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Brief Report

Iron Supplementation in Pregnancy and Breastfeeding and Iron, Copper and Zinc Status of Lactating Women From a Human Milk Bank

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Summary

This study evaluated the influence of iron supplementation in pregnancy and breastfeeding on iron status of lactating women from a Brazilian Human Milk Bank. Blood and mature breast milk samples were collected from 145 women for assessment of iron status, as well as copper and zinc status. Haemoglobin, serum iron and ferritin were determined, respectively, by electronic counting, colorimetry and chemiluminescence. Transferrin and ceruloplasmin were analysed by nephelometry. Serum copper and zinc were measured by atomic absorption spectrophotometry, and serum alkaline phosphatase was measured by a colorimetric method. Iron, zinc and copper in breast milk were determined by spectrometry. Mean values of iron, copper and zinc (blood and breast milk) were compared by ANOVA, followed by Tukey's test. Iron supplementation was beneficial to prevent anaemia in pregnancy but not effective to treat anaemia. During breastfeeding, iron supplementation had a negative effect on maternal copper status, confirming an interaction between these micronutrients.

Key words: iron supplementation, breast milk, copper, zinc, human milk bank.

Introduction

Iron supplementation is recommended to minimize the harmful effects of iron-deficiency anaemia in pregnant and lactating women [1], and in their breastfed infants [2, 3]. However, the issue remains controversial considering that a potentially negative aspect of iron supplementation is the risk of inhibiting the absorption of other micronutrients, like copper and zinc [4–11].

The objective of this study was to evaluate the influence of iron supplementation, on iron, copper and zinc concentrations in blood and breast milk of lactating women.

Materials and methods

This study is part of a larger project involving healthy women who donated breast milk to a

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human milk bank in Marília city, Brazil [12]. A total of 161 donors producing mature milk (20–62 days after delivery) participated in the study in October 2003–August 2004.

The donors were asked whether they had used iron supplements in pregnancy and/or breastfeeding to prevent or treat anaemia. They were divided into three groups, according to the use of iron supplements in pregnancy ($FeST_{treat}$—to treat anaemia; $FeSP_{prevent}$—to prevent anaemia; noFeS—no iron supplementation), and in two groups, according to the use of iron supplements in breastfeeding (FeSBF—iron supplementation; noFeSBF—no iron supplementation).

Breast milk samples were collected manually, by the lactating women, over a period of 24 h. The women were instructed to collect small samples of milk, from both breasts, before, during and after feedings. On the morning of the day of breast milk collection, blood samples were withdrawn for biochemical and haematological analyses.

The minerals were determined in breast milk by spectrometry [13]. The cut-off points to define low levels of iron, copper and zinc in breast milk were <0.27 mg/L, <0.2 mg/L and <2 mg/L, respectively [14]. Haemoglobin (Hb) was measured by electronic counting, and the cut-off point to define anaemia was <120 g/L [1]. Serum iron (references 9–27 µmol/L) and ferritin (references 10–291 µg/L) were measured by colorimetric assay and chemiluminescence, respectively. Transferrin and ceruloplasmin were analysed by nephelometry, reference values 2.12–3.60 g/L and 250–630 mg/L, respectively. Serum concentrations of copper (references 13–24 µmol/L) and zinc (references 11–18 µmol/L) were determined by graphite furnace atomic absorption spectrophotometry. Alkaline phosphatase was measured by a colorimetric method, reference value up to 270 U/L.

The mean values for iron, copper and zinc parameters (in blood and breast milk) were compared by analysis of variance (ANOVA), followed by Tukey's test. Comparison between two means was determined by the student's *t*-test. The level of significance was set at 5%.

The study was approved by the Ethics Committee of the School of Public Health, USP, Brazil. All participants gave written informed consent.

Results

The final sample consisted of 145 women: 2 were lost to follow-up, and 14 had incomplete data.

The characteristics of the women are shown in Table 1. Table 2 shows mean concentrations of the haematological and biochemical parameters investigated for the $FeST_{treat}$, $FeST_{prevent}$ and noFeST groups. Mean levels of Hb were significantly different among the groups. Table 3 shows mean concentrations of the same parameters cited in Table 2 for the FeBF and noFeBF groups. Mean serum levels of ceruloplasmin ($p = 0.03$) and copper ($p = 0.02$) were significantly lower in the FeBF women than in the noFeBF women.

TABLE 1
Characteristics of the lactating women

Variables	*N* (%)
Age (years)	
16–19	20 (12.6)
20–29	90 (56.6)
30–41	49 (30.8)
Marital status	
Married/Stable union	134 (84.3)
Single/Divorced	25 (15.7)
Occupation	
Worker	84 (52.8)
Unemployed or house keeper	75 (47.2)
Education (years)	
0–8.0	54 (34.0)
8.1–11.0	83 (52.2)
>11	22 (13.8)
Family income (BMW[a])	
0–4.0	60 (37.7)
>4.0	99 (62.3)
Parity	
Primiparous	90 (56.6)
Multiparous	69 (43.4)
Smoking	
Yes	23 (14.5)
No	136 (83.5)
BMI (kg/m^2)	
17.0–25.0	91 (57.2)
25.1–30.0	50 (31.4)
30.1–41.0	18 (11.3)
Timing of breast milk donation (days)	
20–30	74 (46.5)
31–40	40 (25.1)
41–65	45 (28.3)
Total	159 (100.0)

[a]BMW, Brazilian minimum wage (1 BMW = 82 US dollars).

Discussion

Anaemia was observed in 21.4% of the women, and 44.8% of them had low iron in breast milk. These data are worrisome because those women, milk bank donors, presented a nutritional risk similar to malnourished lactating women from rural Africa [15].

