nial cerebrovascular disease. In: Recent Advances in the Study of Cerebral Circulation. Tarases J. M., Fischgold H., and Dilenge D., (eds) pp. 175 (Charles C. Thomas, Springfield, IL, 1980).
8. Hauge, A., Thoresen, M., and Walloe, L.: Changes in cerebral blood flow during hyperventilation and CO_2-breathing measured transcutaneously in humans by a bidirectional, pulsed, ultrasound doppler blood velocitymeter. Acta Physiol. Scand., *110:* 167 (1980).
9. Hernandez, M. J., Brennan, R. W., and Bowman, G. S.: Autoregulation of cerebral blood flow in the newborn dog. Brain Res., *184:* 199 (1980).
10. Hernandez, M. J., Brennan, R. W., and Hawkins, R. A.: Regional cerebral blood flow during neonatal asphyxia. In: Passoneau J. V., Hawkins R. A., Lust W. D., Welsh F. A.: p. 196 (Williams and Wilkins, Baltimore, 1980).
11. Hill, A., Perlman, J. M., and Volpe, J. J.: Relationship of pneumothroax to occurrence of intraventricular hemorrhage in the premature newborn. Pediatrics, *69:* 144 (1982).
12. Hill, A. and Volpe, J. J.: Decrease in pulsatile flow in the anterior cerebral arteries in infantile hydrocephalus. Pediatrics, *69:* 4 (1982).
13. Hoedt-Rasmussen, K., Sveinsdottir, E., and Lassen, N. A.: Regional cerebral blood flow in man determined by intra-arterial injection of radioactive inert gas. Circ. Res., *18:* 237 (1966).
14. Joyner, C. R.: Transcutaneous Doppler detector in the study of arterial and venous flow patterns. Cardiovasc. Clin., *6(3):* 385 (1975).
15. Karickhoff, A.: Simultaneous blood pressure and anterior cerebral artery flow velocity wave form analysis in the preterm infant. Pediatr. Res. (Abstract), *15:* 666 (1980).
16. Kennedy, C., Grave, G. D., Jehle J. W., and Sokoloff, L.: Changes in blood flow in the component structures of the dog brain during postnatal maturation. J. Neurochem. *19:* 2423 (1972).
17. Leahy, F. A. N., Sankaran, K., Cates, D., MacCallum, M., and Rigatto, H.: Quantitative noninvasive method to measure cerebral blood flow in newborn infants. Pediatrics, *64:* 277 (1979).
18. Lipman, B., Serwer, G., and Brazy, J. E.: Abnormal cerebral hemodynamics in preterm infants with patent ductus arteriosus. Pediatrics, *69:* 778 (1982).
19. Lou, H. C., Lassen, N. A., and Friis-Hansen, B.: Impaired autoregulation of cerebral blood flow in the distressed newborn infant. J. Pediatr., *94:* 118 (1979).
20. Ment, L. R., Ehrenkranz, E. R., Lange, R. C., Rothstein, P. T., and Duncan, C. C.: Alterations in cerebral blood flow in preterm infants with intraventricular hemorrhage. Pediatrics, *68:* 763 (1981).
21. Mills, C. J.: Measurement of pulsatile flow and flow velocity. In: Bergel D. H.: Cardiovascular Fluid Dynamics. Vol. 1, p. 51. (Academic Press, Inc., London, 1972).
22. Obrist, W., Thompson, H. K., King, H., and Wang, H.: Determination of regional cerebral blood flow by inhalation of 133-Xenon. Circ. Res., *20:* 124 (1967).
23. Peabody, J. L.: Mechanical ventilation of the newborn: . . . Good news . . . bad news. Crit. Care Med., *9:* 710 (1981).
24. Perlman, J. M., Hill, A., and Volpe, J. J.: The effect of patent ductus arteriosus on flow velocity in the anterior cerebral arteries: Ductal steal in the premature newborn infant. J. Pediatr., *99:* 767 (1981).
25. Perlman, J. M. and Volpe, J. J.: The effects of oral suctioning and endotracheal suctioning on cerebral blood flow velocity and intracranial pressure in the preterm infant. Pediatr. Res. (Abstract) *16(4):* 303A (1982).
26. Perlman, J. M. and Volpe, J. J.: The effects of seizures on cerebral blood flow velocity, intracranial pressure, and systemic blood pressure in the preterm infant. Pediatr. Res., (Abstract) *16(4):* 339A (1982).
27. Pourcelot, L.: Applications cliniques de l'examen Doppler transcutane. In: Peronneau P. Velocimetre Ultrasonore Doppler. p. 213. (INSERM, Paris, 1975).
28. Risberg, J. and Smith, P.: Prediction of hemispheric blood flow from carotid velocity measurements. Stroke, *11:* 399 (1980).
29. Rosenkrantz, T. S. and Oh, W.: Cerebral blood flow velocity in infants with polycythemia and hyperviscosity: efects of partial exchange transfusion with plasmanate. J. Pediatr., *101:* 94 (1982).
30. Rudolph, A. M. and Heymann, M. A.: The circulation of the fetus in utero. Circ. Res., *21:* 163 (1967).
31. Sakurada, O., Kennedy, C., Jehle, J., Brown, J. D., Carbin, G. L., and Sokoloff, L. Measurement of local cerebral blood flow with iodo[^{14}C]antipyrine. Am. J. Physiol., *234(1):* H59 (1978).
32. Shapiro, H. M., Greenberg, J. H., Van Horn Naughton, K., and Reivich, M.: Heterogeneity of local cerebral blood flow—$Paco_2$ sensitivity in neonatal dogs. J. Appl. Physiol., *49(1):* 113 (1980).
33. Strandness, D. E.: The use of ultrasound in the evaluation of peripheral vascular disease. Prog. Cardiovasc. Dis., *20:* 403 (1978).
34. Uematsu, S.: Determination of volume of arterial blood flow by an ultrasonic device. J. Clin. Ultrasound, *9:* 209 (1981).
35. Volpe, J. J., Perlman, J. M., Hill, A., and McMenamin, J. B.: Cerebral blood flow velocity in the human newborn: the value of its determination. Pediatrics, *70:* 147 (1982).
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37. Requests for reprints should be addressed to: Dr. Daniel Batton, Department of Pediatrics, The M. S. Hershey Medical Center, Hershey, PA 17033.
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Distribution of Trace Elements and Minerals in Human and Cow's Milk

