Nutritional Status of Lactating Mothers and Their Breast Milk Concentration of Iron, Zinc and Copper in Rural Vietnam

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Summary Breast milk is considered to be the best nutrient source for infants. However, nutritional compositions of breast milk in developing countries, especially among malnourished women, have not been fully investigated. This study aimed to assess nutritional status and nutrient composition of breast milk in lactating mothers in rural Vietnam. Sixty breastfeeding mothers at 6 to 12 mo postpartum, free from any medical disorder and/or medication, and not pregnant were randomly selected in Yen The, Bac Giang, Vietnam. Their nutritional status, breast milk concentration and dietary intakes were assessed. Among the study participants, anemia (39.0%) and low serum zinc concentration (55.4%) were frequently observed. Dietary assessment revealed lower intakes of iron (10.2±2.5 mg/d) and zinc (10.4±2.2 mg/d) than estimated requirements. The breast milk concentration of iron, zinc and copper was 0.43±0.15 mg/L, 0.56 (0.37, 0.82) mg/L and 0.19±0.05 mg/L, respectively. The breast milk concentration of iron, zinc and copper was not correlated to the serum concentration or dietary intakes. In conclusion, we uncovered a high prevalence of anemia and zinc deficiency in lactating mothers in rural Vietnam. The findings demonstrate a low breast milk zinc concentration among the participants, but need further investigation.

Key Words Vietnam, iron, zinc, copper, human milk

Deficiencies of iron, zinc and copper often coexist and have independent and interacting effects on health, growth and immunocompetence (1). Pregnant women, lactating women and young children are considered the most vulnerable population to this problem because of their high requirements. During infancy, iron, zinc and copper are essential for normal growth and development, therefore, deficiency of these nutrients is of public health concern (2–4). Since filling the nutritional requirement only from complementary food is difficult and non-breast milk food involves several problems in most of developed countries, World Health Organization (WHO) (5) and United Nations Children’s Fund (UNICEF) (6) have recommended continuous breastfeeding until for 2 years. However, information on the nutrition quality of breast milk has been not sufficient, especially among lactating mothers after 6 mo postpartum in developing countries.

The nutritional status of lactating mothers is an important health issue since their nutrition status may influence the nutrient concentration of breast milk (7), while maintaining the nutrients in the breast milk further depletes their own body stores (8). In Vietnam, however, no data has been reported on the nutrient composition of breast milk or nutrition status among lactating mothers after 6 mo postpartum.

Thus the present study was undertaken to analyze the breast milk concentration of iron, zinc and copper at 6 to 12 mo postpartum, and to analyze the nutrition adequacy among lactating mothers by anthropometric measurement, blood biochemical measurements and dietary assessment in rural Vietnam.

MATERIALS AND METHODS

Study participants. In May 2005, a cross-sectional survey was conducted in 4 communes in Yen The, Bac Giang, Vietnam. Bac Giang Province, a rural mountainous area, is located in the North-East region and is 51 km from Hanoi, the capital of Vietnam. A census was carried out in these 4 communes and 98 households having children aged 6 to 12 mo were identified. Among the households, 95 mothers met the following inclusion criteria: 1) currently breast feeding, 2) free from medical disorder and/or medication, 3) not pregnant, 4) not having other breastfed children, and 60 mothers were randomly selected for the study. Mothers who had serious disease, medication or the use of hormonal contraceptives were not found among the study participants.
The protocol of the study was approved by the Scientific Board of the National Institute of Nutrition of Vietnam and the Ethical Committee of the Tokushima University. Before conducting the study, written informed consent concerning the procedure and purpose of the study was obtained from every participant.

Data collection and analysis. General information such as demographic profiles, coverage of iron supplementation during pregnancy and after delivery, child’s birth weight and compliance with exclusive breast feeding were asked. Low birth weight was defined as the birth weight <2,500 g. Data collection concerning exclusive breast feeding status was based on the guideline of WHO (5, 9) and exclusive breast feeding was classified as giving only breast milk plus medical drops and syrups to the child.

Anthropometric measurements comprising height and weight were collected. Mothers were weighed with light clothes on a scale. Height was measured to the nearest 0.1 cm. Infants were weighed with light clothes on an infant scale. Recumbent length was measured to the nearest 0.1 cm by a portable infant measuring board. Anthropometric measurements were taken by 2 trained assistants.

