Concentration of Fluoride in Cow's and Buffalo's Milk in Relation to Varying Levels of Fluoride Concentration in Drinking Water of Mathura City in India– A Pilot Study

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ABSTRACT

Aim: To estimate fluoride concentration in drinking water, cow's milk and buffalo's milk and to correlate the concentration of fluoride in cow's milk and buffalo's milk with varying levels of fluoride concentration in drinking water.

Materials and Methods: Ten households having both cows and buffalo's were selected by convenience in each of the 3 zones (below optimum fluoride <0.7 ppm (parts per million), optimum fluoride 0.7-1.2 ppm and above optimum fluoride areas > 1.2 ppm). From these selected households, 200 ml of fresh milk of both cows and buffaloes was collected along with 200 ml of drinking water for estimation of fluoride concentration by using a fluoride ion selective electrode method. The data was analysed using SPSS, version 11.5 for windows.

Results: The mean fluoride concentration of drinking water, cow's milk and buffalo's milk in three different fluoride zones was 0.89 ± 0.39 , 0.09 ± 0.07 , 0.09 ± 0.08 respectively. Pearson's correlation found a statistically significant correlation between fluoride concentrations in cow's and buffalo's milk with varying levels of fluoride concentration in drinking water in zone B and zone C. However, this correlation was not statistically significant in zone A.

Conclusion: With an increase in fluoride concentration in drinking water there was an increase in concentration of fluoride in cow's and buffalo's milk. We conclude that this association is seen in conjunction to not only a single factor but rather due to culmination of several other aspects. So, there is a need to elucidate the other factors that might be contributing to this increase and dental fluorosis.

Keywords: Dental fluorosis, Drinking water fluoride, Milk

INTRODUCTION

Fluorine being most electronegative of all chemical elements; it rarely exist in elemental form. It is largely found in chemically combined state known as "Fluoride" and is the 13th most prevalent naturally occurring element in earth's crust. Fluoride is found in both igneous and sedimentary rocks and its concentration varies at different geographical locations on the earth. Fluoride plays a key role in the prevention and control of dental caries [1]. World Health Organization has also approved fluoride as a preventive measure for dental caries and recommended its appropriate use [2].

Fluoride is a double edged sword. Fluoride on one side, at optimal level gives its beneficial effects as prevention of dental caries and helps maintaining the integrity of oral tissues whereas on other side fluoride exerts its adverse effect such as dental and skeletal fluorosis when ingested continuously and un- interruptly [3].

It was known that dental caries prevalence was less among individuals living in communities where fluoride concentration in drinking water was 1mg/lt. The cariostatic effect of fluoride was predominantly systemic rather than local effect at surface of the tooth. It is also documented that higher level of fluoride concentration in drinking water has shown to be associated with development of dental fluorosis [4].

In a recent unpublished study in Mathura city, it was observed that there is a decrease in incidence of dental caries with increase in concentration of fluoride in drinking water but it was worthy to note that mean fluoride concentration in drinking water ranging from 0.2 ppm to 1.5 ppm, there is varying degree of dental fluorosis though mild to very mild in nature [5]. This observation substantially supports that fluoride consumption from other sources must be playing an important role in increasing the fluoride availability for consumption leading to dental fluorosis. Taking clue from this study, an attempt was made to find out the concentration of fluoride in cow's milk and buffaloes milk living in the same locality with varying levels of fluoride concentration in drinking water. Since, many people in this Holy Mathura city are vegetarians and consume milk regularly the concentration of fluoride in milk might contribute significantly to the total daily consumption of fluoride. In Mathura city, cow's and buffalo's consume water from the same source as of the household thus the fluoride concentration in milk might be one of the contributing factor to total daily fluoride intake leading to the increased prevalence of dental fluorosis.

Keeping this observation in view, an attempt was made to find out the concentration of fluoride in cow's and buffalo's milk in relation to varying levels of fluoride concentration in drinking water of different localities of Mathura city.

