Breastfeeding Attenuates Reductions in Energy Intake Induced by a Mild Immunologic Stimulus Represented by DPTH Immunization: Possible Roles of Interleukin-1β, Tumor Necrosis Factor-α and Leptin[1]

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ABSTRACT An attenuated severity of infections is among the well-documented benefits of breast-feeding. The degree to which this attenuated severity extends to the amelioration of anorexia is understood incompletely, and possible underlying mechanisms have received limited evaluation. This study was designed to test whether breast-feeding attenuates reductions in energy intake associated with a mild immunologic stimulus and to assess poststimulus relationships among putative reductions in energy intake and serum interleukin (IL)-1β, tumor necrosis factor (TNF)-α and leptin concentrations. A quasi-experimental, hospital-based study was conducted in 23 healthy fully breast- (BF) and formula-fed (FF) infants who received the quadruple diphtheria, pertussis, tetanus and hemophilus influenza (DPTH) immunization as an immunologic challenge. Only FF infants had decreased energy intakes (12 $\pm$ 2%, $P = 0.001$) after immunization. Leptin concentrations increased after immunization only in FF infants (30 $\pm$ 7%, $P = 0.03$). Correlations between postimmunization increases in IL-β and reductions in energy intake were of borderline significance ($r = -0.56$, $P = 0.08$). These findings support the view that breast-feeding protects against anorectic responses to mild immunologic stimuli. Increases in leptin are associated with reductions in energy consumption in the postimmunization period in FF infants and postimmunization changes in IL-1β concentrations likely are related to reductions in energy intake in response to immunologic stimuli. J. Nutr. 132: 1293–1298, 2002.

KEY WORDS: • *breast-feeding* • *infants* • *interleukin-1β* • *tumor necrosis factor-α* • *leptin* • *infections* • *anorexia*

Infants receive numerous benefits from breast-feeding. Human milk provides nutrients of high biological value and numerous bioactive factors that may prevent infections and/or attenuate their severity when they occur (1,2). Evidence from selected studies supports the view that breast-feeding also protects the recipient infant against infection-induced anorexia (3–5). However, the interpretations of such published comparisons are not straightforward. Severity of illness often has not been controlled and age disparities between feeding groups commonly confound the interpretation of differences in energy intake. This is important because the intensity of infection-induced reductions in energy intake, or anorexia, has been associated with the severity of concomitant infectious episodes (6). Although it is not possible to isolate the benefits of feeding behaviors that accompany breast-feeding from those due to components specific to human milk, improving our understanding of the basis of this putative response is of inherent biologic interest and may improve nutritional management protocols during illness.

Observations relating inflammatory cytokines directly or indirectly to illness-induced anorexia in animal studies and in humans with cancer (7–11), the established role of serum leptin in food regulation, leptin responses to intravenous doses of interleukin (IL)-1β and tumor necrosis factor (TNF)-α in cancer patients (12,13) and the presence of cytokines and leptin in human milk (14) suggest that the proinflammatory cytokines IL-1β, TNF-α and leptin may mediate putative differential anorectic responses of breast-fed (BF) and formula-fed (FF) infants.

Thus, a quasi-experimental study was carried out in healthy infants who received the first or second administration of diphtheria, pertussis, tetanus and hemophilus influenza (DPTH) immunization. Our objective was to assess whether anorexia induced by an immunologic stimulus is attenuated in BF infants compared with FF infants and to evaluate relationships among changes in energy intake and serum IL-1β, TNF-α and leptin concentrations.

SUBJECTS AND METHODS

The study was conducted in a pediatric hospital of the Mexican Institute of Social Security (IMSS) in Mexico City and in a hospital of the Integrative Family Development Institute (DIFEM) in Toluca,

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[3] Abbreviations used: BF, breast-fed; DPTH, diphtheria, pertussis, tetanus and hemophilus influenza; FF, formula-fed; IL, interleukin; IL-1ra, IL-1 receptor antagonist; TNF, tumor necrosis factor.

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Mexico. Data collection started in June 1997 and ended in September 1999. Parents gave informed consent after written and spoken explanations of the study's benefits and risks to the infants. The right to leave the study at any time was explained carefully to all caregivers. Cornell University's Human Subject Use Institutional Review Board and the Ethics Committees of the IMSS and DIFEM approved the study protocol.

