Adequacy of energy intake among breast-fed infants in the DARLING study: Relationships to growth velocity, morbidity, and activity levels

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Breast-fed infants grow less rapidly after the first 2 to 3 months of age than current standards. The DARLING study (Davis Area Research on Lactation, Infant Nutrition and Growth) was designed to evaluate whether this pattern should be considered "faltering" or is a normal outcome even under optimal conditions. Data on intake, growth, morbidity, activity, and motor development were collected longitudinally from infants who were breast fed for at least 12 months. Gross energy intake, calculated from 4-day records of milk and food intake at 3, 6, 9, and 12 months, averaged 91.4, 84.1, 86.7, and 91.8 kcal/kg per day, respectively, well below recommended amounts of metabolizable energy. Nevertheless, infants usually left some food unconsumed. Growth velocity was also below current reference data and was weakly correlated with energy intake. There were no significant negative associations between energy intake at any time and incidence, prevalence, or duration of any category of morbidity during the subsequent 3 months. There were no consistent associations between energy intake and activity level, time spent sleeping, or achievement of key developmental milestones. Similarly, infants with slower growth velocity were just as active and were ill no more often in subsequent months than infants who were growing more rapidly. Thus the deviation from current recommendations for energy intake and growth can be considered a normal pattern with no apparent deleterious consequences in our population of breast-fed infants. (J Pediatr 1991;119:538-47)

Energy intake by breast-fed infants has been reported to be considerably below current recommendations.[1-5] Prentice et al.[6] argued that present estimates of energy requirements during infancy are excessive because they are based in part on intake by infants given formulas developed in past decades, which tended to be higher in energy than current infant formulas. They cite evidence of a secular trend toward lower energy intake as feeding practices and composition of formulas have changed. However, intake of breast-fed infants appears low even in comparison with intake of infants fed modern infant formulas.[4,5,7,8] Part of this difference may be attributable to difficulties in assessing energy intake of breast-fed infants; few studies have included measurements of both milk intake and the energy density of milk in each mother-infant pair, and even fewer have made adjustments for systematic bias introduced by the test-weighing method.[9,10]

Resolution of this question requires not only accurate measures of infant energy intake but some consideration of the outcome variables by which adequacy of intake can be

IWL	Insensible water loss

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judged. There is now considerable evidence that breast-fed infants gain weight less rapidly than formula-fed infants after the first 2 to 3 months, but there is no self-evident reason to conclude that these slower growth rates are detrimental or represent suboptimal intake. What is needed, therefore, are alternative indexes of functional outcomes to judge whether a particular pattern of intake or growth is advantageous in a given environment. We present data on several such indexes, specifically morbidity, activity level, and developmental milestones, along with carefully collected data on intake and growth during the first 12 months of life from a cohort of breast-fed infants in the DARLING study (Davis Area Research on Lactation, Infant Nutrition and Growth).

METHODS

Study design. The DARLING study, begun in 1986, was designed to collect prospective data on growth patterns, nutrient intake, morbidity, and activity levels of infants who were breast fed for at least 12 months. Selection criteria for the study were as follows: (1) mothers did not plan to feed more than 120 ml/day of other milk or formula throughout the first 12 months, (2) solid foods would not be introduced before 4 months of age, (3) infants were healthy, were of normal gestational age (38- to 42-week gestation), and weighed 2500 to 5000 gm at birth, and (4) mothers did not have any chronic illness and were not taking any medication on a regular basis. Subjects were recruited primarily through letters to new parents, as indicated by birth listing.

The desired sample size at 12 months was 40. To allow for attrition, we recruited 92 subjects; the numbers of infants who remained in the study at 3, 6, 9 and 12 months were 73, 60, 51, and 46, respectively. The reasons for dropping out before 3 months are described elsewhere.[11] Only one subject dropped out because of perceived insufficient milk production. Those who left the study before 3 months of age were no different from those who remained, in terms of maternal age, educational level, income, ethnic group, weight gain during pregnancy, or infant gender or birth weight, but there was a significant difference in maternal prepregnancy percentage of ideal body weight (113 ± 21% vs 100 ± 14%, respectively; $p < 0.05$). Of the subjects who left the study between 3 and 12 months, 12 did so because their infants weaned themselves before 12 months, nine left because the measurements were inconvenient, and six dropped out for other reasons. Characteristics of the subjects participating at 3 months were listed in a previous report.[11] The majority of mothers were of relatively high socioeconomic status, with an average educational level of 16.2 years. Infant birth weight was also relatively high, averaging 3.69 kg.

