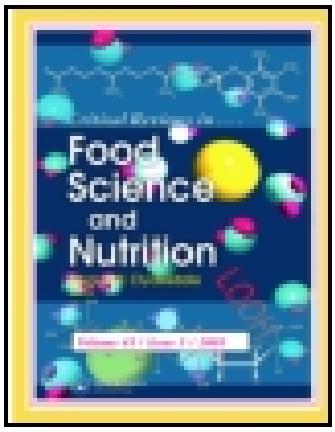

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Palmitic Acid in Early Human Development

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Palmitic Acid In Early Human Development

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Abstract

Palmitic acid (16:0) is a saturated fatty acid present in the diet and synthesized endogenously. Although often considered to have adverse effects on chronic disease in adults, 16:0 is an essential component of membrane, secretory and transport lipids, with crucial roles in protein palmitoylation and signal molecules. At birth, the term infant is 13-15% body fat, with 45-50% 16:0, much of which is derived from endogenous synthesis in the fetus. After birth, the infant accumulates adipose tissue at high rates, reaching 25% body weight as fat by 4-5 months age. Over this time, human milk provides 10% dietary energy as 16:0, but in unusual triglycerides with 16:0 on the glycerol center carbon. This paper reviews the synthesis and oxidation of 16:0

and possible reasons why the infant is endowed with large amounts of fat and 16:0. The marked deviations in tissues with displacement of 16:0 that can occur in infants fed vegetable oil formulas is introduced. Assuming fetal fatty acid synthesis and the unusual delivery of 16:0 in human milk evolved to afford survival advantage to the neonate, it is timely to question if 16:0 is an essential component of tissue lipids whereby both deficiency and excess are detrimental.

Introduction

The third trimester human fetus accumulates substantial amounts of the saturated fatty acid palmitic acid (16:0), much of which arises from endogenous synthesis in the fetus (Dancis, et al. 1973; Herrera, 2000; Herrera, et al. 2000; Widdowson, 1975). After birth, human milk is a rich source of 16:0, providing about 10% of the breast-fed infantôs energy intake although in triglycerides with an unusual arrangement of fatty acids (Innis 2011). The availability of human milk substitutes (formulas) and intravenous lipids containing vegetable oils rather than milk fats since the 1970s has enabled the feeding of preterm and term infants with fats low in 16:0, and this alters the composition of developing tissues and lowers plasma cholesterol (Fuch, et al. 1994; Farquharson, et al. 1993; Hashim, 1983; Hayes, et al. 1992; Sweeney, et al., 1963; Widdowson, et al. 1975). Newer research emphasizes that fatty acids are more complicated than simple, equivalent sources of energy, and this extends to palmitic acid (16:0) which is both a major metabolic fuel and component of membrane structural lipids, with critical roles in protein palmitoylation and palmitoylated signal molecules. This paper reviews 16:0 accumulation before birth, the unusual aspects of 16:0 in human milk, possible reasons why the human fetus and infant are endowed with large amounts of 16:0, and recent advances that may shed light on the possible implications of low 16:0 intakes during early development. Like energy and other nutrients, the relationship between 16:0 and optimal development may well be U-shaped, with adverse effects associated with both low and high intakes. A brief review of fatty acid synthesis and oxidation, placental lipid transfer and human milk fatty acids is included to assist readers of diverse background.

Fatty acid synthesis and oxidation in development.

Palmitic acid (16:0) is a straight chain saturated fatty acid, which can be provided in the diet or synthesized endogenously from other fatty acids, carbohydrate and some amino acids. Although often considered as a component of adipose tissue triglycerides, 16:0 is also abundant in membrane lipids. Whereas triglycerides are composed of a glycerol to which three fatty acids are esterified, membrane and plasma phospholipids are comprised of a glycerol with two fatty acids, one esterified at each of the *sn*-1 (outer) and *sn*-2 (center) carbons, with a polar group, such as choline or ethanolamine, linked through a phosphate at the glycerol sn-3 carbon. Sphingolipids, such as sphingomyelin, are a second major class of membrane and plasma lipids that differ from classical phospholipids in having sphingosine (synthesized from palmitoyl CoA and serine) rather than glycerol as the backbone. The n-6 and n-3 polyunsaturated fatty acids, which have been the subject of much of the recent work on fatty acid nutrition in pregnancy, lactation and infancy are primarily esterified at the sn-2 carbon of glycerol in phospholipids. The sn-1 position of phospholipids and the sphingolipids, however, are rich in saturated fatty acids, including 16:0. Thus, not only n-6 and n-3 fatty acids, but also saturated fatty acids are needed during development for growth of membrane lipids, secretory lipids such as lung surfactant, plasma lipoproteins and bile, as well as for storage in adipose tissue. Important to remember, the fetus and infant are anabolic and nourished continuously or at frequent intervals via the placenta or with human milk, respectively. The supply of protein, fat and mono (glucose) or disaccharide (lactose) is also distinctly different from the usual diet of the adult. Consequently, information from studies in adults, particularly in the context of excess fat or carbohydrate intakes, needs to considered carefully before extrapolation to the fetus or infant.

Fatty acid synthesis and oxidation undergo marked changes from intra-uterine to extra-uterine life that closely parallel the delivery of low amounts of fatty acids by placental transfer before birth, and feeding with milk which is high in fat after birth. With the exception of lactating women, the major organ of fatty acid synthesis in humans, in contrast to rodents, is the liver**.** Fatty acid synthesis is accomplished by the fatty acid synthase complex by addition of seven malonyl CoAs to acetyl CoA, generating 16:0. Insertion of a double bond by æ-9 desaturase (stearoyl CoA desaturase) at carbon 9 of 16:0 forms 16:1n-7 (palmitoleic acid). Elongation of 16:0 to stearic acid (18:0) followed by æ-9 desaturation generates oleic acid (18:1n-9). Insertion of a double bond requires oxygen and NADH. Humans, like other animals lack the æ-12 desaturase and æ-15 desaturase needed to form n-6 or n-3 fatty acids, respectively (Innis, 2003). This means that whereas humans can synthesize saturated and n-9 and n-7 unsaturated fatty acids, all of the n-6 and n-3 fatty acids needed by the fetus and breast-fed infant must come from the mother, by placental transfer or breast milk, respectively.

