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## Original Research Article

## Case study of temporal changes in maternal dietary intake and the association with breast milk mineral contents



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## ABSTRACT

Minerals play important roles in infant growth and development, even though they only make up to 0.2 g% of the mature breast milk contents. Limited studies examined the association between maternal dietary intake and breast milk mineral contents in a temporal manner. Twenty Malaysian Malay postpartum mothers were recruited by either convenience or snow balling sampling from the urban lower middle income residential areas. Dietary intake of the subjects was obtained by 24-hour recall during each breast milk collection. Adequacy of maternal mineral intake was compared with the latest Recommended Nutrient Intake for Malaysia. Each of the subject provided breast milk samples for three times (T1, T2, and T3) at consecutive 2-week intervals. Breast milk concentrations of selected macro- and micro-minerals, including Ca, P, K, Na, Mg, Fe, Zn, Cu, Mn, Se, I, Cr and Mo were determined by inductively coupled plasma mass spectrometry (ICP-MS). Subjects were aged  $31.4 \pm 6.1$  years with a majority (60 %) having post-secondary school/ college education. While maternal intake of macro-minerals, Ca, P, K and Mg, did not display a significant temporal changes from T1 to T3, the intake of micro-minerals, Cu, Mn and I decreased significantly over time from T1 to T3. Breast milk K, Fe, Zn and Cu concentrations showed a significant decreased with the progression of lactation from transitional (2–3 weeks) to established stage (> 8–12 weeks). Significant correlations were established between maternal intake of K, Na, Fe and Se and their respective concentrations in breast milk in the present study. This case study revealed an inadequate maternal intake of several key micro-minerals (Cu, Mn, I) among the postpartum Malay mothers and a decreasing concentrations of certain essential minerals (K, Fe, Zn and Cu) in breast milk with lactation stage.

## 1. Introduction

Human milk is recommended as the sole dietary source for the first 6 months of life and an important complementary food for the first 1–2 years (WHO, 2002). Nutritionally adequate mothers are able to provide breast milk with sufficient energy and a proper array of nutrients to support the normal growth and development of her child through the first 6 months of life, without the need for complementary feeding (Picciano, 2001; Butte et al., 2002). Besides, breast milk also confers other benefits including reduced risk of allergies and gastrointestinal infections (Le Huërou-Luron et al., 2010), improved neuro development

(Isaacs et al., 2010), and decreased risks of obesity, diabetes and hypertension in long term (Parikh et al., 2009).

The influence of maternal nutrition on breast milk nutrients has been the subject of concern in several investigations over the years (Deodhar et al., 1964; Allen, 1994; Chapman and Nommsen-Rivers, 2012). While minerals make up a small proportion (0.2 g%) of the contents in mature breast milk, they play important functions in skeletal and neuromuscular growth and development, in numerous essential enzymatic systems, and in providing immunological protection against infections of the infant (Soetan et al., 2010). Nonetheless, relatively less attention has been accorded to the micronutrients in breast

**Abbreviations:** ICP-MS, Inductively Coupled Plasma Mass Spectrometry; Ca, Calcium; P, Phosphorus; K, Potassium; Na, Sodium; Mg, Magnesium; Fe, Iron; Zn, Zinc; Cu, Copper; Mn, Manganese; Se, Selenium; I, Iodine; Cr, Chromium; Mo, Molybdenum; RNI, Recommended Nutrient Intakes; SPSS, Statistical Package for Social Sciences; SD, Standard deviation; UHT, Ultra-heat treatment

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milk, despite the fact that breast milk has long been recommended as an optimum food for the infant (Allen et al., 2016). There is a dearth of information on the influence of maternal dietary intake on breast milk mineral contents with progressive lactation. A recent systematic review on the impact of maternal nutrition on breast milk composition concluded the overall evidences on the quantitative relations between maternal diet and breast milk composition remains scattered and weak (Bravi et al., 2016).

