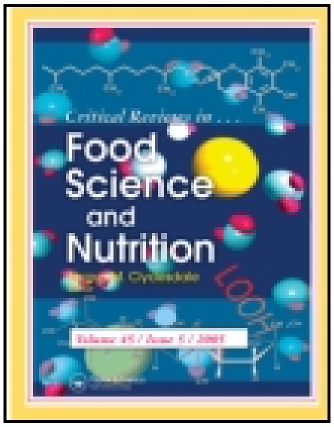

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Longitudinal Changes in Lactoferrin Concentrations in Human Milk: A Global Systematic Review,

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Longitudinal Changes in Lactoferrin Concentrations in Human Milk: A Global Systematic Review*,†

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Lactoferrin is the second most abundant whey protein in human milk and is known for its functional benefits, particularly antimicrobial activities. We report a comprehensive evaluation of the published literature on quantitative changes in lactoferrin in term and preterm human milk through the course of lactation. We also considered methods used to quantify lactoferrin. We critically evaluated 94 articles on human milk with 52 meeting study inclusion criteria (2724 women). A descriptive analysis of the data was performed. Lactoferrin concentration was highest during early lactation and rapidly declined to remain relatively unchanged from 1 month to 2 years of lactation. The unweighted mean of mean (±SEM) concentrations of lactoferrin in early milk (<28 days lactation) was 4.91 ± 0.31 g/L (range of means 0.34–17.94 g/L; median 4.03). For mature milk, the mean of means was 2.10 ± 0.87 g/L (range of means 0.44–4.4 g/L; median 1.91). The majority of data were derived from Europe with fewer studies from Africa and South America. There was a paucity of data on preterm milk. This comprehensive dataset explains in detail the longitudinal changes of lactoferrin concentrations in human milk throughout the world and briefly describes factors that may influence these concentrations.

Keywords Breast milk, protein, infant, nutrition, preterm, term

INTRODUCTION

Lactoferrin is a nonheme iron-binding protein that is part of the transferrin protein family (Gonzalez-Chavez et al., 2009). It is the second most abundant functional protein present in the whey fraction of human milk, comprising around 25% of the whey (de Wit, 1998; Conneely, 2001). The structural characteristics of this major human milk protein allow significant amounts to escape proteolytic digestion within the infant gastrointestinal tract and lactoferrin can be found intact in the stools of breastfed infants. The functional roles of lactoferrin include iron homeostasis, regulation of immune cell function and immune responses and, most notably, antimicrobial effects (Lönnerdal and Iyer, 1995; Yamauchi et al., 2000; Okuda et al., 2005; Ochoa et al., 2008; Egashira et al., 2009).

Human milk is the optimal source of nutrition (or feeding) for infants (Kleinman, 2009). Understanding human milk composition, and how it changes over the course of lactation, is a key first step in defining the nutritional needs of infants. While lactoferrin in human milk has been studied for nearly five decades (Nagasawa et al., 1972; Lönnerdal and Iyer, 1995), we know of no extant review of lactoferrin concentrations throughout the duration of lactation in peer-reviewed literature. More specifically, information is lacking on whether the concentration of lactoferrin through lactation varies with gestation (i.e., term versus preterm milk), geography, and method of analysis. Our goal was to describe the distribution of lactoferrin concentrations in breast milk from free-living mothers from around the world, who delivered preterm or at term, throughout the course of lactation. Our approach was to exhaustively and rigorously consider all

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available studies providing lactoferrin concentration in human milk.

MATERIALS AND METHODS

Data origin. A search for English articles using Scopus, PubMed, and Google Scholar was performed using keywords "breast milk" or "human milk" and "lactoferrin" periodically over several years, most recently in July 2010. Data pertaining to the lactoferrin concentration in human milk were categorized by continent, country, stage of lactation, gestational age, and method of analysis. Stages of lactation were defined as 0–5 days, 6–10 days, 11–30 days, 31–90 days, 91–180 days, 6–12 months, and >12 months to 2 years. Early human milk was defined as milk collected at <28 days lactation and mature human milk was collected ≥28 days lactation (Picciano, 2001; Kleinman, 2009). Term versus preterm classification was assigned according to the definition provided in each study. If gestational age was not specified in a study, values were categorized as "not stated" and grouped with term studies. We grouped study locations into wide geographical regions: Asia (AP), Europe (EU), North America (NA), South America (SA), and Africa (AF).

Inclusion Criteria. All data were derived from mothers in good health, and experimental groups with special diets were excluded. Selected studies clearly stated the method used to quantify lactoferrin. Sufficient information on milk sampling, including stage of lactation, units used to express lactoferrin concentration, and location also had to be stated. Studies were excluded for the following reasons: data were from only one mother, stage of lactation was too broad, study reported a mean derived from a collection of studies, data were duplicated, or studies combined results from preterm and term human milk. Milk could be collected at various times throughout the day, including before and after feeding. Other variables such as age, ethnicity, body weight, parity, and socioeconomic status were not considered.

Data Analysis. Lactoferrin concentrations are most often reported as grams per liter in the published literature. Therefore, data presented in this systematic review are also in grams per liter. Descriptive analyses were completed by the stage of lactation, gestational age, region, country, and method of analysis. We calculated unweighted means from the simple means reported in the papers. We also calculated weighted means which weighed according to the number of women in the study. Summary statistics are provided.

RESULTS

We reviewed 94 articles dating from 1966 to 2010 that provided values for lactoferrin for one or more lactation stages in preterm and/or term human milk. Fifty-two articles providing 228 mean human milk values (from 2724 women) for preterm and/or term milk were deemed to be within the selection criteria (Table 1); excluded articles and reasons for exclusion are listed in Table 2.

Table 1 Included studies in term and preterm lactoferrin analysis

References	
Butte et al., 1984b	Lönnerdal et al., 1980
Cuilliere et al., 1997	Lönnerdal and Forsum, 1985
Dawarkadas et al., 1991	Lönnerdal et al., 1996
Davidson and Lönnerdal, 1987	Lovelady et al., 2003
Donangelo et al., 1991	Marquis et al., 2003
Donangelo et al., 1989	Mathur et al., 1990
Donovan et al., 1989	McClelland et al., 1978
Duncan et al., 1983	Michalke et al., 1991
de Ferrer et al., 2000	Milnerowicz and Chmarek, 2005
Filteau et al., 1999	Montagne et al., 1999
Garg et al., 1988	Montagne et al., 2001
Goldblum et al., 1981	Motil et al., 1995
Goldblum et al., 1982	Nagasawa et al., 1972
Goldman et al., 1982a	Nagasawa et al., 1974
Goldman et al., 1983	Pamblanco et al., 1986
Goldsmith et al., 1983	Prentice et al., 1984
Hennart et al., 1991	Prentice et al., 1987
Hibberd et al., 1981	Raghuvanshi et al., 1988
Hirai et al., 1990	Reddy et al., 1977
Houghton et al., 1985b	Sanchez-Pozo et al., 1986
Houghton et al., 1985a	Sanchez-Pozo et al., 1987
Leelahakul et al., 2009	Semba et al., 1999
Lewis-Jones and Reynolds, 1983	Velona et al., 1999
Lewis-Jones et al., 1985	Yap et al., 1980
Lönnerdal et al., 1976a	Zapata et al., 1994
Lönnerdal et al., 1976b	Zavaleta et al., 1995a

Lactation Stage. Lactoferrin levels (weighted and unweighted) were greatest in the first 5 days of lactation, but steeply declined by up to 50% in the second week of lactation (Table 3). Past 1 month of lactation, lactoferrin concentrations remained stable for both weighted and unweighted means. There were no published data evaluating lactoferrin levels in preterm milk beyond 3 months of lactation (Table 3).

