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Fluoride concentrations in typical Brazilian foods and in infant foods

ABSTRACT

OBJECTIVE: To determine fluoride concentrations in the typical Brazilian meal (rice with beans) and in processed infant foods, and to estimate their contribution towards dental fluorosis.

METHODS: The foods were purchased at supermarkets in the cities of Piracicaba and Campinas, Southeastern Brazil. The processed infant foods were bought in 2001 and the rice and beans in 2003, and they were analyzed immediately. Three brands of rice, three brands of beans and 36 samples of infant foods were analyzed, divided into five groups: ready-to-eat, porridges, formulated foods, powdered milk and others. For the rice and beans, fluoride concentrations were determined in the raw grains and after they were cooked with fluoridated (0.7 ppm) or distilled water. All the fluoride analyses were performed using a specific electrode. A dose of 0.07 mg/kg/day was considered to be the upper limit of fluoride exposure in terms of fluorosis risks.

RESULTS: The fluoride concentrations found in the grains of rice and beans were low. However, they increased 100 to 200-fold after cooking in fluoridated water. Even so, they were lower than what is found in some processed foods. A meal of rice and beans prepared with fluoridated water would be responsible for 29% of the threshold dose for fluoride intake in terms of acceptable fluorosis; the contribution from some processed foods reaches 45%.

CONCLUSIONS: The typical Brazilian food, even when prepared with fluoridated water, is safer in terms of the risk of dental fluorosis than are some processed infant foods.

KEY WORDS: Food analysis. Infant food, analysis. Fluorides, analysis. Fluorosis, Dental, prevention & control.

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INTRODUCTION

Fluoride use has been the main public health measure for controlling dental caries both in developed and in developing countries.[3,15] However it has led to an increase in dental fluorosis.[1,5]

Dental fluorosis is a deficiency in enamel mineralization due to daily fluoride intake during tooth development.[7] Since the dose-effect relationship is not precisely known, the dose of 0.07 mg F/day/kg of body weight has been accepted as the upper limit in terms of the clinically acceptable risk of dental fluorosis.[2]

The main sources of fluoride that have been associated with increased dental fluorosis are fluoridated water, fluoridated supplements, fluoridated dentifrices and processed baby foods consumed before the age of six years.[14] Although the consumption of processed foods is low in developed countries, their contribution

towards the risk of dental fluorosis has been extensively studied in these countries,[11] but little is known in developing countries.[4] Besides the consumption of processed foods, the contribution of the typical diet of each country towards the risk of dental fluorosis should be taken into account. As regards diet, data from two Brazilian cities with fluoridated water has shown that diet contributes 31-44% to the daily intake of fluoride by children aged 2-3 years.[17]

Rice with beans (in the approximate proportion of 2:1) is the typical Brazilian dish, and its contribution towards daily fluoride consumption, *per se*, or when cooked with fluoridated water, is unknown. A low fluoride concentration was found in Brazilian beans bought on the Japanese market,[16] but the sample chosen was unrepresentative of the product that the Brazilian population consumes, and the accompanying rice was not analyzed.

Although dental fluorosis is the only side-effect attributed to exposure to low fluoride concentrations such as water fluoridation,[13] other effects due to exposure to multiple sources should not be ignored.

Therefore, the present study aimed at analyzing the fluoride concentrations in rice, beans and processed baby foods sold in Brazil, and to estimate their contributions towards the development of dental fluorosis.

METHODS

Six 1 kg packs of rice grains and beans of the three brands most sold in the stores in Piracicaba, State of São Paulo, Brazil, were bought at supermarkets. The processed baby foods were bought in the city of Campinas, State of São Paulo, Brazil, and were grouped into five categories: ready-to-eat foods (N=9), foods for porridge (N=11), baby formulas (N=9), powdered milk (N=4) and other foods (N=3). For each food, within each category, two to six samples were bought, in up to three commercial stores. The brand names of the products were omitted and were replaced by codes in the results tables, since the aim of this study was not to compare the products within the same category but between categories.

