

Maternal Milk Concentration of Zinc, Iron, Selenium, and Iodine and Its Relationship to Dietary Intakes

Mohammad A. Hannan · Bahram Faraji ·
Jesus Tanguma · Norma Longoria · R. C. Rodriguez

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Abstract The dietary intake of zinc (Zn), iron (Fe), selenium (Se), and iodine (I) of 31 lactating Mexican–American women attending the Hidalgo County WIC program in Rio Grande Valley (RGV), Texas was estimated from 24-h dietary recall interviews. Milk samples were obtained from lactating mothers who had infants 3 months of age and younger. Milk samples were collected in two visits to assess change in breast milk composition after 1–3 months postpartum: group A—after 30–45 days and group B—75–90 days. Dietary intakes indicated that the study participants had significantly inadequate percent energy intakes than the DRI (Dietary Recommended Intakes) percent recommended kilocalorie values but protein intakes were substantially higher than the percent recommended values. The estimated percent Zn, Fe, Se, and I intakes were also significantly lower than the DRI percent recommended values. The lactating mothers consumed significantly less Zn, Se, and I when compared to the Recommended Dietary Allowances (RDA) even though Fe intake was higher than the RDA value. Breast milk concentration of Zn, Fe, and Se were in agreement within the range of representative values for Constituents of Human Milk but I has significantly less concentration than the representative value. There was no statistically significant correlation observed between dietary intake and milk concentration of Zn, Fe, Se, and I. This study compares the estimated dietary intake of zinc, iron, selenium, and iodine to the concentration of these trace elements in the maternal milk of lactating women of Mexican–American heritage who attend the Rio Grande Valley WIC clinic.

M. A. Hannan (✉)

Department of Physics and Geology, The University of Texas—Pan American, Edinburg, TX, USA
e-mail: hannan@panam.edu

B. Faraji · R. C. Rodriguez

Coordinated Dietetics Program, The University of Texas—Pan American, Edinburg, TX, USA

J. Tanguma

Department of Computer Information System, The University of Texas—Pan American,
Edinburg, TX, USA

N. Longoria

Hidalgo County WIC Program, Edinburg, TX, USA

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Introduction

Human milk is considered to be the optimal source of nutrition for the infant [1]. Nutrient levels in milk-based formulas and milk substitutes for infants are generally modeled on the composition of human milk. Breast milk from a healthy well-nourished mother is nutritionally superior to formulas and can provide the newborn with adequate nutrition and bioactive components for the first 4–6 months of life. Thus, the American Academy of Pediatrics recommends exclusive breast-feeding for the first 6 months after birth and continued breast-feeding along with complementary foods for 12 months or longer [2]. For exclusively breast-fed infants, milk must be nutritionally adequate, and provide the trace metals needed for normal growth and development [3]. Although trace mineral concentration is relatively low in human milk and blood, they play a critical role in many physiological processes [4]. Reports in the literature indicate that important trace elements may be found in less than optimal amounts in milk of low-income lactating mothers. Typically, each trace element exhibits certain functions in the body that depends on its dosage and its nutritional status of the individual with respect to that element. Many trace metals such as Zn and Se act as cofactors in enzymatic systems that control key reactions in the body. Therefore, deficiency or excess of these metals can alter enzyme activities and influence important biological processes in the body. Another trace element of interest is Fe which is needed for hemoglobin and red blood cell formation, as well as some other important functions in the body. Iodine is needed for regulation of basal metabolism and its deficiency may bring about mental and physical retardation in the body (cretinism) [5–9].

Considering that in exclusively breast-fed infants, breast milk is the sole source of nutrition for the first few months of life, it is important to have accurate data on its composition and the factors that influence its composition, such as the mother's dietary intake. Mineral concentrations in human milk generally do not correlate well with maternal intake, with the exception of selenium and iodine [10]. Some investigators report a positive correlation between dietary intake of selenium and iodine and the concentration of these trace elements in maternal milk [11, 12], while others have been unable to demonstrate such a relationship [13]. Since there is no nutritional data available about the dietary intake of trace elements and their relationships to maternal milk composition of lactating mothers in Rio Grande Valley, this study was conducted to find relationships between estimated dietary intake of zinc, iron, selenium, and iodine and concentration of these elements in the maternal milk of lactating mothers.