The distributions of lactoferrin concentrations for term milk are presented as histograms. The mean of mean (±SEM) concentrations of lactoferrin in early milk (<28 days lactation) was 4.91 ± 0.31 g/L (range of means 0.34–17.94 g/L; median 4.03). For mature milk (≥28 days lactation), the mean of means was 2.10 ± 0.87 g/L (range of means 0.44–4.4 g/L; median 1.91) (Figs. 1 and 2).

Geographical Distribution. The majority of the term milk data were derived from Europe ($n = 77$ values) with the least from Africa ($n = 16$ values) (Table 4). Term milk from South America had the highest overall lactoferrin concentration due to the high number of means from the first 5 days of lactation. We also characterized the regional distribution of lactoferrin by the stage of lactation in term milk (Fig. 3). Through the 2 week to >12 months of lactation, lactoferrin means were consistently greater in term milk from Asia compared to other areas of the world (Fig. 3). For preterm human milk, samples from Asia also had somewhat higher lactoferrin concentrations than other parts of the world (Fig. 4). However, this higher value was driven by studies conducted in the first week of lactation.

Table 2 Studies excluded based on study criteria for human milk lactoferrin

Reference	Reason for exclusion
Afify et al., 2003	Unit unclear
Akinbi et al., 2010	Lactoferrin graphed
Bindels and Harzer, 1985	Same as Harzer and Bindels, (1985); theoretical calculation
Britton, 1986	Lactoferrin graphed
Brooke et al., 1980	No lactoferrin method
Brooke et al., 1981	No lactoferrin method (same as Brooke et al., 1980)
Butte et al., 1984b	Values unconvertible and broad lactation stage (only 50–200 days)
Czank et al., 2009	No stage of lactation
Davidson and Lönnerdal, 1987	One woman
Donovan et al., 1987	Values not convertible (no weight and volume given)
Duncan et al., 1983	Only leprosy mothers
Evans et al., 1978	No stage of lactation
Filteau and Tomkins, 1994	Review
Ford et al., 1977	No stage of lactation and exact number of mothers
Forsum and Lönnerdal, 1980	Mothers on special diet
Fransson, 1983	No lactoferrin method, no information on milk sample
Goldman and Smith, 1973	Review
Goldman et al., 1982b	Lactoferrin graphed
Goldman et al., 1983	Lactoferrin graphed
Goldman and Goldblum, 1989	Review
Goldman, 1993	Review
Hambraeus, 1977	Review
Hambraeus et al., 1978	Review
Hartmann and Kulski, 1978	One woman and lactoferrin graphed
Harzer and Bindels, 1985	Theoretical calculation of lactoferrin
Hennart et al., 1991	Broad stage of lactation (12 mo)
Herias et al., 1993	Mothers on special diet
Houghton et al., 1985b	Broad lactation stage (only >15 days)
Houghton et al., 1985a	Broad lactation stage (only >15 days)
Kulski and Hartmann, 1981	Lactoferrin graphed
Kunz and Lönnerdal, 1993	Picture of gel, no values
Kunz and Lönnerdal, 1989	Picture of gel, no values
Larson, 1985	Lactoferrin values from another source
Lien et al., 2004	No stage of lactation, only mean of all countries
Lönnerdal et al., 1976c	Same as Lönnerdal et al. (1976a)
Levay and Viljoen, 1995	Review
Masson and Heremans, 1966	No stage of lactation
Masson et al., 1966	No stage of lactation and estimate of lactoferrin concentration
Mathur and Mathur, 1988	Editorial regarding Raghuvanshi et al. (1988)
Montagne et al., 2000a	Preterm and term milk combined
Montagne et al., 2000b	Lactoferrin graphed
Montgomery et al., 1987	Picture of gel, no values
Murakami et al., 1998	Picture of gel, no values
Prentice et al., 1985	No stage of lactation and milk from mothers with mastitis
Prentice et al., 1984	Broad lactation stage (<9 mo)
Prentice et al., 1983	Lactoferrin graphed
Semba et al., 1999	Only women with mastitis
Slutzah et al., 2010	Preterm and term milk combined
Welsh and May, 1979	Review
Uechi et al., 1982	No sample number and no form of SD or range

Table 3 Mean of means, median, standard deviation, and range of means of term and preterm human milk lactoferrin content according to lactation stage (values in g/L; *n* refers to the number of means)

	n	Weighted mean	Unweighted mean	Median	Range of means
0–5 days					
Term	43	6.63 ± 3.74	6.37 ± 3.19	5.51	0.8–17.94
Preterm	7	5.93 ± 1.39	5.06 ± 2.04	5.75	2.19–6.77
6–10 days					
Term	23	3.45 ± 1.46	3.75 ± 1.44	3.27	1.0–9.36
Preterm	5	3.62 ± 0.76	3.15 ± 1.13	3.14	2.18–4.32
11–30 days					
Term	27	4.24 ± 2.17	3.12 ± 0.99	2.92	0.92–8.04
Preterm	8	3.55 ± 0.90	3.01 ± 1.02	2.86	1.76–4.59
31–90 days					
Term	36	2.32 ± 1.01	2.10 ± 0.96	1.92	0.57–3.85
Preterm	2	3.07 ± 0.08	3.00 ± 1.11	3.00	2.90–3.09
91–180 days					
Term	19	2.03 ± 0.79	1.84 ± 1.03	1.54	0.58–4.28
6–12 months					
Term	12	2.19 ± 0.84	1.90 ± 0.67	1.57	0.44–3.63
>12 months					
Term	9	2.10 ± 0.547	1.87 ± 0.72	2.19	0.62–3.63

Gestational Age (preterm versus term human milk). The dynamics and content of lactoferrin in preterm and term milk were similar (Table 3).

Methods Used for Lactoferrin Analysis. Radial immunodiffusion, immunoelectrophoresis, ELISA, and SDS-PAGE were the most commonly used analytical methods to quantitate lactoferrin in human milk and generated similar mean of means (Table 5). Although microparticle enhanced nephelometric immunoassay was less commonly used, it yielded similar results as those mentioned above (Table 5).