Iron supplementation apparently was not effective in treating anaemia in pregnant women, although effective in preventing it. Iron supplements are given free of charge to Brazilian pregnant women, but we do not know about compliance to the treatment.

There were no differences in the concentrations of iron in breast milk among the women supplemented and not supplemented with iron in pregnancy and

TABLE 2
Comparison of haematological and biochemical variables in three groups of lactating women, according to iron supplementation in pregnancy

Haematological and biochemical variables	Groups of women			p-value[a]
	$FeST_{treat}$	$FeST_{prevent}$	noFeST	
Hb (g/L)	119 ± 12 (A)[a]	133 ± 13 (B)	126 ± 14 (B)	<0.001
Serum ferritin (μg/L)	56.1 ± 43.5	53.8 ± 33.1	44.8 ± 38.3	0.57
Serum iron (μmol/L)	13.6 ± 4.2 (A)	17.1 ± 7.0 (B)	14.6 ± 6.0	0.06
Transferrin (g/L)	2.6 ± 0.4 (A)	2.4 ± 0.3	2.3 ± 0.5 (B)	0.06
Ceruloplasmin (ml/L)	359 ± 55	340 ± 54	353 ± 61	0.27
Serum copper (μmol/L)	17.7 ± 2.9	17.2 ± 2.7	16.8 ± 2	0.54
Serum zinc (μmol/L)	13.8 ± 2.1	14.1 ± 1.4	14 ± 1.6	0.61
Alkaline phosphatase (U/L)	0.25 ± 0.16	0.4 ± 0.4	0.3 ± 0.2	0.08
Milk iron (mg/L)	0.25 ± 0.16	0.4 ± 0.4	0.3 ± 0.2	0.08
Milk copper (mg/L)	0.3 ± 0.1	0.3 ± 0.1	0.3 ± 0.1	0.93
Milk zinc (mg/L)	2.8 ± 1.1	2.6 ± 1.1	2.7 ± 1.3	0.85

Data are expressed as mean ± standard deviation.
[a]Values followed by the same letter are not significantly different (ANOVA and Tukey's test).
$FeST_{treat}$, iron supplementation to treat anaemia; $FeST_{prevent}$, iron supplementation to prevent anaemia; noFeST, no iron supplementation.

TABLE 3
Comparison of haematological and biochemical variables in two groups of lactating women, according to iron supplementation during breastfeeding

Haematological and biochemical variables	Groups of women		p-value[a]
	FeBF ($n = 29$)	noFeBF ($n = 116$)	
Hb (g/L)	131 ± 13	130 ± 14	0.28
Serum ferritin (μg/L)	58.6 ± 43.8	51.5 ± 32.9	0.17
Serum iron (μmol/L)	15 ± 5.4	16.5 ± 6.9	0.14
Transferrin (g/L)	2.4 ± 0.3	2.4 ± 0.3	0.24
Ceruloplasmin (ml/L)	327 ± 50	348 ± 55	0.03
Serum copper (μmol/L)	16.3 ± 2.4	17.4 ± 2.7	0.02
Serum zinc (μmol/L)	14.1 ± 1.5	14.1 ± 1.6	0.46
Alkaline phosphatase (U/L)	159.8 ± 43.62	161.0 ± 45.38	0.79
Milk iron (mg/L)	0.3 ± 0.2	0.4 ± 0.4	0.37
Milk copper (mg/L)	0.3 ± 0.1	0.3 ± 0.1	0.41
Milk zinc (mg/L)	2.5 ± 1.1	2.7 ± 1.1	0.16

Data are expressed as mean ± standard deviation.
[a]Student's *t*-test.
FeBF, iron supplementation during breastfeeding; noFeBF, no iron supplementation during breastfeeding.

breastfeeding, and no associations between those concentrations and any of the iron, copper and zinc parameters in blood (data not shown). Nakamori *et al.* [16] referred that iron in breast milk of Vietnamese women was not associated with serum iron and ferritin. Domellöf *et al.* [17] reported that iron, zinc and copper in breast milk in postpartum month 9 were not associated with maternal mineral status. Apparently iron in breast milk is controlled by a mechanism that seems to prioritize the babies in detriment of the mothers [16, 18–20], at least in mothers with no moderate/severe anaemia [21].

There were lower concentrations of serum ceruloplasmin and copper in the FeBF compared with the noFeBF women. Iron metabolism interacts with the metabolism of zinc and copper, mainly through the divalent metal transporter 1 (DMT1) [22–25] and human copper transporter hCTR1 [26]. In humans, iron–folate supplements decreased serum copper concentrations [27]. However, in an earlier study from

Nigeria [28], iron supplementation in pregnancy was not associated with copper and zinc status.

There was no difference in zinc status, assessed by serum zinc and alkaline phosphatase concentrations in women supplemented and not supplemented with iron in pregnancy. Our data are similar to the study of Sheldon *et al.* [29], in which women followed before conception throughout pregnancy and at 12 weeks postpartum showed no impact of iron supplementation on zinc status. However, our data are in disagreement with the study of O'Brien *et al.* [9], who found that women receiving iron supplements presented with zinc malabsorption and decreased serum concentrations of zinc.

Iron supplementation did not seem to influence the levels of copper and zinc in breast milk. In a study involving children 6–9 months of age (rather than lactating women), receiving long-term iron supplementation, Domellöf *et al.* [30] did not find associations between iron supplementation and absorption of zinc and copper.

Limitations of this study are the cross-sectional design, and that we had to rely on mothers' recall of iron supplements use in pregnancy and breastfeeding. However, it is important to emphasize that a high percentage of them had anaemia and low concentrations of iron in breast milk. In pregnancy, iron supplementation was apparently beneficial to prevent anaemia, and in lactation, supplementation had a negative effect on maternal copper status, confirming an interaction between those micronutrients.

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