Participants were visited in their homes once a month, generally within 3 days of the child's birth date, to weigh and measure infants and their mothers and to record infant feeding patterns. Details of the anthropometric measures were described previously.[11] At 3, 6, 9, 12, 15, and 18 months, measurements of infant intake were completed and samples of milk were collected, as described below. Results presented herein are based on data of infants up to 15 months of age. The protocol for the study was approved by the human subjects review committee at the University of California, Davis, and informed consent was obtained from all mothers.

Energy intake. Every 3 months, mothers were asked to complete a 4-day weighed record of all foods and fluids consumed by the infant. Milk intake was determined by test weighing with a Sartorius 3826 electronic balance (distributed by Brinkmann Instruments, Inc., Westbury, N.Y.) accurate to the nearest gram and programed to average 40 consecutive weights, as described elsewhere.[11]

Because there is a systematic error in test weighing because of insensible water loss during feedings, we corrected milk intake by estimating IWL for each infant. An adjustment factor was determined by measuring IWL directly during both summer and winter months in a subsample of infants. From a total of 342 measurements of 37 infants, an average loss of 0.05 gm/kg per minute was calculated; this loss was relatively constant from 3 to 12 months of age. This factor was then multiplied by each infant's weight (in kilograms) and by the total time of nursing (in minutes) determined for each infant from the 4-day test-weighing record, and the product (total IWL during feedings, in grams) was added to the milk intake measured by test weighing.

Samples of milk representative of a 24-hour period were collected from each mother after the 4-day test-weighing period, with the alternate breast–expression method described previously.[11, 12] Milk was analyzed for protein by a modified Lowry technique,[13] fat by the modified Folch extraction procedure,[14, 15] and lactose by the method described by Dahlquist.[16] Gross energy content was calculated from values for protein, fat, and lactose by using the factors of 5.65, 9.25, and 3.95 kcal/gm, respectively.[17] Gross energy intake from milk was calculated by multiplying the gross energy density of each mother's milk by the corrected milk intake.

Intake of other foods and fluids was measured to the nearest 1 to 2 gm with an electronic balance (Sartorius Gmbh., Göttingen, Germany, or Ohaus Scale Corp., Florham Park, N.J.). All food offered and the amount left unconsumed were measured at each feeding. Detailed information was obtained on the type, brand, and method of preparation of each food, and nutrient composition was determined by using the FOOD PROCESSOR II computer program (ESHA Research, Salem, Ore.) when possible. In the case of food mixtures or family recipes, samples of each food were collected and frozen for analysis of protein content (by the Kjeldahl method) and fat content (by Folch extraction), with carbohydrate content calculated by difference (based on dry weight). Gross energy intake from non-human-milk foods was calculated by using the factors of 9.4 kcal/gm for fat, 5.65 kcal/gm for protein, and 4.15 kcal/gm for carbohydrate.[18]

Morbidity. All mothers were contacted once a week to record the nature and duration of all symptoms of illness of the infant and mother. In addition, any diagnoses made by personal physicians were recorded. Morbidity data were subsequently coded and grouped into four major categories: respiratory illness (coded as an illness if any upper or lower respiratory tract symptoms other than a clear nasal discharge were present), diarrhea (defined as two or more runny stools per day that differed from the child's usual stools), otitis media (diagnosed by a physician, with duration defined on the basis of symptoms of fever or continued discomfort), and "other" (unexplained fevers, vomiting, chickenpox, and other nonrespiratory, presumably viral infections). For the present analysis, the total number of days with any illness, and by each category of illness, was calculated in 3-month intervals: 0 to 3, 3 to 6, 6 to 9, 9 to 12, and 12 to 15 months. Incidence was calculated as the number of episodes per 100 days at risk (i.e., not including days when that illness was already present), but is also presented as the crude rate of episodes in each 3-month interval for convenience.

Assessment of activity and developmental milestones. At 9 (and 18) months of age, infant activity level was assessed in two ways: (1) by having each mother record all time that her infant spent sleeping for 7 consecutive days, and (2) by observation of each infant for 30 minutes when the infant was awake on 3 separate days. The 7-day record was selected to obtain an overall picture of time awake versus time asleep; the second method was chosen to reflect spontaneous activity level while an infant was awake. The latter procedure was adapted from an activity assessment method for preschool children developed by Klesges et al.[19] A trained assistant went to the home to observe the infant at a time when the infant was not ill and was rested and recently fed. The time selected was mutually decided by the mother and the observer on the basis of the child's usual napping and feeding patterns. Mothers were instructed not to hold or constrain the child more than necessary, so that the infant's voluntary level of motion could be observed. After the infant became accustomed to the presence of the observer, recording was begun on a standardized form. During the 30-minute period, activity was observed for 10 seconds and then recorded for 10 seconds alternately for a total of 90 ratings each time. Activity was recorded by six categories of behavior (lying down, sitting supported, sitting unsupported, creeping, crawling, and cruising/walking), each with three levels of movement, for a total of 18 subcategories. A score was calculated on the basis of time spent in each subcategory and averaged during the 3 days of observation. Observers were trained by one of us (J.H.) and practiced until the coefficient of variation of the observer's and trainer's scores for the same subject was less than 3% on three consecutive occasions. Further details of this method and the scoring system utilized are available from the authors on request.

