Fat and Energy Contents of Expressed Human Breast Milk in Prolonged Lactation

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ABSTRACT. ***Objective.*** **To estimate fat and energy contents of human milk during prolonged lactation.**

Methods. **Thirty-four mothers, of term, healthy, growing children, who had been lactating for >1 year (12–39 months) were recruited. Control subjects were 27 mothers, of term infants, who had been lactating for 2 to 6 months. Fat contents of the milk samples were estimated as creamatocrit (CMT) levels. Energy contents of the milk were measured with a bomb calorimeter.**

Results. **The groups did not differ in terms of maternal height and diet, infant birth weight, gestational age, or breastfeeding frequency. They differed significantly in terms of maternal age, maternal weight, and BMI. The mean CMT levels were 7.36 ± 2.65% in the short-duration group and 10.65 ± 5.07% in the long-lactation group. The mean energy contents were 3103.7 ± 863.2 kJ/L in the short-duration group and 3683.2 ± 1032.2 kJ/L in the long-duration group. The mean CMT levels and mean energy contents were correlated significantly with the duration of lactation ($R^2 = 0.22$ and $R^2 = 0.23$, respectively). In multivariate regression analysis, CMT levels (or energy contents) were not influenced by maternal age, diet, BMI, or number of daily feedings but remained significantly influenced by the duration of lactation.**

Conclusions. **Human milk expressed by mothers who have been lactating for >1 year has significantly increased fat and energy contents, compared with milk expressed by women who have been lactating for shorter periods. During prolonged lactation, the fat energy contribution of breast milk to the infant diet might be significant. *Pediatrics* 2005;116:e432–e435. URL: www.pediatrics.org/cgi/doi/10.1542/peds.2005-0313; *breast milk, breastfeeding, lactation, energy intake.***

ABBREVIATIONS. HM, human milk; CMT, creamatocrit.

The optimal duration of breastfeeding is unknown. The American Academy of Pediatrics recommends exclusive breastfeeding for 6 months and a total duration of ≥1 year to obtain the "full benefits of breastfeeding."[1] Among frequently recognized long-term benefits of breastfeeding are reductions in cardiovascular risks in adulthood.[1,2] These reductions were challenged by a retrospective epidemiologic study of men born in 1920 to 1930 in Hertfordshire, England, which suggested that the beneficial effects of breastfeeding on cardiovascular risks existed as long as weaning was performed before 1 year of age; after that time, continued breastfeeding was associated paradoxically with increased cardiovascular risks.[2] Moreover, a study by Leeson et al[3] suggested that prolonged breastfeeding might lead to unwelcome outcomes and might even increase cardiovascular risks in adulthood.

In developed countries, a minority of women continue to lactate for >1 year; in one study from Italy, 17% of mothers were still breastfeeding at 12 months after delivery.[4] The energy contribution of human milk (HM) to the diet of partially breastfed children beyond the first year of life is unknown, because there are no data on the amounts of HM consumed by these children and the nutritional content of HM after prolonged breastfeeding is little known. In particular, the fat and energy contents of HM after prolonged breastfeeding have not been analyzed systematically. We therefore conducted this cross-sectional study of 61 women who had been lactating for periods of 2 to 6 months (short lactation duration) or >1 year (long lactation duration), to estimate the fat and energy contents of HM.

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METHODS

Patients

Thirty-four mothers, of term, healthy, growing children, who had been lactating for >1 year were recruited and compared with 27 mothers, of term infants, who had been lactating for 2 to 6 months. All infants were healthy, were free of congenital malformations, and had been delivered after a normal pregnancy, labor, and delivery. To control for possible diurnal variations,[5,6] for every subject we used for analyses the mean of 2 samples, one collected at 6:00 to 9:00 AM and the other collected at 9:00 PM to midnight. The mothers' diet type (omnivorous versus lacto-ovo-vegetarian) was recorded. Our study was approved by our local institutional review board, and informed consent was obtained from all of the mothers in this study.

