Availability and stability of soluble fluoride content in commercial fluoride dentifrices available in Uruguay

Disponibilidade e estabilidade do conteúdo de flúor solúvel em dentífricos fluoretados comercialmente disponíveis no Uruguai

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ABSTRACT

Objective: The aim of this study was to evaluate the availability and stability of soluble fluoride fraction in commercial fluoride toothpastes available in Uruguay. Methods: Fourteen fluoride toothpastes from four different manufacturers were analyzed. Randomized and blinded analyses were performed in duplicate for each dentifrice at the time of purchase (fresh samples) and after 12 months of storage at room temperature (aged samples). Total fluoride and total soluble fluoride concentrations were measured using a fluoride specific electrode. Results: Total fluoride concentrations in all of the products were lower than the F levels specified by the manufacturers. Total soluble fluoride fractions were lower than the total fluoride concentrations in fresh samples of five toothpastes and in aged samples of ten toothpastes (p < 0.05). Three toothpastes had insufficient and unstable total soluble fluoride fractions (< 60%) and five toothpastes had only unstable total soluble fluoride fractions (<1000 ppm) after 12 months. Conclusion: Based on the results of this study, it can be concluded that from fourteen Uruguayan commercial fluoride toothpastes analyzed in this study, three toothpastes have insufficient and unstable chemically active F fractions and five other toothpastes have the lack of stability which may compromise their efficacies. Review the guidelines on fluoride dentifrices in Uruguay is necessary, in order to ensure optimum benefit for population.

Indexing terms: Dental caries. Dentifrices. Fluorine. Toothpastes. Uruguay.

RESUMO

Objetivo: O objetivo deste estudo foi avaliar a disponibilidade e a estabilidade da fração solúvel de flúor nos dentífricos fluoretados comercialmente disponíveis no Uruguai. Métodos: Quatorze dentífricos fluoretados de quatro diferentes fabricantes foram analisados. Análises randomizadas e cegas foram realizadas em duplicata para cada dentífrico no momento da aquisição (amostras frescas) e depois de 12 meses de armazenamento em temperatura ambiente (amostras envelhecidas). As concentrações de flúor total e flúor solúvel total foram determinadas usando um elektrodo específico para flúor. Resultados: As concentrações de flúor total de todos os...
produtos testados foram menores do que os níveis de flúor especificados pelos fabricantes. Os valores das frações de flúor solúvel total foram menores do que as concentrações de flúor total nas amostras frescas de cinco dentífricos e nas amostras envelhecidas de 10 dentífricos (p < 0.05). Três dentífricos apresentaram as frações de flúor solúvel total insuficientes e instáveis (< 60%) e cinco dentífricos tiveram apenas frações instáveis de flúor solúvel total (<1,000 ppm) após 12 meses. **Conclusões:** Baseado nos resultados deste estudo pode-se concluir que dos quatorze dentífricos fluorados comercialmente disponíveis no Uruguai analisados neste estudo, três produtos apresentaram insuficientes e instáveis frações quimicamente ativas de flúor e cinco outros dentífricos tiveram falta de estabilidade o que pode comprometer suas eficácia. É necessário revisar as orientações sobre dentífricos fluorados no Uruguai a fim de garantir um benefício ótimo para a população.

**Termos de indexação:** Cárie dentária. Dentífricos. Flúor. Cremes dentais. Uruguai.

**INTRODUCTION**

Despite a significant improvement in oral health within industrialized and some developing countries during the last decades, dental caries still remains as a major oral health problem, especially among socioeconomically disadvantaged groups [1-3]. Efficient and widespread use of fluoride toothpastes continues to be a priority to improve worldwide oral health according World Health Organization [3].

Systematic reviews have shown that the use of standard fluoride toothpastes containing 1000 - 1500 ppmF reduces the incidence of dental caries in permanent teeth by approximately 24 - 29% [4-6]. However, the effect of fluoride toothpaste becomes significant only when fluoride is present at a concentration of 1000 ppm or more [6-8].