Study design. A quasi-experimental study was conducted. Healthy term infants ($n = 23$) who had not received either the first or second DPTH immunization (Tetramune, Pasteur Merieux; Paris, France) were hospitalized for 48 h. Infants were divided into two groups. Group assignment was determined by the mothers' previous decisions to fully breast- ($n = 12$) or formula-feed ($n = 11$). These decisions were made before mothers were approached for consent to participate in this study.

Mothers of infants < 6 mo old were interviewed in their family physician's offices. Infants who were either fully BF or FF from birth until the day of interview and whose mothers planned to continue the same feeding mode were eligible. Infants were considered fully BF if they were provided human milk as the only source of milk, with or without other liquids and solids, from birth until the study's completion. FF infants were fed artificial formula as the only source of milk since birth, also with or without other liquids and solids. Peripheral blood samples were obtained at admission and on d 2 at 90, 180 and 240 min postimmunization, and when a rise in body temperature (>37°C) was detected. Blood samples also were obtained 30 min before and after the first feeding the day before immunization and 30 min after the second postimmunization feeding.

Energy intakes were measured for 24 h before and after DPTH immunization. Anthropometric measurements were obtained at admission. Axillary temperatures were obtained at baseline and at 30-min intervals thereafter during d 2 of hospitalization. A standard mercury thermometer was read after being held in the axilla for 3 min.

Energy intake determinations. The 24-h test weighing procedure was used to estimate energy intakes. Formula, liquids other than human milk, and solid foods were weighed in food containers before and after each feeding. The weight difference was assumed to be the amount of food consumed after accounting for spills and regurgitation. Duplicates of all consumed foods other than human and artificial milk were obtained for subsequent calorimetric determinations (Parr Instruments Co Model 1266, Moline, Ill). The energy content of formulas was obtained from information provided by the manufacturers.

Human milk intakes were estimated by weighing infants before and after each nursing on electronic balances (model 3862MP8, Sartorius, Gottingen, Germany). Weight differences were assumed to represent the amount of milk consumed. No correction was made for respiratory or other evaporative water losses. Energy from human milk was calculated by multiplying the volume (L) ingested in 24 h by 650 kcal/L (2.7 MJ/L), the average energy concentration provided by human milk of women in developing countries (15). The number, duration and type of feedings, i.e., human milk, formula and/or solid foods, were recorded.

Blood sampling procedures and laboratory determinations. An indwelling catheter was placed in a peripheral vein at admission to obtain serial blood samples. The catheter was kept open with a heparin lock to avoid coagulation. Blood samples were centrifuged and frozen within 30 min of collection. After blood clotting, samples were centrifuged at 4°C and 2500 × *g* for 15 min and the serum was frozen in aliquots at −60°C until determinations.

Blood samples collected at admission were analyzed for basal glucose, leptin, IL-1β and TNF-α. Postimmunization serial blood samples were collected to identify cytokine surges. Blood glucose concentrations were determined in all samples.

Serum IL-1β and TNF-α were determined with an ultrasensitive ELISA commercial kit that reads in the range of 0.31 and 20 ng/L (Cytoscreen, Immunoassay, Biosource International, Camarillo, CA). Samples with concentrations above this range were diluted and reanalyzed. Cytokines were determined in duplicate. Duplicates with CV > 20% were repeated. Leptin concentrations were determined in duplicate by RIA. Commercial kits were used for human leptin determinations (sensitivity of 0.05 μg/L) (LINCO Research, St. Charles, MO). Glucose was determined by a glucose reactive strip method (Haemo-glucotest, Boehringer Mannheim, Mannheim, Germany).

Data analysis. Unadjusted means of energy intake were expressed as energy intake per kg body [kJ/(kg · d)]. Energy intakes and number and total duration of feedings on d 1 of hospitalization were compared with measurements obtained on the day after immunization. Intake differences between d 1 and 2 were compared in BF and FF infants. Paired and Student *t* tests were used to assess intake differences. Serum cytokine and leptin concentrations were transformed logarithmically for statistical analysis. Paired *t* tests were used to compare serum leptin and cytokine concentrations pre- and postimmunization. Student's *t* tests were used to compare BF and FF groups. Correlations were evaluated between pre- and postimmunization changes in energy intakes, leptin responses and various anthropometric measurements and indicators. Unless indicated otherwise, all values are expressed as means ± 1 SEM.