Blood samples (6 mL) were collected in the morning under fasting conditions. Blood was obtained from the vein with a trace metal-free vacutube with a butterfly Luer-lock adapter (Becton, Dickinson Co., Japan). For the Hemoglobin (Hb) analysis, 200 μL of blood was placed into a plastic tube containing EDTA. Another blood sample was allowed to clot for 1 to 3 h at room temperature and centrifuged at 3,250 rpm for 10 min to obtain serum. The sera were stored at −70°C until the biochemical analysis. Breast milk samples (20 mL) were collected in the morning after the blood collection, and were expressed by hand into a plastic tube after wiping the hands and breast with 70% alcohol. Plastic tubes were washed in 0.1% nitric acid before being used and were tested and found negative for mineral contamination after being washed. The samples were frozen at −20°C until analysis.

Blood samples were analyzed for Hb, serum ferritin (SF), serum iron, serum zinc, serum copper and C-reactive protein (CRP). Hb was measured by the cyanmethemoglobin method within 12 h (Boehringer kits). SF was measured by a two-site enzyme-linked immunosorbent assay (Ramco, Houston, TX, USA) using monoclonal reagents for both the capture and indicator antibodies. CRP was measured by latex agglutination-turbidimetric immunoassay. Iron, zinc and copper concentration in serum and breast milk were measured by Inductively Coupled Plasma Atomic Emission Spectrometry (ICP-AES) using a sequential plasma spectrometer ICP-7500 system for simultaneous determination of multi-elements (10). Serum samples and delipidated milk samples were aliquoted into a teflon tube and covered with a teflon ball. After adding HNO₃ (Wako Pure Chemical Industries, Ltd., Osaka, Japan), the tube was heated on an aluminum-heating block (IWAKI, Asahi Techno Glass, Japan) at 120°C for 5 h. The samples were further heated almost to dryness at 200°C after removing the teflon ball. Finally, the residue was dissolved with 0.1 M HNO₃, which contained 10 ng/mL of internal standard elements (In, Re, Tl). The diluted solution was used for analysis by the ICP-AES. The multi-standard solutions for standardization of calibration curves were prepared from the single-element standard solutions (1,000 μg/mL), purchased from Wako Pure Chemical Industries, Ltd. All specimens were analyzed three times and the mean concentration was used. The CV among the analyses was <6.5%. The Second Generation Human Serum Reference Material (11) and NIST reference standard (SRM 8435-powdered dry whole milk) were activated along with samples for quality control. Milk samples were also analyzed for their total nitrogen by the Kjeldahl method. Protein contents were calculated from total nitrogen ×6.28. Lipid contents were analyzed by the Rose-Gottlieb method. Total solids in whole milk were measured after drying 1 mL aliquots of thoroughly mixed samples at 100±2°C for 3 h in porcelain crucibles until they reached constant weight. Ash was further incinerated at 450°C for 12 h. Carbohydrates were calculated as: Carbohydrates=total solids−proteins−lipids−ash. Gross total energy content was calculated as: Energy=protein×4+fat×9+carbohydrates×4 according to the Atwater general factor system.

Dietary intakes were collected by 24-h recall method for 3 non-consecutive days. The day of unusual food intake, feast day and the day of sickness which affected appetite were not included in the days of survey. Interviewers were centrally trained and certified in data collection according to standardized methods (12, 13). The level of detail needed to adequately describe the foods eaten in any 24-h recall was specified for the interviewers by using an integrated form so that information was obtained and recorded consistently and thoroughly. Serving size was determined by dimensional pictures developed for the food intake analysis system by the National Institute of Nutrition, Vietnam. Portion size and ingredients of local food specialized in the research area were determined by visiting their local markets and weighing foods shelved at the markets.