Fluoride toothpastes must contain an adequate and stable concentration of soluble fluoride, as ion F- or sodium monofluorophosphate ion in its formulation to control dental caries effectively [9,10]. However, its bioavailability may decrease over time due to the undesirable reactions of the fluoride compound if the abrasive used is calcium based [11].

Fluoride toothpastes must be present in a soluble form to guarantee its anti-caries efficacy. Recently, American Dental Association (ADA) published acceptance program requirements for fluoride containing dentifrices to address requirements necessary to determine their anti-caries efficacy. They recommend that at least 90% of the labeled amount of fluoride must be available in both fresh and aged samples [9]. However, Mercosur (Southern America common market that includes Argentina, Brazil, Paraguay and Uruguay) just states that toothpastes must not contain more than 1500 ppm of total F concentration, but do not indicate the soluble F fraction [12].

Uruguay provides a salt fluoridation program for population since 1991. However, difficulties in monitoring the salt distribution and due to recent campaigns to avoid excess of salt consumption, fluoride dentifrice have become an important public health measure in preventing caries.

Therefore, the aim of this study was to evaluate the availability and stability of soluble fluoride in commercial fluoride toothpastes available in Uruguay.

**METHODS**

Fourteen fluoride toothpastes, commercially available in Uruguay, were tested in the present study. Three tubes of each toothpaste were purchased (n = 42) in supermarkets and drugstores located in different areas of Montevideo, Uruguay.

Toothpastes were coded with different letters to blind the analysis. Table 1 shows the codes and characteristics of the toothpastes according to the package information.

The analysis was carried out immediately upon acquisition of the dentifrices (fresh samples) and after 12 months of storage at room temperature (aged samples).

The concentrations of three forms of fluoride available in the toothpastes were determined: fluoride ion (FI), total soluble fluoride (TSF) which includes FI and fluoride as monofluorophosphate (MFP) and total fluoride (TF). Total fluoride is the sum of TSF plus insoluble F (InsF) that is the F bound to the abrasive. From these analyses, the concentrations of F as MFP and the percentage of IF were calculated. Analyses were carried out in duplicates according the modified procedure described by Cury et al [13]. An amount of 90 to 110 mg of toothpaste was weighted (±0.01 mg), homogenized in 10 mL of deionized water and duplicates of 0.25 mL of the suspension were transferred to test tubes for TF analysis. The remaining of the suspension was centrifuged (3,000 x g, 10 min) to remove InsF bound to the abrasive. Duplicates of 0.25 mL of the supernatant were transferred to assay tubes to determine TSF and FI concentrations. For the TF and TSF
tubes, 0.25 mL of 2.0 M HCl was added, and after 1 h at 45°C, the samples were neutralized with 0.5 mL 1.0 M NaOH 1.0 M and buffered with 1.0 mL of Total Ionic Strength Adjustor Buffer II (TISAB II, 1.0 M acetate buffer, pH 5.0, containing 1.0 M NaCl and 0.4% cyclo-hexylenedinitrilo-tetraacetic acid). To the FI tubes, 0.50 mL of 1.0 M NaOH was added and buffered with 1.0 mL of TISAB II and 0.25 mL of 2.0 M HCl. The analyses were carried out using a fluoride ion specific electrode (Orion model 96-09, Orion Research, Cambridge, MA, USA) connected to an ion analyzer (Procyon Instrumentos Científicos, São Paulo, SP, Brazil). Previously calibration was done in triplicate with fluoride standard solutions containing 0.8, 1.6, 3.2 and 6.4 μg F/ml, prepared with the same reagents as the samples. The analyses were validated using internal standards and a coefficient variation lower than 3% was considered as acceptable. The readings were expressed in millivolt (mV) and transformed to ppm F by linear regression curve.

Descriptive analysis was performed and means and standard deviation of the concentrations of TF, TSF and %InsF of fresh and aged samples, of the three samples for each toothpaste, were calculated using Excel software (Microsoft Corporation).