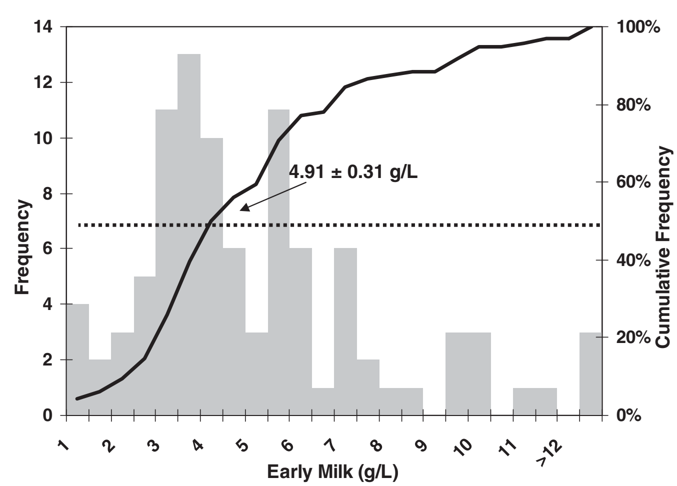

Figure 1 A distribution of lactoferrin in early-term breast milk in included studies presented as a histogram. The arrow refers to the mean at the 50th percentile (mean ± SEM).

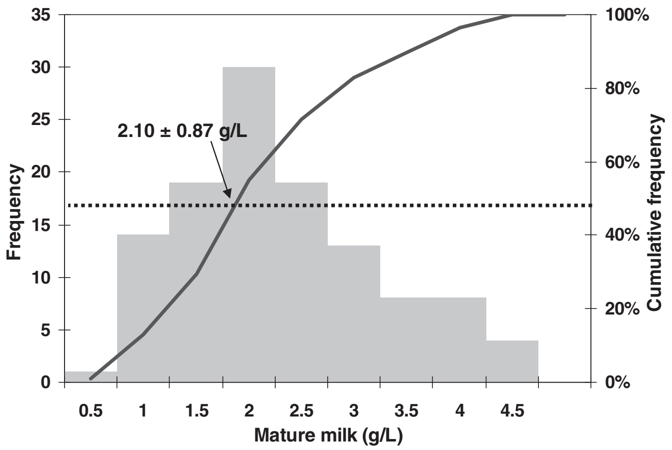

Figure 2 A distribution of lactoferrin in mature-term breast milk in included studies presented as a histogram. The arrow refers to the mean at the 50th percentile (mean ± SEM).

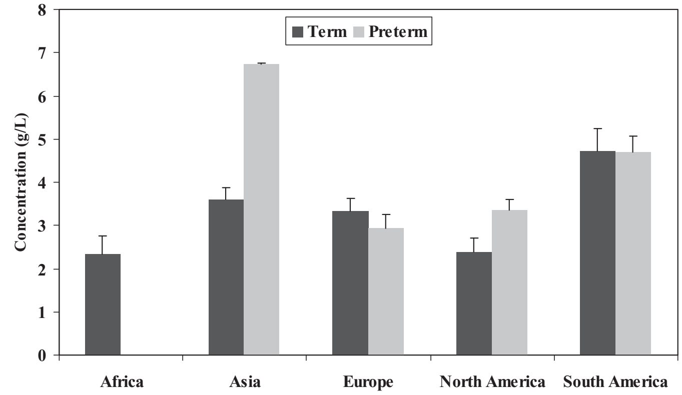

Figure 4 Regional distributions in preterm and term milk (values in g/L; mean ± SEM).

DISCUSSION

Human milk is the ideal food for infants. Expanding our understanding of the dynamics of human milk composition and functionality is an important guide to advancing our knowledge of infant nutrition. Lactoferrin is the second most abundant whey protein in human milk and has been studied for nearly five decades (Nagasawa et al., 1972; Lönnerdal and Iyer, 1995). This, however, is the first extant review of lactoferrin concentrations in human milk. After application of strict selection criteria to maintain data quality, this descriptive analysis revealed that the variability in human milk lactoferrin concentrations is primarily driven by the lactation stage.

Lactoferrin concentrations were highest in colostrum and decreased by about 50% in the first 5 days of lactation. Levels remained relatively constant after the first 30 days of lactation. The mean of mean (±SEM) concentrations of lactoferrin in early milk (<28 days lactation) was 4.91 ± 0.31 g/L (range of means 0.34–17.94 g/L; median 4.03). For mature milk (≥28 days lactation), the mean of means was 2.10 ± 0.87 g/L (range of means 0.44–4.4 g/L; median 1.91). This temporal pattern of change in lactoferrin concentrations parallels that observed for total protein levels in human milk through lactation (Hibberd et al.,

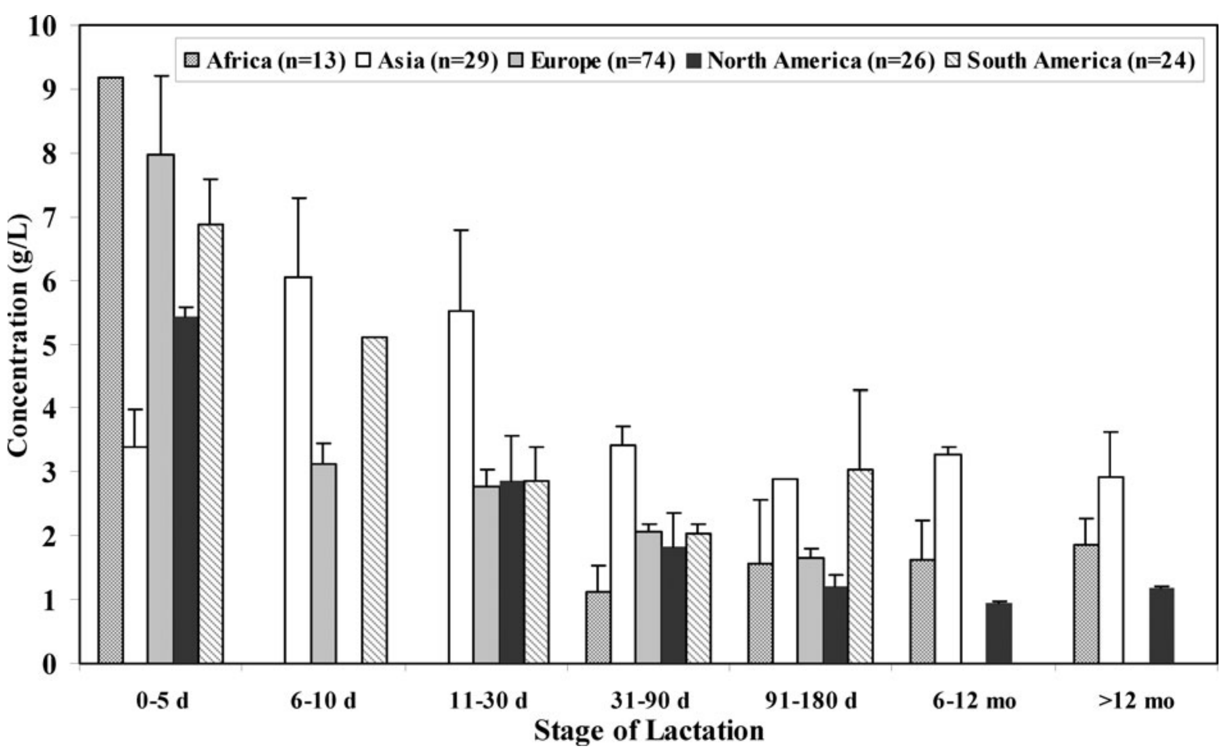

Figure 3 Term lactoferrin concentration by stage of lactation presented according to region (values in g/L; mean ± SEM).