GUN-BRITT FRANSSON[37] AND BO LÖNNERDAL[38,40]

Institute of Nutrition, University of Uppsala, Sweden and Department of Nutrition, University of California, Davis, California, USA

Summary

The concentration of iron in cow's milk, 0.40–0.59 μg/ml, was found to be very similar to that of human milk, 0.20–0.69 μg/ml. The copper concentration of cow's milk (0.06–0.09 μg/ml) is lower than in human milk (0.24–0.50 μg/ml) whereas the concentration of zinc is higher in cow's milk (3.23–5.15 μg/ml) than in human milk (1.16–3.83 μg/ml). Cow's milk contains about 4–5 times more calcium and magnesium, 854–1430 μg/ml and 87–131 μg/ml, respectively, than human milk (220–252 μg/ml and 26–35 μg/ml). Cow's milk was fractionated and the trace element and mineral contents of the different fractions were compared to results from human milk. The casein fraction in cow's milk contains a large proportion of the total amounts of the elements cited above (Fe 24%, Cu 44%, Zn 84%, Ca 41%, Mg 25%) whereas human casein only binds minor amounts (Fe 9%, Cu 7%, Zn 8%, Ca 6%, Mg 6%). Whey proteins bind a major part of these elements in human milk, but not in cow's milk. Significant amounts of iron are bound to the lipid fraction in both cow's and human milk (14 and 33%, respectively), predominantly bound to the outer fat globule membrane. Low molecular weight compounds (ligands) bind significant proportions of all the elements investigated in both cow's and human milk, with the exception of zinc in cow's milk, of which only 2% is associated with this fraction.