Fatty acid oxidation is achieved by sequential release of acetyl CoA from the fatty acid carbon chain. Release of each acetyl CoA by mitochondrial ß-oxidation involves dehydrogenation to create a *trans* double bond between carbon 2 and 3, hydration, then oxidation of the double bond. Finally, insertion of a thiol between carbon 2 and 3 releases acetyl CoA and a fatty acyl CoA which is now two carbons shorter. When a *cis* double bond is on an odd numbered carbon (such as oleic acid, 18:1n-9), enoyl CoA isomerase switches the *cis* bond to the *trans* configuration, and mitochondrial ß-oxidation proceeds. A *cis* double bond on an even carbon requires 2,4 dienoyl-CoA reductase to reduce the double bond using NADPH before ß-oxidation can proceed. To summarize, synthesis of 16:0 rather than unsaturated fatty acids

reduces the demand for NADH; oxidation of 16:0, 16:1n-7 and 18:1n-9 reduces demands for NADPH when compared to n-6 and n-3 fatty acids as sources of metabolic energy.

At birth, the term gestation infant is about 13-15% body fat and one of the fattest known newborns (Kuzawa, 1998). Figure 1 illustrates human body fat content as percent of body weight from the first trimester of gestation through early infancy to five years of age, together with the rate of fat gain/month from birth to 5 years of age. For reference, non-human primates and rodents are about 2-3% body fat at birth, and newborn seals and sea-lions are about 5-10% body fat, although the latter cold climate species have large amounts of brown adipose and relatively little subcutaneous adipose at birth (Kuzawa, 1998). Using data from Widdowson (1950), a 3 kg human newborn has approximately 450 g adipose, which is much more than the 122 g predicted for a mammalian newborn of that weight (Kuzawa, 1998). Not only is the human newborn endowed with an unexpected and unprecedented amount of white adipose tissue, this adipose tissue contains a remarkable 45-50% 16:0, 13-15% 16:1 and 30-34% 18:1 (Baker, 1969; Boersma, 1979; Farquharson, et al., 1993; Hashim, 1963; Sweeney, et al. 1963; Widdowson, et al. 1975), which is summarized in Table 1. Sweeney, et al. (1963) included comparison of adipose tissue fatty acids from newborn infants and their mothers, and this shows two fold high 16:0 and 16:1n-7, but substantially lower 18:1 and 18:2 in the fetal than maternal tissue (Table 1). Comparative studies of adipose tissue form newborn term, appropriate-weight-for-gestational age preterm infants and small-for-gestational age infants, on the other hand, show no differences in the high proportion of 16:0 and 16:1, or in 18:0, 18:1 and 18:2 (Boersma, 1979). From the preceding section, we understand that saturated, n-9 and n-7 monounsaturated fatty acids, which represent about 95% of human adipose fatty acids at birth, are readily oxidized to provide energy

avoiding needs for NADPH to reduce the double bonds in polyunsaturated fatty acids. In healthy women, third trimester adipose tissue accumulation in the fetus is enabled by placental transfer of glucose and its use as a lipogenic substrate, and to a smaller extent by placental transfer of fatty acids and their low oxidation, as discussed in the next section (Herrera, 2000; Herrera & Amusquivar, 2000).

After birth, the nutrient supply switches from primarily glucose and amino acids to milk. Mature human milk produced after the first 2-3 weeks of lactation provides about 70 kcal/dL, with 50% energy from fat, 8-10% from protein and 40% from carbohydrate, primarily lactose (Jensen, 1999; Nommsen, et al. 1991). With birth, hepatic fatty acid synthesis decreases, and the use of exogenous (dietary) rather than endogenously synthesized fatty acids for expansion of adipose tissue reserves increases. Growth of adipose continues rapidly during the first year after birth, with the infant reaching a remarkable 25% body weight as fat by 4-5 months of age (Fomon, 1967; Fomon, et al. 1982). As shown in Figure 1, rates of fat gain are particularly high between 1 and 4 months of age, with peak rates of fat accretion of about 400 g/month at this time.

The question of why humans invest in large adipose tissue reserves in the third trimester of gestation and first months after birth is intriguing. The large brain to body weight ratio of the newborn is a second distinguishing feature of humans, and this brings high metabolic demands to support the brain compared to the rest of the body. Kuzawa (1998) forwarded an hypothesis linking the accumulation of white adipose with the ability to mobilize energy and thus maintain the metabolic demands of the vulnerable infant brain during periods of negative energy balance. Immediately after birth when lactation is becoming established, illness secondary to

immunological immaturity and infectious disease, and introduction of solid foods during weaning are all periods often associated with negative energy balance when the ability to mobilize fatty acids from adipose tissue would be beneficial. Ketone body turnover (about 13-26 µmol/kg/min) in newborn and 6 month old infants fasted less than 8-10 hours are exceeding high, equivalent to about 25% of the newbornôs basal energy requirement, and in the range achieved in adults only after several days total fasting (Bougneres, et al. 1986). While the ability of the young infant to quickly mount ketogenesis to support the energy needs of the brain during periods of negative energy balance is clearly advantageous, the potential benefit of 16:0 as a substrate for oxidation may worth considering.