Laboratory Methods

Milk was collected at home by mothers through manual expression in mid-breastfeeding, stored at 3°C to 5°C, and analyzed (after homogenization) for fat content within 24 hours. The fat content of the milk samples was measured with the creamatocrit (CMT) method, as described previously.[7–11] Briefly, 75-μL aliquots were placed into 2 glass capillary tubes, which were sealed at one end and centrifuged in a hematocrit centrifuge for 5 minutes at 9000 rpm. The CMT level was recorded to the nearest 0.5 mm and expressed as a percentage of the length of the milk column in the tube. Each reading was performed in duplicate in a blinded manner, by an investigator who was not aware of the origin and time of sampling. The final result was the average of the 2 readings. A random subset of samples (n = 30) was also analyzed directly for energy content with the static bomb calorimetry method, as described by Garza et al,[12] with a Parr 1261 bomb calorimeter (Parr Instruments, Moline, IL); results were expressed as kilojoules per liter of milk.

Statistical Analyses

Results were expressed as mean ± SD or number and percentage. Student's t tests were used to determine the differences between the 2 groups (2–6 months and >1 year) for continuous variables, with χ^2 tests for categorical variables. Linear regression analysis was used to determine the correlation between CMT levels and energy contents. Backward, stepwise, multivariate regression analyses were used to determine the effects of lactation duration, maternal weight or BMI, and maternal age (independent variables) on CMT levels and energy contents (dependent variables).

RESULTS

Demographic and maternal characteristics of the participants in this study are presented in Table 1. Briefly, mothers were healthy and had not suffered from hypertensive disorders of pregnancy or diabetes mellitus. Most were omnivorous (Mediterranean-type diet), except for 2 mothers who were lacto-ovo-vegetarians (1 in each group).

There were 61 mothers, who had been breastfeeding for either 2 to 6 months (short-duration group, n = 27) or 12 to 39 months (long-duration group, n = 34). The 2 groups (short-duration versus long-duration lactation) did not differ in terms of maternal height and diet, infant birth weight, gestational age, or breastfeeding frequency. They differed significantly, however, in terms of maternal age, maternal weight, and BMI (Table 1). In addition, there were significant differences between the 2 groups in terms of CMT and energy content values (Table 1). The energy contents were correlated significantly with the CMT levels (energy [kilojoules per liter] = 2288 + 117 · CMT [percent]; $R^2 = 0.37$, $P < .0001$). Mean CMT levels and mean energy contents were correlated significantly with the duration of lactation ($R^2 = 0.22$, $P < .0001$, and $R^2 = 0.23$, $P < .0001$, respectively) (Fig 1). In multivariate regression analysis, CMT levels (or energy contents) were not influenced by maternal age, diet type, BMI, or number of daily feedings but remained significantly influenced by the duration of lactation.

DISCUSSION

We demonstrated that, in HM expressed by mothers who had lactated for >1 year, the fat and energy contents were increased significantly, compared with HM expressed by women who had lactated for a shorter period (2–6 months). In fact, CMT levels for the long-duration group were at times extraordinarily high (up to 28%), compared with the short-duration group or values in previous publications, which reported ranges of 5% to 17.5%.[13]

These results could be explained theoretically by a direct effect of the lactation duration on CMT levels and energy contents. Alternatively, inherent differences between the 2 groups in the study might also have played a role. Indeed, our study was limited by the fact that it was not a longitudinal one, and many subtle or less-subtle differences between the 2 groups other than the lactation duration might have influenced the CMT levels and energy contents. In our study, maternal diet was not studied systematically from a dietary recall standpoint. However, most women in the study ate a Mediterranean-type diet, and only 1 woman in each group was lacto-ovo-vegetarian, which did not allow us to speculate in any manner on dietary influences. The 2 groups were also similar in terms of maternal height, infant birth weight, gestational age, and breastfeeding frequency. They were, however, significantly different in terms of maternal age (older mothers in the long-duration group), maternal weight, and BMI (both higher in the short-duration group). The presence of older mothers in the long-duration group might indicate significant socioeconomic differences between the 2 groups, which theoretically might have influenced the results. However, in the multivariate regression analysis, maternal age did not influence CMT levels. The same was true for maternal weight and maternal BMI, which also were found not to influence CMT levels. Higher maternal weight and BMI in the short-duration group were probably the result of the more recent pregnancies in this group (women are known to lose most of the weight accumulated during pregnancy during the first year of life of the infant[14]).