The differences in trace element bioavailability observed in

infants fed human milk versus cow's milk (formula) may therefore be explained, in part, by differences in ligand binding; the differences in distribution between the casein and whey proteins may be especially important. It is possible that casein may act as a limiting factor for the absorption of trace elements and minerals in the newborn infant with limited digestive function.

Abbreviations

IFGM, inner fat globule membrane
LMW, low molecular weight
OFGM, outer fat globule membrane
SDS, sodium dodecyl sulfate

The trace elements iron, copper and zinc are essential for normal growth and development in infants (18, 32, 33). Calcium and magnesium are likewise of critical importance for the growing infant. Because the only food for the newborn infant is breast milk or a formula, usually based on cow's milk, it is important that the milk or formula provide adequate amounts of these elements in order to prevent deficiency. Trace element deficiency is very rare in breast-fed infants, compared with infants fed cow's milk or an unsupplemented formula (6, 7, 8). Because the concentration of trace elements in cow's milk or formula is similar or higher than in human milk (23, 24), it is reasonable to assume a higher availability of these elements from human milk than from cow's milk or formula. This has been demonstrated for iron by using radioisotope methods (27, 30). Some reports suggest that this is also the case for zinc (4, 14, 31). We have previously described the distribution of iron (10), copper, zinc, calcium, and magnesium (11) in human milk and in human milk fat (12). Those studies were carried out as an attempt to identify the compound or compounds associated with these trace elements and minerals, which are at least partly responsible for their high bioavailability. In this study we have determined the distribution of iron, copper, zinc, calcium, and magnesium among different fractions of cow's milk and compared the results with extended fractionations of human milk.

MATERIALS AND METHODS

Milk samples. Bovine milk samples (0.5–4 months of lactation) were obtained from lactating Holstein cows at the Department of Animal Science, University of California at Davis. Human milk samples were obtained from healthy mothers at different stages of lactation (0.5–12 months) by emptying one breast with a manual pump (Egnell's Hand Pump, Eklund & Co., Davis, CA) at the second nursing of the day, mixing the milk and taking an aliquot. Fresh milk samples were fractionated and analyzed immediately after being taken.

Fractionation procedure. Milk samples were fractionated into fat and skim milk as earlier described (10) (Fig. 1), and the skim milk was ultracentrifuged to obtain a casein pellet. The whey was subsequently ultrafiltered to obtain a fraction with molecular weight less than 10,000 (LMW fraction) and a whey protein fraction. The fat was separated into OFGM, IFGM, and triglycerides. This was achieved by treating the fat fraction with an equal amount of 2% SDS at 40°C for 1 h. This method is a slight modification of the procedure used by Bianco (2). After centrifugation at 15,000 × *g* for one h at 37°C, the SDS solution containing the OFGM could be separated by gentle pipetting. The flotation layer was melted at 60°C for 1 h and then centrifuged at 47°C for 20 min at 15,000 × *g*. After cooling, the triglycerides could be separated from the IFGM fraction with a spatula.

Analytical procedure. Duplicate samples of whole milk and the various fractions were wet-ashed (5) and analyzed by atomic absorption spectrophotometry (Perkin-Elmer 370 and Unicam 2900). Recovery was 92–103% and the coefficient of variation 0.4–5.8%.

RESULTS

The concentration of iron in cow's milk is very similar to that of human milk (Table 1). The copper content of cow's milk is lower than in human milk whereas the concentration of zinc is higher in cow's milk than in human milk. As can be seen, cow's milk contains about 4–5 times more calcium and magnesium, respectively, than human milk.

The milk fat contains a considerable amount of iron both in cow's and human milk, 14% and 33%, respectively (Table 2). Only 2% of the copper in cow's milk is found in the fat whereas in human milk 15% is found in this fraction. Comparatively small proportions of zinc, calcium, and magnesium are found in the fat fraction.

The pellet fraction, which contains mostly casein, contains considerable proportions of all the elements analyzed in cow's milk whereas this fraction contains minor parts of these elements in human milk. The whey proteins, on the other hand, bind comparatively smaller proportions of trace elements and minerals in cow's milk than in human milk.