RESULTS

Table 1 shows the characteristics (manufacturer; type and concentration of F; expiration date and abrasive system) of the 14 toothpastes analyzed. According to the fluoride concentration specified on the packages, 13 toothpastes contained the standard concentration of 1,000 - 1,500 ppmF (A, B, C, D, E, F, G, H, J, K, M, and N) and one product had 2,500 ppmF (I). Nine toothpastes (64.3%) contained NaF and five (35.7%) contained MFP as fluoride compound. Toothpastes formulated with MFP contained calcium carbonate (n = 3) or calcium carbonate + silica (n = 2) as the abrasive; while those with NaF contained silica as the abrasive (n = 5); the others have no information available on package (n = 4).

Table 1. Characteristics of toothpastes analyzed in the study.

| Commercial brand | Manufacturer | Code | Fluoride agent | Fluoride expected (ppm) | Abrasive system | Lot number | Expiration date |
|------------------|--------------|------|----------------|--------------------------|----------------|------------|-----------------|
| Aquafresh Triple Protection | Glaxo Smith Kline | A | MFP | 1,100 | CaCo3 + SiO2 | 1A03C | 01/2013 |
|                    | USA          |      | NaF  | 1,500 | SiO2 | 97521 | 07/2014 |
| PicoJenner Plus   | Laboratorios Abarly | B | NaF | 1,500 | SiO2 | 988861 | 09/2014 |
| Colgate Palmolive | Argentina    | C | MFP | 1,450 | CaCo3 | 1181AR11801 | 06/2014 |
| Periodont Gingival | Laboratorios Abarly | D | NaF | 1,000 | Not declared | 97081 | 06/2014 |
| Colgate Máxima Protección | Unilever Brasil Higiene Pessoal e Limpeza Ltda, Brazil | F | MFP | 1,450 | CaCo3 + SiO2 | 11358BR12BC | 05/2014 |
| Close up Triple Menta | Laboratorios Abarly | G | NaF | 1,500 | Not declared | 89501 | 08/2013 |
| Periodont Zeta | Laboratorios Abarly | H | NaF | 1,500 | Not declared | 91441 | 10/2013 |
| Laboratorios Abarly | Uruguay |      | | | | | |
| Periodont Pro | Laboratorios Abarly |  | | | | | |
|                | Uruguay |      | | | | | |

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Table 1. Characteristics of toothpastes analyzed in the study.

| Commercial brand                  | Manufacturer            | Country | Code | Fluoride agent | Fluoride expected (ppm) | Abrasive system | Lot number | Expiration date |
|-----------------------------------|-------------------------|---------|------|----------------|-------------------------|----------------|------------|----------------|-----------------|
| Biodent 2500                      | Laboratorios Abarly     | Uruguay | I    | NaF            | 2,500                   | Not declared   | 88262      | 06/2013         |
| Colgate Triple Acción             | Colgate Palmolive       | Argentina | J    | NaF            | 1,450                   | SiO₂          | 1170AR11801 | 06/2014         |
| Colgate Herbal                    | Colgate Palmolive       | Argentina | K    | MFP            | 1,450                   | CaCO₃         | 1124AR11802 | 05/2014         |
| Colgate 12 Clean Mint             | Colgate Palmolive       | Brazil   | L    | NaF            | 1,450                   | SiO₂          | 0268BR1228  | 09/2013         |
| Colgate 12 Professional Whitening | Colgate Palmolive       | Brazil   | M    | NaF            | 1,450                   | SiO₂          | 1144BR122D  | 05/2014         |
| Colgate 12 Professional Clean     | Colgate Palmolive       | Brazil   | N    | NaF            | 1,450                   | SiO₂          | 0268BR122D  | 05/2014         |

Note: MFP: sodium monofluorophosphate; SiO₂: silica; CaCO₃: calcium carbonate.

TF concentration in fluoride toothpastes varied from 914.54 to 2122.34 ppmF in fresh samples, and from 860.45 to 2070.53 ppmF in aged samples (Table 2). Fresh and aged samples of all tested toothpastes showed a TF concentration below the fluoride specified and their labels and only 46.1% of the toothpastes (n = 6) had a TF concentration ≥ 89% of the value specified (A, C, D, E, F, and K) in fresh samples (Table 2).