Subjects and Methods

The protocol for the research was approved by the University of Texas—Pan American Institutional Review Board (IRB) for Human subjects and The Texas Department of State Health Services IRB. The study participants were recruited from the Hidalgo County WIC program in the Rio Grande Valley, South Texas. The WIC program is a special supplemental nutrition program for women, infants, and children of low-income families. The participants were women of Mexican–American heritage who had recently given birth to an infant and were lactating. They were mostly house-wives/stay-at-home-mothers and apparently healthy. Informed consent was gained from each participant and was explained to them in their own language before they were asked to sign the consent form.

The 24-h dietary recall interviews were conducted at the beginning of each visit by a trained bilingual (English–Spanish) senior dietetic student. After explaining the purpose and procedure to the participant, a quick list of foods consumed over the preceding 24 h (midnight to midnight of the day before the study [14]) was obtained. Information on the time and place of eating was also gathered. Then, the interviewer read the list back to the participant in order to ensure nothing had been missed, and to obtain additional information such as food components, additions, brand names, type, and supplement use. Serving sizes and amount consumed were estimated using plastic food models and measuring cups. The list was read back to the participant once more for further clarification and confirmation. Finally, the participant was asked if the intake reported reflected the participant's usual intake. If the intake was not typical, the participant was asked to provide an explanation. Each interview took approximately 20–30 min. Demographic and anthropometric data on the mother and infants as well as duration and frequency of breast-feeding were collected by the same interviewer. The mother's body weight was measured on a beam scale with an upright extension meter which was used to measure height. The body mass index (BMI) was calculated as the ratio of body weight to height in meters squared (kg/m^2).

Human milk samples were collected from the study participants on two occasions. The first samples were obtained from 31 mothers 30–45 days postpartum. This group of participants is designated "Group A". Seventeen participants returned to provide the second sample 75–90 days postpartum ("Group B"). Two occasions were chosen to assess change in mature milk composition after 1–3 months postpartum. On both occasions, approximately 20–30 ml of milk was collected either by manual expression into pre-washed plastic vials, or via a pre-washed trace mineral-free breast pump and container. Precautionary measures were followed to prevent contamination of the samples with environmental trace elements. Each sample was divided into two portions and frozen at -20°C until analysis by (1) Flame Atomic Absorption Spectrometry (AAS) and (2) Neutron Activation Analysis (NAA).

Trace Element Analysis

Zinc and Iron by AAS Three milliliters of milk and 10 ml of $\text{HNO}_3/\text{HClO}_4$ (9:1) acid mixture were placed in an Erlenmeyer flask, which was closed and left overnight under a perchloric fume hood. The next morning, the sample was slowly heated on a hot plate to $160^\circ\text{--}170^\circ\text{C}$ until fumes of HClO_4 appeared and the volume of each flask reduced to 2–3 ml. The solution was quantitatively transferred and adjusted to 10 ml flask with Millipore-Milli-Q water. Samples were then measured against zinc and iron standards by the Flame Atomic Absorption Spectrometry (A Analyst 800, Perkin Elmer). Multi-element reference standards (SRM 1549-Powdered non-fat dry milk and SRM 8435-powdered dry whole milk) from the National Institute of Standards and Technology (NIST) were digested and measured in the same manner as the samples [15, 16]). The mean (\pm SD) zinc concentrations for SRM 1549 and SRM 8435 were 44.7 ± 3.2 mg/kg (certified value is 46.1 ± 2.2 mg/kg) and 27.9 ± 4.0 mg/kg (certified value is 28.0 ± 3.1 mg/kg), respectively. The measured SRM 1549 iron concentration value was 1.72 ± 0.22 mg/kg as compared to the certified value of 1.78 ± 0.10 mg/kg. The SRM 8435 iron concentration was measured as 1.78 ± 0.11 mg/kg as compared to the certified value of 1.80 ± 0.11 mg/kg.