Therefore, despite the cross-sectional aspect of our study, we think that differences in the length of lactation per se explained the striking differences in CMT levels and energy contents that we found in this study. The reasons for the differences in energy and fat contents are not found readily in the available literature. Indeed, there are very few data on the nutritional value of breast milk during prolonged (>1-year) lactation. A Medline search using the key words "lactation" or "breast milk" or "human milk" or "breastfeeding" and "duration" and "composition" revealed only 8 articles, of which 7 originated

TABLE 1. Subject Characteristics and CMT and Energy Data

	Short Lactation (2–6 mo) (n = 27)	Long Lactation (>1 y) (n = 34)	P
Maternal age, y	30.7 ± 2.9	34.4 ± 5.1	.004
Maternal weight, kg	66.3 ± 11.8	59.8 ± 8.7	.03
Maternal BMI, kg/m^2	24.5 ± 3.9	22.1 ± 3.0	.01
Breastfeeding frequency, times per d	7.1 ± 1.9	5.9 ± 3.3	NS
CMT, %	7.36 ± 2.65 (2–12)	10.65 ± 5.07 (2–28)	<.0001
Energy content, kJ/L	3103.7 ± 863.2 (1075–4652)	3683.2 ± 1032.2 (682–5887)	<.0001

Data are expressed as mean ± SD (range). NS indicates not significant.

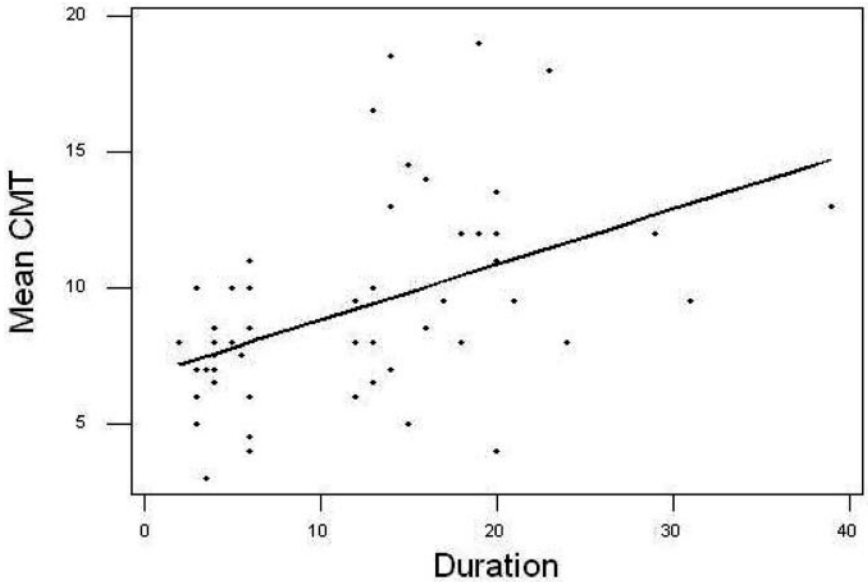

Fig 1. Correlation between mean CMT values (percent) and duration of lactation (months).

from developing countries[15–21] and only 2 dealt with issues of milk composition.[20,21] The largest study on the topic, by Dewey et al,[22] found that "fat . . . concentrations" (in late lactation) "were similar to those in earlier stages of lactation." However, careful examination of the data reported by Dewey et al[22] revealed that the aforementioned statement in their article's abstract was not supported by their actual data, which showed a significant increase in fat content in late lactation, compared with early lactation (6.22 ± 2.87 g/100 mL vs 4.03 ± 2.17 g/100 mL). This increase of ~50% in fat content from early to late lactation is very similar to the ~50% increase in CMT levels that we found in our study. In a longitudinal study by Nommsen et al,[23] there were no changes in lipid or energy contents over time, but that study was limited to the first year of life and did not assess prolonged lactation beyond the first year. Moreover, the contribution of fat content to energy content was very significant in our study. Others found that CMT levels were correlated very well with energy contents of milk.[7,8] Our study confirmed this and was the only study of its type that examined the relationship between CMT and energy contents measured with direct bomb calorimetry, rather than calculated from nutritional equations.[11]