Low molecular weight compounds (ligands) bind significant proportions of the elements investigated both in cow's and human milk, with the exception of zinc in cow's milk, of which only 2% is associated with this fraction.

The majority of the elements found in the milk fat fraction was found in the OFGM in both cow's and human milk (Table 3). Only-minute amounts of these elements were found in the triglyceride ("core") fraction.

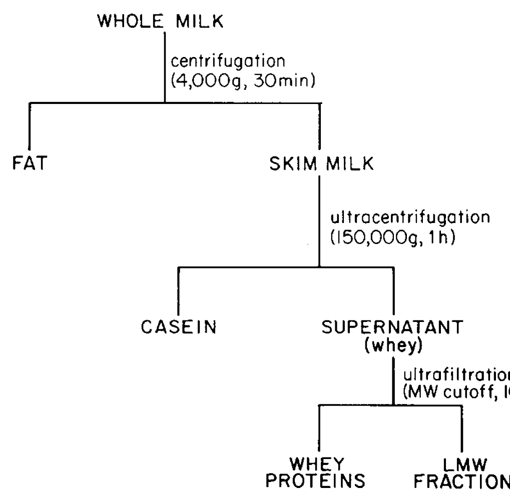

Fig. 1. Fractionation procedure.

Table 1. *Total concentration of trace elements and minerals in human and cow's milk (µg/ml)*

	(*n*)	x̄ ± S.E.M.	Range
Fe			
Human	(41)	0.36 ± 0.03	0.10–0.82
Cow's	(9)	0.29 ± 0.06	0.11–0.59
Cu			
Human	(30)	0.27 ± 0.02	0.11–0.50
Cow's	(9)	0.10 ± 0.02	0.02–0.21
Zn			
Human	(30)	1.45 ± 0.25	0.35–5.70
Cow's	(9)	3.50 ± 0.32	2.02–5.15
Ca			
Human	(20)	279.20 ± 28.60	172.31–772.14
Cow's	(9)	1170.60 ± 68.50	854.50–1430.20
Mg			
Human	(23)	35.04 ± 2.46	18.50–63.60
Cow's	(9)	117.39 ± 5.10	86.96–131.65

Table 2. *Distribution of trace elements and minerals among major milk fractions*

	Fe		Cu		Zn		Ca		Mg	
Fat (%)										
Human	(37)[1]	33 ± 2[2]	(31)	15 ± 1	(32)	18 ± 2	(26)	16 ± 4	(23)	2 ± 0
Cow's	(9)	14 ± 2	(6)	2 ± 1	(9)	1 ± 0	(9)	1 ± 0	(9)	0
Pellet (%)										
Human	(30)	9 ± 2	(27)	7 ± 2	(29)	8 ± 2	(25)	6 ± 2	(20)	6 ± 1
Cow's	(9)	24 ± 5	(6)	44 ± 8	(9)	84 ± 3	(9)	41 ± 4	(9)	25 ± 3
Whey protein (%)										
Human	(20)	26 ± 2	(18)	56 ± 4	(18)	51 ± 4	(20)	41 ± 3	(19)	36 ± 3
Cow's	(9)	29 ± 9	(6)	8 ± 4	(9)	13 ± 3	(9)	30 ± 3	(9)	31 ± 3
LMW (%)										
Human	(20)	32 ± 3	(17)	21 ± 3	(18)	19 ± 3	(20)	38 ± 2	(19)	58 ± 3
Cow's	(9)	32 ± 5	(6)	47 ± 11	(9)	2 ± 1	(9)	28 ± 3	(9)	44 ± 3

[1] Total number of samples (n).
[2] $\bar{x}$ ± S.E.M.