Selenium and Iodine by NAA The second half of milk samples were freeze dried at the USDA Subtropical Agricultural Research Center, Weslaco, Texas. Two hundred fifty milligrams of freeze dried milk samples were packed in trace element free polyethylene vials and sent to the University of Austin TRIGA Nuclear Reactor for irradiation. Samples

were irradiated eight hours (8 h) at the epithermal neutron flux of $1 \times 10^{11} \text{ ncm}^{-2} \text{ s}^{-1}$ for selenium. For iodine, samples were irradiated two minutes (2 min) at the thermal neutron flux of $1 \times 10^{12} \text{ ncm}^{-2} \text{ s}^{-1}$. Neutron fluxes were measured using sulfur wire monitors. NIST reference standards [SRM1632 b-Trace elements in coal (bituminous) and SRM1635-Trace elements in coal (sub bituminous)] were activated along with samples for quality control. Samples and standards were counted one hour (1 hr) several times after a cooling period of 21–26 days for selenium. Iodine samples and standards were counted ten minutes (10 min) several times after a cooling period of ten minutes (10 min). Gamma spectra were collected in two modes: normal and Compton suppression HPGe (High Purity Germanium) gamma spectrometer. The Compton spectrometer greatly reduces background radiation. The signal to noise ratio significantly improved the detection of small peaks [17]. The complete description of the set up of Compton suppression is given elsewhere [18, 19]. Spectral data processing and quantification by the relative method was performed using the software NADA developed by the UT Austin Nuclear Engineering Laboratory [20]. Measured selenium concentration values of NIST 1632b and 1635 were $1.29 \pm 0.21 \text{ mg/kg}$, and $0.89 \pm 0.14 \text{ mg/kg}$ as compared to certified values of $1.29 \pm 0.11 \text{ mg/kg}$, and $0.9 \pm 0.3 \text{ mg/kg}$, respectively. The measured iodine standard values for NIST 1570 and 1572 were $1.11 \pm 0.07 \text{ mg/kg}$, and $1.77 \pm 0.05 \text{ mg/kg}$ (certified values for iodine are $1.20 \pm 0.12 \text{ mg/kg}$, and $1.84 \pm 0.03 \text{ mg/kg}$), respectively.

Dietary and Statistical Analysis

The 24-h food recalls were analyzed using Food Processor SQL, a version 9.6 (ESHA Research Inc, Salem, Oregon). Energy, protein and nutrient intakes were expressed for 24 h and were compared to the DRI (Dietary Recommended Intakes) values. The results from Food Processor SQL were transferred to the Statistical Analysis Software SPSS version 15.0 (SPSS, Inc., Chicago, IL) for statistical analysis using Pearson product moment correlation coefficients between dietary intakes and milk concentrations [21, 22].

Results

Participants' age, weight and body mass index (BMI) for groups A and B are presented in Table 1. The mean (\pm SD) age of the 31 lactating women in group A was 28.2 (\pm 5) years. Subjects' age ranged from 20–38 years. Fourteen mothers declined to participate in the second interview. The mean age of the group B participants was 28.4 (\pm 6) years. Mean (\pm SD) weight of group A participants was 71 (\pm 15) kg, and 69 (\pm 14) kg for group B. The mean (\pm SD) body mass index in group A was 29 (\pm 6) kg/m^2 with the range of 19–42 kg/m^2 and for group B, the BMI mean (\pm SD) was 29 (\pm 6) kg/m^2 with the range of 18–42 kg/m^2 .

Table 1 Participants, Age, Weight, and BMI for Groups A and B

Group (Number of subjects)	Age (years)		Weight (kg)		BMI (kg/m^2)	
	Mean (\pm SD)	Range	Mean (\pm SD)	Range	Mean (\pm SD)	Range
A (n=31)	28.2 (\pm 5)	20–38	71 (\pm 15)	46–102	29 (\pm 6)	19–42
B (n=17)	28.4 (\pm 6)	20–38	69 (\pm 14)	44–101	29 (\pm 6)	18–42

Table 2 Energy, Protein, Zinc, Iron, Selenium, and Iodine Dietary Intakes of RGV Lactating Mothers

Group (Number of subjects)	Energy Intake as % DRI ^a	Protein Intake as % DRI	Zinc Intake as % DRI	Iron Intake % DRI	Selenium Intake % DRI	Iodine Intake % DRI
A (n=31)	78.6±28.1 ^b	139.5±57.5	76.1±58.0	86.8±68.2	70.6±47.8	50.5±49.0 ^c
B (n=17)	68.4±18.9	125.0±55.0	62.3±38.5	59.8±32.4	65.1±40.5	34.3±35.1

^a Dietary Reference Intakes^b Mean ± SD^c Significantly different from group B (p<0.05)

Energy, protein, zinc, iron, selenium, and iodine intakes were expressed as percentages of Dietary Reference Intakes (DRI) in Table 2. With the exception of protein, mean energy and other nutrients were less than the DRI recommendations as indicated by percentage below 100%. Protein mean (±SD) intakes for groups A and B were estimated as 139.5±57.5% and 125±55.0% DRI, respectively.