Therefore, it seems theoretically that, during prolonged lactation, the contribution of breast milk to the infant diet might be significant, from an energy intake standpoint. Indeed, a reduction in the volume of milk consumed by a child who is also eating solid foods might well be counterbalanced by the increase in energy concentration. A limitation of this statement is that, similar to the study by Dewey et al,[22] we were not able to measure accurately the volumes consumed at the breast by each infant, for obvious technical reasons. Mothers in our study were not instructed to double-weigh the infant before and after every feeding, because of the cumbersome aspects. Nevertheless, it seemed that some women in the long-duration lactation group were able to reach extraordinarily high CMT values, such as a woman whose expressed breast milk had a CMT level of 28%.

The long-term effects of such high fat intake have not been studied. We did not analyze the fat qualitatively in this study, but it is known that, compared with infant formula, breast milk is much richer in cholesterol and saturated fat.[24] The effects of high intake of saturated fat and cholesterol on infant cholesterol metabolism are consequential. We showed that, in the first few months of life, higher cholesterol intake among breastfed infants led to downregulation of endogenous cholesterol production.[25] Whether continued high saturated fat and cholesterol intake through breastfeeding beyond the first year of life is beneficial is unknown. As quoted earlier, prolonged breastfeeding has been "accused" of possibly inducing long-term endothelial damage and of decreasing arterial distensibility,[2,3,26–28] but many objections have been raised against this accusation.[29–31] The type of fats present in the milk of mothers lactating for >1 year needs to be measured before suggestions of the role in adult heart disease can be mentioned. Also, because of the changes in diets from childhood to adult years, it may not be possible to determine the influence of prolonged breastfeeding on cardiovascular disease.

We must point out that, at the present time, the official policy of the American Academy of Pediatrics is not to put any limit on the duration of lactation.[1] Moreover, a recent review of biological versus cultural aspects of weaning suggested that, from an anthropologic standpoint based on primates studies, "breastfeeding a child for 2.5 to 7 years is normal for our species."[32]