All mothers were asked to complete the Carey Infant Temperament Questionnaire[20] when their infants were 9 months of age. This 95-item questionnaire is designed to rate infants on the basis of nine different categories of temperament. One of the categories is intended to reflect activity level, as perceived by the mother. From the 13 questions in this category, a score from 1 to 4 is calculated, representing low to high activity.

The age at which infants achieved each of six developmental milestones was recorded during interviews with mothers at the monthly visits. The milestones chosen were those which reflected major changes in behavior and were least subject to subjective impressions of when the child first began the new behavior. These included the age (in weeks) at which the child started creeping, sitting unsupported, crawling, "pulling to stand" at tables or other surfaces, "cruising" (i.e., walking while holding onto a table or other surface), and walking independently.

Data analysis. Data were analyzed with the Statistical Package for the Social Sciences.[21] Energy intake data for infants whose mothers had difficulty with either the test-weighing record or the milk sample collection procedure were excluded from analysis (n = 2, 4, 5, and 6 at 3, 6, 9, and 12 months, respectively). Some of the mothers did not consistently record container weight when completing the 4-day record of nonmilk foods offered; in these cases the net amount of food consumed could be calculated, but not the amount left unconsumed. Therefore the sample sizes for this variable are somewhat lower than for other variables.

Because birth weight and length values were reported on the basis of measurements taken at the hospital, rather than by our team, growth velocity variables reported herein were generally calculated beginning at 1 month of age, rather than at birth. Rates of weight and length gain were determined by dividing the difference between measurements at different time points by the actual time between measurements. No interpolated values were used.

Characteristics of subjects who left the study at various time points after 3 months were compared with those of subjects remaining in the study. There were no significant differences in any of the maternal characteristics. Infants of mothers who dropped out before 12 months (n = 27) had significantly lower weight-for-length values at 2 months (z

Table I. Intake of human milk and total energy

	At 3 mo of age			At 6 mo of age			At 9 mo of age			At 12 mo of age		
	Total	Male infants	Female infants	Total	Male infants	Female infants	Total	Male infants	Female infants	Total	Male infants	Female infants
Human milk intake	812	856	775*	769	814	733	646	687	605	448	499	402
(gm/day)	(133)	(129)	(125)	(171)	(183)	(155)	(217)	(233)	(197)	(251)	(270)	(228)
n	73	34	39	60	27	33	50	25	25	42	20	22
Milk energy density	0.70	0.70	0.70	0.72	0.72	0.72	0.74	0.75	0.74	0.74	0.75	0.73
(kcal/gm)	(0.08)	(0.08)	(0.08)	(0.10)	(0.11)	(0.09)	(0.10)	(0.10)	(0.11)	(0.15)	(0.12)	(0.17)
n	71	33	38	56	26	30	46	22	24	40	19	21
Energy intake (n)	71	33	38	56	26	30	46	22	24	40	19	21
In kcal/day												
All sources	569	605	537*	642	683	606*	738	819	651*	844	888	803
	(86)	(76)	(83)	(103)	(87)	(105)	(164)	(161)	(118)	(158)	(149)	(159)
Human milk	569	605	537*	549	570	530	466	498	432	322	366	282
	(86)	(76)	(83)	(120)	(114)	(125)	(152)	(153)	(146)	(181)	(193)	(163)
Other	0	0	0	93	113	76	272	321	219	522	522	522
				(105)	(131)	(73)	(190)	(214)	(146)	(222)	(208)	(239)
In kcal/kg/day												
All sources	91.4	92.1	90.8	84.1	85.0	83.3	86.7	92.0	80.9*	91.8	91.6	92.0
	(11.7)	(10.2)	(13.0)	(13.3)	(11.9)	(14.5)	(18.9)	(19.9)	(16.2)	(18.2)	(18.9)	(18.1)
Human milk	91.4	92.1	90.8	71.6	70.3	72.8	54.3	55.1	53.4	34.7	37.0	32.7
	(11.7)	(10.2)	(13.0)	(15.2)	(11.9)	(17.6)	(17.3)	(15.5)	(19.5)	(19.9)	(19.6)	(20.4)
Other	0	0	0	12.4	14.7	10.5	32.3	36.8	27.4	57.0	54.6	59.3
				(14.3)	(17.6)	(10.7)	(23.6)	(26.4)	(19.5)	(25.9)	(25.8)	(26.5)

Values are expressed as means, with standard deviations in parentheses.
*Difference between male and female subjects is significant at $p < 0.05$.

scores: 0.03 ± 0.84 vs 0.40 ± 0.61; $p < 0.05$), but not at any subsequent time point; there were no significant differences in growth velocity, energy intake at 3, 6, or 9 months, or morbidity between dropouts and those who remained in the study through 12 months.