To summarize, the fetus accumulates large amounts of 16:0 and 16:1n-7 during gestation. Adipose tissue increases from 2% body weight at 12 weeks gestation, to 8% fat at 33 weeks gestation, and about 13-15% fat at term gestation (Fomon, et al., 1982; Widdowson, 1950; Widdowson and Spay, 1951), as illustrated in Figure 1. Close to 95% of the fatty acids in infant adipose tissue reserves are saturated and monounsaturated fatty acids, with 16:0 representing about half of all the fatty acids present (Baker, 1969; Boersma, 1979, Farquharson, et al., 1993; Hashim, 1963; Sweeney, et al. 1963; Widdowson, et al. 1975), with much higher 16:0 than in the mother (Table 1). Oxidation of 16:0, or its metabolite 16:1n-7, as an energy source does not require NADPH for isomerization of double bonds as found in polyunsaturated fatty acids, and after conversion to ketones, may also provide energy for the brain.

Placental fatty acid transfer and palmitic acid accretion

As introduced, large amounts of lipid and in particular 16:0 and 16:1n-7 are accumulated in fetal tissues during the last trimester of gestation. Placental fatty acid transfer increases from

about 2% fetal energy requirements in early gestation to 11% close to term gestation, with glucose and amino acids providing much of the substrate for saturated fatty acid synthesis in the fetus (Dancis, et al. 1973; Herrera, et al. 2000; Herrera 2000). Intact triglycerides and phospholipids do not cross the placenta, but require hydrolysis by placental lipases (Lager and Powell 2012). Unesterified fatty acids released from maternal plasma esterified lipids mix with maternal plasma unesterified fatty acids for placental uptake. The hydrophobic nature of fatty acids means that uptake and transport mechanisms are needed to enable transfer of fatty acids from the maternal circulation across the placenta prior to release to the umbilical vein. In the fetus, the umbilical vein flows into left portal vein, from which the ductus venosus channels blood to the left hepatic vein and thence the inferior vena cava; this allows much of the placental blood to initially bypass the fetal liver. The umbilical vein and ductus venosus close shortly after birth.

Interpretation of relative (percent) differences in fatty acids in maternal and fetal (or newborn infant cord) blood is complicated by the marked differences in intra- and extra-uterine fatty acid supply, metabolism, and of course plasma lipid transport and concentrations. The plasma triglycerides, phospholipids, cholesterol esters and unesterified fatty acid concentrations are considerably lower in fetal than maternal plasma, with a much higher phospholipid/triglyceride and unesterified fatty acid/triglyceride ratio in the fetus (2.58 and 0.39, respectively) than mother (1.20 and 0.18, respectively) (Berghaus, et al. 1998). Triglycerides and phospholipids are synthesized in the fetal liver then secreted in lipoproteins for transfer to the organs. Fetal plasma unesterified fatty acids presumably reflect the interplay of maternal transfer and recycling of fatty acids from fetal tissues and plasma lipoproteins. Studies on human

placental fatty acid transfer indicate that the net transfer of 16:0 is proportional to its concentration in the maternal unesterified fatty acids, and lower than required for deposition in the fetus in the last trimester of gestation (Booth, et al. 1981; Dancis, et al. 1973; Hersfield and Nemeth, 1968). Although 16:1n-7 is abundant in fetal adipose (Baker, 1969; Boersma, 1979; Farquharson, et al., 1993; Sweeney, et al. 1963; Widdowson, et al. 1975), the low 16:1n-7 in maternal plasma lipids suggests 16:1n-7 arises from desaturation of 16:0 in fetal tissues. The 2-fold higher 16:0 and 16:1, but substantially lower 18:1 and 18:2 in maternal than infant adipose tissue (Table 1) is also consistent with an interpretation that much of the 16-carbon chain fatty acids in the fetus are derived from endogenous synthesis in fetal tissues rather than transfer from the mother.

In summary, the human fetus accumulates large amounts of 16:0 during the last trimester of gestation, much of which originates from synthesis in the fetal liver. High amounts of 16:0 are needed for growth of membrane lipids, including the brain and other organs, but particularly adipose tissue, where high amounts of 16:1n-7 are also present. The extent to which the maternal fatty acid supply in healthy gestation, or nutritional care of low birthweight infants with intravenous lipids or enteral formulas low in 16:0 limit the normal synthesis and accumulation of 16:0 and 16:1n-7, and any metabolic or physiological implications of this are unclear.

Palmitic Acid in Human milk

Lactation is a unique mammalian feature with an evolutionary history extending over 150 million years (Lefèvre, et al. 2010; Innis, 2013). Current theories suggest an evolutionary advantage grounded in the motherôs ability to provide the infant born into pathogen-laden,

nutrient-poor environments with a secretory fluid that affords crucial immunological protection and nutrition. Over time, these secretions evolved into species-specific, highly complex mammalian milks, although they retain several unifying characteristics. Relevant to 16:0, the human mammary gland has evolved with unique pathways of both fatty acid synthesis and triglyceride synthesis (Innis, 2011; Innis, 2013); presumably the gland would not invest in expression of such unusual enzymatic pathways unless of benefit to the infant, mother or both.

Human milk lipid is comprised of 98% triglyceride which is present in globules surrounded by the milk fat globule membranes (Garcia and Innis, 2013). Fatty acids taken up from the maternal plasma by the mammary gland are combined with fatty acids synthesized in the gland itself for esterification into triglycerides and secretion in milk. Together the milk fatty acids contribute about 50% of the exclusively breastï fed infantôs energy intake, with 16:0 representing about 20% of the milk fatty acids, equivalent to about 10% of energy (Innis, 2013; Jensen, 1999). The medium chain, monounsaturated, n-6 and n-3 fatty acids in human milk vary widely and are readily and rapidly altered by changing the fat in the lactating motherôs diet. In contrast, the amounts of 16:0 in human milk seems to be quite constant among women from different regions of the world and following very different diets (Innis, 2004; Innis, 2011; Jensen, 1999). Current evidence suggests human milk 16:0 is maintained by combining and balancing uptake of 16:0 from the maternal plasma with synthesis by elongation of 14:0 in the mammary gland (Novak and Innis, 2011; Innis, 2013).