Table 3. *Distribution of trace elements and minerals within the fat fraction (% of total amount in fat fraction)*

	Fe		Cu		Zn		Ca		Mg	
OFGM[1] (%)										
Human	(25)[2]	64 ± 3[3]	(18)	72 ± 3	(17)	64 ± 3	(17)	67 ± 3	(16)	76 ± 4
Cow's	(9)	69 ± 5	(6)	71 ± 7	(9)	70 ± 9	(6)	74 ± 3	(6)	0
IFGM[4] (%)										
Human	(25)	26 ± 3	(18)	16 ± 2	(19)	25 ± 3	(17)	24 ± 3	(16)	20 ± 4
Cow's	(9)	26 ± 6	(6)	24 ± 3	(9)	26 ± 9	(6)	26 ± 3	(6)	0
Core (%)										
Human	(25)	10 ± 2	(18)	12 ± 3	(19)	11 ± 3	(17)	9 ± 3	(16)	4 ± 2
Cow's	(9)	5 ± 2	(6)	5 ± 2	(9)	4 ± 2	(6)	0	(6)	0

[1] Outer fat globule membrane.
[2] Total number of samples (n).
[3] $\bar{x}$ ± S.E.M.
[4] Inner fat globule membrane.

DISCUSSION

The values obtained for iron, copper, zinc, calcium, and magnesium in cow's milk in this study, are very similar to those reported by other investigators (23, 28, 34, 35, 36). The wide variation in concentration for each element among the samples most likely reflect individual differences and varying lactation time. There are pronounced changes in concentrations of iron, copper, and zinc during the lactation period (23); however, as no significant changes in distribution among fractions was found, data were grouped. It should be noted that the major changes in mineral concentrations occurs during early lactation (28, 29, 30) and that in this study only mature milk was studied. In order to study the distribution of trace elements and minerals in milk, a new type of fractionation procedure was developed (Fig. 1). Classical procedures for milk fractionation (skimming, churning, acid-precipitation etc.) are often inadequate and cause re-distribution of these elements. The methods used in this paper are non-invasive and give the separations required. We have previously reported that a significant percentage (30%) of the iron in human milk is found in the fat fraction (10). Similar observations have been made for cow's milk, which is in agreement with the findings of King *et al.* (20). By using the solubilization procedure for the fat globules, we found that iron is not associated with the fat *per se*, but rather to the membrane proteins of the outer membrane surrounding the fat globule. We have preliminary evidence that a major part of the iron in the fat fraction of human milk is bound to the enzyme xanthine oxidase (12); it has been suggested that iron in cow's milk fat is also bound to this enzyme (29). In addition, our preliminary studies indicate that zinc in this membrane is bound to alkaline phosphatase (12), a zinc metalloenzyme also known to be present in the human milk fat.

Casein, which is by far the major component of the pellet fraction, binds significant amounts of trace elements and minerals in cow's milk, but only very minor amounts in human milk. The procedure used in this study (ultracentrifugation) separates casein in the form of micelles only. Using this method, no detectable amount of free casein subunits was found in the supernatants from the ultracentrifugation of human milk whereas in cow's milk very small amounts of casein subunits were found. The method commonly used to isolate casein, by acid precipitation, was found to be inappropriate for this study because the change in pH causes re-distribution of metal ions (16, 21). The binding of these elements to casein is most likely explained by the presence of phosphoserine groups on the casein molecule, which have been demonstrated to bind cations like calcium (13) and iron (17). The larger proportions of the elements bound to casein in cow's milk can most likely be explained by the considerably higher concentration of casein in cow's milk than in human milk (15).

A considerably smaller proportion of the iron in cow's milk than in human milk is bound to the whey protein fraction. We have previously shown that the iron in the whey protein fraction of human milk is bound to lactoferrin (10). This protein is present in much lower concentration in cow's milk than in human milk, which may partly explain why less iron is bound to this fraction in cow's milk. Zinc and copper in human milk is to a large extent bound to whey proteins. We have recently shown that the protein in whey binding zinc and copper is identical to serum albumin (22), which is a non milk-specific protein passively transferred to the milk from the blood. Despite the fact that serum albumin is of the same concentration in both milks, this protein is only of significance as a zinc binding compound in human milk. These observations, together with the difference in casein content, may explain the larger proportion of zinc and copper in the whey protein fraction of human milk.