There are limited studies in Malaysia on breast milk compositions, and they have focused on lipids (Ma et al., 2015) and fatty acids (Kneebone et al., 1985; Daud et al., 2013), but not the minerals. There is increasing evidence that mineral deficiency in growing children can cause multiple problems, including retarded physical growth and cognitive performance, poor bone development and impaired immunological functions (Andrade et al., 2014). This investigation was undertaken to obtain an understanding and correlation on the changes in breast milk mineral concentrations with different lactation stages in relations to maternal dietary intake in a temporal manner.

## 2. Methods

### 2.1. Study design

This is a cross-sectional purposive study conducted within the urban residential areas of Kajang and Serdang. These two locations were purposively selected for their large populations of Malay residents from the lower-middle income background. These densely populated locations provided a convenient and snow balling sampling sites to recruit potential subjects from a similar socio-economic background.

### 2.2. Ethics approval and consent to participate

This study was undertaken in agreement with the Helsinki declaration of 1975 as revised in 2013. The International Medical University Joint Committee on Research and Ethics gave approval for this study with the approval number of IMU R123. The International Medical University Joint Committee on Research and Ethics operates in accordance to the International Conference of Harmonization/ Good Clinical Practice Guidelines (ICH-GCP) Guidelines, the Declaration of Helsinki 2000 and Malaysian Good Clinical Practice. The study proposal was also registered with the Netherlands National Trial Register with the registration number of NTR 4404.

### 2.3. Inclusion and exclusion criteria

The inclusion criteria of the subjects are apparently healthy Malay women aged between 20–40 years, practising breastfeeding throughout the study period, not taking any dietary supplements, singleton infant with gestational age  $\geq 37$  weeks and birth weight  $\geq 2500$  g, infant older than 2 weeks at recruitment to exclude colostrum milk feeding and also family from middle income status, based on the Malaysia Department of Statistics median monthly income of urban households as RM5,156 (Department of Statistics Malaysia, 2014).

On the other hand, the exclusion criteria of the subjects are breastfeeding mothers on special diets or fasting during study period and mother or infant fall sick at recruitment.

### 2.4. Subject recruitment process

Recruitment of subjects was based on the community feedback and knocking on door-to-door. Several potentially candidates who fulfilled the inclusion criteria did not agree to participate, giving reasons such as too busy to commit, husband did not agree and working outside the home. Approximately, out of every 5 potential subjects encountered, only one person agreed to participate. Thus, through the 'snowball' approach, a total of 20 mothers who fulfilled the study criteria were

recruited during the entire study duration. The study protocol was explained to the participants and all the participants signed the written consent forms having agreed to be part of this study. Appointments were scheduled for three consecutive collections of breast milk samples and 24-hour dietary recall at two week intervals.

### 2.5. Collection of milk samples

Breast milk samples were collected three times (T1, T2, T3) from each subject at consecutive 2-week intervals. Milk samples were collected between 9–11 a.m. each collection to minimise circadian influence. Before collection, the nipple and areola of a single breast were cleaned with 70 % ethyl alcohol. As much milk was expressed from one breast by a trained research assistant using a manual breast pump (Phillips Avent model SCF 330-13). At least 25 ml of breast milk was collected each time into a sterile tube and stored at  $-80^{\circ}\text{C}$  until further analysis.

### 2.6. Milk mineral contents analysis

The milk samples were air freighted on dry ice to the University of Groningen, Netherlands for milk composition analysis. Mineral concentrations of the breast milk samples were determined at the European Laboratory of Nutrients (ELN). A total of 13 minerals were analysed, including the macro-minerals: calcium (Ca), phosphorus (P), potassium (K), sodium (Na) and magnesium (Mg), as well as micro-minerals of dietary importance: iron (Fe), zinc (Zn), copper (Cu), manganese (Mn), selenium (Se), iodine (I), chromium (Cr) and molybdenum (Mo). The above mentioned macro- and micro-minerals were selected as they are included in the Recommended Nutrient Intakes (RNI) for Malaysia (National Coordinating Committee on Food and Nutrition, 2017).