Dietary zinc, iron, selenium, and iodine intakes based on the 24-hour recall, and their breast milk concentrations are presented in Table 3. Generally, dietary intakes and breast milk concentrations were reduced from the first visit (group A) to the second visit (group B). Comparing the values from group A to their respective values from group B revealed no statistically significant difference between the two groups with the exception of mean dietary intake of iodine which was significantly higher in group A when compared to group B (p<0.05).

Correlations of dietary intakes of trace elements to their breast milk concentrations within each group and groups A and B together is presented in Table 4. Pearson's product moment correlation coefficients for the four trace elements were not statistically significant with the exception of group B dietary intake of zinc versus milk zinc concentration, which showed a statistically significant correlation (p<0.05). When groups A and B are combined, Pearson's product moment correlation coefficients for these four elements were not statistically significant.

Discussion

The results of this study are in agreement with the general finding that mineral content of human milk does not correlate well with maternal intake [10] but does not show correlation

Table 3 Zinc, Iron, Selenium, and Iodine Dietary Intakes and Human Milk Concentration Values of RGV Lactating Mothers

Group (Range of lactatingDays)	Dietary Zinc ^a (mg/d)	Human milk Zinc (mg/L)	Dietary Iron ^a (mg/d)	Human milk Iron (mg/L)	Dietary Selenium (µg/d)	Human milk Selenium (µg/L)	Dietary Iodine (µg/d)	Human milk Iodine (µg/L)
A (30–45 days)	6.1±4.6 ^b (n=31)	2.1±1.4 (n=30)	15.6±12.3 (n=31)	0.5±1.0 (n=30)	38.8±26.3 (n=30)	15.9±4.1 (n=20)	75.7±73.5 (n=25)	47.8±17.1 ^c (n=30)
B 75–90 (days)	5.0±3.1 (n=17)	2.0±1.7 (n=17)	10.8±5.8 (n=17)	0.4±0.3 (n=17)	35.8±22.3 (n=17)	15.7±5.3 (n=10)	51.5±52.7 (n=14)	42.3±8.71 (n=16)

^a Dietary intakes include prenatal zinc and iron supplements^b Mean ± SD^c Significantly different from group B (p<0.05)

Table 4 Pearson's Product Moment Correlation Coefficients for Dietary Intake Versus Milk Concentration of Trace Elements

	Dietary Zinc Vs. Milk Zinc	Dietary Iron Vs. Milk Iron	Dietary Selenium Vs. Milk Selenium	Dietary Iodine Vs. Milk Iodine
Group A	r=-0.03 p=0.86, NS*	r=-0.07 p=0.73, NS	r=-0.10 p=0.67, NS	r=0.29 p=0.18, NS
Group B	r=0.48 p<0.05	r=-0.37 p=0.15, NS	r=-0.39 p=0.30, NS	r=0.09 p=0.73, NS
Groups A&B Together	r=0.12 p=0.41, NS	r=-0.08 p=0.63, NS	r=-0.18 p=0.35, NS	r=0.22 p=0.19, NS

* NS—Not Significant

between dietary selenium and iodine with breast milk content of the two elements as reported by others [11, 12]. The amounts of trace elements measured in breast milk by atomic absorption and neutron activation are in agreement with values reported in the literature and food composition tables. One limitation of the study is estimation of dietary intakes of nutrients by a single 24-hour recall for every occasion. Also, the study would have benefited from larger sample size. The lactating mothers maintained same mean (\pm SD) weight and body mass index from first visit (group A) to the second visit (group B) as shown in Table 1 which is expected due to short time span between two visits (average of 45 days).