RESULTS

Subject characteristics. Healthy infants ($n = 23$) immunized against DPTH were followed; 12 were BF and 11 FF. Anthropometric characteristics of infants stratified by feeding mode are summarized in **Table 1**. There were no age differences between feeding groups. Anthropometric Z-scores were higher for BF than for FF infants; however, no values at or below Z-scores of −2 were noted in any of the subjects. Of all infants, 35% received solid foods (<10% of total energy as vegetables and fruit purees); 41% of these were BF and 27% FF infants. Neither group consumed any other milk than that designated by feeding group. Half of the BF and FF infants received the first vaccine in the DPTH series of three. The others received the second in the series.

The mean body temperature (°C) was lower on admission in BF than in FF infants (36.3 ± 0.27 vs. 36.8 ± 0.42, $P = 0.002$). All but two infants experienced a rise in body temperature (°C) after immunization, but no difference was detected between feeding groups (1.5 ± 0.6 vs. 1.2 ± 0.5, $P = 0.10$ for FF and BF, respectively). However, FF infants had increased body temperature earlier than did BF infants (430 ± 48 min after immunization vs. 570 ± 53 min, $P = 0.04$).

Postimmunization blood glucose concentrations rose in 15 of the 19 infants for whom this information was available. No differences were noted between feeding groups.

Energy intake pattern. Baseline and postimmunization energy intakes are recorded in **Table 2**. As expected (1), baseline energy intakes were higher for FF than for BF infants. Mean energy intakes by FF infants fell by 12% in the postim-

TABLE 1

Nutritional characteristics of breast- (BP) and formula-fed (FP) study infants[1]

	BF ($n = 12$)	FF ($n = 11$)
Age, *mo*	3.9 ± 0.4	3.8 ± 0.4
Nutritional indicators		
Weight-for-age[2]	0.7 ± 0.3	−0.5 ± 0.1[3]
Length-for-age[2]	0.2 ± 0.3	−0.4 ± 0.3
Weight-for-length[2]	0.6 ± 0.2	−0.2 ± 0.3[3]
Arm circumference, *mm*	14 ± 0.5	13 ± 0.4[3]
Triceps skinfold, *mm*	11 ± 0.4	8 ± 0.7[3]

[1] Values are means ± SEM.
[2] Z-scores.
[3] Student *t* test, $P < 0.05$.

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TABLE 2

Pre- and post-diphtheria, pertussis, tetanus and hemophilus influenza (DPTH) immunization energy intakes, cytokine and leptin serum concentrations in breast- (BF) and formula- (FF) fed infants[1]

	BF (n = 12)	FF (n = 11)	P-value[2]
Energy intake, *kJ/(kg · d)*			
Baseline	353 ± 26	495 ± 28	0.0008
Postimmunization	354 ± 32	437 ± 24	0.03
Relative change	1.00 ± 0.2	0.88 ± 0.2[3]	0.03
Interleukin-1β, *ng/L*			
Baseline	0.90 ± 1.5	0.52 ± 1.4	0.87
Postimmunization	1.61 ± 1.4	1.17 ± 1.4	0.77
Relative change	1.79 ± 1.2[3]	2.26 ± 1.3[3]	0.22
Tumor necrosis factor-α, *ng/L*			
Baseline	6.15 ± 1.4	5.18 ± 1.2	0.69
Postimmunization	10.12 ± 1.5	9.23 ± 1.2	0.58
Relative change	1.64 ± 1.1[3]	1.78 ± 1.2	0.33
Leptin, *μg/L*			
Baseline	3.47 ± 1.2	1.37 ± 1.4	0.007
Postimmunization	3.18 ± 1.2	1.82 ± 1.4	0.09
Relative change	0.92 ± 1.1	1.31 ± 1.2[3]	0.03

[1] Values are means ± SEM.
[2] Student's t test comparing FF and BF infants.
[3] Paired t test, comparing baseline and postimmunization values, $P < 0.05$.

munization period (paired t test, $P = 0.001$). Reductions in energy intake in the postimmunization phase were totally from milk. Nonsignificant changes ($P = 0.19$) of energy from solid foods were noted in this period, (348 ± 162 kJ/d at baseline vs. 444 ± 218 kJ/d at postimmunization).