Significant proportions of the elements studied were found in the LMW fraction, with the exception of zinc in cow's milk. Within this fraction, the trace elements are associated to LMW ligands; this form has been suggested to render these elements easily accessible for absorption (23). The LMW ligand binding zinc in human milk is citrate (25). It is possible that citrate, which has high binding constants for calcium, iron, copper, and magnesium as well, is the LMW ligand binding these cations too. A high binding constant of a LMW ligand can cause less zinc to be bound to proteins which may inhibit absorption (16, 20). Although the identification of the LMW ligand in human milk has been controversial (19), the initial report of citrate being this ligand has been verified by several other investigators (1, 3, 26). The presence or zinc-binding properties in milk of other suggested ligands, such as picolinic acid (9), has not been confirmed outside the laboratories of their initial report.

A thorough investigation of the characteristics of the different compounds binding trace elements and minerals in cow's milk versus human milk may aid in the elucidation of the mechanisms of trace element absorption. Such knowledge is essential for the design of formulas in order to optimize the availability of trace elements and minerals.

REFERENCES AND NOTES

1. Arver, S.: Zinc and zinc ligands in human seminal plasma. III. The principal low molecular weight zinc ligands in prostatic excretion and seminal plasma. Acta Physiol. Scand., *116:* 67 (1982).
2. Bianco, D. H.: Lipid composition of polyunsaturated and conventional bovine milk fat globules. Masters Thesis, Univ. of California, 1972.
3. Blakeborough, P., Salter, D. N., and Gurr, W. I.: Zinc binding in human and cow's milk. Biochem. J., *209:* 505 (1983).
4. Casey, C. E., Walravens, P. A., and Hambidge, K. M.: Availability of zinc: loading tests with human milk, cow's milk, and infant formulas. Pediatrics, *68:* 394 (1981).
5. Clegg, M. S., Keen, C. L., Lönnerdal, B., and Hurley, L. S.: Influence of ashing techniques on the analysis of trace elements in animal tissue. I. Wet ashing. Biol. Trace Elem. Res., *3:* 107 (1981).
6. Committee on Nutrition, American Academy of Pediatrics: Iron supplementation for infants. Pediatrics, *58:* 765 (1976).
7. Committee on Nutrition, American Academy of Pediatrics: Zinc. Pediatrics, *61:* 408 (1978).
8. Cordano, A.: Copper deficiency in clinical medicine. in *Zinc and Copper in Clinical Medicine*, Hambidge K. M., Nichols, B. L., Jr. (eds). p. 119 (SP Medical and Scientific Books, New York, NY, 1978).
9. Evans, G. W. and Johnson, P. E.: Characterization and quantitation of a zinc binding ligand in human milk. Pediatr. Res., *14:* 876 (1980).
10. Fransson, G.-B. and Lönnerdal, B.: Iron in human milk. J. Pediatr., *96:* 380 (1980).
11. Fransson, G.-B. and Lönnerdal, B.: Copper, zinc, calcium and magnesium in human milk. J. Pediatr., *101:* 504 (1982).
12. Fransson, G.-B. and Lönnerdal, B.: Iron, copper, zinc, calcium and magnesium in human milk fat. Am. J. Clin. Nutr., (in press).
13. Greenberg, R., Groves, M. L., and Peterson, R. F.: Amino terminal sequence and location of phosphate groups of the major human casein. J. Dairy Sci., *59:* 1016 (1976).
14. Hambidge, K. M., Walravens, P. A., Casey, C. E., Brown, R. M., and Bender, C.: Plasma zinc concentrations of breast-fed infants. J. Pediatr., *94:* 607 (1979).
15. Hambraeus, L., Lönnerdal, B., Forsum, E., and Gebre-Medhin, M.: Nitrogen and protein components of human milk. Acta Paediatr. Scand., *67:* 561 (1978).