For calculating the nutrients contents of the diet, the nutritive composition table of Vietnamese foods (14); the Standard Tables of Food Composition in Japan, 5th edition (15) and the USDA National Nutrient Database for Standard Reference (16) were used. Daily energy, protein, zinc, calcium and vitamin C were compared with dietary reference intakes (DRIs) in Vietnam (17, 18). Since an iron requirement for lactating women has not been established in Vietnam, DRI for iron was referred from FAO/WHO in 2002 (19). Potential bioavailability of iron in participant’s diet was categorized by the model of FAO/WHO in 1988 (20) with criteria of DRIs in Vietnam (17, 18). The FAO/WHO model (20) is based on estimates of iron absorption from a typical meal in Asia, India, Latin America, and Western countries and classifies these typical meals into three broad
Iron deficiency was defined as SF concentration below assuming 15% of iron amount absorbed among healthy adults, is a simple, monotonous diet containing cereals, roots and/or tubers and negligible quantities of meat, fish (<30 g/d), or ascorbic acid-rich foods (<25 mg/d); an intermediate-bioavailability diet, assuming 10% of iron absorption, is a diet consisting mainly cereals, roots, and/or tubers and minimal quantities of food of animal origin (30–90 g/d) and ascorbic acid (25–75 mg/d), both of which promote iron availability; a high-bioavailability diet, assuming 15% of iron absorption, is a diversified diet containing generous quantities of meat, poultry, fish (>90 g/d), and/or foods containing high amounts of ascorbic acid (>75 mg/d). Potential bioavailability of zinc was categorized by the model of FAO/WHO in 2002 (19) with criteria of DRIs in Vietnam (16, 17): a low-bioavailability diet, assuming 15% of zinc amount absorbed among healthy adults, is when animal food intake is low and phytate-zinc molar ratio of total diet exceeds 15; a moderate-bioavailability diet, assuming 30% of zinc amount absorbed, is when a diet contains moderate amounts of animal food and the phytate-zinc molar ratio is 5 to 15; a high-bioavailability diet, assuming 50% of zinc amount absorbed, is when a diet contains adequate protein from non-vegetable sources and with the phytate-zinc molar ratio less than 5.

Statistical analysis. A database was established using Epi info version 6 (CDC, Atlanta, GA, USA). All data were checked for missing data and outliers, and cleaned before data analysis. Statistical analysis was performed by using SPSS version 11.5J (Statistical Package for Social Science, Inc.). A one-sample Kolmogorov-Smirnov test was used to assess whether the data were normally distributed. When the data were not normally distributed, statistical analysis was carried out after log transformation. Results are presented as means and standard deviation (SD) or median and 25th, 75th percentile. To compare the breast milk concentration between different times of lactation, an unpaired t-test was used. Correlation of breast milk concentration, nutrient status and nutrient intakes was determined by Pearson’s correlation coefficient. The anthropometric indicators among children, Z-scores of weight-for-age (WAZ), height-for-age (HAZ) and weight-for-height (WHZ), were calculated on the basis of recent WHO growth references (21). Malnutrition was classified according to the cut-off shown by WHO (21): underweight was determined as WAZ < -2, stunting as HAZ < -2, wasting as WHZ < -2. Chronic Energy Deficiency (CED) among the mothers was defined as Body Mass Index (BMI) below 18.5 kg/m². Anemia was defined as Hb level below 120 g/L (22). Iron deficiency was defined as SF concentration below 15 μg/L (23). Deficiencies of zinc and copper were defined when their serum level were below 70 mg/L (24) and 0.75 mg/L (24), respectively. Elevation for CRP was considered when the value was >10 mg/L.

RESULTS

Fifty-nine mothers participated in the survey and 90% of mothers were Kinh ethnics. Mean age of mothers was 25.0 ± 4.4 y (range 19 to 37 y) and mean age of their infants was 7.9 ± 1.6 mo (range 5.5 to 12.2 mo). Seven children (11.9%) were born at low birth weight. Approximately 17% of infants were exclusively breastfed in the first 4 mo of their life, but no one had continued it until the first 6 mo. Most of the mothers had less than 9 y of education (74.6%). Although distribution of 60 mg iron and 250 μg folic acid supplementation tablets was done weekly to women through pregnancy and the first month after delivery in these communities, only 25.4% mothers took the supplement by the 1st mo postpartum.

Nutrient status including anthropometric measurements and biochemical analysis of mothers are shown in Table 1. High prevalence of malnutrition among the study participants (CED 23.7%) and their infants (wasting 10.2%, underweight 18.6% and stunting 20.3%) was observed. Anemia was found in 39.0% of study participants. The percentage of subjects having lower serum zinc was 55.4% and lower serum copper was 21.4%. Among anemic women, 8.7% women concurrently had lower SF. 60.0% had lower serum zinc and 20.0% had lower serum copper. There was no difference in the mean concentration of Hb, SF, serum zinc and serum copper between mothers at 6 to 8 mo postpartum and mothers at 9 to 12 mo postpartum (p=0.159, 0.385, 0.676 and 0.346, respectively). No one had elevated CRP.