The breast milk samples were thawed at room temperature prior to analysis. The thawed samples were digested in a microwave digestion system and analysed by inductively coupled plasma mass spectrometry (ICP-MS). Briefly, 0.5 ml of breast milk sample was digested at  $180^{\circ}\text{C}$  for 25 min with 7 ml of 65 % nitric acid ( $\text{HNO}_3$ ) and 1 ml of 30 % hydrogen peroxide ( $\text{H}_2\text{O}_2$ ). Digested samples were then cooled to  $50^{\circ}\text{C}$  for 30 min. Quality is instituted by the use of control cards by the NEN norm 6603 internally, and additionally, with the laboratory participation of an external quality surveillance for ICP, organized by the Dutch Foundation for Quality Assessment in Medical Laboratories (SKML).

### 2.7. Maternal dietary intake

During each milk collection, the subjects were interviewed face-to-face for dietary intake recording using the 24-hour recall method. The mineral contents of various food consumed by the subjects were computed using the Malaysian Food Composition (Tee et al., 1997), USDA Nutrient Database (Agriculture Research Service, 2016) and AUSNUT Food Nutrient Database (Foods Standards Australia New Zealand, 2015) in the above order of priority. Following that, the mineral contents were expressed as percentages of meeting the Recommended Nutrient Intakes (RNI) for Malaysia women during the first 6 months of lactation (National Coordinating Committee on Food and Nutrition, 2017).

### 2.8. Statistical analysis

Statistical analyses were conducted using the SPSS (Statistical Package for Social Sciences) version 20. Descriptive statistics were expressed as mean  $\pm$  standard deviation (SD). The mineral contents in breast milk at different lactation stages were analysed by Kruskal-Wallis H test. On the other hand, maternal dietary intake of minerals at the three collection times were analysed using one-way ANOVA. The correlations between maternal dietary intake of minerals and breast milk mineral contents at different lactation stages were carried out using

Pearson correlation test. Significance level was set at  $p < 0.05$  for all analyses.

### 3. Results

#### 3.1. Socio-economic background

The study subjects and spouses had a mean age of  $31.4 \pm 6.1$  and  $33.8 \pm 7.4$  years, respectively (Table 1). The majority of them had more than 10 years of formal schooling (post-secondary). While most of the subjects were housewives, the spouses were employed in the construction and manufacturing industry. There were more female infants (70 %) than males (30 %), and on average, the study infant was the third child among siblings. Data on birth weight indicates none were born with low birth weight (range 2840 g–3780 g).

At the first milk collection, eight out of the 20 infants were aged 2–3 weeks, another eight were between > 3–8 weeks old, and the remaining four were aged between > 8–11 weeks. At the second milk sample collection, 10 infants were between > 3–8 weeks old while another 10 were aged between > 8–14 weeks. By the third milk collection, eight of the infants were between > 3–8 weeks old while the rest of the 12 infants were aged between > 8–16 weeks.

#### 3.2. Breast milk mineral contents

Based on the age of the infants when breast milk was collected, the lactation stage was categorised according to 'transitional' (2–3 weeks), 'early' (> 3–8 weeks) and 'established' (> 8–16 weeks) (Li et al., 2016). Table 2 shows the breast milk mineral concentrations at each lactation stage, as well as the percentage of meeting the daily dietary recommendations for infants at aged 0–5 months (National Coordinating Committee on Food and Nutrition, 2017).

The present study demonstrated significant decreasing trends on the concentrations of K, Fe, Zn and Cu with the progression of lactation. However, the majority of the minerals in consideration did not display a similar downward trend. In considering the adequacy of milk minerals for breastfed infants, it was found that all the minerals met the

