Fatty acid composition of mature human milk of Egyptian and American women1–4

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ABSTRACT Fatty acid composition of mature human milk of rural Egyptian and American women was determined by gas-liquid chromatography. Milk of Egyptian women contained significantly higher percentages of capric, lauric, myristic, linoleic and arachidonic acids, saturated fatty acids (SFA), and polyunsaturated fatty acids (PUFA). Conversely, milk of American women contained higher percentages of stearic and oleic acids, total unsaturated fatty acids, and monounsaturated fatty acids. The PUFA:SFA ratio in Egyptian samples was 0.54 ± 0.18 compared to 0.47 ± 0.22 in American samples. Increased percentages of medium-chain SFA in Egyptian milk suggested increased mammary gland lipids synthesis. Analysis of Egyptian diets indicated high-carbohydrate and low-fat intakes may have resulted in limited availability and incorporation of dietary fatty acids into milk triglycerides. Thus, increased percentages of medium-chain SFA observed in Egyptian milk may reflect mammary gland synthesis in an attempt to maintain lipid concentrations in milk. Am J Clin Nutr 1986;44: 330–335.

KEY WORDS Fatty acids, human milk, lactation, infant nutrition, lipid

Introduction

Lipids are the most variable constituent in human milk (1). Examination of milk-lipid concentration and composition is of interest because milk-lipid provides the major fraction of calories in human milk (1), contains fat-soluble vitamins (2), and provides essential fatty acids necessary for growth and development of the central nervous system (CNS) (3). Although milk-lipid concentrations show considerable variation both during a feeding and diurnally, the fatty acid pattern remains constant (4, 5) and, cross-culturally, milk fatty acids are more remarkable for their similarity than dissimilarity (3). However, the fatty acid composition of human milk responds rapidly and markedly to dietary changes (6, 7).

The purpose of this study was to compare the fatty acid composition of lipid in the milk of rural Egyptian women, some of whom may have been marginally malnourished, with that of well-nourished American women.

Materials and methods

Description of subjects

Egyptian subjects included 22 rural women residing in the village of Kalama. This village is currently being examined to determine the effects of marginal malnutrition on human function. In preliminary studies assessing body size and composition (Galal O, Harrison GG, Jerome NW, and Kirksey A. Unpublished draft report, Phase I research...

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on food intake and function, September 1983), anthropometric measurements, including height (length for infants), weight, midarm circumferences, skinfolds at three sites, and for infants only, head circumference, were utilized to assess those aspects of body size and composition most sensitive to alterations in energy balance. Data collected in Kalama documented a range of normal through moderately malnourished status in relation to international reference data (8, 9). Stunting was the most common pattern of growth alteration observed in Kalama and was typical of that reported in the Egyptian National Nutrition Survey (10). Stunting was defined as a deficit in linear growth for age and appeared to be reflective of a nutritional deficit over an unspecified time during the past. In Kalama, stunting appeared to occur predominantly in the first 2 yr of life. Substantial catch up growth was not demonstrated in the ages surveyed in the preliminary study, in spite of appropriate weight for height values.

Egyptian women were 18 to 45 yr of age (x = 28 ± 6.5 yr). These women had an average of four living children (ranging from one to nine children) and only one woman was a primipara. Weight and height measurements were available for all except one woman and the mean weight was 65.2 ± 12.0 kg and height was 155.6 ± 6.1 cm.

American subjects included 21 nonvegetarian women from a university community participating in a larger study. American women were 25 to 44 yr of age (x = 30 ± 5 yr). These women had an average of two living children (ranging from one to four children) and nine were primiparas. Mean weight of these women was 60.8 ± 7.4 kg and height was 165.5 ± 5.7 cm. All procedures were approved by the University Committee for Use of Human Subjects in Research.

Dietary intakes

Food consumption of all participants in the study was ad libitum. Preliminary analysis of Egyptian diets in a village of Kalama indicated that these women likely consumed 2400 to 2600 kcal/day with ~18 to 20% of the total calories from dietary fat, 70% of total calories from carbohydrate (CHO), and 10 to 12% of total calories from protein. The major source of dietary fats appeared to be cooking fats and oils and cheese since meat and poultry were consumed only once weekly. Cheese (whole and skim) was produced from buffalo milk. Unhydrogenated cottonseed oil comprised ~90% of the fat used in the village. Linoleic acid accounted for 48.9% of the fatty acids in cottonseed oil (11). Hydrogenated vegetable oil, beef lard, cow ghee (butter oil), and buffalo butter, cream, and ghee were also consumed.