The breast milk nutrient analysis revealed that one litter of breast milk consists of 611 ± 117 kcal, 10.5 ± 1.7 g protein and 30.8 ± 12.6 g lipid. Breast milk concentrations of iron, zinc and copper are shown in Table 1. There was slight correlation between child’s age and breast milk concentration of zinc (r = 0.242, p = 0.073), and the breast milk concentration of zinc was significantly lower among the mothers at 9 to 12 mo postpartum than that of the mothers at 6 to 8 mo postpartum (p = 0.034).

The energy and nutrients intakes of the study participants along with DRI are given in Table 2. According to the criteria for categorizing potential availability of iron and zinc in a diet (17–20), bioavailability of iron in the participants’ diet was assumed as “intermediate” based on their median (25, 75 percentile) intake of meat; 85.7 (51.6, 135.0) g/d, while that of zinc was assumed as “low” based on a previous report on phytate-zinc molar ratio in Vietnam: 21.6 (23). Compared to the estimated requirements, iron and zinc intakes were insufficient among the study participants.

Breast milk iron concentration was not correlated with concentration of SF nor serum iron, nor iron intakes (r = -0.035, p = 0.793 and r = 0.037, p = 0.784 and r = -0.112, p = 0.398, respectively). Breast milk zinc concentration was not correlated with serum zinc concentration (r = 0.008, p = 0.954), but there was a slight correlation with zinc intake (r = 0.239,
Iron, Zinc and Copper in Breast Milk, Vietnam

Table 1. Anthropometry, blood analysis and breast milk analysis of the study participants.

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>6–8 mo (n=37)</th>
<th>9–12 mo (n=22)</th>
<th>Total (n=59)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Weight (kg)</td>
<td>45.4±5.0</td>
<td>44.4±4.5</td>
<td>45.0±4.8</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>151.9±4.7</td>
<td>149.9±3.9</td>
<td>151.2±4.5</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>19.7±1.9</td>
<td>19.7±1.6</td>
<td>19.7±1.8</td>
</tr>
<tr>
<td>%CED*</td>
<td>27.0 (10)</td>
<td>18.2 (4)</td>
<td>23.7 (14)</td>
</tr>
<tr>
<td>Blood analysis</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hb (g/L)</td>
<td>124.3±11.8</td>
<td>119.8±11.0</td>
<td>122.6±11.6</td>
</tr>
<tr>
<td>%&lt;120 g/L</td>
<td>35.1 (13)</td>
<td>45.5 (10)</td>
<td>39.0 (23)</td>
</tr>
<tr>
<td>SF (ug/L)</td>
<td>35.4±17.7</td>
<td>36.1±29.9</td>
<td>35.7±22.8</td>
</tr>
<tr>
<td>%&lt;15 μg/L</td>
<td>0.0 (0)</td>
<td>18.2 (4)</td>
<td>6.8 (4)</td>
</tr>
<tr>
<td>Serum Fe (mg/L)²</td>
<td>2.18±0.76</td>
<td>1.62±0.67</td>
<td>1.98±0.77</td>
</tr>
<tr>
<td>Serum Zn (mg/L)⁻¹</td>
<td>0.67 (0.61, 0.79)</td>
<td>0.70 (0.55, 0.80)</td>
<td>0.68 (0.60, 0.79)</td>
</tr>
<tr>
<td>%&lt;0.70 mg/L</td>
<td>61.1 (22)</td>
<td>45.0 (9)</td>
<td>55.4 (31)</td>
</tr>
<tr>
<td>Serum Cu (mg/L)</td>
<td>0.94±0.21</td>
<td>0.88±0.24</td>
<td>0.91±0.22</td>
</tr>
<tr>
<td>%&lt;0.75 mg/L</td>
<td>16.7 (6)</td>
<td>30.0 (6)</td>
<td>21.4 (12)</td>
</tr>
<tr>
<td>Breastmilk analysis</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Breastmilk Fe (mg/L)</td>
<td>0.40±0.14</td>
<td>0.46±0.15</td>
<td>0.43±0.15</td>
</tr>
<tr>
<td>Breastmilk Zn (mg/L)³</td>
<td>0.59 (0.47, 0.85)</td>
<td>0.38 (0.20, 0.73)</td>
<td>0.56 (0.37, 0.82)</td>
</tr>
<tr>
<td>Breastmilk Cu (mg/L)</td>
<td>0.19±0.05</td>
<td>0.18±0.05</td>
<td>0.19±0.05</td>
</tr>
</tbody>
</table>

*Mean±SD, ²Percent (number), ³Geometric mean (95%CI).