For analysis of the raw grains, these were ground up in a domestic food processor until homogeneous powders were obtained. Triplicate samples of 600 mg were weighed (± 0.01 mg) on petri dishes (Falcon 1007), to which 2.4 mL of deionized water was added.

To analyze the fluoride in cooked rice and beans, six samples of each brand of grain were cooked in deionized or fluoridated water (0.7 ppm F). The cooking conditions were standardized, particularly the ratio between grain weight and water volume (weight/volume, w/v), which was 1:2 and 1:4, respectively for the rice

and beans. The cooked foods were homogenized in a blender with deionized water in the proportions (w/v) of 1:2 for the rice and 1:1 for the beans. A volume of 3.0 mL of each homogenized food was transferred to petri dishes.

Triplicate samples of each processed foods were weighed (± 0.01 mg) in petri dishes to which 3.0 ml of deionized water was added.

The samples of rice and beans (raw and cooked) and the samples of processed foods placed in the petri dishes were subjected to microdiffusion facilitated by hexamethyldisiloxane (HMDS),[21] to separate and subsequently analyze the fluoride present.

Before sample weighing, a cap from a plastic tube (Falcon 2030) was placed using vaseline in the center of a plastic petri dish. After sample weighing, 0.10 mL f 1.65N NaOH[12] was placed in the cap. The petri dish was sealed with vaseline and 1.0 mL of 6N HCl saturated with HMDS added to the sample through a hole made in the petri dish cap. The hole was sealed with vaseline and the assemblage was shaken at room temperature. After 12-18h the Falcon plastic cap containing fluoride diffused from the sample was put to dry at 60°C. This cap was then put into a plastic assay tube (Falcon 2017) containing 0.40 mL of 0.66N acetic acid. The tube was inverted and vigorously shaken to dissolve the fluoride crystals present in the cap. The fluoride concentration was determined by means of a specific electrode (Orion Research Inc., Model 96-09; Boston, United States) coupled to an ion analyzer (Orion Research Inc., Model EA 940, Boston, United States). Standard solutions (Orion #940907) in triplicates, at concentrations from 0.05 to 2.5 mg F/mL, were also acid-diffused. Likewise, blanks were subjected to this procedure.

With the aim of ensuring accurate determination, increasing quantities of powdered grains were subjected to acid-diffusion and the degree of linearity between the weight and fluoride concentration was determined.

Table 1. Concentration of fluoride (μg F/g, dry weight) in raw grains of rice and beans. Piracicaba, Southeastern Brazil, 2003. N=6

Grain	Brand name	Mean concentration (μg F/g)
Rice	A	0.031±0.011*
	B	0.012±0.003*
	C	0.277±0.164*
Beans	D	0.043±0.030
	E	0.086±0.045
	F	0.042±0.011

* Values differ statistically (p<0.05) among brands (for the same type of food – Kruskal-Wallis test)

Table 2. Fluoride concentrations in brands of rice and beans cooked either in fluoridated or non-fluoridated water, and daily estimated percentage contributions towards the threshold dose for the risk of dental fluorosis. Piracicaba, Southeastern Brazil, 2003. N=6

Food brand	Mean concentration (μg F/g)	Estimated dose (mg F/kg/day)	%*
Cooked with non-fluoridated water			
Rice A	0.015±0.006	0.00014	0.2
Rice B	0.019±0.003	0.00017	0.2
Rice C	0.100±0.033**	0.00090	1
Beans D	0.077±0.084	0.00061	0.9
Beans E	0.301±0.151**	0.00340	5
Beans F	0.069±0.043	0.00055	0.8
Cooked with fluoridated water			
Rice A	1.046±0.124	0.00943	13
Rice B	0.836±0.104**	0.00754	11
Rice C	1.014±0.072	0.00914	13
Beans D	1.476±0.238	0.01170	17
Beans E	1.538±0.238	0.01219	17
Beans F	1.469±0.125	0.01164	16

* Percentage contributions towards threshold dose (0.07 mg F/kg/day) of dental fluorosis.
** Values that differ statistically (p<0.05) from the others within the same category (same kind of food and cooking conditions)

Thus, it was found that, for the raw foods, 600 mg was the ideal weight of powdered grains for use in the analysis, since this quantity in 3.0 mL of water enabled adequate homogenization during microdiffusion and the quantity of fluoride extracted satisfied the sensitivity limit of the electrode.