MATERIALS AND METHODS

A pilot study was designed and ethical permission form was taken from institutional ethical committee before the commencement of study. Purpose of study was explained to all individual households and their consent was taken before collecting milk sample. The study was conducted from June 2012 to July 2012 over the period of one month.

Sampling Design: In the present study, Mathura city was divided into 3 fluoride zones based on concentration of fluoride in drinking water obtained from previous study and as reported by Public Water Supply Department of Mathura City [5].

The zones were: Zone A - Below optimum fluoride (<0.7 ppm); Zone B - Optimum fluoride (0.7-1.2 ppm); Zone C - Above optimum fluoride areas (> 1.2 ppm).

From each of these 3 zones, 10 households having both cows and buffaloes were selected by convenient sampling.

Milk Sample Collection: From 10 selected households each in 3 zones, 200 ml fresh milk sample of both cows and buffaloes were collected from the udder of cow and buffalo respectively and was poured into a plastic bottle.

Water Sample Collection: Likewise, 200 ml of water samples were collected from the same 10 households each in 3 zones from where milk samples were collected. The water samples were taken for estimation of fluoride concentration in drinking water.

Storage and Transportation of Milk Sample: The milk in the plastic bottle from 10 selected households each in 3 zones was then separately poured into the steel container for boiling and was then cooled to room temperature before pouring it back into the plastic bottle. The milk was then placed in the icebox overnight and was transported to the laboratory for fluoride estimation.

Estimation of Fluoride Concentration in Milk: Fluoride estimation in milk was determined by electrochemical means using a fluoride ion selective electrode (Orion-9609BNWP) coupled to an ion analyzer (Orion-4 Star). Before estimating the fluoride content in milk, milk was brought to a constant temperature of 25°C in a water bath and the instrument was calibrated using fluoride standards containing TISAB III which encompassed the range of the measurement to be made (fluoride standards were 1ppm F and 10 ppm F). After the calibration was done, 50 ml of milk was measured using a measuring beaker and to this 5 ml of TISAB III was added using a pipette which standardized the ionic strength and pH of the medium. The solution was stirred at a uniform rate until equilibrium was reached. Once the equilibrium was reached, the reading was recorded from the ion meter in ppm [6] [Table/Fig-1].

Estimation of Fluoride Concentration in Drinking Water: Same method was employed for the estimation of fluoride concentration in drinking water.



RESULTS

[Table/Fig-2] shows that when independent t-test was used to compare fluoride concentration in cow's and buffalo's milk in relation to varying levels of fluoride concentration in drinking water, no statistically significant difference was observed between the fluoride concentration of cow's milk and the fluoride concentration of buffalo's milk.

[Table/Fig-3] show that when Pearson's correlation was used to correlate fluoride concentration in cow's and buffalo's milk with different levels of fluoride concentration in drinking water in 3 zones. It was found that there was a significant increase in fluoride concentration in cow's and buffalo's milk with increase in fluoride concentration in drinking water. A positive correlation ranging from weak to strong exist between fluoride concentration in cow's and buffalo's milk with different levels of fluoride concentration in cow's and buffalo's milk with different levels of fluoride concentration in drinking water in 3 zones. However, this correlation was not significant statistically in Zone A as compared to Zone B and C.

Zones	Fluoride Concentration (Mean ± SD) (ppm)			t	p-value	Significance
	Drinking Water	Cow's milk	Buffalo's milk			
Zone A	0.47 ± 0.11	0.016 ± 0.006	0.019 ± 0.007	0.93	0.36	Not significant
Zone B	0.82 ± 0.13	0.074 ± 0.01	0.078 ± 0.01	0.71	0.48	Not significant
Zone C	1.38 ± 0.09	0.18 ± 0.06	0.18 ± 0.07	0.00	1.00	Not significant
Total	0.89 ± 0.39	0.09 ± 0.07	0.09 ± 0.08			