Several methods were used to determine whether the outcome measures of morbidity, activity, and achievement of developmental milestones showed any relationship to prior energy intake or growth. First, scatterplots were examined to determine whether any trends were evident and, if so, whether the relationships were linear or nonlinear. Second, the distribution of each outcome variable was examined to determine whether the assumption of normality was valid. The variables of activity, developmental milestones, and growth velocity were normally distributed, but many of the morbidity variables were highly skewed. Natural log transformations were effective in normalizing the distributions of the illness duration variables, but for some incidence and prevalence variables, no transformation yielded a normal distribution. In these latter cases, dichotomous outcome variables (e.g., whether or not an infant was ill during the interval in question) were created. Third, at each time point, infants were divided into groups on the basis of low versus higher energy intake (divided at the median). Both parametric (Student t tests) and nonparametric (Mann-Whitney U tests; chi-square tests) methods were used to compare subsequent growth velocity, morbidity, and activity of groups. Finally, depending on whether the outcome variable was continuous or dichotomous, multiple linear regression analysis or logistic regression, respectively, was used to examine associations between energy intake or growth and the outcome variables. Potentially confounding variables were included in all analyses as appropriate. For the morbidity comparisons, these included season, time spent in day care, number of siblings, infant gender, and weight or length at the beginning of the interval. Because only two mothers smoked, this variable was not included in the analysis. There were no significant associations between the activity or developmental milestone variables and time spent in day care or number of siblings; therefore only initial weight or length, infant gender, and season were included as covariables in the analyses relating energy intake or growth velocity to these outcomes. In the linear regression analyses, log transformations were used when necessary and analysis of the residuals was completed to check for normality and heteroscedasticity. Because of the large number of analyses performed, some of the significant associations observed would be due to chance (e.g., for morbidity outcomes there were generally 15 variables [total plus four categories of illness, each expressed as incidence, prevalence, and duration] considered with six main effects [kilocalories per day, kilocalories per kilogram per day,

Table II. Morbidity from birth to 15 months of age

	Interval (age)				
	At 0-3 mo	At 3-6 mo	At 6-9 mo	At 9-12 mo	At 12-15 mo
No. of infants	72	66	58	47	45
Total days of observation	6229	5473	5021	4031	3756
Total morbidity					
Incidence per 100 days	1.2	3.2	4.2	4.3	4.5
(SD)	(1.3)	(2.7)	(3.7)	(3.5)	(2.7)
Episodes per child	0.9	2.0	2.6	2.7	3.1
(SD)	(0.9)	(1.4)	(1.6)	(1.8)	(1.5)
Total No. of days	7.2	16.7	21.7	18.7	19.1
(SD)	(8.5)	(14.2)	(14.3)	(13.6)	(10.2)
Respiratory infection					
Incidence per 100 days	1.0	2.4	2.6	2.2	12.3
(SD)	(1.1)	(2.0)	(2.2)	(2.0)	(1.7)
Episodes per child	0.8	1.6	1.7	1.5	1.7
(SD)	(0.8)	(1.1)	(1.1)	(1.1)	(1.1)
Total No. of days	6.3	15.9	18.2	14.0	13.5
(SD)	(8.4)	(14.4)	(13.6)	(13.0)	(9.4)
Diarrhea					
Incidence per 100 days	0.05	0.09	0.20	0.31	0.61
(SD)	(0.24)	(0.30)	(0.44)	(0.60)	(0.97)
Episodes per child	0.04	0.08	0.17	0.26	0.49
(SD)	(0.20)	(0.27)	(0.41)	(0.57)	(0.70)
Total No. of days	0.3	0.2	0.8	1.9	3.0
(SD)	(1.5)	(0.7)	(2.1)	(5.0)	(6.3)
Otitis					
Incidence per 100 days	0.05	0.37	0.53	0.62	0.37
(SD)	(0.24)	(0.62)	(0.93)	(1.08)	(0.87)
Episodes per child	0.04	0.23	0.43	0.49	0.29
(SD)	(0.20)	(0.52)	(0.82)	(0.82)	(0.59)
Total No. of days	0.3	1.2	2.5	2.9	2.1
(SD)	(1.3)	(2.7)	(4.6)	(5.8)	(5.2)
Other					
Incidence per 100 days	0.06	0.18	0.33	0.62	0.71
(SD)	(0.26)	(0.47)	(0.73)	(0.87)	(0.71)
Episodes per child	0.06	0.15	0.28	0.53	0.62
(SD)	(0.23)	(0.40)	(0.31)	(0.67)	(0.61)
Total No. of days	0.2	0.7	1.2	2.0	2.3
(SD)	(1.0)	(2.0)	(2.7)	(3.1)	(3.0)

achieved weight and length, and gain in weight and length] at each of four time points, for a total of approximately 360 analyses).