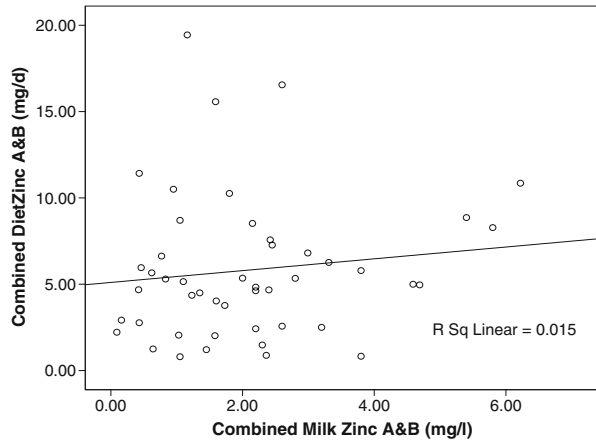
Zinc

The mean dietary Zn intake of the RGV lactating women, 6.1 ± 4.6 (group A) and 5.0 ± 3.1 (group B) mg/d in Table 3, were significantly less than the DRI recommended allowance of 12 mg of the National Academy of Sciences [23]. The mean milk concentration of zinc, 2.1 ± 1.4 (group A) mg/L and 2.0 ± 1.7 (group B) mg/L, are in agreement within the range of 1–3 mg of the representative values for Constituents of Human Milk (Table 3) [10]. This study found no significant correlation between dietary zinc and milk zinc in their concentration values for group A but observed significant correlation for group B ($p < 0.05$). But significant correlation was not observed between dietary zinc intake and milk zinc concentration for combined groups A and B in Table 4 (Fig. 1). Most previous studies found no significant correlation between maternal dietary zinc intake and milk zinc concentrations [24–26]. A slight increase in milk zinc after supplementation was observed in a single study [27] but in the larger study in subsequent period by the same researcher found no effect on milk zinc [28]. This may be due to adequate amount of zinc status in most of the women in those studies. Only few studies have been done on milk zinc in women with low zinc status, but extensive animal studies have been done but no effect on milk zinc has been found [29].

Iron

The mean Fe intakes of the RGV lactating women, 15.6 ± 12.3 (group A) and 10.8 ± 5.8 (group B) mg/d, were significantly higher than the DRI recommended allowance of 9 mg [23]. The use of supplement Fe may have contributed to higher intake value. The mean iron milk values of 0.5 ± 1.0 mg/L and 0.4 ± 0.3 mg/L were within the range value of 0.3–0.9 mg of the representative values for Constituents of Human milk (Table 3) [10]. There was no statistically significant correlation between maternal iron status and milk iron concentration

Fig. 1 Correlation between combined (groups A and B) dietary zinc and milk zinc ($r=0.12$, $p=0.41$)



observed in this study both for group A and B individually and also in combined groups A and B in Table 4 (Fig. 2). Previous studies have shown no statistically significant correlation between maternal dietary iron intake and human milk iron concentration [30, 31]. Maternal dietary iron has little effect on milk iron concentration [11].

Selenium

The RGV lactating women consumed mean dietary selenium intake of 38.8 ± 26.3 $\mu\text{g/d}$ (group A) and 35.8 ± 22.3 $\mu\text{g/d}$ (group B) compared to the DRI Recommended Allowance of 70 μg [23] which indicates almost 50% less intake values than the recommended value. The mean milk Se concentrations in the two groups, 15.9 ± 4.1 $\mu\text{g/L}$ and 15.7 ± 5.3 $\mu\text{g/L}$ respectively, were at the lower end of the representative values of 7–33 μg of Human milk (Table 3) [10]. In selenium study, no statistically significant correlation was observed between dietary selenium and milk selenium values both in groups A and B individually and also in combined groups A and B. in Table 4 (Fig. 3). Some researchers did not also

Fig. 2 Correlation between combined (groups A and B) dietary iron and milk iron ($r=-0.08$, $p=0.63$)

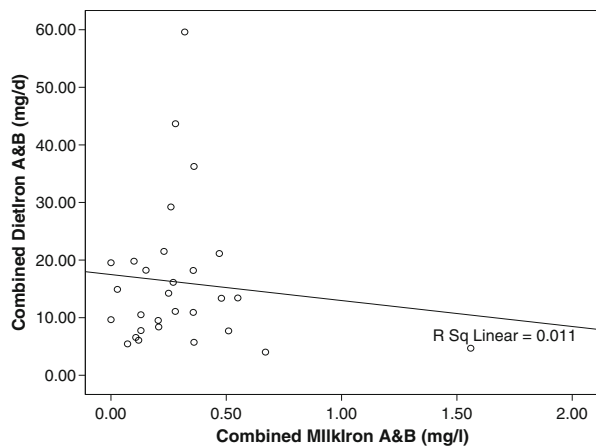
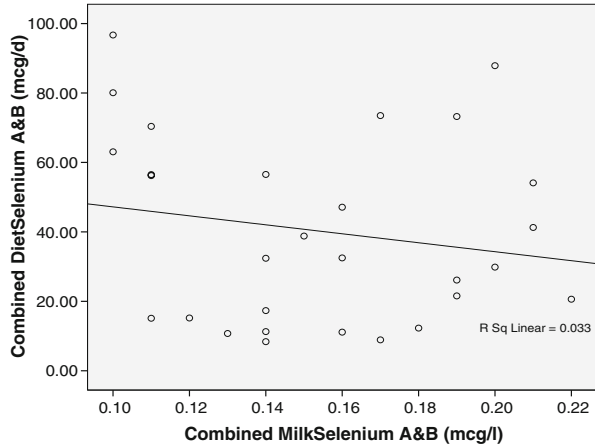