Table 4 Lactoferrin content in term human milk by origin of milk collection (values in g/L; * denotes not applicable; *n* refers to number of means)

Country	*n*	Unweighted mean	SEM
Africa			
Ethiopia	8	2.67	1.03
Gambia	6	2.30	0.06
Malawi	1	0.57	*
Congo (Zaire)	1	0.90	*
Mean	16	2.29	0.52
Asia			
Bangladesh	2	5.72	2.32
India	20	3.71	0.41
Japan	6	4.17	0.63
Thailand	1	2.27	*
Mean	29	3.90	0.35
Europe			
Belgium	1	1.00	*
France	11	3.61	0.43
Germany	1	2.81	*
Italy	2	6.09	3.66
Poland	2	1.75	0.05
Spain	23	2.70	0.20
Sweden	11	2.00	0.22
UK	26	4.83	0.94
Mean	77	3.49	0.36
North America			
USA	26	2.41	0.36
Mean	26	2.41	0.36
South America			
Argentina	4	5.25	1.56
Brazil	9	4.81	0.79
Peru	11	4.87	0.97
Mean	24	4.91	0.57

1982; Butte et al., 1984a). Based on the limited number of studies conducted with preterm human milk, we were unable to confirm if lactoferrin levels differed between preterm and term human milk.

It is unclear if geography is important in dictating lactoferrin concentrations due to the unequal distribution of data among different geographical locations. Interestingly for term milk, there was a striking difference in the number of samples derived from Europe (close to three times as much data) compared with the other regions. Milk from South America had the highest overall lactoferrin concentration (Table 4), but half of the mean values for this region were from the first 5 days of lactation (colostrum) (Fig. 3). On average, lactoferrin concentrations by the stage of lactation in term human milk from Asia were consistently greater than values from the other regions after the first 5 days of lactation. Again, the data were unbalanced among lactation stages and regions. The overall mean lactoferrin concentration in preterm milk from Asia was also greater than all other regions. More data are needed to determine if these geographical differences are real and the reasons for the differences.

Table 5 Lactoferrin mean of means in term human milk by commonly cited methods to measure lactoferrin (values in g/L; *n* refers to the number of means)

Method	*n*	Unweighted mean	SEM
EIA	2	1.75	0.05
ELISA	27	3.65	0.59
Electroimmunodiffusion	17	2.47	0.49
IEF	1	2.81	*
Immunoelectrophoresis	38	3.87	0.36
Microparticle enhanced nephelometric immunoassay	10	3.73	0.43
Radial immunodiffusion	45	3.72	0.53
Radioimmunoassay	2	0.95	0.05
SDS-PAGE	24	3.93	0.43

It is unclear if dietary, cultural, genetic, or other health factors play a role in these differences in lactoferrin concentrations. Parity, age, ethnicity, and socioeconomic status do not appear to influence the concentration of lactoferrin in human milk (Houghton et al., 1985a; Donangelo et al., 1991; Hennart et al., 1991; Lien et al., 2004). We did not attempt to further explore how these variables might affect lactoferrin concentrations since many studies did not include specific information and sorting the data in this way would have resulted in eliminating several studies. Further research on how these differences may affect lactoferrin concentrations is warranted. In addition, lactoferrin concentration in human milk does not appear to depend on maternal iron status and is not influenced by iron supplementation (Lönnerdal and Iyer, 1995). Some research indicates that maternal malnutrition may negatively affect lactoferrin levels (Houghton et al., 1985a), while other research does not (Reddy et al., 1977; Hennart et al., 1991). Maternal infections may influence lactoferrin concentrations. Acute febrile illnesses from urinary tract, upper respiratory, gastrointestinal, or skin infections during labor or early postpartum have been associated with significantly lower concentrations in colostrum and early milk (Lönnerdal et al., 1996). These illnesses did not influence lactoferrin concentrations or measured milk volumes after lactation was established (Zavaleta et al., 1995b). Duncan and colleagues (1983) reported no differences in lactoferrin concentrations when comparing milk from women with leprosy and those without leprosy over 24 months of lactation. Mastitis, however, has been associated with higher concentrations (Semba et al., 1999). We limited our review to data from apparently healthy women, but it is possible that women with unknown health issues were included in the original studies thus influencing our findings. More study is needed to determine how exposure to pathogens, illness, and other health issues affect lactoferrin concentrations.

Radial immunodiffusion, immunoelectrophoresis, ELISA, and SDS-PAGE were the most commonly used analytical methods to determine lactoferrin concentrations. These methods, as well as the less commonly used microparticle enhanced nephelometric immunoassay, generated similar values for lactoferrin concentrations (Table 5). It was beyond the scope of our review to determine or comment on the accuracy of particular analytical methods due to limited information or small sample sizes reported with some of the methods. The method variation, however, would have been incorporated in the analytical values reported in the individual studies or sample populations analyzed. Based on the consistency of values, it is reasonable to conclude that these methods of analysis do not contribute to the variability of lactoferrin content in human milk. EIA, IEF, and radioimmunoassay were not frequently used and generated values lower than the more commonly cited methods. Only five means were generated with these methods so analytical method does not appear to contribute to the variability of lactoferrin concentrations that we found. Immunoassays appear to be good and reliable techniques for measuring lactoferrin as the values and ranges are quite comparable among the different studies we evaluated.

We calculated both unweighted means and weighted means from the data reported in the papers we reviewed. The unweighted means are simple means of the reported mean values. Unweighted means weigh each study evenly, regardless of the number of women in each study. This method is biased toward regions or countries in which more studies have been conducted (Brenna et al., 2007). We also calculated means that were weighted according to the number of subjects in each study (weighted means). These values are biased toward studies in which more subjects were enrolled (Brenna et al., 2007). For term milk, values for unweighted and weighted means differed by only $\leq$0.3 g/L for all stages of lactation except for 11–30 days. For this stage of lactation, the difference between the unweighted and weighted means was 1.12 g/L. It is possible that the high level (mean 8.04 g/L) found in the large study ($n =$ 212) by Filteau et al. (1999) may have influenced this weighted mean value (Filteau et al., 1999).