RESULTS

Milk intake, milk energy density, and total energy intake of all infants at 3, 6, 9, and 12 months of age are shown in Table I. Milk intake declined gradually from 812 gm/day at 3 months to 448 gm/day at 12 months. The percentage of total energy intake provided by human milk was 100 ± 0%, 86 ± 15%, 65 ± 22%, and 39 ± 23% at 3, 6, 9, and 12 months, respectively. Infants were fed solid foods two or three times per day beginning at 4 to 7 months (mean 5.3 ± 1.1 months). At 6 months of age these foods generally consisted of infant cereals, fruits, and vegetables. By 9 months, 52% of the infants also received meats (including fish and poultry), 63% received dairy products (other than milk or formula), and 83% received other grain products; at 12 months of age these proportions were 81%, 83%, and 93% for meat, dairy, and grain products, respectively.

Total (gross) energy intake from all sources increased from 569 kcal/day at 3 months to 844 kcal/day at 12 months of age; intake per kilogram of body weight averaged 91.4, 84.1, 86.7, and 91.8 kcal/kg per day at 3, 6, 9, and 12 months, respectively. These values are less than the current United States Recommended Daily Allowances[22] of 108 kcal/kg per day at 1 to 6 months and 98 kcal/kg per day at 6 to 12 months, despite the fact that the recommended values are for metabolizable energy and our data are for gross energy. Nevertheless, infants generally did not con-

Table III. Growth velocity, morbidity, and activity level by energy intake

	Energy intake (per kg)*	
	Low	Higher
At 3 mo		
n	34	31
Weight gain 3-6 mo (gm/mo)	460 ± 138†	501 ± 137
Length gain 3-6 mo (cm/mo)	1.87 ± 0.35	1.91 ± 0.43
Total morbidity 3-6 mo		
Incidence per 100 days	3.1 ± 2.6	3.3 ± 2.7
No. of episodes	2.0 ± 1.4	2.1 ± 1.4
Total No. of days	15.7 ± 14.0	18.4 ± 14.3
At 6 mo		
n	27	25
Weight gain 6-9 mo (gm/mo)	283 ± 106	303 ± 113
Length gain 6-9 mo (cm/mo)	1.29 ± 0.36	1.36 ± 0.36
Total morbidity 6-9 mo		
Incidence per 100 days	4.3 ± 4.9	3.8 ± 2.1
No. of episodes	2.5 ± 2.0	2.6 ± 1.3
Total No. of days	19.1 ± 14.2	20.1 ± 10.4
At 9 mo		
n	22	23
Weight gain 9-12 mo (gm/mo)	224 ± 90	254 ± 130
Length gain 9-12 mo (cm/mo)	1.36 ± 0.32	1.29 ± 0.31
Total morbidity 9-12 mo		
Incidence per 100 days	4.0 ± 3.1	4.7 ± 4.0
No. of episodes	2.6 ± 1.8	2.8 ± 1.7
Total No. of days	18.0 ± 11.8	19.3 ± 15.7
Activity score, 9 mo‡	391 ± 23	379 ± 3.0
Activity rating (temperament), 9 mo§	2.8 ± 1.0	2.1 ± 0.8
Time spent sleeping, 9 mo (hr)‖	12.1 ± 1.0	12.1 ± 1.2
At 12 mo		
n	17	21
Weight gain 12-15 mo	244 ± 122	229 ± 106
Length gain 12-15 mo (cm/mo)	1.12 ± 0.29	1.23 ± 0.37
Total morbidity 12-15 mo		
Incidence per 100 days	4.8 ± 3.8	4.0 ± 1.9
No. of episodes	3.1 ± 1.9	2.8 ± 1.2
Total No. of days	18.2 ± 11.8	18.5 ± 9.0

*Groups of low versus higher intake divided at median: 3 months (<91.2 vs ≥91.2 kcal/kg); 6 months (<84.3 vs ≥84.3 kcal/kg); 9 months (<84.7 vs ≥84.7 kcal/kg); 12 months (<92.2 vs ≥92.2 kcal/kg).
†Values are expressed as mean ± SD.
‡n = 21 versus 22.
§n = 19 versus 17.
‖n = 19 versus 19.

sume all of the food available to them: the percentage of nonmilk foods left unconsumed at 6, 9, and 12 months of age was 23 ± 15%, 33 ± 21%, and 25 ± 15%, respectively.