The American women consumed calories sufficient for weight maintenance. Examination of food frequency patterns indicated that 20 of the 21 American women consumed 2% fat or skim milk, 18 of 21 used margarine rather than butter, and all used vegetable fats and oils for cooking. Generally four servings of beef and/or pork and two servings of each of poultry and fish were consumed per week.

Milk sampling

Milk samples (~5 ml) obtained from Egyptian and American women were collected manually at a midmorning feeding. Twelve of the Egyptian samples were fore milk and 10 were hind milk. All American samples were fore milk. Due to the preliminary nature of the study, it was not possible to obtain a 24-h expression or milk volume assessment for Egyptian women. The stage of lactation at the time of sampling ranged from 0.5 to 29 mo. Mean stages of lactation were comparable between the Egyptian (7.5 mo) and American women (6.6 mo) as an attempt was made to select matched controls for the Egyptian women. In both groups, 13 samples were collected during the first 6 mo of lactation. Egyptian milk samples were immediately placed on ice until transport to Cairo where they were stored at −30°C. Within a month after collection, samples were transported to the United States for analysis. Milk samples from American women were stored in a similar manner.

Fatty acid analysis

Lipids in 0.2 ml of milk were extracted by the Bligh-Dyer method (12). An appropriate amount of heptadecanoic acid (17:0) was added to the milk prior to the extraction procedure and served as an internal standard. Fatty acids were methylated (13) and analyzed using gas-liquid chromatography. The separation was achieved by using a Model 5700A Hewlett Packard (Palo Alto, CA) gas chromatograph equipped with a flame ionization detector and a Model 3390A Hewlett Packard integrator. A 2 mm × 1829 mm silanized glass column was packed with 5% DEGS-PS on 100/120 Supelcopor (Supelco, Inc, Bellefonte, PA). The injection port and detector were each maintained at 250°C while the samples were run isothermally at an oven temperature of 175°C with a nitrogen carrier gas flow rate of 40 ml/min. Hydrogen and air flow rates were 40 and 160 ml/min, respectively. Identification of the milk fatty acids was based on retention times of known fatty acids. Although arachidic acid (20:0) was included in the standard mix, it was detected in only one American sample. The 5% DEGS-PS column used in this study did not enable the selective resolution of cis and trans isomers.

Statistical analysis

Means of fatty acids in milk of Egyptian and American women were compared by an analysis of variance (14). Pearson correlations (14) were used to determine relationships between various fatty acids.

Results

Data for fatty acid composition of milk samples are shown in Table 1. Milk of Egyptian women contained significantly greater percentages of total fatty acids as capric (10:0), lauric (12:0), myristic (14:0), linoleic (18:2) and arachidonic (20:4) acids, saturated fatty acids (SFA), and polyunsaturated fatty acids (PUFA) compared to milk of American women. Conversely, milk of American women contained significantly higher percentages of fatty acids as stearic (18:0) and oleic (18:1)
acids, unsaturated fatty acids (UFA), and monounsaturated fatty acids (MUFA) than milk of Egyptian women. Percentages of fatty acids as palmitic (16:0), palmitoleic (16:1), and linolenic (18:3) acids were similar for both groups. The PUFA:SFA ratio in Egyptian milk samples was 0.54 ± 0.18 compared to 0.47 ± 0.22 in American milk samples. The differences were not statistically significant (p > 0.05).

Medium-chain SFA (capric, lauric, and myristic) comprised 19.8% of the total fatty acids in Egyptian milk whereas they accounted for only 13.7% of the total fatty acids in the American milk samples. Frequency distributions of lauric and myristic acids indicated that milk of American women generally contained small percentages (< 10%) of these fatty acids whereas milk of Egyptian women often showed significant elevations in percentage of fatty acids as lauric and myristic acids (10-20%) (Figures 1 and 2). Percentage of fatty acids as medium-chain SFA was inversely correlated to the percentage of total fatty acids as oleic acid in Egyptian (r = −0.83, p < 0.001) and American milk (r = −0.66, p < 0.001). A similar relationship was not found for palmitic acid; however, the percentage of total fatty acids as palmitic acid was inversely related to the percentage of total fatty acids as long-chain fatty acids (stearic, oleic, linoleic, linolenic, and arachidonic) in American milk (r = −0.66, p < 0.001) but not in Egyptian milk (r = −0.28, p > 0.05).