In addition to a high 10% of energy from 16:0, the human milk fed-infant consumes 16:0 in an unusual arrangement in the milk triglycerides (Innis, 2011). The triglycerides in human tissues and plasma, like those of other animals and vegetable oils, typically have an unsaturated

fatty acid esterified at the *sn*-2 glycerol carbon, with saturated fatty acids like 16:0 on the *sn*-1 carbon. Distinctly different from this, about 70% of the 16:0 in human milk is esterified on the *sn*-2 carbon of the triglyceride glycerol, known as 2-palmitate (Innis, 2011). Cowsô and rodent milk fats have 20-25% 16:0 and 15% 16:0, respectively, with 45% or 58% esterified on the *sn*-2 glycerol carbon of the their milk triglycerides, respectively, (Freeman, et al., 1965; Jensen, 1999; Innis, 2011). Lard (pig fat) is a notable exception to dietary fats and oils with about 85% of the 16:0 in the fat esterified at *sn*-2 carbon of the triglyceride glycerol. Early studies showing that the position of 16:0 in dietary triglycerides influences 16:0 absorption in infants involved formulas with natural lard (Filer, et al. 1969; Innis, 2011).

To summarize, human milk has evolved to provide the infant with a rich source of 16:0 and to do so in triglycerides with an unusual fatty acid pattern in which 16:0 is primarily esterified at the center carbon of the glycerol backbone. Dietary fats and oils vary widely in their saturated and unsaturated fatty acid mix which affords the ability not only to replace 16:0 with other fatty acids, but to also provide the same amount of 16:0 in triglycerides with distribution of fatty acids distinctly different from human milk.

Implications of dietary 16:0 in human development

The effects of 16:0 in the infant diet have been most extensively studied with respect to its absorption, and the implications for infant plasma cholesterol and adipose tissue fatty acids when 16:0 is replaced with unsaturated fatty acids. It is well-known that the absorption of unesterified fatty acids from the intestine decreases with increasing carbon length, and increases with increasing unsaturation. The estimated average coefficient of absorption (% of intake absorbed)

of unesterified fatty acids in infants is >88% for 14:0, 90-92% for 18:1n-9 and 92-94% for 18:2n-6, but 74% for 16:0 and 63% for 18:0 (Jensen, et al. 1986). Early studies noted the efficient absorption of fat from human milk by young infants, despite its high 16:0 content (Innis, 2011). In insightful work, these early studies showed the importance of the distribution of 16:0 in triglycerides for its absorption (Filer, et al., 1969). The lower absorption of 16:0 than shorter chain saturated or long-chain unsaturated fatty acids is explained by a phase transition (melting point) above body temperature, and greater tendency to form insoluble soaps with divalent cations, such as calcium and magnesium, at alkaline pHs such as found in the intestine (Innis, 2011). Digestion of dietary triglycerides by gastric and pancreatic lipases releases *sn*-2 monoglycerides and unesterified fatty acids cleaved from the glycerol *sn*-1 and *sn*-3 positions (Innis, 2011). When esterified at the sn-2 position of dietary triglycerides, 16:0 is released into the intestine in *sn*-2 monoglycerides (2-palmitate) rather than as unesterified fatty acids. The more polar nature of monoglycerides facilitates micellarization and transfer through unstirred water layer, improving absorption. In the enterocyte, 2-monoglycerides are re-esterified to triglycerides for secretion in chylomicrons, and these triglycerides retain the characteristics of the dietary triglycerides with respect to fatty acid arrangement. By minimizing release into the intestinal lumen as an unesterified fatty acid, dietary triglycerides with 2-palmitate reduce malabsorption of 16:0 in insoluble complexes with calcium or other minerals (Innis, 2011). Several studies with term and preterm infants have shown that feeding triglycerides with 2-palmitate rather than 1,3 palmitate decreases malabsorption of 16:0 and calcium, and increases bone mineral content and density (Innis 2011). The implications for bone health beyond the period of feeding, however, are unclear. The broader implication is that human milk is designed

to provide the infants with a relatively large amount of 16:0 and to do so in an usual way, also associated with efficient absorption. It remains possible, and is worth considering, whether the implications of 16:0 for mineral absorption particularly from human milk is a secondary variable, with the true nature of the importance of dietary 16:0 for the young infant lying beyond that of the intestinal lumen.

Re-esterification of 2-monoglycerides in the intestinal enterocytes after their absorption means that the arrangement of fatty acids in dietary triglycerides is largely retained in the triglycerides secreted into the blood stream in chylomicrons (Innis, 2011). Studies stemming from the 1970s, coincident with the change from cowsô milk fats to vegetable oils in infant formulas in many countries, were interested in the effects of replacing saturated fatty acids in the diet of young infants with vegetable oils lacking cholesterol, low in 16:0 and high in 18:2n-6. Infants fed formulas rich in 18:2n-6 and low in 16:0 have lower plasma total and LDL-cholesterol, lower triglycerides and phospholipids, and altered HDL subclasses when compared to breast-fed infants or infants fed formula with cowsô milk fat (Fuch, et al., 1994; Hayes, et al., 1992; Van Biervliet, et al., 1977; Van Biervliet, et al., 1986). Regardless of concerns that cholesterol in human milk expands the LDL pool with an "unfavorable" increase in the LDL/HDL ratio in the breast-fed infant (Hayes, et al., 1992), it is now clear that adults who were breast-fed as infants do not show increased risk factors for cardiovascular disease (blood pressure, elevated blood lipids, insulin resistance, or increased waist circumference) (Pirilä, et al., 2013). The importance of the plasma lipoprotein transport system for delivering essential fatty acids or other lipophilic nutrients to developing tissues and the implications of decreasing

the liver-derived apo B containing LDL secondary to replacing 16:0 in infant diets with unsaturated fatty acids offers many opportunities for research.