Potential influences of age or whether infants received the first or second DPTH immunization also were explored. Ages of FF and BF infants who received the first or second dose did not differ. The mean relative reduction in energy intakes of FF infants who received the first immunization dose was 18%. Energy intakes remained stable in FF infants who received the second dose and in all BF infants.

Serum IL-1β and TNF-α concentrations. Preimmunization serum IL-1β concentrations were below detectable limits (0.31 ng/L) in five infants. Only two infants did not experience increased (relative to baseline) postimmunization serum IL-1β concentrations. Postimmunization serum IL-1β concentrations tended to increase ($P = 0.22$) more in FF than in BF infants (1.3-fold vs. 0.8-fold, respectively, Table 2). TNF-α was detected at baseline in all infants (2.6–111 ng/L). Postimmunization serum TNF-α concentrations increased in 21 of the 23 subjects. The remaining two were represented in the BF and FF groups. TNF-α almost doubled ($P = 0.004$) in the entire sample but no differences were noted between feeding groups, (80 vs. 60% for FF and BF, respectively, $P = 0.33$, Table 2). One infant presented baseline TNF-α values that were 3 SD above the mean. Results of analyses with and without this infant's data were similar.

Pre- and postimmunization serum concentrations of TNF-α were correlated positively with pre- and postimmunization serum IL-1β concentrations ($r = 0.61$ and 0.60, respectively, $P = 0.003$).

Separate univariate regression analyses were conducted to assess relationships between pre- and postimmunization differences in energy intake and cytokine concentrations. Only increases in serum IL-1β concentrations among FF infants were associated with changes in energy intake, but the significance of this association was borderline ($r = -0.56$, $P = 0.08$, **Fig. 1**).

Differences in cytokine responses between infants who received the first or second DPTH immunization also were evaluated. FF infants who received the first dose tended ($P = 0.11$) to have greater increases in serum IL-1β concentrations than did FF infants who received the second dose (3.1 ± 1.3 vs. 1.7 ± 1.4 ng/L, respectively). No similar trend was detected among BF infants.

Serum leptin concentration. Leptin was detected in the serum of all infants. BF infants presented with significantly higher leptin concentrations at baseline (3.5 ± 1.2 vs. 1.4 ± 1.4 μg/L, $P = 0.007$). A net increase in postimmunization serum leptin concentrations was noted only in the FF group ($P = 0.01$). No association was noted between baseline energy intakes and leptin concentrations in either feeding group. Similarly, no associations were detected between postimmunization changes in leptin concentrations and energy intakes of FF infants.

Baseline serum leptin concentrations were correlated positively with weight-for-age and triceps skinfold thickness in the FF group (**Table 3**). Absolute ($r = 0.77$, $P = 0.009$) and relative ($r = 0.61$, $P = 0.06$) increments in postimmunization serum leptin concentrations also were correlated positively