TF concentration ranged from 330.63 to 1327.20 ppmF in all fresh samples, and from 288.81 to 1321.10 ppmF in all aged samples (Table 2).

Table 2. Concentrations (ppmF) of total fluoride and total soluble fluoride in fresh and aged samples of toothpastes tested. Each value corresponds to the mean of three different tubes made in triplicate ± standard deviation.

| Code | Fluoride Expected | Fresh samples | Aged samples |
|------|-------------------|---------------|--------------|
|      | Total fluoride    | Total soluble fluoride | Total fluoride | Total soluble fluoride |
| A    | 1,100             | 1067.99 ± 23.42 | 1091.38 ± 57.34 | 860.45 ± 43.37 | 776.87 ± 55.61 |
| B    | 1,500             | 1120.20 ± 175.72| 520.90 ± 136.56| 1064.02 ± 171.79| 926.69 ± 39.86 |
| C    | 1,450             | 1133.77 ± 12.47 | 1240.08 ± 8.27 | 1238.54 ± 24.40 | 1219.14 ± 50.07 |
| D    | 1,000             | 914.54 ± 30.84  | 962.60 ± 35.01 | 905.49 ± 19.84  | 881.22 ± 40.42 |
| E    | 1,450             | 1359.51 ± 69.14 | 1319.03 ± 7.64 | 1304.73 ± 25.04 | 1013.36 ± 43.25 |
| F    | 1,450             | 1293.55 ± 39.66 | 1327.20 ± 27.90| 1258.69 ± 83.10 | 1219.14 ± 50.07 |
| G    | 1,500             | 1284.00 ± 86.97 | 1313.40 ± 106.78| 1324.46 ± 32.72 | 1321.10 ± 35.81 |
| H    | 1,500             | 981.18 ± 78.89  | 330.63 ± 7.89 | 1340.46 ± 182.95| 288.81 ± 6.86 |
| I    | 2,500             | 2122.34 ± 84.68 | 696.29 ± 577.89| 2070.53 ± 186.51| 634.45 ± 417.58 |
| J    | 1,450             | 1235.37 ± 46.53 | 1233.62 ± 20.47| 1207.34 ± 2.83  | 1256.00 ± 28.22 |
| K    | 1,450             | 1304.04 ± 18.08 | 1286.21 ± 48.24| 1172.45 ± 13.33 | 990.57 ± 56.57 |
| L    | 1,450             | 1234.91 ± 36.72 | 1286.21 ± 27.49| 1094.08 ± 55.78 | 1196.57 ± 17.79 |
| M    | 1,450             | 1279.48 ± 112.87| 1260.63 ± 28.19| 1155.33 ± 69.66 | 1164.36 ± 40.25 |
| N    | 1,450             | 1240.28 ± 20.63 | 1179.48 ± 29.66| 1063.79 ± 45.30 | 1133.81 ± 20.80 |
For fresh samples, TSF concentrations were lower than the TF in 57.1% of the toothpastes (B, C, E, H, I, K, M, and N) indicating that fluoride was partially insoluble (Figure 1). Ten toothpastes (A, B, C, D, E, F, G, H, I, and K) showed TSF concentrations lower than TF concentrations in aged samples (table 2).

TSF concentrations were lower in the aged samples than in the fresh samples in almost all the toothpastes. This can be explained due to the increase in % insF concentration. The % insF concentration in fresh samples ranged from 0 to 66.4% and from 0 to 78.2% in aged samples (mean ± SD, 21.3 ± 29.0). The % insF concentration increased in 78.6% of aged samples, and was > 60% of TF in toothpastes B, H, and I (figure 1).

**DISCUSSION**

The widespread use of fluoride is considered the main reason for the worldwide decline in dental caries prevalence, and fluoride toothpastes are the most widely used forms [4,14,15].