Due to lack of blood samples, n=36 in the 6–8 mo group and n=20 in the 9–12 mo group for the analysis of serum Fe, Zn and Cu.

³p<0.01 compared to women at 9–12 mo of lactation.

BMI, body mass index; CED, chronic energy deficiency; Hb, hemoglobin; SF, serum ferritin; Fe, iron; Zn, zinc; Cu, copper.

Table 2. Energy and nutrition intakes.

<table>
<thead>
<tr>
<th>Indicators</th>
<th>6–8 mo (n=37)</th>
<th>9–12 mo (n=22)</th>
<th>Total (n=59)</th>
<th>DRIs*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Energy (kcal/d)</td>
<td>2,051±277</td>
<td>1,962±336</td>
<td>2,018±301</td>
<td>2,975</td>
</tr>
<tr>
<td>Protein (g/d)</td>
<td>74.1±16.5</td>
<td>71.4±23.2</td>
<td>73.1±19.1</td>
<td>86–97</td>
</tr>
<tr>
<td>Animal protein (g/d)</td>
<td>26.7±15.0</td>
<td>27.3±17.4</td>
<td>27.0±15.8</td>
<td>30–35% of total protein</td>
</tr>
<tr>
<td>Iron (mg/d)</td>
<td>10.4±2.1</td>
<td>9.9±3.1</td>
<td>10.2±2.5</td>
<td>15</td>
</tr>
<tr>
<td>Iron from animal source (mg/d)</td>
<td>2.3±1.6</td>
<td>2.3±1.6</td>
<td>2.3±1.6</td>
<td>ND</td>
</tr>
<tr>
<td>Zinc (mg/d)</td>
<td>10.6±2.0</td>
<td>10.2±2.5</td>
<td>10.4±2.2</td>
<td>14.4</td>
</tr>
<tr>
<td>Copper (mg/d)</td>
<td>1.24±0.16</td>
<td>1.16±0.22</td>
<td>1.21±0.19</td>
<td>ND</td>
</tr>
<tr>
<td>Calcium (mg/d)</td>
<td>517.0±179.2</td>
<td>511.6±208.4</td>
<td>515.0±188.8</td>
<td>1,300</td>
</tr>
<tr>
<td>Vitamin C (mg/d)</td>
<td>123.5±77.2</td>
<td>112.6±62.4</td>
<td>119.4±71.7</td>
<td>95</td>
</tr>
</tbody>
</table>

*DRIs was referred by DRIs in Vietnam (18) except for iron (19).

DRIs, dietary reference intakes; ND, not defined.

There was no correlation between breast milk copper concentration and serum copper concentration (r=-0.211, p=0.118). Neither zinc intake nor serum zinc concentration was correlated with breast milk copper concentration (r=-0.040, p=0.954, r=-0.166, p=0.210, respectively).

DISCUSSION

This study was the first report determining the concentration of iron, zinc and copper in breast milk along with assessments of nutrition status and nutrient intakes of Vietnamese women. Anemia, and low concentrations of serum zinc and copper were highly prevalent among the study participants and their iron and zinc intakes were lower than those of estimated requirements.

Breast milk concentrations are affected by the time of day and the time of sampling during feeding; for example, iron content in breast milk was reported to be higher at the nighttime feeding and in hind-milk samples (25); therefore, breast milk is ideally collected in the morning more than an hour after the previous breastfeeding. In this study, we collected breast milk between 7 and 9 am; however breastfeeding status could not be identified. Variation of breast milk iron concentration in our study participants showed relatively lower SD (0.14 mg/L) and CV (34.9%) compared to the other studies regulated by design on the timing of

p=0.068. There was no correlation between breast milk copper concentration and serum copper concentration (r=-0.211, p=0.118). Neither zinc intake nor serum zinc concentration was correlated with breast milk copper concentration (r=-0.040, p=0.954, r=-0.166, p=0.210, respectively).
collecting breast milk (26). On the other hand, breast milk concentrations of zinc and copper were reported to have no significant diurnal variations (25, 27).