For the cooked foods, standardization was done with homogenized preparations using different proportions of water (1:1 to 1:9; w/v). The best results were 1:1 for the beans (r=1.00) and 1:2 (r=0.99) for the rice.

Due to the very low concentrations of fluoride in the group of processed foods, the maximum amount of sample to be weighed was also standardized, which would enable sample homogenization and accurate fluoride determination. It was found that a mean of 103% of the fluoride present in the sample was recovered.

Taking into consideration the upper limit of 0.07 mg F/kg/day for the risk of fluorosis,[3] the amount of food intake per day was estimated. The amount of rice and beans consumed by 27 children, aged from 20 to 30 months, who were attending the São Vicente de Paulo public daycare center in Piracicaba, State of São Paulo, was determined by the duplicated-diet technique.[12] The mean amounts (±SD) found were 119.1 ± 17.0 and 104.6 ± 14.8 g/day of rice and beans, respectively.

For the group of processed foods, the estimates were based on the recommended intake indicated on the product package, considering two-year-old children weighing approximately 13.2 kg.

The results found from the raw rice and bean samples and from the cooked beans were analyzed using the Kruskal-Wallis test and the cooked rice data were analyzed using the Tukey test. For all statistical analyses, the Bioestat* software was used and the significance limit was set at 5%. The results from the processed foods were analyzed descriptively.

RESULTS

The fluoride concentrations in the rice grains of the different brands analyzed differed statistically (p<0.05), and were highest in the A brand rice (Table 1), but among the bean samples no significant difference was found (p>0.05).

Table 2 shows that the fluoride concentrations found both in rice and in beans cooked with deionized water differed statistically among the samples analyzed (p<0.05). Nonetheless, the contribution of meals based on rice and beans, cooked with deionized water, towards the threshold dose for the risk of dental fluorosis, should be considered negligible (<1%). However, when the

Table 3. Fluoride concentration in the group of ready-to-eat foods, in daily intake dose and percentage contributions towards the threshold dose for dental fluorosis. Piracicaba, Southeastern Brazil, 2001.

Food (N)	Mean concentration* (µg F/g)	Estimated dose (mg F/Kg/day)	%**
Apple (N=5)	0.009 ± 0.0060	<0.001	<1
Pear (N=5)	0.017 ± 0.0154	<0.001	<1
Apple and orange (N=3)	0.010 ± 0.0020	<0.001	<1
Assorted fruits (N=4)	0.016 ± 0.0023	<0.001	<1
Banana with oats (N=6)	0.067 ± 0.0286	0.0010	1.5
Chicken and vegetables (N=4)	0.082 ± 0.0187	0.0012	1.8
Meat, vegetables, rice and egg yolks (N=3)	0.055 ± 0.0105	0.0008	1.2
Meat, vegetables and cereals (N=6)	0.084 ± 0.0258	0.0012	1.8
Meat, vegetables and macaroni (N=4)	0.019 ± 0.0116	<0.001	<1

* wet weight
** Percentage contributions towards threshold dose (0.07 mg F/kg/day), in terms of the acceptable dental fluorosis.

Table 4. Fluoride concentrations in the group of formulated foods, estimated daily intake dose per child and percentage contributions towards the threshold dose for dental fluorosis. Campinas, Southeastern Brazil, 2001.