Little information is available on whether milk triglycerides with their unusual 2-palmitate alters lipoprotein metabolism or tissue fatty acid delivery in young infants. Higher plasma apo B and apo A-1, the primary apo-proteins in LDL and HDL, respectively, in infants fed triglycerides with 2-palmitate rather than native palm olein oil (Nelson and Innis, 1999) suggests that the both the amount and distribution of 16:0 in the infant diet is important to the metabolic effects postabsorption. Consistent with this, gene expression differs when the same fatty acids are provided to hepatocytes in chylomicron remnants rather than as unesterified fatty acids (Kohan, et al. 2011). Chylomicron remnants from animals fed lard, which is rich in 2-palmitate, where much less effective in suppressing fatty acid synthesis or æ-9 desaturase than chylomicron remnants from animal fed safflower oil, which is rich in 18:2n-6 (Kohan et al 2011). Whether 16:0 in human milk contributes to higher hepatic fatty acid synthesis, perhaps from the milk medium-chain fatty acids, æ-9 desaturase, or secretion of apo B-containing VLDL from the liver of the breast-fed infant is not known. Such an interpretation is consistent with the known differences in plasma lipoprotein patterns between breast-fed and formula fed infants, the effects of replacing dietary 16:0 with unsaturated fatty acids, and the limited data on triglycerides with 2-palmitate in infants (Nelson and Innis, 1999). Relevant to the 16:0 supply, recent studies have reported that 16:1n-7 functions as a lipokin linking adipose tissue with hepatic metabolism, improving insulin sensitivity and reducing inflammation (Cao, et al., 2008), although evidence for this in humans is limited (Hodson and Karpe, 2013).

Early nutrition is one of the most important factors determining the composition of microbiota that colonize the intestine, and this can have both short- and long- term implications for metabolic and immunologic development (Natua, et al., 2013). Clinical studies have provided some evidence that formula containing triglycerides with 2-palmitate rather than unmodified palm oil may favor higher colonization of the young infant intestine with Lactobacillus and Bifidobacteria (Schmelzle, et al., 2003; Yaron, et al., 2013). Also relevant, dietary 16:0 increased growth and adaptation of the jejeunum and ileum in a rat model of short bowel syndrome, with 75% bowel resection, through decreased cell apoptosis and increased cell proliferation (Sukhotnik, et al., 2008). While the molecular mechanisms are unknown, the roles of 16:0 not only in membrane phospholipids and sphingolipids, but in several palmitoylated proteins offers plausible mechanisms through which inadequate 16:0 could be detrimental by reducing tissue repair and regrowth, and enabling unrestrained inflammation. Palmitoylated proteins include the transmembrane claudins, which are tight junction proteins involved in regulation of paracellular permeability in the intestine (Findley and Koval, 2009), Mucin 2, a major component of mucus and the dominant mucin in the intestine (Wei, et al. 2012), and endothelial nitric-oxide synthase (eNOS) which is involved in endothelial permeability (Wei, et al. 2011). Palmitoylethanolamide (PEA), which is synthesized from phosphatidylethanolamine and palmitic acid, is a lipid mediator best-known for its role in reducing pain and inflammation (Bradshaw, et al., 2009; Lambert, et al., 2002). Information to show that PEA is altered by dietary fatty acids is limited, although evidence that intestinal, but not brain, 16:0 and PEA are altered in animals fed diets containing different types of vegetable oil have been published in animals (Artmann, et al., 2008).

It is well-known that adipose tissue fatty acids in humans and other non-ruminants are readily altered by the amount and type of fat in the diet, although the large adipose tissue mass in adults results in a relatively slow turnover. Infants, on the other hand, accumulate large amounts of adipose tissue during the first few months after birth (Figure 1) and this offers the potential for rapid effects of diet on the infantôs adipose tissue fatty acids. After birth, 16:0 decreases from about 46% to 30-32% of the adipose tissue fatty acids, while 18:2n-6 increases from about 2% to 6-8% fatty acids in breast-fed infants between birth and six to seven months of age (Boersma, 1979; Farquharson, et al., 1993; Table 1) . Classic studies by Widdowson and colleagues are one of the most illustrative demonstrations of the dependence of the type of fatty acids accumulated in infant adipose tissues on the fatty acid in the milk or formula diet (Widdowson, et al., 1975). Their study described the adipose tissue fatty acids of British and Dutch infants at birth, Dutch infants after several weeks feeding with formula containing corn oil (58% 18:2n-6) and British infants who were breast-fed or fed formula which at that time still contained cowsô milk fat. These classic studies showed infants fed formula with corn oil accumulated 18:2n-6 to reach about 26% adipose tissue fatty acids with loss of 16:0, while the breast-fed infants increased 18:2n-6 to about 6% adipose tissue fatty acids while maintaining about 30 % 16:0. Similar data from studies in several countries are available to show adipose tissues of infants fed formula with vegetable oils achieve high amounts of high 18:2n-6 or 18:1n-9, depending on the oils in the products, with lower 16:0 than in breast-fed infants or infants fed formulas with cowsô milk fat (Farquharson, et al, 1963; Hashim, 1983; Sweeny, et al., 1963). Although the growth of adipose tissue that differs markedly in infants fed formulas from the biological norm of the breast-fed

infants has been known for over two decades (Widdowson, 1989), the implications remain unknown.

