the triacylglyceride, which is different from the triglycerides in other human tissues and plasma. The development of structured triglycerides enables mimicking the composition as well as structure of human milk fat in infant formulas. Studies conducted two decades ago, together with very recent studies, have provided increasing evidence that this unusual positioning of 16:0 in human milk triglycerides has a significant role for infant health in different directions, such as fat and calcium absorption, bone health, intestinal flora and infant comfort. This review aims to unravel the relevance of human milk triglyceride *sn*-2 16:0 for intestinal health and inflammatory pathways and for other post-absorption effects.

1. Introduction

Human breast milk provides the optimum nutrition for infants, and is being designed to provide perfectly balanced nutrition to meet the needs of the growing infant in the first months after birth. About 50% of energy in human milk is provided by fatty acids in the milk triglycerides, which themselves are molecules comprised of mixtures of three fatty acids esterified to a glycerol backbone [1]. Triglyceride synthesis, rather than involving random esterification of three fatty acids to glycerol, involves specific positioning of fatty acids at the outer *sn*-1 and *sn*-3, and center *sn*-2 positions of the triacylglyceride. The human mammary gland has evolved with unusual pathways for acylation of fatty acids into triglycerides for secretion in milk, with these pathways resulting in a different triglyceride structure (triglyceride fatty acid arrangement) from the triglycerides in other human tissues and plasma [2], or common dietary fats and oils. This stereospecific positioning of fatty acids in human milk triglycerides involves preferential positioning of the saturated fatty acid palmitic acid (16:0) at the *sn*-2 position, rather than at the *sn*-1,3 positions, as is typical of human tissue and plasma lipids, and vegetable oils common in human diets, and in the fat blends used in the manufacture of infant formula [3]. Studies over the last two to three decades have provided increasing evidence that this usual positioning of 16:0 in human milk triglycerides promotes the absorption of both 16:0 and calcium in term and preterm infants [4–8]. However, the importance of the *sn*-2 positioning of 16:0 in human milk triglycerides is expected to extend beyond saturated fatty acids and calcium absorption, since it also impacts the composition of unesterified fatty acids in the intestinal lumen, and the composition of unesterified fatty acids and *sn*-2 monoglycerides that enter the intestinal enterocyte. Recently, there has been a growing understanding that dietary fatty acids may influence the intestinal microbiome and contribute in complex ways to neurological and immune system development through roles in cell signaling and regulation of gene expression. This paper provides a review of new studies with triglycerides synthesized to contain 16:0 on the *sn*-2 (also termed β-16:0) and 18:1n-9 on the *sn*-1, 3 position to resemble the major 16:0 containing triglyceride species in human milk. Our purpose is to begin to unravel the relevance of human milk triglyceride *sn*-2 16:0 for intestinal health and inflammatory pathways and for other post-absorption effects.

2. Structured triglycerides

Palmitic acid (C16:0) is the major saturated fatty acid in human milk, accounting for 17–25% of the total fatty acids [2]. Equally important, over 70% of 16:0 is esterified at the milk triglyceride *sn*-2 position [2,9]. The major unsaturated fatty acid in human milk is oleic acid (18:1n-9) and this is mostly esterified at the triglyceride *sn*-1,3 (outer) positions, with the result that triglycerides with the structure 18:1n-9—16:0—18:1n-9 are a major triglyceride species in human milk and represent an estimated 11.8% of the total triglyceride species [2]. Early studies addressing the importance of the human milk triglyceride structure compared fat absorption in infants fed human milk with

* Corresponding author.
E-mail address: fabiana@ENZYMOTEC.COM (F. Bar-Yoseph).

0952-3278/$ - see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.plefa.2013.03.002

infants fed formulas containing lard (an unusual animal fat in which 16:0 is also high and esterified in the triglyceride *sn*-2 position), and infants fed lard that had been randomized to redistribute 16:0 equally across all three carbons on the triacylglyceride [10]. The latter studies showed that redistributing 16:0 from the *sn*-2 position of the formula triglyceride led to decreased fat absorption. Although two or more vegetable oils can be blended to give the same average amounts of 16:0, 18:1n-9 and 18:2n-6 in an infant formula as in human milk, the stereo-specific arrangement of vegetables oil triglycerides means that the 16:0 will be present almost entirely on the triglyceride *sn*-1,3 positions [3]. The development of structured triglycerides enables mimicking both the composition as well as the structure of human milk fat for infant formulas. Structured TG are achieved through an enzymatic process by which the 16:0—18:1n-9—16:0 is transformed to 18:1n-9—16:0—18:1n-9. The resulting product contains 17–25% palmitic acid with above 40% located at the center *sn*-2 position.