The concentration of iron in breast milk is considered as low in relation to serum iron since milk iron concentration is 20 to 30% of serum iron. Most previous studies showed no correlation between maternal iron status and breast milk concentration of iron (26, 28, 29), while one study showed it was increased among severely anemic mothers, Hb <80 g/L (30). Although anemia was highly prevalent among the study participants, no severely anemic woman was included: the lowest Hb concentration was seen to be 94.5 g/L. Iron intakes of our study participants were observed to be less than their requirements; however, it was not found to be related to the breast milk concentration. Their breast milk iron concentration was found to be similar to values for American lactating mothers with higher iron intakes; 0.42 mg/L and 47.1 mg/d among women at 7 to 9 mo postpartum, while 0.38 mg/L and 40.8 mg/d among women at 10 to 12 mo postpartum, respectively (31).

The concentration of copper in breast milk is 20 to 25% of that in serum. Some studies reported dietary copper intakes did not affect the concentrations of copper in breast milk (32). In our study, we found no correlation of maternal serum copper to the breast milk copper concentration. On the other hand, breast milk copper concentration may be affected by other nutrition status such as zinc. It was suggested that marginal zinc intake during pregnancy and lactation increases the abundance of mammary gland Cu transporters and alters their localization, resulting in high milk copper concentration (33). In our study, breast milk copper concentration had a negative association to serum zinc concentration and zinc intake, though it was not statistically significant (p>0.05). Compared to the breast milk copper concentration of US women (31); 0.30 mg/L and 0.24 mg/L among the women at 7 to 9 mo and 10 to 12 mo postpartum, respectively, that of our study participants showed relatively lower levels. However, breast milk concentration of copper has greater inter- and intra- individual differences (34) and we presumed that it was also influenced by the other factors besides their nutrient status of copper or zinc.

In contrast to iron and copper, whose concentration in human milk are a fraction of those in serum, breast milk zinc concentration is approximately 1 to 2 times higher than in serum in healthy lactating women. Thus, there is more possibility that the maternal zinc status influences the breast milk concentration if the lactating women are zinc deficient. In fact, breast milk zinc concentration among the study participants, approximately half of whom had low serum zinc concentration, was 1 to 2 times lower than their serum zinc concentration. Numerous cross-sectional and limited longitudinal studies have shown no consistent correlation between maternal dietary zinc intake and milk zinc concentration; however, it is only when maternal intakes are relatively high (33). Although animal studies have suggested marginal zinc intakes decreased zinc transfer to mammary gland and lower milk zinc concentration (36–38), the effect of low maternal zinc intakes during lactation has not been well determined in humans. Data from several developing countries where zinc intakes presumed to be low, such as Bangladesh (39), India (40), Egypt (41), Nigeria (42), South Brazil (43) and Amazon (44) indicated that the breast milk zinc concentrations were relatively lower than those of well-nourished US women (45), though the comparison needs caution for interpretation since there were different methodologies used among the studies. Comparing our data and data for US women (45); 0.83 mg/L and 0.53 mg/L among the women at 6 to 9 mo and 10 to 12 mo postpartum, respectively, our data were found to be relatively lower breast milk zinc concentration. Among the data from developing countries, the data from Bangladesh (39) had comparable postpartum time with our data, and the breast milk zinc concentration was also as low as ours: 0.73 mg/L at 7 to 9 mo postpartum and 0.54 mg/L at 10 to 12 mo postpartum. In the present study, we did not observe correlation between the breast milk zinc concentration and zinc intakes or zinc status, but it might be because their zinc intakes were below the estimated requirements (95% CI: 6.8–14.2 mg/d) and their zinc status was shifted to a lower level (95% CI: 0.60–0.79 mg/L).

Although the lower level of the breast milk zinc concentration may not greatly affect the total zinc intake of infants aged at 6 to 12 mo as the infants start complementary food and contribution of breast milk is only 30% of the total requirement (46), it of greater effect during the first 6 mo after birth as the infants are recommended to be given only breast milk. As reported in the national survey 2004 (47), stunting was highly prevalent among the infants of present study participants. Some previous studies have also shown high prevalence of zinc deficiencies among infants in rural Vietnam (48, 49). Low breast milk zinc concentration, therefore, could be a cause for infant malnutrition. Since the present study design was a cross-sectional observational study and had limitations to interpret the situation, a longitudinal intervention study is required to determine whether chronically low zinc intake in lactating women is associated with lower breast milk zinc concentration.