Male and female infants differed significantly in milk intake at 3 months of age and in total energy intake at 3, 6, and 9 months, primarily because of differences in body weight. However, energy intake per kilogram of body weight was significantly lower among female than among male infants at 9 months: 81 versus 92 kcal/kg per day. There was "tracking" in total energy intake throughout the first year of life: rank-order correlations of energy intake at different time points were statistically significant ($p < 0.05$) for all comparisons, with coefficients ranging from a high of 0.62 for the correlation between 3 and 6 months to a low of 0.26 for the correlation between 3 and 12 months.

As described elsewhere,[23] growth patterns of these breast-fed infants differed from current reference data. These differences were most evident when presented as growth velocity.[24] The percentage of infants whose data fell below the fifth percentile of current reference data for growth velocity[25] ranged from 8% at birth to 3 months of age to 57% at 9 to 12 months for weight gain, and from 12% at birth to 3 months of age to 54% at 6 to 9 months for length gain.

The mean age at which the infants achieved each of the

Table IV. Percentage of nonmilk foods left unconsumed and nursing frequency by energy intake in previous quarter

	Energy intake (kcal/kg)	
	Low	Higher
At 6 mo		
Food unconsumed (%)*	18 ± 13†	28 ± 16
(n)	(11)	(11)
Nursing frequency	6.7 ± 1.9	6.9 ± 1.8
(n)	(28)	(28)
At 9 mo		
Food unconsumed (%)	33 ± 21	29 ± 21
(n)	(17)	(17)
Nursing frequency	6.2 ± 1.8	6.0 ± 2.1
(n)	(23)	(23)
At 12 mo		
Food unconsumed (%)	25 ± 18	24 ± 13
(n)	(10)	(15)
Nursing frequency	4.9 ± 2.7	4.5 ± 2.1
(n)	(17)	(23)

*Not all infants received solid food at this age.
†Values are expressed as mean ± SD.

developmental milestones was 23.0 ± 7.9 weeks for creeping, 32.3 ± 6.9 weeks for crawling, 32.9 ± 5.8 weeks for pulling to stand, 36.4 ± 7.0 weeks for cruising, and 47.5 ± 5.8 weeks for walking.

Morbidity data for the first 15 months of life are shown in Table II. There were few significant differences by gender except at 12 to 15 months, when the total morbidity rate was higher among female infants. Infants were rarely ill during the first 3 months. Most of the illnesses were respiratory infections: the percentage of episodes categorized as respiratory declined from 89% at birth to 3 months of age, to 56% at 9 to 12 months; the percentage categorized as diarrheal illness increased from 4% to 16% and as otitis from 4% to 18% during the first 12 months. Among those infants with complete data throughout the first year of life (n = 44), the total number of days of illness averaged 63 ± 32 (17% of 365 days), and the total number of episodes averaged 8.2 ± 3.8, of which 5.4 ± 2.7 were respiratory illnesses, 0.6 ± 0.6 were diarrhea, 1.2 ± 1.4 were otitis, and 1.0 ± 1.3 were other illnesses.

Comparisons of subsequent growth velocity, morbidity, and activity, including time spent sleeping, between groups divided at the median for energy intake per kilogram at each time point are shown in Table III; the results were nearly identical when categorized by absolute energy intake. Regression analysis was also performed with energy intake as a continuous variable in each case. There were no significant differences in growth velocity between groups, although there were significant correlations between energy intake at 3 months and weight gain from 3 to 6 mo ($r = 0.30$; n = 60; $p < 0.01$), between energy intake per kilogram at 9 months and weight gain from 9 to 12 months ($r = 0.33$; n = 43; $p > 0.05$), and between energy intake at 12 months and length gain from 12 to 15 months ($r = 0.36$; n = 37; $p < 0.05$).

None of the morbidity variables was significantly associated with energy intake or intake per kilogram at any time point, and there were no differences between groups divided at the median, except that infants with lower energy intakes at 9 months had a lower incidence of ear infections from 9 to 12 months (chi-square statistic from logistic regression = 16.1; $p < 0.05$). There were no negative associations between energy intake and any category of morbidity, whether expressed as incidence, prevalence (number of days), or duration.

Activity score, activity rating from the Infant Temperament Questionnaire, and time spent sleeping at 9 months also did not differ between groups and were not associated with energy intake in regression analyses controlling for infant gender and season. Neither absolute energy intake nor energy intake per kilogram at any time point was significantly correlated with the age at which any of the developmental milestones was achieved. When divided at the median for energy intake per kilogram at 6 months of age, infants with higher intake began to "pull up to stand" at an earlier age than those with lower intake (31.3 ± 6.1 vs 35.2 ± 4.9 weeks; $p < 0.05$), but there was no difference in the age at which they first walked (47.4 ± 6.0 vs 48.0 ± 5.9 weeks). Likewise, infants with higher intake at 9 months were no different from those with lower intake in the age when they first walked (48.6 ± 6.8 vs 47.3 ± 5.0 weeks).