Food (N)	Mean concentration* (µg F/g)	Estimated dose (mg F/Kg/day)	%**
Milk-based formulas***			
A (N=4)	0.52 ± 0.011	0.0080	11
B (N=5)	1.67 ± 0.199	0.0126	18
C (N=3)	0.47 ± 0.043	0.0043	6
D (N=2)	0.84 ± 0.063	0.0054	8
Soybean-based formulas			
A (N=2)	3.84 ± 0.036	0.0093	13
B (N=4)	2.92 ± 0.079	0.0314	45
C (N=2)	1.41 ± 0.195	0.0037	5
D (N=3)	0.87 ± 0.174	0.0046	7
E (N=3)	2.80 ± 0.509	0.0200	29

* wet weight
** Percentage contributions towards dose (0.07 mg F/kg/day) in terms of acceptable dental fluorosis.
*** Reconstituted with non-fluoridated water

meals were prepared with fluoridated water, the fluoride concentrations were much higher, and it was estimated that a meal with rice and beans would contribute 11% to 13% and 16% to 17%, respectively, to the threshold dose for the risk of dental fluorosis (Table 2).

The fluoride concentrations in the ready-to-eat meals were very low, and their contributions towards the risk of dental fluorosis were less than 2% (Table 3).

Table 4 shows that the fluoride concentrations in the formulated baby foods and milk-based foods would contribute 6 to 18% towards the threshold dose for the risk of dental fluorosis. On the other hand, in the soybean-based formulas, higher concentrations of fluoride were found, and some products could contribute up to 45% of the threshold dose of daily fluoride intake in terms of the clinically acceptable dental fluorosis.

The fluoride concentrations in the powdered milk porridges and other foods (Table 5) were low, the porridge A presented the higher fluoride concentration (1.63-0.04 mg F/kg). Their contribution towards undesirable dental fluorosis doses would be lower than 10%.

DISCUSSION

The solid and liquid diet is a significant source for the daily intake of fluoride in both developed[19] and developing countries.[17] It must therefore be considered to be risk factor for dental fluorosis.[14]

Fluoride has widespread distribution in nature, and this makes it difficult to identify its origin in the diet. Its presence in the diet depends on regional characteristics, such as the typical dish of each population. There are

Table 5. Fluoride concentrations in the porridges, foods formulated with powdered milk and other foods, estimated daily dose per child and percentage contributions towards the threshold dose for dental fluorosis. Campinas, SP, 2001.

Food (N)*	Mean concentration (µg F/g)	Estimated dose (mg F/Kg/day)	%**
Porridges***			
A (N=6)	0.47 ± 0.089	0.0035	5
B (N=4)	1.19 ± 0.266	0.0054	8
C (N=2)	1.06 ± 0.071	0.0048	7
D (N=6)	0.0 ± 0.010	0	0
E (N=5)	0.21 ± 0.088	0.0016	2
F (N=6)	0.27 ± 0.136	0.0020	3
G (N=3)	0.12 ± 0.041	0.0013	2
H (N=5)	1.63 ± 0.509	0.0069	10
I (N=5)	1.26 ± 0.429	0.0053	8
J (N=5)	1.09 ± 0.273	0.0035	5
K (N=6)	1.12 ± 0.216	0.0036	5
Powdered Milk			
A (N=5)	0.18 ± 0.042	0.0012	2
B (N=6)	0.07 ± 0.018	0.0005	<1
C (N=6)	0.15 ± 0.032	0.0010	1.4
D (N=3)	0.37 ± 0.071	0.0025	4
Other Foods			
A (N=6)	0.22 ± 0.014	0.0015	2
B (N=6)	0.04 ± 0.028	0.0003	<1
C (N=5)	0.54 ± 0.073	0.0037	5

* Reconstituted with water or milk, with a concentration <0.1 ppm F, in accordance with the manufacturers' recommendations and considering the number of glasses or feeding bottles recommended by each manufacturer.
**Contribution to threshold dose(0.07mgF/kg/day) for dental fluorosis.
*** Two meals daily, prepared in accordance with the manufacturers' instructions.

some foods that are recognized as being rich in fluoride, such as black tea-based beverages.[8]

The fluoride concentrations found in raw grains of rice and beans were low, and those in beans confirmed what had been observed in Brazilian seeds sold on the Japanese market.[16] Investigation of the reasons for the variability between brands was not the purpose of this study, but such variability may be related to fertilization, cultivation medium, plant type or irrigation with water containing natural fluoride.