Fig. 3 Correlation between combined (groups A and B) dietary selenium and milk selenium ($r=-0.18$, $p=0.35$)

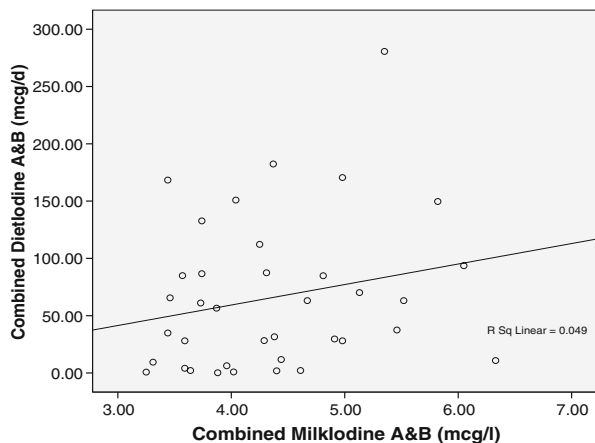


find statistically significant correlation between dietary selenium and milk selenium values [13] but some investigators found selenium concentration in breast milk to be closely related to dietary selenium intake [32–34].

Iodine

The lactating mothers' daily mean dietary iodine intakes were 75.7 ± 73.5 $\mu\text{g/d}$ (group A) and 51.5 ± 52.7 $\mu\text{g/d}$ (group B) which were significantly less (~75–80%) than the DRI Recommended intake value of 290 mcg for lactating women [23]. The mean milk iodine values in the two groups were obtained as 47.8 ± 17.1 $\mu\text{g/L}$ and 42.3 ± 8.7 $\mu\text{g/L}$ respectively, and were also significantly less (~70%) than 150 μg in the representative values for Constituents of Human Milk (Table 3) [10]. The maternal iodine status reduced significantly in group B than group A. Significant statistical correlations were observed both for dietary iodine intake for groups A and B ($p < 0.05$) and milk iodine concentration for groups A and B ($p < 0.05$) but no statistical correlation was observed between dietary iodine intake and milk iodine concentrations for groups A and B individually and combined

Fig. 4 Correlation between combined (groups A and B) dietary iodine and milk iodine ($r=0.22$, $p=0.19$)



groups A and B in Table 4 (Fig. 4). Some investigators found statistically significant correlation between dietary iodine intake and milk iodine concentration [12].

Conclusions

The energy intakes of RGV lactating mothers were substantially lower than the recommended percent values. The low energy reserves may be due to malnutrition. Protein intakes were substantially higher than the percent recommended values. The dietary percent zinc, iron, selenium, and iodine intakes have significantly lower values than the recommended percent values as shown in Table 2. This suggests low consumption of dietary intakes for these lactating mothers.

The daily dietary intake of Zn, Se, and I were significantly less than the DRI Recommended Dietary Allowances though the Fe intake was higher than the DRI value. The dietary intake values were estimated by a single 24-hour recall for every occasion. These values may accurately be evaluated by estimating several 24-hour recalls during the lactating period. Breast milk concentration values of Zn, Fe, and Se were within the range of representative values for Constituents of Human Milk though iodine was significantly less than the representative value.

It appears that the diet consumed by the study participant in the Rio Grande Valley for Zn, Fe, and Se was adequate to support the health and growth of their breast-fed infants except Iodine has substantially low concentration value in the mother milk.

In this work, there was no statistically significant correlation obtained between dietary intake and milk concentration for zinc, iron, selenium, and iodine for combined groups A and B but other researchers have found correlation between selenium and iodine. The authors recommend larger sample size to accurately evaluate statistically significant correlation between dietary intake and maternal milk.

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