Infants with lower energy intake left behind just as much food as infants with higher energy intake: Table IV illustrates that the percentage of nonmilk foods offered but not consumed was not significantly different between groups at 6, 9, or 12 months. Likewise, nursing frequency was similar between groups at these time points.

Associations of subsequent morbidity, activity level, and time spent sleeping with growth velocity in the preceding quarter were analyzed by regression analysis. Significant *positive* associations were found between weight gain from 9 to 12 months of age and both the total number of days of illness and the number of days of respiratory illness from 12 to 15 months ($r = 0.34$ and 0.36, respectively; n = 43; $p < 0.05$). These associations remained significant when infant gender, season, time spent in day care, number of siblings, and initial weight were considered. There were no other significant relationships between growth velocity and morbidity, activity level, or time spent sleeping, nor between achieved weight or length (expressed either in absolute units or as z scores) at 3, 6, 9, or 12 months and any of these outcomes in the subsequent quarter.

With respect to the developmental milestones, the variables considered in the analysis included not only growth velocity during each quarter but also weight and length gain during longer intervals (i.e., 1 to 6, 6 to 12, 1 to 12, and 3 to 12 months). In the first 3 months, infants with higher weight gains began creeping later than those with lower weight gains ($r = 0.27$; n = 63; $p < 0.05$), even when birth weight was considered. In contrast, infants with higher weight gains from 6 to 9 months began "pulling to stand" and "cruising" earlier than those with lower weight gains ($r = -0.35$ [n = 50] and $r = -0.28$ [n = 49], respectively; $p < 0.05$), but the correlation with age at which the infant began to walk was not significant. Length gain from 6 to 12 months, although not weight gain, was associated with walking at an earlier age ($r = -0.36$; n = 39; $p < 0.05$), but the reason for this relationship appeared to be that the infants with more rapid length gain during this interval were developmentally more advanced even before 6 months of age: when the age at which the infant began creeping (which generally occurred before 6 months) was entered into the regression as an indicator of early motor skills, the association between length gain from 6 to 12 months and age when the infant began walking was no longer significant. There were no other significant correlations between weight or length gain during any of the intervals and the developmental milestones. Achieved weight or length, whether expressed in absolute units or as *z* scores, was not significantly associated with the age when the developmental milestones were achieved, except that infants who were heavier at 5 months of age began creeping later than those who weighed less ($r = 0.31$; n = 57; $p = 0.01$).

Infants with slower growth velocities left behind just as much food at 6 and 9 months of age as infants with more rapid weight or length gain during the previous 3 months. Infants with slower weight gain from 9 to 12 months left behind more food at 12 months than those with more rapid gain ($r = 0.47$; $p < 0.05$). There was no association between growth velocity and nursing frequency at any time point.

DISCUSSION

The results of this study confirm the observation that energy intakes of infants exclusively fed human milk are considerably less than current recommendations,[22] even with a rigorous method that includes adjustment for ILW during test weighing and calculations of milk energy density based on a 24-hour sampling procedure. This pattern continues throughout the first year of life, well after the introduction of complementary foods at 4 to 6 months of age. These subjects were not a random or representative sample of breast-feeding mothers and their infants: all the mothers intended to breast-feed for at least 12 months, which is relatively uncommon in the United States.[26] In fact, human milk represented a considerable proportion (39%) of total energy intake at 12 months. Furthermore, the average educational level of mothers was high (16.2 years), the majority were of relatively high socioeconomic status, and all of them reported that they breast fed on demand. We previously reported that maternal nutritional status was not a limiting factor to milk production in this group.[11] Thus we consider the patterns exhibited by these infants to represent optimal environmental conditions for breast-feeding. With this in mind, the discrepancy between current recommendations and actual energy intakes by breast-fed infants deserves attention.

If metabolizable energy averages 92% of gross energy intake,[27] the estimated mean metabolizable energy intakes by infants in this study were 84, 77, 80, and 84 kcal/kg per day, or 78%, 79%, 82%, and 86% of the recommended allowances at 3, 6, 9, and 12 months of age, respectively. Although few data are available on infant energy metabolism, Butte et al.[7] found that total daily energy expenditure of exclusively breast-fed infants at 4 months of age, measured by the double-labeled water method, averages 64 kcal/kg per day, not including the energy used for growth. Among infants in the DARLING study, average weight gain from 2 to 4 months was 21 gm/day. With a value of 5.0 kcal/gm for the energy content of tissues deposited during growth,[28] this would represent approximately 105 kcal/day, or 17 kcal/kg based on the average weight of 6.2 kg among DARLING study infants at 3 months of age. The sum of estimated total daily energy expenditure and energy for growth would thus be 81 kcal/kg per day, which is close to the estimated metabolizable energy intake of 84 kcal/kg per day among DARLING study infants at 3 months.