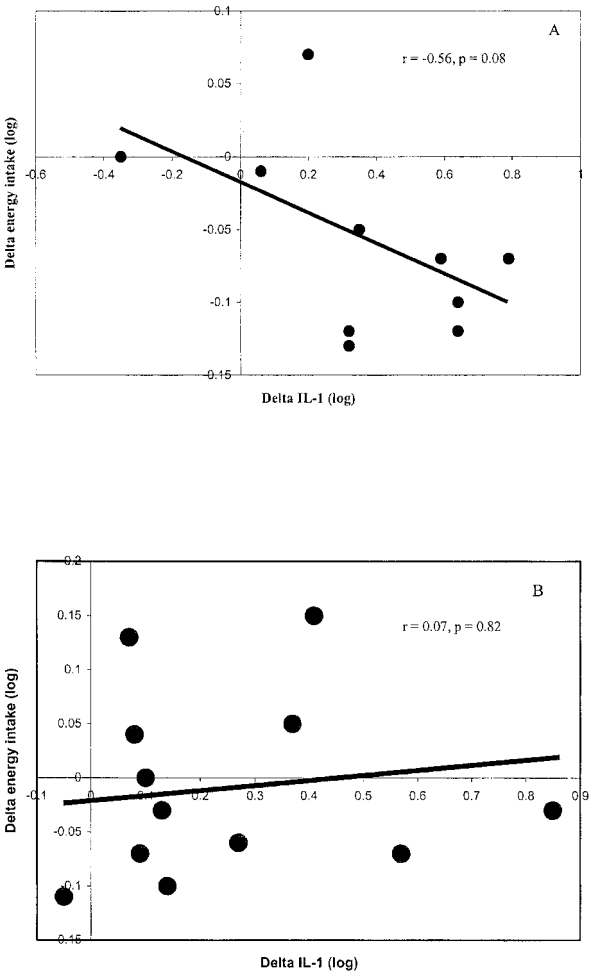

FIGURE 1 Postimmunization changes in energy intake [kJ/(kg · d)] vs. postimmunization changes in serum interleukin (IL)-1β (ng/L), in formula-fed (FF) infants (*panel A*) and in breast-fed (BF) infants (*panel B*).

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with weight-for-length Z-scores, but only in FF infants. Similar relationships were not observed between either absolute or relative increments in postimmunization serum leptin concentrations and any anthropometric value in BF infants.

Serum leptin concentrations also were determined before and 30 min after the first meal preceding immunization. No differences were noted between pre- and postprandial leptin concentrations (3.6 ± 0.6 vs. 3.5 ± 0.6 μg/L).

No differences were noted in the relative or absolute increments in postimmunization serum leptin and cytokines concentrations among infants who received the first or second immunization dose. Nor were there any associations detected between postimmunization increases in serum IL-1β and leptin ($P = 0.71$) concentrations.

DISCUSSION

These results support the assertion that breast-feeding protects against reductions in energy intake that accompany mild illnesses associated with immunologic and/or inflammatory responses and previous reports that energy regulation differs between breast- and formula-fed infants. Disparate basal metabolic rates, core body temperatures (16) and ad libitum energy intakes (1) have been noted previously between feeding groups. In this study, reductions in energy intake were associated with increases in serum leptin concentrations and very likely with increases in serum IL-1β concentrations, but both observations were noted only in FF infants. The net increase in serum leptin concentrations in the FF group following an immunologic challenge also suggests a role for leptin in the changes in energy intake experienced by FF infants in this study.

Breast-feeding is reported to attenuate the severity of infectious illnesses presumably because of the likelihood of decreased inoculums and/or more active immune responses by the breast-fed infant. Lower reductions in energy intake among breast-fed infants compared with those fed formula are cited as an example of the attenuated response to illness. Age, however, must be considered carefully in this association from at least two perspectives. Age is associated negatively with energy intake normalized per body weight and the likelihood of formula feeding increases at older ages. Hence, differences in energy intake between young and older infants may be misinterpreted as an effect of feeding mode.

The design used in this study provided an opportunity to analyze the anorectic effects of a controlled immunologic challenge in BF and FF infants of similar ages. These two design features are important because previous studies of BF and completely weaned infants often resulted in comparisons of infants of different ages. That is, BF infants were younger than those who were weaned and, thus, observed differences might have reflected disparities in responses due to age and not to differences in feeding mode. Selecting infants who were fed human or artificial milk from birth also creates comparisons between feeding groups more robust than is possible by comparisons that include weaned infants who received human milk at earlier ages.

The provision of a controlled stimulus to both feeding groups also strengthened the evaluation of the effect of feeding mode. Postimmunization reductions in energy intake were not observed in BF infants. FF infants decreased energy intake in the postimmunization phase by an average 12%, but it is important to note that the magnitude of the mean likely underestimates the anorexia associated with mild immunologic stimuli in FF infants. We did not anticipate that differences between the reactogenicity of the first and second dose would result in disparities in energy intakes as great as those indicated by these findings, i.e., those who received the first DPTH dose decreased energy intake by 18% (13–25%). These reductions are similar to those reported from community-based studies. No net reductions were detected in FF infants who received the second DPTH dose.