**Table 1**  
Characteristics of subjects and infants (N = 20).

Characteristics	N (%)	Mean $\pm$ SD
<b>Subject</b>		
Age (years)	20 (100)	$31.4 \pm 6.1$
Body mass index ( $\text{kg}/\text{m}^2$ )	20 (100)	$26.0 \pm 5.1$
<b>Education level</b>		
Secondary school and below	8 (40)	
Post-secondary/ college	12 (60)	
<b>Occupation</b>		
Housewife	13 (65)	
Employees <sup>†</sup>	7 (35)	
<b>Husband</b>		
Age (years)	20 (100)	$33.8 \pm 7.4$
<b>Education level</b>		
Secondary school and below	7 (35)	
Post-secondary/ college	13 (65)	
<b>Occupation</b>		
Employees <sup>‡</sup>	20 (100)	
<b>Infant</b>		
<b>Male</b>		
Male	6 (30)	
<b>Female</b>		
Female	14 (70)	
Birth weight (g)	20 (100)	$3298 \pm 458$
<b>Age at recruitment (months)</b>		
1.0 and below	8 (40)	
1.1 – 2.0	8 (40)	
2.1 – 3.0	4 (20)	

<sup>†</sup> service and production industry.

<sup>‡</sup> construction and production industry.

requirement levels at each lactation stage in the present study, with the exceptions of micro-minerals, Mg (T1), Cr (T1-T3) and Mo (T3).

Out of the macro-minerals, Na concentration in breast milk at each lactation stage exceeded its recommendation level by approximately 1.5–2.5 times and Na was the only macro-mineral that showed wide variations in concentration during lactation, as indicated by its high standard deviation values. On the other hand, among the breast milk micro-minerals that displayed wide variations at each lactation stage were Fe, Zn, Cr and Mo (average value for Fe =  $374 \pm 220 \mu\text{g}/\text{L}$ , Zn =  $2330 \pm 1300 \mu\text{g}/\text{L}$ , Cr =  $0.10 \pm 0.10 \mu\text{g}/\text{L}$ , Mo =  $3.6 \pm 4.6 \mu\text{g}/\text{L}$ ).

#### 3.3. Quality control for ICP-MS

The intra-assay coefficient of variation of the pooled breast milk samples for the micro-minerals at the lower mean level is within the range of 1.4%–6.3% (Table 3) while at the higher mean level is within the range of 0.9%–4.9%. On the other hand, the inter-assay coefficient of variation at the lower mean level is within the range of 5.3%–9.9% and at the higher mean level is within the range of 0.5%–9.6%.

#### 3.4. Maternal mineral intake

Maternal mineral intake at each of the three points of assessment (T1, T2 & T3) are expressed as mean amount consumed and percentage of meeting the daily recommendation intake (National Coordinating Committee on Food and Nutrition, 2017) (Table 4). Intake of the major macro-minerals (Ca, P, K and Mg) did not display a significant temporal changes over the assessment points, except for Na, which displayed significant variations over time. Among the micro-minerals, only Cu, Mn and I intake decreased significantly from T1 to T3. In addition, data that showed wide subject variations at each point of assessment, as indicated by high standard deviation values, were found for Ca, Na, Fe and Zn intake (average value for Ca =  $940 \pm 540 \text{ mg}$ , Na =  $1330 \pm 240 \text{ mg}$ , Fe =  $26 \pm 11 \text{ mg}$ , Zn =  $8.5 \pm 5.2 \text{ mg}$ ).

In terms of meeting the dietary recommendations (National Coordinating Committee on Food and Nutrition, 2017) for the first 6 months of lactation, the intake of the macro-minerals appeared adequate in general, either being close to or exceeded the respective recommendation levels. In contrast, intake levels of all the micro-minerals in the present study did not meet the requirements for lactation, except for Fe intake, which markedly exceeded its daily recommendation level.

In considering the food sources of the subjects' mineral intake, results from the 24-hour dietary recall revealed that milk, beef and chicken were the main sources of Ca, P, Fe and Zn (Table 5). The popular forms of milk consumed by the postpartum subjects include milk powder, ultra-heat treatment milk (UHT) and fresh milk. The subjects also reported taking a variety of vegetables, with the main types being mustard green (*Brassica juncea*) and spinach (*Spinacia oleracea*), as well as legumes especially beans and soy products. These foods are rich sources of both macro-minerals like K and Mg, and micro-minerals including Cu, Mn, Cr and Mo.