The concentration of most minerals in breast milk remains fairly constant throughout the course of lactation; however iron, zinc and copper are the exceptions. These minerals have their highest concentrations immediately after parturition, and fall several months thereafter (50). Concentration of iron and copper in breast milk tends to remain constant after 3 mo of lactation, while concentration of zinc in breast milk significantly declines at 6 mo postpartum and even after the 6 mo (50). We also observed a significant fall in zinc concentration from 6 to 8 mo postpartum to 9 to 12 mo postpartum. Krebs et al. (45) found that a more rapid fall in milk zinc concentrations was seen in mothers who had lower zinc intakes; moreover, the difference in
milk zinc concentrations was greater in mothers after 6 mo postpartum. It would be interesting to determine the effect of nutrient deficiency on the fall in the breast milk concentration by a longitudinal intervention study.

Although there has been no report on the prevalence of anemia in lactating women in Vietnam, anemia is one of the important health issues in Vietnam. A national report in 2000 indicated that 32.2% of pregnant women were anemic (51). In the present study, the percentage of anemia was high. It was observed that the study participants had lower iron intakes than their requirements; though further consideration is needed to estimate iron requirements for Vietnamese lactating women including the assessment of iron absorption in their diet, and their usage of iron supplementation was decreased with the course of pregnancy. Iron deficiency is the most common cause of anemia, but it is not the only cause of anemia (22). It is of interest that little of the observed anemia reported here appeared to be associated with storage iron depletion. Recent studies in Vietnam among reproductive aged women have shown that incidence of anemia was strongly related with intestinal infection (52), and was also related with the other micronutrient deficiencies such as folic acid, vitamin B12, Vitamin A, riboflavin and Vitamin C (53). Thus, prevention of anemia should be ensured from all these aspects in early pregnancy, throughout pregnancy and during the postpartum period.

There is not much data among Vietnamese adults about zinc and copper nutrient status. The risk of zinc deficiency among Vietnamese has been estimated by International Zinc Nutrition Consultative Group (23) based on the per capita amounts of selected nutrients such as energy, zinc, phytate, and food components including absorbable zinc content of national food supplies. It was estimated that the percentage of the population at risk of inadequate zinc intake was 27.8%, and the risk of zinc deficiency was categorized to be high. Our data also indicated a high risk of zinc deficiency among the lactating women. Since the present study has the limitation of small sample size and insufficient information on their related factors, further research on a large scale including the other factors is needed.

Additionally, we had a limitation in the design of the dietary intake survey to estimate usual intake for all individuals in the sample. While several quantitative methods for assessing usual dietary intakes of individuals exist, we used 24-h dietary recall in the present study because an interactive 24-h dietary recall method has been specially designed for measuring usual intakes of total and absorbable iron and zinc in developing countries (1, 3). However, dietary intakes have inter- and intra-individual variances and the distribution of observed dietary intakes in a group must be corrected for the variation in intake. To estimate usual intakes for all individuals in the sample, the number of days of dietary data of individuals required to derive this estimate can be calculated using the intra-individual coefficient of variation (CV_intra) (23). We observed that CV_intra for intakes of iron, zinc and copper were 24.5%, 21.1% and 15.7%, respectively, and calculated approximately 6 d, 5 d and 3 d are needed for estimating iron, zinc and copper intakes using 24-h dietary recalls within 20% of long-term true intake, 95% of the time (54, 55).

Although the 3 non-consecutive day 24-h dietary recall method was designed in the present study, the number of days of the dietary intake survey was short to estimate individual iron and zinc intakes. On the other hand, Gersovitz et al. (56) suggested that validity of the 24-h dietary recalls declined by the fifth consecutive record day and the demographic nature of the sample became biased due to drop-outs and decreased usability of the records as the record progressed to the seventh day. A design of the dietary intake survey to estimate these micronutrients in Vietnamese lactating women should be considered in a future study.

In conclusion, the present study indicated a high prevalence of anemia and zinc deficiency among lactating mothers in rural Vietnam. In addition, the subjects had lower breast milk zinc concentration, though no correlation between breast milk concentration of zinc and zinc status was observed. Further research needs to be carried out to confirm the observation.

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