Discussion

Fatty acids appear in milk as a result of dietary intake, mobilization from fat depots, and endogenous synthesis by the mammary gland. The human mammary gland is capable of synthesizing SFA, primarily 10 to 14 carbons in length. Fatty acids of chain lengths > 16 carbons are not synthesized in the mammary gland and must be obtained either from the diet or mobilized from fat depots (3, 15, 16). Thus, the fatty acid composition of milk is dependent on the amount and type of dietary fat consumed, the total caloric intake, and, most importantly, the CHO intake of the lactating woman in relation to the total caloric intake (16–19).
Fatty acid composition of mature milk of American women in this study was similar to reports by other researchers (5, 20–23) except that linoleic acid comprised 17.2% of the fatty acids (Table 1). This was slightly higher than that observed by others (5, 20–23) in milk of women consuming ad libitum diets. Increased percentages of fatty acids as linoleic acid resulted in a slightly increased PUFA:SFA ratio of 0.47 observed in the milk of these women. Dietary intakes of linoleic acid have been reported to influence the content of this fatty acid in milk (4, 6, 7, 17, 18). Therefore, it seemed likely that the increase in the percentage of linoleic acid in the milk of American women in the present study was a result of increased dietary intakes of linoleic acid. During the past 25 years, there has been a marked increase in the consumption of vegetable fats relative to animal fats by the American population (20). American women in this study consumed primarily low fat or skim milk, margarine, included poultry and fish as frequently as beef and pork in their weekly diets, and used vegetable fats and oils for cooking. Therefore, fat contributed ~35–45% of the total calories in their diets, and in conjunction with other sources of energy was sufficient for weight maintenance. As a result a high percentage of the total fatty acids in the milk of these women appeared to have been of dietary origin. This was supported by the fact that medium-chain SFA comprised only 13.7% of their milk fatty acids. Palmitic acid can either be synthesized by the mammary gland or obtained from the diet. The percent of palmitic acid in the milk of American women was significantly correlated to the percentage of long-chain fatty acids in their milk. These findings, similar to those of Read et al (19), indicated that the content of palmitic acid in milk probably was related to those fatty acids derived from the diet rather than from synthesis by the mammary gland.

Oleic acid represented the largest percentage of total fatty acid analyzed in both American and Egyptian milk samples; however, levels of oleic acid in Egyptian milk were significantly less than those in American milk (Table 1). Levels of oleic acid in American milk samples were within the range of 33 to 40% of total, reported by others (4, 20, 21, 23). The percentage of oleic acid most likely represented not only oleic acid but also its trans isomer, elaidic acid, since the 5% DEGS-PS column used in this study did not separate cis and trans isomers. American women were consuming ad libitum diets containing significant amounts of hydrogenated fats. Dietary intake of hydrogenated fat has been reported to significantly influence the levels of elaidic acid in human milk (24). Dietary fat intakes of the residents of Kalama suggested that Egyptian women consumed diets containing limited amounts of hydrogenated fat. Unhydrogenated cottonseed oil contributed ~90% of the total fat intake. This difference most likely contributed to the higher levels of oleic acid observed in American milk compared to Egyptian milk.

The fatty acid composition of the milk of Egyptian women was, with few exceptions, markedly different from that of American women in this study. This was not unexpected since the diet of Egyptian women differed considerably from that of American women. In addition, the possibility exists that some of the Egyptian women were marginally malnourished since this condition was observed in the village of Kalama. Due to the preliminary nature of the study, it was only possible to obtain information regarding dietary con-
Linoleic acid accounted for 23.8% of the fatty acids in milk of the Egyptian women. These findings were contrary to those of Soliman et al (25) who reported that linoleic acid comprised 7.65% and 7.97% of the fatty acids in the milk of rural and urban Egyptian women, respectively. UFA comprised 40 to 43% of the fatty acids in the milk of these women. These researchers attributed the lower levels of UFA in the milk of these women to their high intakes of hydrogenated oils as the primary source of dietary fat. Precise caloric intakes of these women were not reported but were said to be generally less than the recommended allowances. Percent of total calories from fat and protein was not included but the percent of total calories from CHO was 68 to 82% and similar to those in the present study. In the present study, Egyptian women consumed diets where unhydrogenated cottonseed oil containing high amounts of linoleic acid, comprised ~90% of the fat. Therefore, increased levels of linoleic acid in the milk of the Egyptian women reported by us can be attributed to dietary intakes of the acid (4, 6, 7, 17, 18) as well as other factors such as caloric intakes.