#### 3.5. Correlations between maternal intake and breast milk minerals at T1, T2 and T3

Significant correlations between maternal mineral intake and breast milk mineral concentrations were observed for the three lactation stages: T1 (values in bold), T2 (values in italic) and T3 (values in non-bold) (Table 6). Overall, out of the 13 minerals investigated in the present study, only K, Na, Fe and Se showed significant correlations between maternal intake of minerals and their counterparts in breast milk, at different lactation stages. For instance, maternal K intake correlated with breast milk K concentration at T1 only, while intake of Fe showed correlation with breast milk Fe at T3 only.

**Table 2**

Mineral concentrations of breast milk according to lactation stage, % RNI (National Coordinating Committee in Food and Nutrition, 2017) and the RNI values for infant aged 0-5 months.

Breast milk mineral contents	Stage of lactation <sup>(Li et al., 2016)</sup>						RNI for infant aged 0-5 months
	Transitional		Early		Established		
	2-3 weeks		> 3-8 weeks		> 8 - 16 weeks		
	(n = 8)		(n = 26)		(n = 26)		
	Mean ± SD	% RNI	Mean ± SD	% RNI	Mean ± SD	% RNI	
Calcium mg/L	248 ± 24	124 ± 12	275 ± 68	138 ± 34	273 ± 51	136 ± 26	200mg
Phosphorus mg/L	152 ± 32	152 ± 32	138 ± 36	138 ± 36	136 ± 41	136 ± 41	100mg
Potassium mg/L	586 ± 88 <sup>a</sup>	146 ± 22	583 ± 120 <sup>a</sup>	146 ± 31	487 ± 130 <sup>b</sup>	122 ± 32	400mg
Sodium mg/L	242 ± 340	202 ± 280	201 ± 110	168 ± 92	173 ± 220	144 ± 180	120mg
Magnesium mg/L	29 ± 8.1	97.3 ± 27	32 ± 7.0	107 ± 23	34.9 ± 8.8	116 ± 29	30mg
Iron µg/L	590 ± 320 <sup>a</sup>	-	281 ± 170 <sup>b</sup>	-	251 ± 170 <sup>b</sup>	-	-
Zinc µg/L	3910 ± 1900 <sup>a</sup>	355 ± 170	1760 ± 1100 <sup>b</sup>	160 ± 95	1320 ± 920 <sup>b</sup>	120 ± 84	1100µg
Copper µg/L	610 ± 160 <sup>a</sup>	305 ± 82	364 ± 130 <sup>b</sup>	182 ± 64	268 ± 110 <sup>c</sup>	134 ± 54	200µg
Manganese µg/L	8.3 ± 2.7 <sup>a</sup>	276 ± 90	4.1 ± 4.2 <sup>b</sup>	137 ± 140	3.7 ± 4.5 <sup>b</sup>	123 ± 150	3µg
Selenium µg/L	24.4 ± 10	406 ± 180	18.9 ± 5.6	315 ± 93	16.7 ± 11	278 ± 180	6µg
Iodine µg/L	141 ± 63	210-224 ± 94-100	124 ± 48	184-198 ± 72-77	104 ± 78	155-166 ± 110-120	63.0-67.5µg
Chromium µg/L	0.05 ± 0.10 <sup>a</sup>	25 ± 50	0.12 ± 0.10 <sup>b</sup>	60 ± 50	0.12 ± 0.10 <sup>a,b</sup>	60 ± 50	0.2µg
Molybdenum µg/L	6.9 ± 6.9	345 ± 345	2.3 ± 3.9	115 ± 190	1.7 ± 3.2	85 ± 160	2µg

Different alphabet in the same row indicate significant difference ( $p < 0.05$ ).

**Table 3**

Analytical intra- and inter- coefficient of variance (CV) for micro-minerals analysed by ICP-MS.