Summary

Foods and nutrition are major modifiable factors able to impact development, with potentially long-lasting effects secondary to alterations in the growth and development of organ systems, biochemical and molecular pathways. During growth, 16:0 is needed for synthesis of new cell membranes, expansion of adipose tissue, secreted lipids, such as lipoproteins and lung surfactant, specific roles in palmitoylated proteins and signal molecules, and not least as a source of metabolic energy. Before birth, fetal adipose tissue has close to 50% 16:0, much of which is synthesized in fetal tissues. After birth, human milk provides the infant with a diet that at other life stages might be deemed undesirable, with 50% energy from fat and 20% fatty acids, equivalent to about 10% energy from 16:0 in triglycerides with an unusual fatty acid distribution with much of the 16:0 at the *sn*-2 position of the triglycerides. Most of the early emphasis on 16:0 in the diet of young infants focused on its absorption, and plasma cholesterol lowering. However, marked alterations in tissue lipids, particularly in organs such as the adipose and intestine, but not brain occur in infants nourished with low 16:0-highly unsaturated fatty acid-rich oils. Why the human fetus synthesizes and accumulates 16:0, and the human mammary gland invests in specific pathways to provide the infant with 16:0 are important questions for developmental biology, with broad implications for the nutritional care of infants that involve fatty acids that alter sources of metabolic energy, and the growing tissue and plasma lipids. It is

timely to consider that 16:0 is a normal, arguably essential component of human fetal and infant tissue lipids, whereby low levels may be detrimental.

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REFERENCES

Artmann, A., Petersen, G.Hellgren, L.I., Boberg, J., Skonberg, C., Nellemann, C., Hansen, S.H. and Hansen, H.S. (2008). Influence of diary fatty acids on endocannabinoid and *N*-acylethanolamine levels in rat brain, liver and small intestine. *Biochim. Biophys. Acta.* **1781**:200-212.

Baker, G.L. (1969). Human adipose tissue composition and age. *Am. J. Clin. Nutr.* **22**:829-835.

Berghaus, T.M., Demmelmair, H. and Koletzko B. (1998). Fatty acid composition of lipid classes in maternal and cord plasma at birth. *Eur. J. Pediatr.* **157**:763-768.

Boersma, E.R. (1979). Changes in fatty acid composition of body fat before and after birth in Tanzania: an international comparative study. *Br. Med. J.* **31**:850-853.

Bougneres, P.F., Lemmel, C., Ferré, P. and Bier, D.M. (1986). Ketone body transport in the human neonate and infant. *J. Clin. Invest.* **77**:42-48.

Booth, C., Elphick, M.C., Hendrickse, W. and Hull, D. (1981). Investigation of [14C] linoleic acid conversion into [14C] arachidonic acid and placental transfer of linoleic and palmitic acids across the perfused human placenta. *J. Dev. Physiol.* **3**:177-189.

Bradshaw, H.B., Rimmerman, N., Hu, S.S., Burstein, S. and Walker, J.M. (2009) Novel endogenous N-acyl glycines identification and characterization. *Vitam. Horm.* **281**:191-205.

Cani, P.D., Osto, M., Geurts, L. and Everard, A. (2012). Involvement of gut microbiota in the development of low-grade inflammation and type 2 diabetes associated with obesity. *Gut Microbes* **3**:279-288.

Cao, H., Gerhold, K., Mayers, J.R., Wiest, M.M., Watkins, S.M. and Hotamisligil GS. (2008). Identification of a lipokine, a lipid hormone linking adipose tissue to systemic metabolism. *Cell* **134**:933-944.

Dancis, J., Jansen, V., Kayden, H.J., Schneider, H. and Levitz M. (1973). Transfer across perfused human placenta. II. Free fatty acids. *Pediatr. Res.* **7**:192-197.

David, L.A., Maurice, C.F., Carmody, R.N., Gootenberg, D.B., Button, J.E., Wolfe, B.E., Ling, A.V., Devlin, A.S., Varma, Y., Fischbach, M.A., Biddinger, S.B., Dutton, R.J. and Turnbaugh, P.J. (2013). Diet rapidly and reproducibly alters the human gut microbiome. *Nature.* Dec 11 [Epub ahead of print].

Farquharson, J., Cockburn, F., Patrick, W.A., Jamieson, E.C. and Logan, R.W. (1993). Effect of diet on infant subcutaneous tissue fatty acids. *Arch. Dis. Childhood* **69**:589-593.

Filer, L.J. Jr., Mattson, F.H. and Fomon, S.J. (1969). Triglyceride configuration and fat absorption by the human infant. *J. Nutr.* **99**:293-298.

Findley, M.K. and Koval, M. (2009). Regulation and roles for caudin-family tight junction proteins. *IUBMB Life* **61**:431-437.

Fomon, S.J. (1967). Body composition of the male reference infant during the first year of life. *Pediatrics* **40**:863-870.

Fomon, S.J., Haschke, F., Ziegler, E.E. and Nelson, S.E. (1982). Body composition of reference children from birth to age 10 years. *Am. J. Clin. Nutr.* **35**:1169-1175.

Freeman ,C.P., Jack, E.L. and Smith, L.M. (1965). Intramolecular fatty acid distribution in the milk fat triglycerides of several species. *J. Dairy Sci.* **48**:853-858.

Fuch, G.J., Farris, R.P., DeWier, M., Hutchinson, S., Strada, R., and Suskind, R.M. (1994). Effect of dietary fat on cardiovascular risk factors in infancy. *Pediatrics* **93**:756-763.

Garcia, C. and Innis, S. (2013). Structure of the human milk fat globule. *Lipid Technology* **25**:223-226.

Hashim, S.A. (1983). Dietary fats and adipose tissue fatty acid composition. *Preventive Medicine* **12**:854-867.

Hayes, K.C., Pronczuk, A., Wood, R.A. and Guy, D.G. (1992). Modulation of infant formula fat profile alters the low-density lipoprotein/high-density lipoprotein ratio and plasma fatty acid distribution relative to those with breast-feeding. *J. Pediatr.* **120**:S109-116.

Herrera, E. and Amusquivar, E. (2000). Lipid metabolism in the fetus and the newborn. *Diabetes Metab. Res. Rev.* **16**:202-210.

Herrera, E. (2000). Metabolic adaptations in pregnancy and their implications for the availability of substrates to the fetus. *Eur. J. Clin. Nutr.* **54**:S47-51.