REFERENCES

1. Gartner LM, Morton J, Lawrence RA, et al. Breastfeeding and the use of human milk. *Pediatrics.* 2005;115:496–506

2. Fall CH, Barker DJ, Osmond C, Winter PD, Clark PM, Hales CN. Relation of infant feeding to adult serum cholesterol concentration and death from ischaemic heart disease. *BMJ.* 1992;304:801–805
3. Leeson CP, Kattenhorn M, Deanfield JE, Lucas A. Duration of breast feeding and arterial distensibility in early adult life: population based study. *BMJ.* 2001;322:643–647
4. Bertini G, Perugi S, Dani C, Pezzati M, Tronchin M, Rubaltelli FF. Maternal education and the incidence and duration of breast feeding: a prospective study. *J Pediatr Gastroenterol Nutr.* 2003;37:447–452
5. Clark RM, Ferris AM, Fey M, Brown PB, Hundrieser KE, Jensen RG. Changes in the lipids of human milk from 2 to 16 weeks postpartum. *J Pediatr Gastroenterol Nutr.* 1982;1:311–315
6. Ruel MT, Dewey KG, Martinez C, Flores R, Brown KH. Validation of single daytime samples of human milk to estimate the 24-h concentration of lipids in urban Guatemalan mothers. *Am J Clin Nutr.* 1997;65:439–444
7. Lucas A, Gibbs JA, Lyster RL, Baum JD. Creamatocrit: simple clinical technique for estimating fat concentration and energy value of human milk. *Br Med J.* 1978;1:1018–1020
8. Lemons JA, Schreiner RL, Gresham EL. Simple method for determining the caloric and fat content of human milk. *Pediatrics.* 1980;66:626–628
9. Lonnerdal B, Smith C, Keen CL. Analysis of breast milk: current methodologies and future needs. *J Pediatr Gastroenterol Nutr.* 1984;3:290–295
10. Jensen RG, Clark RM. Methods of lipid analysis. *J Pediatr Gastroenterol Nutr.* 1984;3:296–299
11. Wang CD, Chu PS, Mellen BG, Shenai JP. Creamatocrit and the nutrient composition of human milk. *J Perinatol.* 1999;19:343–346
12. Garza C, Butte NF, Dewey KG. Determination of the energy content of human milk. In: Jensen RG, Neville MC, eds. *Human Lactation, Milk Components and Methodologies*. New York, NY: Plenum Press; 1985:121–125
13. Meier PP, Engstrom JL, Murtaugh MA, Vasan U, Meier WA, Schanler RJ. Mothers' milk feedings in the neonatal intensive care unit: accuracy of the creamatocrit technique. *J Perinatol.* 2002;22:646–649
14. Dewey KG, Heinig MJ, Nommsen LA. Maternal weight-loss patterns during prolonged lactation. *Am J Clin Nutr.* 1993;58:162–166
15. Gopalan C. Studies on lactation in poor Indian communities. *J Trop Pediatr.* 1958;4:87–97
16. Hanafy MM, Morsey MR, Seddick Y, Habib YA, el-Lozy M. Maternal nutrition and lactation performance: a study in urban Alexandria. *J Trop Pediatr Environ Child Health.* 1972;18:187–191
17. Reinhardt MC, Lauber E. Maternal diet, breast feeding and infants' growth: a field study in the Ivory Coast (West Africa). *J Trop Pediatr.* 1981;27:229–236
18. van Steenbergen WM, Mossel DA, Kusin JA, Jansen AA. Lactation performance of mothers with contrasting nutritional status in rural Kenya. *Acta Paediatr Scand.* 1983;72:805–810
19. Prentice AM, Whitehead RG, Roberts SB, Paul AA. Dietary supplementation of lactating Gambian women, I: effect on breast-milk volume and quality. *Hum Nutr Clin Nutr.* 1983;37:53–64
20. Brown KH, Black RE, Becker S, Nahar S, Sawyer J. Consumption of foods and nutrients by weaning in rural Bangladesh. *Am J Clin Nutr.* 1983;37:61–65
21. Boediman D, Ismail D, Iman S, Ismangoen, Ismadi SD. Composition of breast milk beyond one year. *J Trop Pediatr Environ Child Health.* 1979;25:107–110
22. Dewey KG, Finley DA, Lonnerdal B. Breast milk volume and composition during late lactation (7–20 months). *J Pediatr Gastroenterol Nutr.* 1984;3:713–720
23. Nommsen LA, Lovelady CA, Heinig MJ, Lonnerdal B, Dewey KG. 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.* 1991;53:457–465
24. Giovannini M, Agostoni C, Riva E. Fat needs of term infants and fat content of milk formulae. *Acta Paediatr Suppl.* 1994;402:59–62
25. Cruz ML, Wong WW, Mimouni F, et al. Effects of infant nutrition on cholesterol synthesis rates. *Pediatr Res.* 1994;35:135–140
26. Singhal A, Lucas A. Early origins of cardiovascular disease: is there a unifying hypothesis? *Lancet.* 2004;363:1642–1645
27. Singhal A, Cole TJ, Fewtrell M, Lucas A. Breastmilk feeding and lipoprotein profile in adolescents born preterm: follow-up of a prospective randomised study. *Lancet.* 2004;363:1571–1578
28. Drake AJ, Walker BR. The intergenerational effects of fetal programming: non-genomic mechanisms for the inheritance of low birth weight and cardiovascular risk. *J Endocrinol.* 2004;180:1–16
29. Chong YS. Human milk is still best. *BMJ.* 2003;327:683
30. Holt A. Duration of breast feeding and adult arterial distensibility: does this study herald the return of national dried milk? *BMJ.* 2001;323:691
31. Dark PM, Rolli MJ. Duration of breast feeding and adult arterial distensibility: statistical analysis was unclear. *BMJ.* 2001;323:691–693
32. Dettwyler KA. When to wean: biological versus cultural perspectives. *Clin Obstet Gynecol.* 2004;47:712–723

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