Lactoferrin in human milk is associated with functional benefits for infants, but the specific level of lactoferrin in human milk that contributes to these benefits has not been defined. In contrast to human milk, the concentrations of lactoferrin in cow milk and typical cow milk-based infant formulas are low (King et al., 2007). Studies have demonstrated beneficial effects of supplementing infants and children with 0.1–1 g/day of bovine milk-derived lactoferrin (Chierici et al., 1992; Okuda et al., 2005; Egashira et al., 2007, 2009). These levels are below or near the ranges for human milk that we reported ($<$28 days lactation range of means 0.34–17.94 g/L; $\geq$28 days lactation range of means 0.44–4.4 g/L). Additional studies are required to determine if specific levels of lactoferrin in human milk are needed to achieve biological effects.

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DR designed the project; DR, ASA, GPR, JB, and BL participated in collection and interpretation of data and writing of the manuscript; ASA and WZ analyzed the data. All authors read and approved the final manuscript. DR, ASA, WZ, GPR, and JB were employed by Mead Johnson Nutrition at the time of the study.

ABBREVIATIONS

AF = Africa
AP = Asia
D = Day
ELISA = Enzyme-linked immunosorbent assay
EU = Europe
HPLC = High-performance liquid chromatography
Mo = Month
NA = North America
PAGE = Polyacrylamide gel electrophoresis
SA = South America
SDS-PAGE = Sodium dodecyl sulfate polyacrylamide gel electrophoresis
SEM = Standard error of mean
Wk = Week
Y = Year

REFERENCES

Afify, A. M., Mohamed, M. A., Abdel-Salam, A. M. and Abd El-Azim, S. A. (2003). Electrophoretic analysis of colostrum and mature Egyptian human milk using SDS-polyacrylamide gel. *Milchwissenschaft*. **58**:583–585.

Akinbi, H., Meinzen-Derr, J., Auer, C., Ma, Y., Pullum, D., Kusano, R., Reszka, K. J. and Zimmerly, K. (2010). Alterations in the host defense properties of human milk following prolonged storage or pasteurization. *J. Pediatr. Gastroenterol. Nutr.* **51**:347–352.

Bindels, J. G. and Harzer, G. (1985). Amino acid and protein composition of human milk over the lactation period. *Ernahrungs-Umschau*. **32**:223–224.

Brenna, J. T., Varamini, B., Jensen, R. G., Diersen-Schade, D. A., Boettcher, J. A. and Arterburn, L. M. (2007). Docosahexaenoic and arachidonic acid concentrations in human breast milk worldwide. *Am. J. Clin. Nutr.* **85**:1457–1464.

Britton, J. R. (1986). Milk protein quality in mothers delivering prematurely: Implications for infants in the intensive care unit nursery setting. *J. Pediatr. Gastroenterol. Nutr.* **5**:116–121.

Brooke, O. G., Carter, N., Hibberd, C., Wood, C. and Brown, I. (1981). Protein concentrations in milk from mothers of preterm and term infants. *Biochem. Soc. Trans.* **9**:69–70.

Brooke, O. G., Carter, N. D., West, C. M. and Wood, C. (1980). Lysozyme, IgA, lactoferrin, and carbonic anhydrase levels in the milk of mothers of preterm and term infants. *Arch. Dis. Child.* **55**:652–653.

Butte, N. F., Garza, C., Johnson, C. A., Smith, E. O. and Nichols, B. L. (1984a). Longitudinal changes in milk composition of mothers delivering preterm and term infants. *Early Hum. Dev.* **9**:153–162.

Butte, N. F., Goldblum, R. M., Fehl, L. M., Loftin, K., Smith, E. O., Garza, C. and Goldman, A. S. (1984b). Daily ingestion of immunologic components in human milk during the first four months of life. *Acta Paediatr. Scand.* **73**:296–301.

Chierici, R., Sawatzki, G., Tamisari, L., Volpato, S. and Vigi, V. (1992). Supplementation of an adapted formula with bovine lactoferrin. 2. Effects on serum iron, ferritin, and zinc levels. *Acta Paediatr*. **81**:475–179.

Conneely, O. M. (2001). Antiinflammatory activities of lactoferrin. *J. Am. Coll. Nutr.* **20**:389S–395S; discussion 96S–97S.

Cuilliere, M. L., Montagne, P., Mole, C., Bene, M. C. and Faure, G. (1997). Microparticle-enhanced nephelometric immunoassay of lactoferrin in human milk. *J. Clin. Lab. Anal.* **11**:239–243.

Czank, C., Prime, D. K., Hartmann, B., Simmer, K. and Hartmann, P. E. (2009). Retention of the immunological proteins of pasteurized human milk in relation to pasteurizer design and practice. *Pediatr. Res.* **66**:374–379.

Davidson, L. A. and Lönnerdal, B. (1987). Persistence of human milk proteins in the breast-fed infant. *Acta Paediatr. Scand.* **76**:733–740.

Dawarkadas, A. M., Saha, K. and Mathur, N. B. (1991). A comparative study of cells and anti-microbial proteins in colostrum of mothers delivering pre- and full-term babies. *J. Trop Pediatr.* **37**:214–219.

de Ferrer, R., Baroni, A., Sambucetti, M., Lopez, N. and Ceriani Ceradas, J. (2000). Lactoferrin levels in term and preterm milk. *J. Am. Coll. Nutr.* **19**:370–373.

de Wit, J. N. (1998). Marschall Rhone-Poulenc Award Lecture. Nutritional and functional characteristics of whey proteins in food products. *J. Dairy Sci.* **81**:597–608.

Donangelo, C. M., Trugo, N. M., Koury, J. C., Barreto Silva, M. I., Freitas, L. A., Feldheim, W. and Barth, C. (1989). Iron, zinc, folate and vitamin B_{12} nutritional status and milk composition of low-income Brazilian mothers. *Eur. J. Clin. Nutr*. **43**:253–266.

Donangelo, C. M., Trugo, N. M., Mesquita, V. L., Rosa, G. and Da-Silva, V. L. (1991). Lactoferrin levels and unsaturated iron-binding capacity in colostrum of Brazilian women of two socioeconomic levels. *Braz. J. Med. Biol. Res.* **24**:889–893.

Donovan, S. M., Atkinson, S. A. and Lönnerdal, B. (1987). Whey protein in the feces of preterm infants receiving preterm milk and infant formula. **In**: Human Lactation 3: Effect of Human Milk Upon the Recipient Infant, pp. 377–378. Goldman, A. S., Atkinson, S. A. and Hanson, L. A., Eds., Plenum Press, New York.

Donovan, S. M. M., Atkinson, S. A., Whyte, R. K. and Lönnerdal, B. (1989). Partition of nitrogen intake and excretion in low-birth-weight infants. *Am. J. Dis. Child.* **143**:1485–1491.