Although the analysis of fluoride concentration in the grains is important, it is even more important to correlate the effect of the cooking water towards increasing the fluoride concentrations in foods to be consumed, since some foods may uptake fluoride more than others. In addition, some foods are consumed in a more liquid form than others. Therefore, the amount of fluoride intake will depend on the final concentration in the product and on the conditions under which the food is cooked.

The fluoride concentrations determined from raw rice and bean samples (Table 1) showed that, when these grains are cooked in non-fluoridated water (Table 2), they do not cause concern regarding the suggested limit for an acceptable level of dental fluorosis.[2] Thus, the mean fluoride concentrations in rice and beans prepared with non-fluoridated water were 0.044 ± 0.047 µg F/g and 0.149 ± 0.131 µg F/g respectively, so that a rice and bean-based meal would subject children to a dose of only 0.0020 mg F/kg of weight per day (Table 2). However, when these foods were prepared with fluoridated water, the dose increased tenfold (to 0.020 mg F/kg of weight per day). On the other hand, the quantity of fluoride consumed by children through rice and beans cooked in optimally fluoridated water is completely safe in terms of fluorosis, since it corresponds to 29% of the upper limit of the acceptable dose in relation to the risk of dental fluorosis.[2]

However, some of the processed baby foods (Table 4) are of concern in terms of the risk of dental fluorosis.[14] The data from the present study have confirmed that soybean-based foods[22] could contribute up to 50% of

the maximum limit of the dose in terms of clinically acceptable fluorosis. This concern about baby formulas dates back to 1979. There have been attempts in the United States to reduce the fluoride concentration in these foods, which however have had little influence on reducing the risk of developing dental fluorosis,[18] particularly in regions with fluoridated water.

Low fluoride concentrations were found in vegetable and fruit-based ready-to-eat foods (Table 3), thus concording with findings from products on the American market.[20] In beef or chicken-based products (data not shown), lower concentrations than in products on the American market were found.[10] High fluoride concentration are often found in foods because of the inclusion of bone fragments in the product, which are relatively rich in fluoride. Neither the fruit-based products nor the vegetable-based products containing chicken or meat were of concern with regard to being a source of fluoride and a risk of dental fluorosis (Table 3).

In the formulated foods (Table 4), the fluoride concentrations ranged from 0.47 to 3.84 µg F/g, and this variability could be explained mainly by the water that was used in the processing.[22] The mean concentration found for milk-based formulas was 0.87 ± 0.48 µg F/g, and this result was higher than what was found by other authors.[10,22] For the soybean-based foods (Table 3), the mean fluoride concentration found in the products on the Brazilian market (2.368 ± 1.206 µg F/g) was lower than in products in the United States.[22]

The fluoride concentrations found in the cereals group (Table 5) were in agreement with what has been observed on the Brazilian market.[4] The fluoride concentrations in products for preparing porridge (Table 5), which are basically cereals, were between 0.0 and 2.11 µg F/g. This mean was lower than what was found in Canadian products,[6] which may be explained by the fluoride concentration in the water used for processing these products.[9,20]

The low fluoride levels found in the samples from various brands of powdered milk, which were mostly less than 0.05 µg F/g (Table 5), are plausible, since milk is a food that naturally contains extremely low fluoride concentrations, without any risk whatsoever of dental fluorosis. The same is valid for the groups of "other foods" analyzed (Table 5).

It is concluded that the majority of the foods studied, including typically Brazilian foods, do not present any cause for concern with regard to the chronic toxicology of fluoride, taking into consideration the concentration in the foods, the amount consumed, and the dose that is considered to be the safe threshold for clinically acceptable dental fluorosis.

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