Hershfield, M.S. and Nemeth A.M. (1968). Placental transport of free palmitic and linoleic acids in the guinea pig. *J. Lipid Res.* **9**:460-468.

Hodson, L. and Karpe, F. (2013). Is there something special about palmitoleate? *Curr. Opin. Clin. Nutr. Metab. Care.* **16**:225-231.

Innis, S.M. (2003). Perinatal biochemistry and physiology of long-chain polyunsaturated fatty acids. *J. Pediatr.* **143**:S1-8.

Innis, S.M. (2004). Polyunsaturated fatty acids in human milk: an essential role in infant development. *Adv. Exp. Med. Biol.* **554**:27-43.

Innis, S.M. (2011). Dietary triacylglycerol structure and its role in infant nutrition. *Adv. Nutr.* **2**:275-283.

Innis, S.M. (2013). Maternal nutrition, genetics, and human milk lipids. *Current Nutrition Reports* **2**:151-158.

Jensen, R.G. (1999). Lipids in human milk. *Lipids* **34**:1243-1271.

Jensen, C., Buist, N.R. and Wilson, T. (1986). Absorption of individual fatty acids from long chain or medium chain triglycerides in very small infants. *Am. J. Clin. Nutr.* **43**:745-751.

Kohan, A.B., Qing, Y., Cyphert, H.A., Tso, P. and Salati, L.M. (2011). Chylomicron remnants and nonesterified fatty acids differ in their ability to inhibit genes involved in lipogenesis in rats. *J. Nutr.* **141**:171-6.

Kuzawa, C.W. (1998). Adipose tissue in human infancy and childhood: an evolutionary perspective. *Yearbook of Physical Anthropol.* **41**:177-209.

Lager, S. and Powell, T.L. (2012). Regulation of nutrient transport across the placenta. *J. Pregnancy* Epub 2012 Dec 10.

Lambert, D.M., Vandevoorde, S., Jonsson, K.O. and Fowler, C.J. (2002). The palmitoylethanolamide family: a new class of anti-inflammatory agents? *Curr. Med. Chem.* **9**:663-674.

Lefèvre, C.M., Sharp, J.A. and Nicholas, K.R. (2010). Evolution of lactation: ancient origin and extreme adaptations of the lactation system. *Annu. Rev. Genomics Hum. Genet.* **11**:219-238.

Malcom, G.T., Bhattacharyya, A.K., Velez-Duran, M., Guzman, M.A., Oalmann, M.C. and Strong, J.P. (1983). Fatty acid composition of adipose tissue in humans: differences between subcutaneous sites. *Am. J. Clin. Nutr.* **50**:288-291.

Nauta, A.J., Ben Amor, K., Knol, J., Garssen, J and van der Beek, E.M. (2013). Relevance of pre- and postnatal nutrition to development and interplay between the microbiota and metabolic and immune systems. *Am. J. Clin. Nutr.* **98**:586S-593S.

Nelson, C.M. and Innis, S.M. (1999). Plasma lipoprotein fatty acids are altered by the positional distribution of fatty acids in infant formula triacylglycerols and human milk. *Am. J. Clin. Nutr.* **70**:62-69.

Nommsen, L.A., Lovelady, C.A., Heinig, M.J., Lönnerdal, B. and Dewey, K.G. (1991). Determinants of energy, protein, lipid, and lactose concentrations in human milk during the first 12 mo of lactation: the DARLING Study. *Am. J. Clin. Nutr.* **53**:457-465.

Novak, E.M. and Innis, S.M. (2011). Impact of maternal dietary n-3 and n-6 fatty acids on milk medium-chain fatty acids and the implications for neonatal liver metabolism. *Am. J. Physiol. Endocrinol. Metab.* **301**:E807-817.

Pirilä, S., Taskinen, M., Viljakainen, H., Mäkitie, O., Kajosaari, M., Saarinen-Pihkala, U.M. and Turanlahti, M. (2013). Breast-fed infants and their later cardiovascular health: a prospective study from birth to age 32 years. *Br. J. Nutr.* **8**:1-8. [Epub ahead of print].

Schmelzle, H., Wirth, S., Skopnik, H., Radke, M., Knol, J., Böckler, H.M., Brönstrup, A., Wells, J. and Fusch, C. (2003). Randomized double-blind study of the nutritional efficacy and bifidogenicity of a new infant formula containing partially hydrolyzed protein, a high beta-palmitic acid level, and nondigestible oligosaccharides. *J. Pediatr. Gastroenterol. Nutr.* **36**:343-351

Sukhotnik, I., Hayari, L., Bashenko, Y., Chemodanov, E., Mogilner, J., Shamir, R., Bar Yosef. F., Shaoul, R. and Coran, A.G. (2008). Dietary palmitic acid modulates intestinal re-growth after massive small bowel resection in a rat. *Pediatr. Surg. Int.* **24**:1313-1321.

Sweeney, M.J., Ettledorf, J.N., Throop, L.J., Timma, D.L. and Wrenn, E.L. (1963). Diet and fatty acid distribution in subcutaneous fat and in the cholesterol-triglyceride fraction of serum of young infants. *J. Clin. Invest.* **42**:1-9.

Van Biervliet, J.P., Rosseneu, M. and Caster, H. (1986). Influence of dietary factors on the plasma lipoprotein composition and content in neonates. *Eur. J. Pediatr.* **144**:489-493.

Van Biervliet, J.P., Vinaimont, N., Caster, H., Vercaemst, R. and Rosseneu, M. (1977). Lipoprotein patterns in newborns. Influence of nutritional factors. *Pediatr. Res.* **11**:744-748.

Wei, X., Schneider, J.G., Shenouda, S.M., Lee, A., Towler, D.A., Chakravarthy, M.V., Vita, J.A. and Semenkovich, C.F. (2011). De novo lipogenesis maintains vascular homeostasis through endothelial nitric-oxide synthase (eNOS) palmitoylation. *J. Biol. Chem.* **286**:2933-2945.