[Table/Fig-2]: Comparison of fluoride concentration in cow's and buffalo's milk in relation to varying levels of fluoride concentration in drinking water of 3 different zones ($p \le 0.05 - Significant, Cl = 95\%$)

Zones	Cow's milk			Buffalo's milk				
	r-value	p-value	Inference	r-value	p-value	Inference		
Zone A	0.52	0.12	Moderate positive	0.48	0.16	Weak positive		
Zone B	0.73	0.01*	Moderate positive	0.92	0.00*	Strong positive		
Zone C	0.68	0.02*	Moderate positive	0.70	0.02*	Moderate positive		

[lable/Fig-3]: Correlation of fluoride concentration in cow's and buffalo's milk with varying level of fluoride concentration in drinking water of 3 different zones (Pearson's correlation) ($p \le 0.05 - Significant$, Cl = 95 %)

$(p \le 0.03 - 3)g(n)(a)(n, 0) = 93 / 6)$

STATISTICAL ANALYSIS

The data obtained was analysed using SPSS (Statistical Package for the Social Sciences), version 11.5 for windows. Mean and standard deviations were calculated for each clinical parameter. Independent t-test was used to compare the fluoride concentration in cow's and buffalo's milk in relation to levels of fluoride concentration in drinking water. The correlation between fluoride concentrations in milk at varying levels of fluoride concentration in water was determined by Pearson's correlation test.

DISCUSSION

There are several methods available to estimate fluoride concentration in water and milk such as spectrophotometry, gas chromatography, capillary electrophoresis, micro diffusion, and ion analysis in conjunction with ion-selective electrodes. However in the present study, "ion selective electrode coupled to an ion analyser" method was used to estimate fluoride concentration in milk and drinking water [7].

The level of fluoride in milk has been a subject of disagreement for many years. With advances in analytical technology, the reported fluoride concentration has steadily declined [8]. Present study showed that the mean fluoride concentration of cow's milk is $0.09 \pm$ 0.07 and buffalo's milk is 0.09 ± 0.08 . Similar fluoride concentration of 0.02-0.8 ppm in cows and milk formulas; bovine milk in the range of 0.007 to 0.08 ppm have also been observed by Tinanoff et al., [9], Vlachou et al., [10], Larsen et al., [11] and Dabeka et al., [12].

In the present study, a weak to strong correlation was found between fluoride concentration in cow's milk and buffalo's milk at different levels of fluoride concentration in drinking water. The study showed that with increase in fluoride concentration in drinking water there was increase in concentration of fluoride in cow's and buffalo's milk. To best of our knowledge there is lack of pertinent studies in literature which attempted to establish relation between fluoride concentration in drinking water and concentration of fluoride in cow's and buffalo's milk. However, we assume that it might be attributed to several factors such as drinking water, fodder consumed and consumption of plantations which are grown in the same area.

Fluoride combines with the calcium component of the milk so amount of fluoride available for absorption is very less. So, consumption of milk alone is not likely to cause dental fluorosis, intake of milk with high fluoride content together with fruit juices, beverages such as tea, coffee etc. the safety level of daily fluoride consumption could easily be exceeded by such combination of diet [13]. So, there is need to do more extensive studies taking accountability of these factors which might be contributing to prevalence of dental fluorosis. Present study have certain limitations that there is small sample size, cow's and buffalo's were of different age, size, consume different amount of water, fodder and the amount of milk produced by them vary and in present study blood fluoride level is also not evaluated of people living there. Since, present study is a pilot study considered being a pioneer study in this region to best of our knowledge; we are in process of conducting a larger dairy based population with a wider geographical distribution by keeping in mind the limitations of present study.

CONCLUSION

With increase in fluoride concentration in drinking water there was increase in concentration of fluoride in cow's and buffalo's milk. We conclude that this association is seen in conjunction to not only a single factor but rather due to culmination of several other aspects. So, there is need to elucidate the other factors that might be contributing to this increase and dental fluorosis.

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