Growth patterns of infants in this study also differed from current reference data, as described previously,[23] especially when expressed as growth velocity. The high birth weight of infants in this study could explain part of the relatively slow growth velocities observed, but this effect would likely be evident only in the first 6 months. Current growth charts are based on infants who were predominantly bottle fed, many of whom were given solid foods well before 4 months of age. Several investigators have suggested that these reference data are inappropriate for judging the adequacy of growth of breast-fed infants.[29-31]

Despite the relatively low energy intakes and growth velocities of infants in this study, there was no evidence that these patterns were associated with any adverse outcomes. Preliminary evidence from a parallel study of 41 formula-fed infants that we are conducting indicates a lower incidence of diarrhea and shorter duration of respiratory illness in the breast-fed cohort than in the formula-fed infants, even though energy intakes and growth velocities in the formula-fed group were greater.[32] The incidence of illness in our breast-fed cohort was similar to or lower than rates reported for the first year of life in other studies of U.S. infants when comparable prospective morbidity surveillance methods were used.[33-38] Two prospective studies of gastrointestinal illness and diarrhea, one from New Zealand[38] and the other from New Mexico,[39] calculated incidence rates separately for breast-fed and non-breast-fed infants. Rates of diarrhea in our cohort were considerably lower than those reported in these two studies. Other studies contrasting morbidity of infants fed human milk and those fed formula (e.g., Rubin et al.[40] and Howie et al.[41]) did not use methods that allow direct comparison of morbidity rates with our data.

Within the breast-fed cohort, there were no significant negative associations between energy intake and subsequent morbidity during any of the intervals in the first 15 months of life. Similarly, there was no evidence that infants with slower weight or length gain were at any greater risk of morbidity during the subsequent 3-month interval, regardless of how morbidity was expressed (i.e., as incidence, prevalence, or duration within any of the categories of illness examined). Achieved weight and length at 3, 6, 9, or 12 months were also unrelated to subsequent morbidity.

If energy intake were limiting, one would expect infant activity to be reduced, and yet there were no indications that infants with lower energy intakes were any less active or slept any longer than infants with higher energy intakes. Likewise, infants with slower growth velocity during months 6 to 9 were no different in activity score or time spent sleeping from those with higher growth velocities, nor was achieved weight or length at 9 months significantly associated with these outcomes. Data from our formula-fed comparison group indicate that they actually spent more time sleeping than the human milk–fed cohort.[32] Activity scores did not differ between the two groups. However, the activity score that we developed has not yet been compared against other measures based on time-motion studies. It may be more indicative of motor development than of total energy expenditure.

The mean ages at which the breast-fed infants achieved each of the key developmental milestones were similar to or earlier than the 50th percentile values listed in the Denver Developmental Screening Test norms.[42] There were no significant correlations between the age at which any of the milestones we evaluated were achieved and either energy intake or attained weight or length. Those who were already more advanced developmentally both walked earlier and had a more rapid length gain during the second 6 months of life. This explanation is consistent with the observation that motor activities involving muscle contraction and weight bearing are important determinants of linear growth.[43]

We showed previously that low milk intakes among breast-fed infants in this population are a result of infant self-regulation of intake rather than inadequate maternal milk production.[11, 44] Data presented herein further support the idea that low energy intakes are attributable to self-regulation, because infants consistently left unconsumed about one fourth of all nonmilk foods offered to them. Furthermore, infants with energy intakes less than the median or with slower growth velocity left behind just as much food as infants with higher intakes or growth velocity. It is difficult to judge the significance of our measure of the amount of food left unconsumed, because it depends on the total amount of food offered. Parents may vary in their ability to estimate their child's needs and in the timing of such meals. Nevertheless, the results suggest that breast-fed infants reject a portion of total calories made available to them. Other investigators have demonstrated that breast-fed infants introduced to solid foods tend to reduce their intake of milk.[3]

The results of this study indicate that the deviation from current recommendations for energy intake and growth can be considered a normal pattern with no apparent deleterious consequences in our population of breast-fed infants. Further research is necessary to establish guidelines for identifying truly inadequate intakes and growth faltering among breast-fed infants in different environments. Meanwhile, the patterns exhibited by infants in the DARLING study may be useful as a reference against which to compare measurements of breast-fed infants in populations subject to less than optimal conditions.

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