Duncan, M. E., Samson, R. R., McGrath, J. and McClelland, D. B. (1983). Humoral defence factors in the breast milk of Ethiopian women with leprosy and healthy controls. *Am. J. Clin. Nutr.* **38**:921–928.

Egashira, M., Moriuchi, M. and Moriuchi, H. (2009). Prevention of the norovirus gastroenteritis in day-care centers with bovine lactoferrin-containing products. *E-PAS*. **65**: Abstract 4315.8.

Egashira, M., Takayanagi, T., Moriuchi, M. and Moriuchi, H. (2007). Does daily intake of bovine lactoferrin-containing products ameliorate rotaviral gastroenteritis? *Acta Paediatr*. **96**:1238–1244.

Evans, T. J., Ryley, H. C., Neale, L. M., Dodge, J. A. and Lewarne, V. M. (1978). Effect of storage and heat on antimicrobial proteins in human milk. *Arch. Dis. Child*. **53**:239–241.

Filteau, S., Rice, A., Ball, J., Chakraborty, J., Stoltzfus, R., de Franciso, A. and Willumsen, J. (1999). Breast milk immune factors in Bangladesh women supplemented postpartum with retinol and beta-carotene. *Am. J. Clin. Nutr.* **69**:953–958.

Filteau, S. and Tomkins, A. (1994). Infant feeding and infectious disease. **In**: Infant Nutrition, pp. 143–162. Walker, A. and Rolls, B., Eds., Chapman and Hall, London.

Ford, J. E., Law, B. A., Marshall, V. M. and Reiter, B. (1977). Influence of the heat treatment of human milk on some of its protective constituents. *J. Pediatr.* **90**:29–35.

Forsum, E. and Lönnerdal, B. (1980). Effect of protein intake on protein and nitrogen composition of breast milk. *Am. J. Clin. Nutr.* **33**:1809–1813.

Fransson, G. B. (1983). The role of lactoferrin in iron absorption and its relation to nutritional status. *Kieler Milchwirtschaftliche Forschungsberichte*. **35**:441–443.

Garg, M., Thirupuram, S. and Saha, K. (1988). Colostrum composition, maternal diet and nutrition in north India. *J. Trop Pediatr.* **34**:79–87.

Goldblum, R. M., Goldman, A. S., Garza, C., Johnson, C. A. and Nichols, B. L. (1982). Human milk banking. II. Relative stability of immunologic factors in stored colostrum. *Acta Paediatr. Scand*. **71**:143–144.

Goldblum, R., Garza, C., Johnson, C., Harrist, R., Nichols, B. and Goldman, A. (1981). Human milk banking. I. Effects of container upon immunologic factors in mature milk. *Nutr. Res.* **1**:449–459.

Goldman, A. S. (1993). The immune system of human milk: Antimicrobial, antiinflammatory and immunomodulating properties. *Pediatr. Infect. Dis. J.* **12**:664–671.

Goldman, A. S., Garza, C., Nichols, B. L. and Goldblum, R. M. (1982a). Immunologic factors in human milk during the first year of lactation. *J. Pediatr.* **100**:563–567.

Goldman, A. S., Garza, C., Nichols, B. L., Johnson, C. A., Smith, E. O. and Goldblum, R. M. (1982b). Effects of prematurity on the immunologic system in human milk. *J. Pediatr.* **101**:901–905.

Goldman, A. S. and Goldblum, R. M. (1989). Immunologic system in human milk: Characteristics and effects. **In**: Textbook of Gastroenterology and Nutrition in Infancy, pp. 135–142. Lebenthal, E., Ed., Raven Press, New York.

Goldman, A. S., Goldblum, R. M. and Garza, C. (1983). Immunologic components in human milk during the second year of lactation. *Acta Paediatr. Scand*. **72**:461–462.

Goldman, A. and Smith, C. (1973). Host resistance factors in human milk. *J. Pediatr.* **82**:1082–1090.

Goldsmith, S. J., Dickson, J. S., Barnhart, H. M. et al. (1983). IgA, IgG, IgM, and lactoferrin contents of human milk during early lactation and the effect of processing and storage. *J. Food Protect*. **46**:4–7.

Gonzalez-Chavez, S. A., Arevalo-Gallegos, S. and Rascon-Cruz, Q. (2009). Lactoferrin: Structure, function and applications. *Int. J. Antimicrob.* Agents. 33:e1–e8.

Hambraeus, L. (1977). Proprietary milk versus human breast milk in infant feeding. A critical appraisal from the nutritional point of view. *Pediatr. Clin. North Am.* **24**:17–36.

Hambraeus, L., Lönnerdal, B., Forsum, E. and Gebre-Medhin, M. (1978). Nitrogen and protein components of human milk. *Acta Paediatr. Scand.* **67**:561–565.

Hartmann, P. E. and Kulski, J. K. (1978). Changes in the composition of the mammary secretion of women after abrupt termination of breast feeding. *J. Physiol*. **275**:1–11.

Harzer, G. and Bindels, J. (1985). Changes in human milk immunoglobulin A and lactoferrin during early lactation. **In**: Composition and Physiological Properties of Human Milk. Amsterdam: Elesevier Science.

Hennart, P. F., Brasseur, D. J., Delogne-Desnoeck, J. B., Dramaix, M. M. and Robyn, C. E. (1991). Lysozyme, lactoferrin, and secretory immunoglobulin A content in breast milk: Influence of duration of lactation, nutrition status, prolactin status, and parity of mother. *Am. J. Clin. Nutr.* **53**:32–39.

Herias, M. V., Cruz, J. R., Gonzalez-Cossio, T., Nave, F., Carlsson, B. and Hanson, L. A. (1993). The effect of caloric supplementation on selected milk protective factors in undernourished Guatemalan mothers. *Pediatr. Res.* **34**:217–221.

Hibberd, C. M., Brooke, O. G., Carter, N. D., Haug, M. and Harzer, G. (1982). Variation in the composition of breast milk during the first 5 weeks of lactation: Implications for the feeding of preterm infants. *Arch. Dis. Child.* **57**:658–662.

Hibberd, C. M., Brooke, O. G., Carter, N. D. and Wood, C. (1981). A comparison of protein concentrations and energy in breast milk from preterm and term mothers. *J. Hum. Nutr.* **35**:189–198.

Hirai, Y., Kawakata, N., Satoh, K., Ikeda, Y., Hisayasu, S., Orimo, H. and Yoshino, Y. (1990). Concentrations of lactoferrin and iron in human milk at different stages of lactation. *J. Nutr. Sci. Vitaminol*. **36**:531–544.

Houghton, M. R., Gracey, M., Burke, V., Bottrell, C. and Spargo, R. M. (1985a). Breast milk lactoferrin levels in relation to maternal nutritional status. *J. Pediatr. Gastroenterol. Nutr.* **4**:230–233.

Houghton, M., Santoso, H., Soetjiningsih and Gracey, M. (1985b). Lactoferrin concentrations in the breast milk of Indonesian mothers. *Paediatrica Indonesiana*. **25**:163–166.

