Correlation between Fluoride in Drinking Water and Its Levels in Breast Milk in Golestan Province, Northern Iran

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Abstract
Background: Fluoride is an essential element for human health. However, excess fluoride in drinking water may cause dental and/or skeletal fluorosis. Drinking water is the main route of fluoride intake. The aim of the present study was to measure fluoride levels in human breast milk collected from two regions of Golestan Province, northern Iran with different amount of fluoride concentration of drinking water in Bandar Gaz and Nokande cities and to correlate it with fluoride concentrations in drinking water used by mothers living in these two areas.
Methods: Twenty samples of water were collected from seven drinking water wells during 2012 from Bandar Gaz and Nokande in Iran during 2012. Fluoride concentration of water samples was measured using SPADNS method. Sixty breast milk samples were collected from lactating mothers of Bandar Gaz and Nokande cities. Content in breast milk was determined using standard F ion-selective electrode. Spearman’s rho correlation analysis was used to assess any possible relationship between fluoride levels in breast milk and in drinking water.
Results: The means and standard deviation for F concentration in breast milk and drinking water were 0.002188±0.00026224 ppm and 0.5850±0.22542 ppm, respectively. Analysis of data showed that the variables were not normally distributed so the Spearman correlation coefficient between two variables calculated (\( \rho_s = 0.65 \)) and it was significant (P=0.002).
Conclusion: Fluoride concentration in water can directly act on its concentration in breast milk. We speculate that modifying F concentration in water can affect accessibility of fluoride for infants.

Keywords: Breast milk, Fluoride, Drinking Water, Iran

Introduction

The relationship between fluoride and human health has been studied for years and scientists have recognized the variable fluoride concentrated in teeth and bones of humans (1). Most health researches focus on chronic fluoride exposure from drinking water, because that is the easiest way to quantify for a community served by a public water supply. However, food provides another potentially important pathway, as do toothpaste and other products used for dental health (2). Fluoride gets concentrated in hard tissues of human body and plays an important role in mineralization of bones and teeth. Excessive intake of F\(^{-1}\) causes dental/or skeletal fluorosis. There are
many studies regarding fluoride in drinking water and tooth DMFT index in children (3).

Fluoride can be absorbed by human body via drinking water, food, toothpaste, mouthwash products and via air. Drinking water is the main source of fluoride intake in most of communities. Food is the other important source of fluoride intake (4-6). Breast milk is considered an important nutrient for infants, especially in the first six months of life, where milk is considered as the main diet. After 6 months of age, the dietary importance of milk gradually changes due to supplementation of other foods. Hence, it is important to identify the potential milk source of fluoride concentration intake in an infant’s diet and to evaluate the need for fluoride supplementation (7).

Knowledge of fluoride content in drinking water, food, tea, etc., across the country is important for health care personnel. There are only few studies on the level of fluoride in drinking water, black tea and tea liquor and powdered milk in Iran (8-12). Fluoride concentration of breast milk displays an increase along with fluoride intake. The level of fluoride in human milk has been the topic of investigation for many years.

We conducted this study to determine levels of fluoride in breast milk of lactating mothers in Golestan province (Bandar Gaz and Nokande cities) and to correlate it with fluoride content in drinking water for domestic use.

**Materials and Methods**

*Sample collection & Extraction and determination of fluoride in breast milk*

Twenty mothers were randomly chosen from 450 lactating mothers, and then breast milk samples were taken three times during every two months and analyzed. Totally 60 samples of breast milk were collected from lactating mothers in Bandar Gaz and Nokande cities during 2012. A total of 20 human milk samples were randomly collected. Samples were analyzed in the Central Nutrition Laboratory of Tehran. All of the reagents used in the analyses of fluoride were reagent grade and supplied by MERCK Inc. Sodium fluoride was used for preparing stock and standard solutions.

The fluoride stock solutions (0.5 mmol/mL) were prepared monthly and stored at 4 °C. Working standard solutions of different concentrations were prepared daily by diluting the intermediate standard solution with de-ionized water. A Metrohm (Model 744) bench top pH/ion meter (fluoride ion-selective electrode as working electrode and silver/silver chloride as reference electrode) was used for potentiometric determination of fluoride in milk samples. The pH of the sample solutions was monitored simultaneously with a conventional glass pH electrode. The fluoride concentration in dry milk samples was determined by comparing the potential of the sample solutions with calibration curve constructed from a series of standard fluoride solutions. The fluoride ISE determined fluoride concentration by measuring the potential difference across an interface that responds only to fluoride. The accuracy of the fluoride ion selective electrode was also evaluated by the recovery test. Recoveries of fluoride from milk sample spiked with three different amounts of sodium fluoride were tested. Recoveries between 101 to 105% were obtained.

This method showed a very good accuracy due to the high recovery (13). Twenty water samples were collected from 7 wells drinking water in 2012 from Bandar Gaz and Nokande cites in Golestan Province Iran.

Fluoride concentration of water samples was measured using SPADNS method. The standard SPADNS method was used with a DR/2000s Spectrophotometer (HACH Company, USA (14). Finally, quantitative data were statistically processed by means of SPSS version 18. The normality of variables was evaluated using Kolmogorov-Smirnov and Shapiro-Wilk tests. Spearman’s rho correlation analysis was used to assess any possible relationship between fluoride levels of breast milk and levels of fluoride in drinking water.