Thus, the anorectic response in FF infants appeared more sensitive to the nature of the immune stimulus than anticipated. There is strong evidence that although the immunogenicity determined by increased titers of antibodies (17) of the DPTH vaccine does increase through the progressive administration of the three doses, reactogenicity seems to decrease as the series progresses. A decrease in reactogenicity was demonstrated in a study that analyzed the adverse effects of the DPTH vaccine. The percentage of infants who presented fever > 38°C decreased from 53% in those who received the first dose, to 31% in infants who received the second and to 24% in those who received the last dose of the primary series. Similarly, a decreasing prevalence was reported for unusual crying and drowsiness (18). It is noteworthy that cytokine release in the current study was not influenced by immunization dose sequence. It is possible, however, that sample size and variability in cytokine increases limited the study's power to detect differences should any exist.

It is important to point out that these data do not permit us to evaluate whether breast-feeding's protection against the anorexia associated with an immunologic stimulus extends to more severe infectious conditions. Indeed, preliminary data (19) obtained in BF and FF infants with severe pneumonia suggest that protection afforded by breast-feeding in milder infections is not sustainable under more severe conditions.

Nonetheless, the protective effect is potentially very important even if it is limited to milder infections. In a study conducted in Mexico by our laboratory, the incidence of acute respiratory infections and diarrhea during the first 6 mo of age was 5 and 3 times higher in FF infants than in infants fed human milk, respectively (2). The mean duration of episodes was ~6.4 d. If we consider a 13–25% reduction in energy intake per illness day, a FF infant suffering 4 episodes of mild infections in his/her first 6 mo of life will accumulate a deficit of at least 2520–4872 kcal (10.5–20.4 MJ) and fail to obtain the micronutrients that normally would accompany that intake. This of course presents a "worst case" scenario with no catch-up in intake during convalescence. On the other hand, it is noteworthy that this estimate also does not include

TABLE 3

Correlation coefficients between selected variables and baseline serum leptin concentrations in formula- (FF) and breast-fed (BF) infants

	FF		BF	
	r-coefficient	*P*-value[1]	*r*-coefficient	*P*-value
Age, *mo*	−0.04	0.84	−0.03	0.86
Weight-for-age[2]	0.76	0.007	0.37	0.24
Length-for-age[2]	0.25	0.48	0.25	0.46
Weight-for-length[2]	0.22	0.55	0.23	0.50
Triceps skinfold	0.58	0.07	0.53	0.10
Arm circumference	0.40	0.26	−0.03	0.93

[1] Pearson analyses.
[2] Z-scores.

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exacerbated needs imposed by higher metabolic rates associated with illness.

Conducting this study in a hospital setting presented the opportunity to control for some factors that often complicate interpretations of field observations. Similar foods were offered to all infants consuming mixed diets and the methods used to estimate energy intakes were similar and applied rigorously to all observations. Withholding food by mothers was avoided because mothers remained with their infants in the hospital and all were encouraged to feed their infants ad libitum. Thus, results from this hospital-based study support the view that illness-related reductions in energy intake reported by field studies are unlikely to have been confounded substantially by these or other similar factors that are much more difficult to control under field conditions (4,5).

Previous studies conducted in animals or in humans with cancer suggest that reductions in energy intakes during illness may be mediated by the release of the proinflammatory cytokines IL-1β and TNF-α in response to infectious stimuli. The few available human studies, however, that have been conducted in healthy subjects have focused on adults, i.e., none have been done in infants. The higher than anticipated variability in cytokine responses and the study's sample size likely precluded the detection of significant associations between the two variables.

Another possibility merits consideration. Possible differences in soluble cytokine receptor concentrations between feeding groups also may have influenced the results of this investigation. IL-1 receptor antagonist (IL-1ra) is a naturally occurring antagonist of IL-1 that binds competitively to the cellular IL-1 receptor and does not result in cell activation (20). Previous studies have shown that human milk contains both IL-1ra and soluble TNF receptors that persist throughout lactation (21). It also has been reported that IL-1ra may contribute to high neonatal morbidity and mortality due to sepsis. Neonates with severe infection or sepsis were reported to present higher serum concentrations of IL-1ra compared with others with mild infections (22). The present study measured only circulating cytokine and detected no significant differences between feeding groups. It is possible that peripheral concentrations of soluble receptors differ between feeding groups, and that differences in energy responses observed between them are influenced, at least in part, by IL-1ra concentrations.