	Low Mean	Intra-CV (%)	Inter-CV (%)	High Mean	Intra-CV (%)	Inter-CV (%)
Calcium (mmol/L)	6.99	1.4	7.6	8.13	4.2	8.1
Phosphorus (mmol/L)	5.17	5.0	8.6	5.62	3.8	8.7
Potassium (mmol/L)	14.4	4.3	7.2	16.2	4.6	8.1
Sodium (mmol/L)	6.61	4.8	7.7	13.3	1.6	6.6
Magnesium (mmol/L)	1.03	3.4	6.6	1.36	4.7	7.4
Iron (µmol/L)	3.51	5.1	8.2	7.88	4.5	7.3
Zinc (µmol/L)	0.03	4.9	5.3	0.06	4.7	6.9
Copper (µmol/L)	6.22	5.0	7.3	8.73	4.5	7.4
Manganese (µmol/L)	0.04	6.3	7.6	0.05	4.2	9.6
Selenium (µmol/L)	0.13	5.0	9.5	0.20	4.9	8.4
Iodine (µmol/L)	0.71	1.9	5.9	2.05	0.9	0.5
Chromium (µmol/L)	ND	ND	ND	ND	ND	ND
Molybdenum (µmol/L)	0.01	6.1	9.9	0.06	1.8	8.8

ND: Not detected.

**Table 4**

Maternal intake of minerals at three consecutive assessments (T1, T2, T3) of a 2-week intervals and % RNI (National Coordinating Committee in Food and Nutrition, 2017) for 1<sup>st</sup> 6 months of lactation.

	T1		T2		T3		Average	
	Mean ± SD	% RNI	Mean ± SD	% RNI	Mean ± SD	% RNI	Mean ± SD	% RNI
Calcium (mg)	878 ± 570	87.8 ± 57	1130 ± 860	113 ± 86	811 ± 690	81.1 ± 68	940 ± 540	94.1 ± 54
Phosphorus (mg)	624 ± 74	89.1 ± 10	682 ± 120	97 ± 16	686 ± 110	98 ± 16	664 ± 41	94.8 ± 5.9
Potassium (g)	7.0 ± 1.6	137 ± 30	8.0 ± 2.0	156 ± 40	6.8 ± 1.6	132 ± 30	7.2 ± 1.0	142 ± 20
Sodium (mg)	1470 ± 460 <sup>a</sup>	98.2 ± 30	1160 ± 500 <sup>b</sup>	77.2 ± 33	1340 ± 390 <sup>a</sup>	89.8 ± 26	1330 ± 240	88.4 ± 16
Magnesium (mg)	360 ± 42	116 ± 14	391 ± 69	126 ± 22	394 ± 68	127 ± 22	382 ± 26	123 ± 8.4
Iron (mg)	24 ± 18	162 ± 120	30 ± 17	198 ± 110	25 ± 13	164 ± 89	26 ± 11	174 ± 76
Zinc (mg)	8.0 ± 6.2	84.4 ± 65	9.9 ± 7.7	104 ± 82	7.5 ± 6.5	78.7 ± 68	8.5 ± 5.2	89 ± 54
Copper (µg)	999 ± 270 <sup>a</sup>	76.9 ± 21	724 ± 79 <sup>b</sup>	55.7 ± 6.1	675 ± 210 <sup>b,c</sup>	51.9 ± 16	800 ± 120	61.5 ± 9.6
Manganese (mg)	2.0 ± 0.6 <sup>a</sup>	75.4 ± 22	2.1 ± 0.7 <sup>a</sup>	80.5 ± 28	1.7 ± 0.5 <sup>b</sup>	65 ± 20	1.9 ± 0.4	73.6 ± 14
Selenium (µg)	27 ± 3.6	79.5 ± 10	27 ± 3.5	77.9 ± 10	26 ± 2.7	75.5 ± 7.8	26 ± 2.1	77.6 ± 6.1
Iodine (µg)	164 ± 53 <sup>a</sup>	82.1 ± 26	124 ± 44 <sup>b</sup>	62.3 ± 22	131 ± 52 <sup>b,c</sup>	65.6 ± 26	140 ± 34	70 ± 17
Chromium (µg)	28 ± 14 <sup>a</sup>	61.3 ± 32	38 ± 12 <sup>b</sup>	83.8 ± 28	30 ± 12 <sup>a,b</sup>	67.3 ± 26	32 ± 8.2	70.8 ± 18
Molybdenum (µg)	36 ± 18	71.4 ± 38	34 ± 17	68.8 ± 34	36 ± 21	71.5 ± 42	35 ± 12	70.6 ± 23

Different alphabet in the same row indicate significant difference ( $p < 0.05$ ).