**Results**

The concentrations of fluoride in breast milk, and in drinking water are presented in Table 1, 2. Fluoride levels of water in Bandar Gaz and Nokande were in range of 0.3-0.5 mg/L and 0.6-0.8
mg/L, respectively. The means and standard deviation for F concentration in breast milk and drinking water were 0.002188±0.00026224 ppm and 0.5850±0.22542 ppm, respectively. The minimum and maximum levels of F were 0.0014 ppm and 0.0025 ppm, respectively. Results are shown in Table 3. Human breast milk contained lower fluoride concentration, considering that mean fluoride in drinking water is about 0.53±0.09 mg/L in this area. Analysis of data showed that the variables were not normally distributed so the Spearman correlation coefficient between two variables calculated (\( \rho_s = 0.65 \)) and it was significant (\( P=0.002 \)).

### Table 1: Concentration of fluoride in breast milk and water in Nokandeh, Golestan Province, Iran

| Number sample | F content in milk (ppm) | F content in water (ppm) |
|---------------|-------------------------|--------------------------|
| 1             | 0.00226                 | 0.8                      |
| 2             | 0.00225                 | 0.8                      |
| 3             | 0.0023                  | 0.8                      |
| 4             | 0.0023                  | 0.8                      |
| 5             | 0.0025                  | 0.8                      |
| 6             | 0.0022                  | 0.7                      |
| 7             | 0.0023                  | 0.8                      |
| 8             | 0.00245                 | 0.8                      |
| 9             | 0.0023                  | 0.8                      |
| 10            | 0.0024                 | 0.8                      |

### Table 2: Concentration of fluoride in breast milk and water in Bandar Gaz, Golestan Province, Iran

| Number sample | F content in milk (ppm) | F content in water (ppm) |
|---------------|-------------------------|--------------------------|
| 1             | 0.00182                 | 0.3                      |
| 2             | 0.00223                 | 0.4                      |
| 3             | 0.00226                 | 0.5                      |
| 4             | 0.0021                  | 0.3                      |
| 5             | 0.00224                 | 0.4                      |
| 6             | 0.0017                  | 0.3                      |
| 7             | 0.00227                 | 0.3                      |
| 8             | 0.00233                 | 0.3                      |
| 9             | 0.0014                  | 0.3                      |
| 10            | 0.0022                  | 0.5                      |

### Table 3: Fluoride concentration of breast milk and drinking water samples (ppm)

|                          | Minimum | Maximum | Mean Std. Deviation | W.H.O guide line for Fluoride concentration |
|--------------------------|---------|---------|---------------------|--------------------------------------------|
| Breast milk (n=20)       | 0.00140 | 0.00250 | 0.002188±0.00026224 | 5–10 µg/l                                   |
| Drinking water (n=20)    | 0.3     | 0.8     | 0.5850±0.22542      | 0.5-1.5 mg/l                                |
| Correlation coefficient  | \( \rho_s = 0.65 \) | p-Value =0.002      |

**Discussion**

Our study revealed the means and standard deviation for F concentration in breast milk and drinking water was in range of 0.002188±0.00026224 ppm and 0.5850±0.22542ppm, respectively. Levels of minimum and maximum of F were 0.0014 ppm and 0.0025 ppm, which comply with WHO guidelines. According to WHO, levels of fluoride in breast milk should be in range 5-10 µg/l (15). Fluoride concentration in studying water samples ranged from 0.3 to 0.8 ppm (Table 1, 2). Forty five percent of the samples had a fluoride level lower than the permissible limit, 55% of the samples had a level within the optimum limit of 0.5 to 1.5 ppm.
Result showed that fluoride content in breast milk had correlation with drinking water ($\rho_s = 0.65$) and ($P=0.002$). Table 3, which is in accordance to similar studies conducted in Iran and worldwide. Dabeka et al. revealed that levels of fluoride in breast milk had correlation to levels of fluoride drinking water consumed by the mothers (17). In Egypt, the fluoride levels of plasma, breast milk and drinking tap water samples were 0.0207±0.012, 0.0111±0.0044 ppm, and 0.482±0.117 ppm, respectively. A significant direct correlation between fluoride content in breast milk and drinking tap water was found ($r=0.858$, $P<0.01$) (7). The mean fluoride concentration in breast milk was 0.017±0.02 ppm. No correlation was found between breast milk fluoride content and fluoride levels in either drinking water or water for domestic use ($r = -0.09$, $P = 0.32$, $r = -0.04$, $P = 0.65$) respectively that disagrees with our finding (18). It is recommended in areas with high concentrations of fluoride in water, in order to reduce the amount of daily intake by mothers, fluoride be removed from drinking water by proper process (19).

**Conclusion**

All the samples had detectable levels of fluoride implying that mother's nutrition in the area studied is widely affected the F concentration of breast milk. The present study was carried out to understand if the low and high F concentration in Bandar Gaz and Nokande water could cause low and high fluoride intake of human breast milk, respectively. The F content in human breast milk depends on various factors, such as drinking tea and other source of fluoride ingestion provides far more fluoride than is contained in drinking water alone. Our study shows that fluoride concentration in water can directly act on its concentration in breast milk. We speculate that modifying the F concentration in water can affect accessibility of fluoride for infants.

**Ethical considerations**

Ethical issues (Including plagiarism, informed consent, misconduct, data fabrication and/or falsification, double publication and/or submission, redundancy, etc.) have been completely observed by the authors.

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