King, J., Cummings, G., Guo, N., Trivedi, L., Readmond, B., Keane, V., Feigelman, S. and de Waard, R. (2007). A double-blind, placebo-controlled, pilot study of bovine lactoferrin supplementation in bottle-fed infants. *J. Pediatr. Gastr. Nutr.* **44**:245–251.

Kleinman, R., Ed. (2009). Pediatric Nutrition Handbook, American Academy of Pediatrics, Elk Grove Village, IL.

Kulski, J. K. and Hartmann, P. E. (1981). Changes in human milk composition during the initiation of lactation. *Aust. J. Exp. Biol. Med. Sci*. **59**:101–114.

Kunz, C. and Lönnerdal, B. (1989). Human milk proteins: Separation of whey proteins and their analysis by polyacrylamide gel electrophoresis, fast protein liquid chromatography (FPLC) gel filtration, and anion-exchange chromatography. *Am. J. Clin. Nutr.* **49**:464–470.

Kunz, C. and Lönnerdal, B. (1993). Protein composition of rhesus monkey milk: Comparison to human milk. *Comp. Biochem. Physiol. Comp. Physiol.* **104**:793–797.

Larson, B. L. (1985). Determination of specific milk proteins. **In**: Human Lactation, pp. 33–38. Jenson, Neille, Ed. Plenum, New York.

Leelahakul, V., Tanaka, F., Sinsuksai, N., Vichitsukon, K., Pinyopasakul, W., Kido, N. and Inukai, S. (2009). Comparison of the protein composition of breast milk and the nutrient intake between Thai and Japanese mothers. *Nurs. Health Sci.* **11**:180–184.

Levay, P. F. and Viljoen, M. (1995). Lactoferrin: A general review. *Haematologica.* **80**:252–267.

Lewis-Jones, D. I., Lewis-Jones, M. S., Connolly, R. C., Lloyd, D. C. and West, C. R. (1985). Sequential changes in the antimicrobial protein concentrations in human milk during lactation and its relevance to banked human milk. *Pediatr. Res.* **19**:561–565.

Lewis-Jones, D. I. and Reynolds, G. J. (1983). A suggested role for precolostrum in preterm and sick newborn infants. *Acta Paediatr. Scand.* **72**:13–17.

Lien, E., Jackson, J., Kuhlman, C., Pramuk, K., Lönnerdal, B. and Janszen, D. (2004). Variations in concentrations of lactoferrin in human milk: A nine country survey. *Adv. Exp. Med. Biol.* **554**:423–426.

Lönnerdal, B. and Forsum, E. (1985). Casein content of human milk. *Am. J. Clin. Nutr.* **41**:113–120.

Lönnerdal, B., Forsum, E., Gebre-Medhin, M. and Hambraeus, L. (1976a). Breast milk composition in Ethiopian and Swedish mothers. II. Lactose, nitrogen and protein contents. *Am. J. Clin. Nutr.* **29**:1134–1141.

Lönnerdal, B., Forsum, E. and Hambraeus, L. (1976b). The protein content of human milk I. Transversal study of Swedish normal material. *Nutr. Rep. Inter.* **13**:125–134.

Lönnerdal, B., Forsum, E. and Hambraeus, L. (1976c). A longitudinal study of the protein, nitrogen, and lactose contents of human milk from Swedish well-nourished mothers. *Am. J. Clin. Nutr.* **29**:1127–1133.

Lönnerdal, B., Forsum, E. and Hambraeus, L. (1980). Effect of oral contraceptives on composition and volume of breast milk. *Am. J. Clin. Nutr.* **33**:816–824.

Lönnerdal, B. and Iyer, S. (1995). Lactoferrin: Molecular structure and biological function. *Ann. Rev. Nutr.* **15**:93–110.

Lönnerdal, B., Zavaleta, N., Kusunoki, L., Lanata, C. F., Peerson, J. M. and Brown, K. H. (1996). Effect of postpartum maternal infection on proteins and trace elements in colostrum and early milk. *Acta Paediatr.* **85**:537–542.

Lovelady, C. A., Hunter, C. P. and Geigerman, C. (2003). Effect of exercise on immunologic factors in breast milk. *Pediatrics.* **111**:E148–E152.

Marquis, G. S., Penny, M. E., Zimmer, J. P., Diaz, J. M. and Marin, R. M. (2003). An overlap of breastfeeding during late pregnancy is associated with subsequent changes in colostrum composition and morbidity rates among Peruvian infants and their mothers. *J. Nutr.* **133**:2585–2591.

Masson, P. L. and Heremans, J. F. (1966). Studies on lactoferrin, the iron-binding protein of secretions. *Protides Biol. Fluids.* **14**:115–124.

Masson, P. L., Heremans, J. F. and Dive, C. H. (1966). An iron-binding protein common to many external secretions. *Clinica Chimica Acta.* **14**:735–739.

Mathur, G. P. and Mathur, S. (1988). Breast milk total nitrogen, non-protein nitrogen and lactoferrin content. *Indian Pediatr.* **25**:701–702.

Mathur, N. B., Dwarkadas, A. M., Sharma, V. K., Saha, K. and Jain, N. (1990). Anti-infective factors in preterm human colostrum. *Acta Paediatr. Scand.* **79**:1039–1044.

McClelland, D. B., McGrath, J. and Samson, R. R. (1978). Antimicrobial factors in human milk. Studies of concentration and transfer to the infant during the early stages of lactation. *Acta Paediatr. Scand Suppl.* **271**:1–20.

Michalke, B., Munch, D. C. and Schramel, P. (1991). Contribution to Zn-speciation in human breast milk: Fractionation of organic compounds by HPLC and subsequent Zn-determination by DCP-AES. *J. Trace Elem. Electrolytes Health Dis.* **5**:251–258.

Milnerowicz, H. and Chmarek, M. (2005). Influence of smoking on metallothionein level and other proteins binding essential metals in human milk. *Acta Paediatr.* **94**:402–406.

Montagne, P., Cuilliere, M. L., Molé, C., Béné, M. C. and Faure, G. C. (1999). Immunological and nutritional composition of human milk in relation to prematurity and mother's parity during the first 2 weeks of lactation. *J. Pediatr. Gastroenterol. Nutr.* **29**:75–80.

Montagne, P. M., Cuillière, M. L., Molé, C. M., Béné, M. C. and Faure, G. C. (2000a). Dynamics of the main immunologically and nutritionally available proteins of human milk during lactation. *J. Food Comp. Anal.* **13**:127–137.

Montagne, P. M., Cuillière, M. L., Molé, C. M., Béné, M. C. and Faure, G. C. (2001). Changes in lactoferrin and lysozyme levels in human milk during the first twelve weeks of lactation. *Adv. Exp. Med. Biol.* **501**:241–247.