Preliminary analysis of Egyptian diets indicated that fat supplied 18–20% of the total calories and CHO supplied 70% of the total calories. Also, the fat content of the Egyptian diet appeared to increase as the socioeconomic status of the family increased. Read et al (19) observed that consumption of diets containing high percentages of CHO resulted in increased lauric and myristic acid levels with a concomitant reduction of oleic and stearic acids in the milk of lactating women. Increases in dietary CHO appear to favor increased synthesis of medium-chain SFA by the mammary gland (18). Milk of Egyptian women in this study contained significantly higher percentages of capric, lauric, and myristic acids and significantly lower percentages of oleic and stearic acids compared to American women. In addition, the percentage of oleic acid in the milk of Egyptian women was inversely related to that of the medium-chain SFA, a finding recently observed by Bitman et al (23) in the milk of mothers of both preterm and term infants. Despite the increased percentages of medium-chain SFA, the PUFA:SFA ratio in Egyptian milk remained significantly higher compared to that in milk of American women due to the considerable amounts of linoleic acid in milk of Egyptian women (Table 1).

Assuming that the total fat concentration of milk from Egyptian and American women was similar and within the normal range, the possible nutritional implications of these findings to the infant is of considerable interest. For the American infant, increased percentages of linoleic acid in milk will result in increased intakes of this fatty acid. The American Academy of Pediatrics, Committee on Nutrition (AAP, CON) has stated that linoleic acid comprises 8 to 10% of the fat in human milk (26); however, results of this study and those of others (5, 20, 23) suggest that linoleic acid may comprise up to 14 to 18% of the fat in milk of American women consuming typical Western diets. Although linoleic acid is an essential fatty acid necessary for growth and development of the CNS (3), excess linoleic acid produces excessive peroxidation and increases the vitamin E requirement of infants (26). The AAP, CON has recommended that infants receive 0.3 IU of vitamin E/100 kcal and at least 0.7 IU of vitamin E/g of linoleic acid (26). However, lactating women who increase their dietary intakes of linoleic acid also increase their intake of tocopherols since foods high in linoleic acid may have higher vitamin E content as well (6). Jansson et al (21) found a significant correlation between total tocopherol content and the linoleic acid content of mature human milk of Swedish women and the AAP, CON recommendations were met in 21 of 24 samples. Although further investigation is needed, it seems unlikely that levels of linoleic acid that comprise up to 18% of the fatty acids in milk place the infant at risk. In contrast, linoleic acid comprised nearly one-fourth of the total fatty acids in the milk of Egyptian women. Despite the naturally high vitamin E content of cottonseed oil (11), some vitamin E may be lost due to storage conditions in Egypt, particularly exposure to sunlight and warm temperature. Thus, the milk of Egyptian women may contain proportionally less vitamin E/g linoleic acid compared to that in milk of American women. This possibility warrants further investigation.

Fat in human milk, especially medium-
chain SFA, is well absorbed by the infant. In addition, medium-chain triglycerides, from which medium-chain SFA are derived, are more easily hydrolyzed than long-chain triglycerides. For this reason, the increased percentages of medium-chain SFA in milk of Egyptian women may be beneficial to their infants. However, Crawford et al. (3) observed both increased levels of medium-chain SFA and lower fat content (<2%) in the milk of malnourished women compared to values for well-nourished women. Also, infants of malnourished women have lower milk volume intakes than those of well-nourished women (2). If Egyptian women were marginally malnourished, possible advantages to the infant resulting from high levels of medium-chain SFA in milk could be compromised by decreased intake of milk containing lower amounts of fat and thus lower energy. These could seriously affect the nutritional status of these infants.

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