**Table 5**  
Main dietary sources of mineral consumed by the study subjects.

Dietary minerals	Main food sources (Tee et al., 1997; Agriculture Research Service, 2016; Foods Standards Australia New Zealand, 2015)
Calcium	Milk
Phosphorus	Milk, meat, eggs, fish
Potassium	Green leafy vegetables, chicken
Sodium	Noodles, sauces
Magnesium	Green leafy vegetables, beans
Iron	Milk, beef, chicken
Zinc	Milk, meat
Copper	Beans, organ meat
Manganese	Spinach, bean curd
Selenium	Meat, eggs
Iodine	Eggs, noodles
Chromium	Meat, beans
Molybdenum	Bean curd, eggs

#### 4. Discussion

The present case study provided observations about maternal dietary and its association with breast milk minerals content in a sample of Malay mothers during the first four months postpartum. Given the small sample size, a wide subject variation in dietary mineral intake were found, which was not unexpected, despite the fact that the subjects were from similar ethnic and socio-economic background.

On average, maternal intake for K, P, Mg, Ca and Fe met the daily dietary recommendations, which were derived mainly from milk, meat, fish, egg, and legumes. This finding of postpartum subjects consuming reportedly more protein foods than usual may reflect in part by the Malay cultural beliefs and traditional food intake practices during the post-delivery period. During this confinement period of about 1–2 months, Malays believe that the mother is weak and vulnerable, and should adhere to food restrictions that would hasten the restoration of her health (Aloysius and Shariffah Suraya, 2015). Mothers are encouraged to consume more “hot” foods, such as meat and fish that will reinstate “internal heat” lost by the body during delivery, while fruits in general and selected vegetables (e.g. cucumber, cabbage, pumpkin) deemed as “cooling” are avoided (Fok et al., 2016). Some Malay mothers also believe that increasing intake of milk, mustard green and spinach will stimulate milk flow.

Compared to the macro-minerals, maternal intake of micro-minerals was generally inadequate, particularly Cu, I, Cr, Mo and Se. Breastfed infants rely on the adequacy of maternal essential micro-minerals

intake for optimum growth and development. For instance, I and Se are required for the production of thyroid hormones and regulating the immune response during pregnancy and the postpartum period (Mao et al., 2016; Negro et al., 2007). Nutrition education on the importance of these micro-minerals should be promoted to both pregnant and postpartum mothers (Henjum et al., 2017). It is encouraging to note that the Ministry of Health Malaysia plans to expand the current mandatory iodisation of salt in Sarawak and Sabah to the rest of the country by 2018 (National Coordinating Committee on Food and Nutrition, 2016).

The stage of lactation is known to influence individual breast milk nutrient composition (Su et al., 2010; Gidrewicz and Fenton, 2014; Grote et al., 2015). In this study, the concentrations of K, Fe, Zn and Cu in breast milk showed a significant decreasing trends from transitional to mature lactation stage. Li et al. (2016) also reported progressively lower concentrations of K, Cu and Zn from transitional to early and established lactation among Guatemalan mothers, in agreement with the findings from the present study. In an analysis of 1197 milk samples of Japanese mothers, several elements including K, Na, Zn, Cu and Se were at significantly lower concentrations between 11–20 days and 21–89 days of lactation, approximating transitional and early milk stage (Yamawaki et al., 2005). Decreasing Zn and Cu contents by the third month of lactation was reported by Silvestre et al. (2000) and Yalcin et al. (2015). However, others reported Zn concentration increased with lactation duration (Domelof et al., 2004; Eneroth et al., 2009). Plausible reasons for the different results include differences in the mineral analysis method, sampling time, foremilk versus mature milk, milk collection method, and inter-individual variability, besides the stage of lactation (Khaghani et al., 2010; Örün et al., 2012).