Montagne, P. M., Tregoat, V. S., Cuillière, M. L., Molé, C. M., Béné, M. C. and Faure, G. C. (2000b). Measurement of nine human milk proteins by nephelometric immunoassays: Application to the determination of mature milk protein profile. *Clin. Biochem.* **33**:181–186.

Montgomery, P. A., Patton, S., Huston, G. E. and Josephson, R. V. (1987). Gel electrophoretic analysis of proteins in human milk and colostrum. *Comp. Biochem. Physiol.* **86**:635–639.

Motil, K. J., Thotathuchery, M., Bahar, A. and Montandon, C. M. (1995). Marginal dietary protein restriction reduced nonprotein nitrogen, but not protein nitrogen, components of human milk. *J. Am. Coll. Nutr.* **14**:184–191.

Murakami, K., Lagarde, M. and Yuki, Y. (1998). Identification of minor proteins of human colostrum and mature milk by two-dimensional electrophoresis. *Electrophoresis.* **19**:2521–2527.

Nagasawa, T., Kiyosawa, I. and Kuwahara, K. (1972). Amounts of lactoferrin in human colostrum and milk. *J. Dairy Sci.* **55**:1651–1659.

Nagasawa, T., Kiyosawa, I. and Takase, M. (1974). Lactoferrin and serum albumin of human casein in colostrum and milk. *J. Dairy Sci.* **57**:1159–1163.

Ochoa, T. J., Chea-Woo, E., Campos, M., Pecho, I., Prada, A., McMahon, R. J. and Cleary, T. G. (2008). Impact of lactoferrin supplementation on growth and prevalence of Giardia colonization in children. *Clin. Infect. Dis.* **46**:1881–1883.

Okuda, M., Nakazawa, T., Yamauchi, K., Miyashiro, E., Koizumi, R., Booka, M., Teraguchi, S., Tamura, Y., Yoshikawa, N., Adachi, Y. and Imoto, I. (2005). Bovine lactoferrin is effective to suppress Helicobacter pylori colonization in the human stomach: A randomized, double-blind, placebo-controlled study. *J. Infect. Chemother.* **11**:265–269.

Pamblanco, M., Ten, A. and Comin, J. (1986). Proteins in preterm and term milk from mothers delivering appropriate or small-for-gestational age infants. *Early Hum. Dev.* **14**:267–272.

Picciano, M. F. (2001). Representative values for constituents of human milk. *Pediatr. Clin. North Am.* **48**:263–264.

Prentice, A., Ewing, G., Roberts, B., Lucas, A., MacCarthy, A., Jarjou, L. M. A. and Whitehead, R. G. (1987). The nutritional role of breast milk IgA and lactoferrin. *Acta Paediatr. Scand.* **76**:592–598.

Prentice, A., Prentice, A. M., Cole, T. J., Paul, A. A. and Whitehead, R. G. (1984). Breast-milk antimicrobial factors of rural Gambian mothers. I. Influence of stage of lactation and maternal plane of nutrition. *Acta Paediatr. Scand.* **73**:796–802.

Prentice, A., Prentice, A. M., Cole, T. J. and Whitehead, R. G. (1983). Determinants of variations in breast milk protective factor concentrations of rural Gambian mothers. *Arch. Dis. Child.* **58**:518–522.

Prentice, A., Prentice, A. M. and Lamb, W. H. (1985). Mastitis in rural Gambian mothers and the protection of the breast by milk antimicrobial factors. *Trans. R Soc. Trop Med. Hyg.* **79**:90–95.

Raghuvanshi, R. S., Agarwal, K. N., Fransson, G. B. and Hambraeus, L. (1988). Breast milk total nitrogen nonprotein nitrogen and lactoferrin content. *Indian Pediatr.* **25**:149–159.

Reddy, V., Bhaskaram, C., Raghuramulu, N. and Jagadeesan, V. (1977). Antimicrobial factors in human milk. *Acta Paediatr. Scand.* **66**:229–232.

Sanchez-Pozo, A., Lopez, J., Pita, M. L., Izquierdo, A., Guerrero, E., Sanchez-Medina, F., Martinez Valverde, A. and Gil, A. (1986). Changes in the protein fractions of human milk during lactation. *Ann. Nutr. Metab.* **30**:15–20.

Sanchez-Pozo, A., Lopez, J., Morales, J., Izquierdo, A., Martinez-Valverde, A. and Gil, A. (1987). Protein composition of human milk in relation to mothers' weight and socioeconomic status. *Hum. Nutr. Clin. Nutr.* **41**:115–125.

Semba, R., Kumwenda, N., Taha, T., Hoover, D., Lan, Y., Eisinger, W., Mtimavalye, L., Broadhea, R., Miotti, P., Van der Hoeven, L. and Chiphangwi, J.

(1999). Mastitis and immunological factors in breast milk of lactating women in Malawi. *Clin. Diagn. Lab. Immunol.* **6**:671–674.

Slutzah, M., Codipilly, C. N., Potak, D., Clark, R. M. and Schanler, R. J. (2010). Refrigerator storage of expressed human milk in the neonatal intensive care unit. *J. Pediatr.* **156**:26–28.

Uechi, M., Ikezawa, Y. and Kosuge, K. (1982). Anti-infective substances in human colostrum and milk. *Pediatr. Int.* **24**:245–251.

Velona, T., Abbiati, L., Beretta, B., Gaiaschi, A., Flauto, U., Tagliabue, P., Galli, C. L. and Restani, P. (1999). Protein profiles in breast milk from mothers delivering term and preterm babies. *Pediatr. Res.* **45**:658–663.

Welsh, J. K. and May, J. T. (1979). Anti-infective properties of breast milk. *J. Pediatr.* **94**:1–9.

Yamauchi, K., Hiruma, M., Yamazaki, N., Wakabayashi, H., Kuwata, H., Teraguchi, S., Hayasawa, H., Suegara, N. and Yamaguchi, H. (2000). Oral administration of bovine lactoferrin for treatment of tinea pedis. A placebo-controlled, double-blind study. *Mycoses.* **43**:197–202.

Yap, P. L., Mirtle, C. L., Harvie, A. and McClelland, D. B. (1980). Milk protein concentrations in neonatal milk (witch's milk). *Clin. Exp. Immunol.* **39**:695–697.

Zapata, C. V., Donangelo, C. M. and Trugo, N. M. F. (1994). Effect of iron supplementation during lactation on human milk composition. *J. Nutr. Biochem.* **5**:331–337.

Zavaleta, N., Lanata, C., Burtron, B., Peerson, J. M., Brown, K. H. and Lönnerdal, B. (1995a). Effect of acute maternal infection on quantity and composition of breast milk. *Am. J. Clin. Nutr.* **62**:559–563.

Zavaleta, N., Nombera, J., Rojas, R., Hambraeus, M., Gislason, J. and Lönnerdal, B. (1995b). Iron and lactoferrin in milk of anemic mothers given iron supplements. *Nutr. Res.* **15**:681–690.