3. Importance of dietary 16:0 positioning in triglycerides for fatty acid and calcium absorption.

Studies done several decades ago have demonstrated the greater efficiency of fat absorption and softer stools in breast-fed infants compared to that of infants fed with formulas containing 16:0 from saturated vegetables; effect that was linked to the large amounts of 16:0 in human milk at the *sn*-2 position of the milk triglycerides [4–8]. Triglyceride digestion by endogenous lipases leads to hydrolysis of fatty acids from the triacylglyceride *sn*-1,3 linkages, to release two unesterified fatty acids and one *sn*-2 monoglyceride from each triglyceride into the intestinal lumen [11]. A role for the milk bile salt-stimulated lipase in completing hydrolysis of *sn*-2 monoglycerides with 16:0 released during triglyceride digestion is unlikely, since unesterified 16:0 is poorly absorbed [3]. Structuring 16:0 on the triglyceride *sn*-2 position of milk or formula fats improves 16:0 absorption [12,13], and plasma chylomicron triglycerides of breast fed infants are high in *sn*-2 16:0 [14,15]. In addition to low intraluminal solubility, unesterified 16:0 has an increased tendency to combine with divalent cations, such as calcium, to form insoluble soaps, which are malabsorbed [16]. Clinical evidence for this has been provided by studies to show increased fecal excretion of fatty acid soaps of 16:0 and calcium, accompanied by harder stools, in infants fed formula containing 16:0 from saturated vegetable oils rather than structured triglycerides containing β-16:0 [4–8]. Fig. 1 shows the correlation between the level of 16:0 in the milk or formula triglyceride sn-2 position and infant fatty acid and calcium absorption calculated as a modified Cohen's effect size (f2) [17] using data from published studies with term [5–7] and preterm infants [4,8]. Since an effect size of over 0.8 is recognized as a large effect [17], β-16:0 with over 40% 16:0 at the *sn*-2 position would have a large beneficial effect on fatty acid and calcium. The results show that progressively increasing 16:0 at the *sn*-2 (and decreasing 16:0 at the *sn*-1,3 positions) of the formula triglyceride leads to a dose-dependent increase in 16:0 and calcium absorption ($r=0.95$ and $r=0.78$ for 16:0 and calcium, respectively). The reduction in fecal calcium and 16:0 as calcium soaps is accompanied by a decrease in the incidence of harder stools [6,18,19].

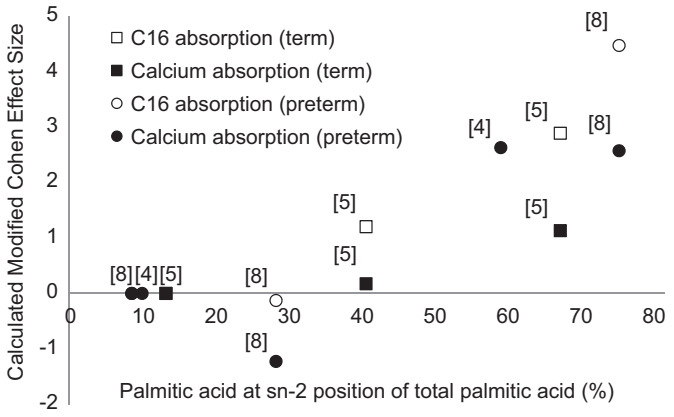

Fig. 1. Modified Cohen's effect size of the effect of the 16:0 position in formula triglycerides on the absorption of 16:0 and calcium in term and preterm infants. The percentage of 16:0 in formula triglycerides (*x*-axis) was plotted against the mean percentage of 16:0 absorption (open circles or squares) or calcium (solid circles or squares) reported in clinical studies with preterm (circles) and term (squares) infants to derive the Cohen effect (f2), as described (Cohen 1988) [17]. The figure shows that progressive enrichment of 16:0 at the *sn*-2 rather than *sn*-1,3 positions of formula triglycerides leads to a dose response increase in calcium and 16:0 absorption. The numbers on the data points on the graph refer to the published studies, as cited in the references.

4. Bone health

Malabsorption of calcium in fatty acid soaps in infants fed formulas containing 16:0 rich vegetable oils has led to interest in the possible effects of milk and formula 16:0 on bone mineralization in young infants [6]. Recent advances in methods for assessment of bone strength parameters with the development of quantitative measures of bone using supersonic speed of sound (SOS) have recently been applied to this direction of research. Ultrasound bone sonometry is a non-invasive technique that enables quantitative longitudinal assessment of changes in bone parameters in the tibia or other bones in term and preterm infants from birth [20–22]. Litmanovitz et al. recently applied the bone SOS technology in a randomized, controlled, double-blind clinical study of bone parameters in term infants fed formula containing triglycerides with *sn*-2 16:0 from InFat® or standard vegetable oil blends with comparison to a non-randomized group of breast-fed infants [23]. Tibia bone SOS decreased during the first 3 months

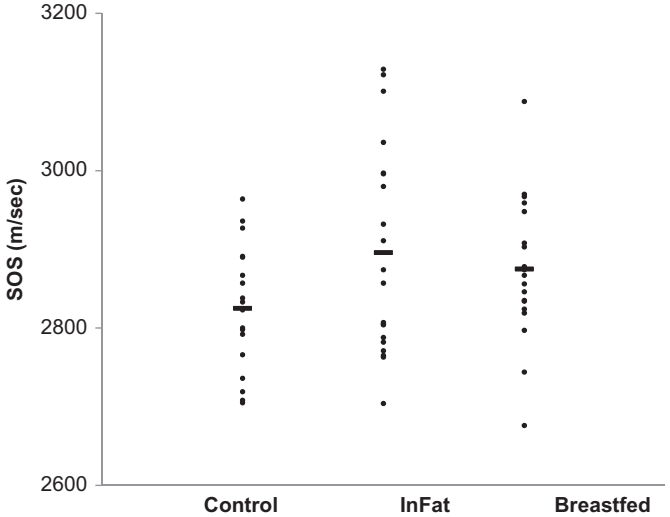

Fig. 2. Ultrasound Speed of Sound (SOS) of the tibia of term infants 3 fed with formula containing 16:0 in structured triglycerides (β-16:0) or unmodified vegetable oil, or who were breast-fed. from birth to 12 weeks of age. Data for each infant within each group is shown by the individual points. The group mean is indicated by the solid line, with the mean for the group fed β-16:0 ($n=20$) significantly higher than for the group fed the standard formula ($n=18$) ($p<0.05$), and not different from the group of the breast-fed infants ($n=22$). The formulas contained about 20% 16:0, with 43% or 14% 16:0 in the *sn*-2 position of the β-16:0 and standard formulas, respectively.