The significant decreasing K, Fe, Zn and Cu concentrations with lactation stages in this study have implications for breast feeding and complementary feeding practices. These minerals are involved in many complex enzyme systems involved in physical and neurological development of infants. They also interact with one another and deficiency in one mineral may impair the absorption of another. For instance, Cu deficiency impairs Fe absorption, while Zn absorption is impaired by Fe deficiency (Özden et al., 2015). In addition, Zn and Cu are known to compete at the level of intestinal absorption, whereby high Zn levels in the diet can reduce Cu absorption (Osredkar and Sustar, 2011). Thus, adequate maternal intake of the abovementioned minerals should be emphasised to pregnant and postpartum mothers.

Out of the 13 minerals investigated, the present case study revealed significant correlations between maternal dietary intake and their

**Table 6**  
Correlations between maternal dietary intake and breast milk mineral contents at T1 (**bold**), T2 (*italic*) and T3 (non-**bold**) (only significant correlations are shown).

Breast milk	Maternal dietary intake												
	Ca	P	K	Na	Mg	Fe	Zn	Cu	Mn	Se	I	Cr	Mo
Ca		0.558		0.446	0.543		−0.517			−0.529			
P						−0.466	−0.488	0.726				0.487	
K			−0.502*					0.459					
Na			−0.484	0.532*				0.545		0.612			
Mg			−0.733					−0.479		−0.519		0.454	
Fe			−0.618			0.519*						0.593	
Zn		0.475		0.581	0.484			0.564					0.486
Cu		0.557	−0.486	0.580	0.525								0.453
Mn			−0.624										
Se	0.456		−0.602							0.485*		0.510	
I				−0.451				−0.602					
Cr													−0.748
Mo			−0.516	0.489						0.488		0.461	

Log transformations were conducted for sodium (T1, T2 and T3), manganese (T3), iodine (T3), chromium (T3) in breast milk and maternal intake of calcium (T3), as the data distributions were skewed substantially (skewness > ± 2).

\* Significant correlation of maternal intake of minerals and their counterpart in breast milk.

counterparts in breast milk for K, Na, Fe and Se only. In general, mineral contents in breast milk are less affected by maternal dietary intakes, unlike vitamins and fatty acids (Butte et al., 2002; Domelof et al., 2004; Innis, 2014; Allen, 2012). However, the results of the current study show otherwise, which warrant further investigations on the relationship between maternal dietary intake and the breast milk micro-nutrients content, particularly for K, Na, Fe and Se.

#### 4.1. Strengths and limitations of study

A principal strength of this study design is the simultaneous collection of maternal dietary intake and breast milk samples at a consecutively 2-week intervals. Through this temporal approach, breast milk mineral concentrations and variations from the transitional to established lactation stages were reported.

On the other hand, as these case study involving a small sample size, the findings do not reflect the maternal dietary intake of the general Malaysian population. The volume of breast milk consumed per day was not estimated in this study and thus, the total intake of minerals per day for infants is not available. Maternal dietary intake assessment was subject to the usual methodological errors inherent with the 24-hour dietary recall method. These include memory lapses, inaccurate estimation of portion size, and truthfulness on the part of the subjects. Reliance on non-Malaysian food composition tables may affect the accuracy of estimating mineral contents of locally produced foods.

## 5. Conclusion

In conclusion, this case study revealed two significant findings, namely inadequate maternal intake of several key micro-minerals among the postpartum Malay mothers, and the progressive decreasing concentrations of essential minerals with lactation stage. In light of the important roles of dietary minerals during the postpartum period for the health of both the mother and child, it is suggested that these findings be verified on a larger sample size of mother-infant pairs from various ethnic and socio-economic backgrounds.

### Ethics approval and consent to participate

The International Medical University Joint Committee on Research and Ethics gave approval for this study with the approval number of IMU R123. The study proposal was registered with the Netherlands National Trial Register with the registration number of NTR 4404 on 21 January 2014. The study protocol was explained to the participants and all the participants signed the written consent forms upon agreed to be part of this study.

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### Declaration of Competing Interest

The authors declare that they have no conflicts of interest.

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## Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:<https://doi.org/10.1016/j.jfca.2020.103468>.

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