Wei, X., Yang, Z., Rey, F.E., Ridaura, V.K., Davidson, N.O., Gordon, J.I. and Semenkovich, C.F. (2012). Fatty acid synthase modulates intestinal barrier function through palmitoylation of mucin 2. *Cell Host Microbe* **11**:140-152.

Widdowson, E. (1950). Chemical composition of newly born mammals. *Nature* **166**:626-629.

Widdowson, E.M. and Spray, C.M. (1951). Chemical development in utero. *Arch. Dis Child.* **26**:205-214.

Widdowson, E.M. (1989). Upper limits of total fat and polyunsaturated fatty acids in infant formula. *J.Nutr*. **119**:1814-1817.

Widdowson, E.M., Dauncey, M.J., Gairdner, D.M., Jonxis, J.H. and Pelikan-Filipkova, M. (1975). Body fat of British and Dutch infants. *Br Med J.* **1**:653-655.

Yaron, S., Shachar, D., Abramas, L., Riskin, A., Bader, D., Litmanovitz, I., Bar-Yoseph, F., Cohen, T., Levi, L., Lifshitz, Y., Shamir, R. and Shaoul. R. (2013). Effect of high ɓ-palmitate content in infant formula on the intestinal microbiota of term infants. *J. Pediatr. Gastroenterol. Nutr.* **56**:376-381.

Table 1. Fatty acid composition of adipose tissue from human newborns at birth, with comparison to mothers, breast-fed infants, and adult humans.

Author, year, country, subjects		Adipose tissue fatty acids, % total					Total
		16:0	16:1	18:0	18:1	18:2	
Widdowson, 1975, England:	newborn	48.9	12.6	4.1	29.6	1.0	96.2
Widdowson, 1975, Netherlôd:	newborn	45.8	15.2	3.8	29.0	2.9	96.7
Farquharson, 1993, Scotland:	newborn	46.2	13.6	4.0	28.9	2.2	94.9
Boersma, 1979, Africa:	newborn	48.2	12.7	4.9	27.8	2.3	95.9
Hashim, 1963, USA:	newborn	51.9	9.4	4.1	26.5	0.8	92.7
Sweeney, 1963:	newborn	45.1	15.3	3.0	29.8	2.3	95.5
Sweeny, 1963:	maternal	23.4	6.7	4.4	50.7	10.6	95.8
Malcolm, 1989, USA	adult	21.8	7.2	4.0	48.3	15.4	96.7
Farquharson, 1993, Scotland:	infant	30.4	7.6	5.0	38.1	6.6	87.7
Boersma, 1979, Africa:	infant	32.2	8.4	3.4	26.8	8.1	78.9

Data from Sweeney et al., 1963 is for matched mother ï newborn infant pairs; data from Malcolm et al., 1989 is for adult men and women; data from Farquharson, et al., 1993 and Boersma, 1979 for breast-fed infants is for infants 5-21 weeks of age and 1-7 months of age, respectively. Netherlôd, The Netherlands.

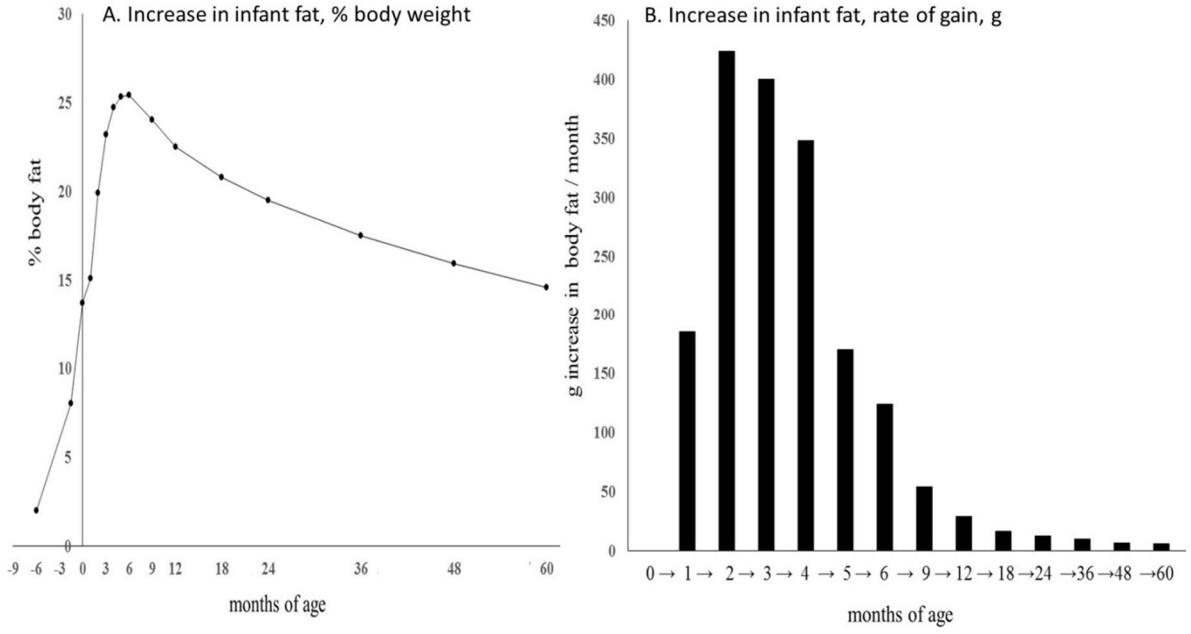

Figure 1. Panel A shows the change in percent body fat from 11 weeks gestation to 5 years of age plotted using reference data for a healthy term boy from Fomon, et al., 1982. Data for the fetus are from File and Weil, 1963. Panel B illustrates the net increase in fat in as g gain/month calculated from the data by Fomon, et al. 1982,