16. Harzer, G. and Kauer, H.: Binding of zinc to casein. Am. J. Clin. Nutr., *35:* 981 (1982).
17. Hegenauer, J., Saltman, P., Ludwig, D., Ripley, L., and Ley, A.: Iron-supplemented cow milk. Identification and spectral properties of iron bound to casein micelles. J. Agric. Food Chem., *27:* 1294 (1979).
18. Hurley, L. S.: Developmental Nutrition. pp. 335 (Prentice-Hill, Englewood Cliffs, NJ 1979).
19. Hurley, L. S. and Lönnerdal, B.: Zinc binding in human milk: Citrate versus picolinate. Nutr. Rev., *40:* 65 (1982).
20. King, R. L., Luick, J. R., Litman, I. I., Jennings, W. G., and Dunkley, W. L.: Distribution of natural and added copper and iron in milk. J. Dairy Sci., *42:* 780 (1959).
21. Lönnerdal, B. and Forsum, E.: Casein content of human milk. Proc. 3rd Eur. Nutr. Conf. Var Foda, (suppl. 3) *31:* 239 (1979).
22. Lönnerdal, B., Hoffman, B., and Hurley, L. S.: Zinc- and copper-binding proteins in human milk. Am. J. Clin. Nutr., *36:* 1170 (1982).
23. Lönnerdal, B., Keen, C. L., and Hurley, L. S.: Iron, copper, zinc and manganese in milk. Ann. Rev. Nutr., *1:* 149 (1981).
24. Lönnerdal, B., Keen, C. L., Ohtake, M., and Tamura, T.: Iron, zinc, copper, and manganese in infant formulas. Am. J. Dis. Child., *137:* 433 (1983).
25. Lönnerdal, B., Stanislowski, A. G., and Hurley, L. S.: Isolation of a low molecular weight zinc binding ligand from human milk. J. Inorg. Biochem., *12:* 71 (1980).
26. Martin, M. T., Licklider, K. F., Brushmiller, J. G., and Jacobs, F. A.: Detection of low molecular weight copper (II) and zinc (II) binding ligands in ultrafiltered milks—the citrate connection. J. Inorg. Biochem., *15:* 55 (1980).
27. McMillan, J. A., Oski, F. A., and Lourie, G.: Iron absorption from human milk, simulated human milk, and proprietory formulas. Pediatrics, *60:* 896 (1977).
28. Picciano, M. F. and Guthrie, H. A.: Copper, iron and zinc contents of mature human milk. Am. J. Clin. Nutr., *29:* 242 (1976).
29. Richardson, T. and Guss, P. S.: Lipids and metals in fat globule membrane fractions. J. Dairy Sci., *48:* 523 (1965).
30. Saarinen, U. M., Siimes, M. A., and Dallman, P. R.: Iron absorption in infants: high bioavailability of breast milk iron as indicated by the extrinsic tag method of iron absorption and by the concentration of serum ferritin. J. Pediatr., *91:* 36 (1977).
31. Sandström, B. M., Cederblad, Å., and Lönnerdal, B.: Zinc absorption from human milk, cow's milk and infant formulas. Am. J. Dis. Child, *137:* 726 (1983).
32. Shaw, J. C. L.: Trace elements in the fetus and young infant. I. Zinc. Am. J. Dis. Child, *133:* 1260 (1979).
33. Shaw, J. C. L.: Trace elements in the fetus and young infant. II. Copper, maganese, selenium and chromium. Am. J. Dis. Child., *134:* 74 (1980).
34. Siimes, M. A., Vuori, E., Kuitunen, P.: Breast milk iron—a declining concentration during the course of lactation. Acta Paediatr. Scand., *68:* 29 (1979).
35. Vaughan, L. A., Weber, C. W., and Kemberling, S. R.: Longitudinal changes in the mineral content of human milk. Am. J. Clin. Nutr., *32:* 2301 (1979).
36. Vuori, E. and Kuitunen, P.: The concentrations of copper and zinc in human milk. Acta Paediatr. Scand., *68:* 33 (1979).
37. Research Assistant, Ph.D., Institute of Nutrition, University of Uppsala, Sweden.
38. Associate Professor, Ph.D., Department of Nutrition, University of California, Davis.
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40. Requests for reprints should be addressed to: Bo Lonnerdal, Ph.D., Department of Nutrition, University of California, Davis, CA 95616 (USA).
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