BF infants presented higher baseline serum leptin concentrations than did FF infants (3.5 ± 1.2 vs. 1.4 ± 1.4 μg/L, $P = 0.007$). These results differ from those reported earlier by Lonnerdal et al. (23), who detected no differences in serum leptin concentrations between breast- and formula-fed infants. Differences between groups, however, may have been blurred in that earlier report because, unlike the present study, neither feeding status nor sampling times were controlled. Augmented serum leptin concentrations in BF infants may reflect an increased production of leptin due to higher adiposity, the acquisition of exogenous leptin through human milk and/or other possibilities.

Postimmunization serum leptin concentrations increased only in the FF group. As is true for other differences related to energy regulation referred to earlier, the basis of differences in leptin responses is unknown. Perhaps an external leptin supply or other bioactive components in human milk dampen leptin responses to stimuli such as infections. It also is of interest that in addition to the changes in serum leptin concentrations that were observed only in FF infants, this group was also the only one that decreased its energy consumption after immunization. This observation and the association between serum leptin concentrations and selected anthropometric values in this study (Table 3) suggest multiple roles for leptin in energy metabolism that intersect with the immune system and infant feeding choices.

The likelihood of differences in energy regulation by both groups also is supported by differences between feeding groups in baseline serum leptin concentrations and body temperatures, i.e., BF infants presented with lower baseline temperatures. However, questions remain. For example, Luheshi and co-workers (24) reported studies in rats demonstrating leptin's central effects on temperature regulation. They injected leptin directly into the brain and noted subsequent increases in IL-1 and core temperatures. These observations are generally consistent with changes in leptin and body temperature in FF infants, but they are not consistent with the differences in temperatures and serum leptin at baseline between formula- and breast-fed infants, unless peripheral leptin concentrations do not reflect leptin concentrations in the central nervous system.

In conclusion, energy intakes are reduced and peripheral leptin concentration increased in FF infants after a mild immunologic stimulus. Although the present design does not permit distinguishing the effect of differences between caring behaviors in both feeding groups from those due to disparities in the composition of human milk and artificial formulas, physiologic feeding responses clearly differed between feeding groups. Reductions in energy intake were associated positively with increases in IL-1β, i.e., the greater the postimmunization increases in serum IL-1β concentrations, the greater the decrease in energy intake. BF infants did not exhibit anorectic responses to a mild immunologic stimulus. This effect of breast-feeding was not explained by differences in the nature or magnitude of the offending antigens. The ability of breast-feeding to protect infants against anorexia in more severe infections remains to be demonstrated convincingly. Therefore, studies designed to evaluate anorectic responses during infectious episodes in BF and FF infants remain of interest.

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LITERATURE CITED

1. Garza, C. & Butte, N. F. (1990) Energy intake of human milk-fed infants during the first year. J. Pediatr. 117: S124–131.
2. López-Alarcón, M., Villalpando, S. & Fajardo, A. (1997) Breast feeding lowers the frequency and duration of acute respiratory infections and diarrhea in infants under six months of age. J. Nutr. 126: 436–443.
3. Hoyle, B. Y., Yunus, M. & Chen, L. C. (1980) Breast-feeding and food intake among children with acute diarrheal diseases. Am. J. Clin. Nutr. 33: 2365–2371.
4. Dickin, K. L., Brown, K. H., Fagbule, D., Adedoyin, M., Gittelsohn, J., Esrey, S. A. & Oni, G. A. (1990) Effect of diarrhea on dietary intake by infants and young children in rural villages of Kwara State, Nigeria. Eur. J. Clin. Nutr. 44: 307–317.
5. Brown, K. H., Stallings, R. Y., Creed de Kanashiro, H., Lopez de Romaña, G. & Black, R. E. (1990) Effect of common illnesses in infant energy intake from breast milk and other foods during longitudinal community-based studies in Huascar (Lima), Peru. Am. J. Clin. Nutr. 52: 1005–1013.
6. Brown, K. H. & Perez, F. (1992) Determinants of dietary intake during childhood diarrhea and implications for appropriate therapy. Acta Pediatr. Suppl. 381: 127–132.
7. McCarthy, D. O., Kluger, M. J. & Vander, A. J. (1985) Suppression of food intake during infection: is interleukin-1 involved? Am. J. Clin. Nutr. 42: 1179–1182.
8. Kapas, L. & Krueger, J. M. (1992) Tumor necrosis factor-beta induces sleep, fever and anorexia. Am. J. Physiol. 263: R703–R707.

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9. Warren, R. S., Starnes, H. F., Gabrilove, J. L., Oettgen, H. F. & Brennan, M. F. (1987) The acute metabolic effects of tumor necrosis factor administration in humans. Arch. Surg. 122: 1396–1400.

10. Klasing, K. C. (1988) Nutritional aspects of leukocytic cytokines. J. Nutr. 118: 1436–1446.

11. Watkins, L. R., Maier, S. F. & Goehler, L. E. (1995) Cytokine-to-brain communication: a review and analysis of alternative mechanisms. Life Sci. 57: 1011–1026.

12. Janik, J. E., Curti, B. D., Considine, R. V., Rager, H. C., Powers, G. C., Alvord, W. G., Smith, J. W., 2nd, Gause, B. L. & Kopp, W. C. (1997) Interleukin 1α increases serum leptin concentrations in humans. J. Clin. Endocrinol. Metab. 82: 3084–3086.

13. Zumbach, M. S., Boehme, M.W.J., Wahl, P., Stremmel, W., Ziegler, R. & Nawroth, P. P. (1997) Tumor necrosis factor increases serum leptin levels in humans. J. Endocrinol. Metab. 82: 4080–4082.

14. Casabiell, X., Piñeiro, V., Tomé, M. A., Peinó, R., Diéguez, C. & Casanueva, F. F. (1997) Presence of leptin in colostrum and/or breast milk from lactating mothers: a potential role in the regulation of neonatal food intake. J. Clin. Endocrinol. Metab. 82: 4270–4273.

15. Jensen, R. G. (1995) Handbook of Milk Composition, pp. 439–443. Academic Press, San Diego, CA.

16. Butte, N. F., Smith, E. D. & Garza, C. (1990) Energy utilization of breast-fed and formula-fed infants. Am. J. Clin. Nutr. 51: 350–358.

17. Ada, G. (1996) Overview of vaccines. In: Methods in Molecular Medicine. Vaccine Protocols (Robinson, A. & Farrar, G. H., eds.), pp. 1–16. Humana Press, Totowa, NJ

18. Bell, F., Heath, P., MacLennan, J., Shackley, F., Shearstone, N., Diggle, L., Thornton, C., Griffiths, H., Moxon, E. R. & Finn, A. (1999) Adverse effects and sero-responses to an acellular pertussis/diphtheria/tetanus vaccine when combined with *Hemophilus influenzae* type B vaccine in accelerated schedule. Eur. J. Pediatr. 158: 329–336.

19. López-Alarcón, M. G. (2000) Illness-Induced Anorexia in Breast-Fed Infants. Role of Interleukin-1β, Tumor Necrosis Factor-α and Leptin. Doctoral thesis, Cornell University, Ithaca, NY.

20. Granowitz, E. V., Clark, B. D., Mancilla, J. & Dinarello, C. (1991) Interleukin-1 receptor antagonist competitively inhibits the binding of interleukin-1 to the type II interleukin-1 receptor. J. Biol. Chem. 266: 141–147.

21. Buescher, E. & Malinowska, I. (1996) Soluble receptors and cytokine antagonists in human milk. Pediatr. Res. 40: 839–844.

22. De Bont, E.S.J.M. De Leij, L.H.F.M., Baarsma, A.O.R. & Kimpen J.L.L. (1995) Increased plasma concentrations of interleukin-1 receptor antagonist in neonatal sepsis. Pediatr. Res. 37: 626–629.

23. Lonnerdal, B. & Havel P. J. (2000) Serum leptin concentrations in infants: effects of diet, sex, and adiposity. Am. J. Clin. Nutr. 72: 484–489.

24. Luheshi, G. N., Gardner, J. D., Rushforth, D. A., Loudon, A. S. & Rothwell, N. J. (1999) Leptin actions on food intake and body temperature are mediated by IL-1. Proc. Natl. Acad. Sci. U.S.A. 96: 7047–7052.

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