Lack of consistency between labeled and actual fluoride content in toothpastes can compromise its effectiveness. Total fluoride concentration may also contain insoluble and other ineffective fractions of fluoride [16,17]. Therefore, fluoride in dentifrices must be present in a soluble form to guarantee its anti-caries efficacy.

In the present study, all tested toothpastes had TF concentration lower than that informed in their packages, in fresh and aged samples (figure 1). Except for toothpaste I, all the products were in accordance with the Mercosur legislation, which states that the maximum fluoride concentration in a dentifrice must be 1500 ppmF. Although toothpaste I had a TF value > 2,000 ppm, being considered therapeutic, it is marketed freely in Uruguay without a need for a professional prescription. In most toothpastes analyzed in the present study, the TF concentration of fresh and aged samples did not vary. It is expected that the TF concentration should remain stable after one year, although there may be variations in the other fluoride forms such as TSF, due to its instability [18,19].

TSF concentration in the most dentifrices tested was lower in aged samples compared to fresh samples. This variation can be explained by the formulation as part of the chemically active F can bind with Ca ions in the abrasive [11]. Although MFP is more compatible with a CaCO₃ or DCPD (dicalcium phosphate dihydrate) abrasive [10,11], later studies showed that part of F can bind with Ca in the abrasive, reducing the amount of soluble F [18-21]. These decreases occur during product storage due to the release of F ions from the hydrolysis of MFP [18,19]. Even toothpaste I has a concentration of TF (> 2,000 ppmF) higher than all other dentifrices, only 32.8% of its TF concentration corresponded to TSF and this value was lower than that found to other tested fluoride toothpastes (table 2).

Three dentifrices (B, H and I) showed % insF higher than 60% of TF concentration in fresh and aged samples.

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**Figure 1.** Percentage of insoluble fluoride in fresh and aged samples of commercial fluoride toothpastes available in Uruguay.
These toothpastes are produced by the same Uruguayan manufacturer and are very popular. They have NaF as fluoride compound and while toothpaste B used SiO$_2$ as abrasive, toothpastes H and I had no information in their packages.

Only one (D) of the fourteen dentifrices analyzed in the present study presented amount of available fluoride in fresh and aged samples, as recommended by the ADA [9]. Other four tested dentifrices (A, E, F and J) presented adequate fluoride concentration only in fresh samples.

Surprisingly, the dentifrices B, H and I showed only 34%, 22.04% and 27.85%, respectively, of total fluoride in form of TSF in fresh samples. These percentages decreased in aged samples showing 21.59%, 19.25% and 25.37%, respectively. Toothpastes B, H and I had TSF values very low that recommended one which could compromise their efficacy against caries (table 2).

Stability was evaluated in this study over a period of one year, however it should be emphasized that the estimated time that dentifrices stay on the shelves of the stores is less than this evaluated period. This shelf time of the dentifrices would be sufficient for no loss of soluble fluoride and indicates that dentifrices can be effective in preventing dental caries.

Community salt fluoridation is available in Uruguay and population has access to domestic fluoridated salt (250 mgF/kg). Comparable to water fluoridation, this alternative demonstrated a similar beneficial reduction in the prevalence of caries, and provided a choice to consumers. Individual choice is an interesting option, but the use of fluoridated salt among individuals is not similar [22]. Because of the difficulties in monitoring the program of fluoridated salt distribution and campaigns to avoid overconsumption of salt, fluoride dentifrice has become an important public health measure in preventing caries.

Almost all commercial toothpastes available in Uruguay are fluoridated, so it is necessary to control the quality of commercial toothpastes and establish the minimum concentration of soluble fluoride to have anti-caries efficacy. Unfortunately, Mercosur just states that toothpastes must not contain more than 1500 ppm F and it does not establish how much of total fluoride content should be maintained soluble in toothpaste formulation. Review the guidelines on fluoride dentifrices in Uruguay and Mercosur is necessary, in order to ensure optimum benefit.

CONCLUSION

Based on the results, it can be concluded that from fourteen Uruguayan commercial fluoride toothpastes analyzed in this study, three toothpastes have insufficient and unstable chemically active F fractions and five other toothpastes have the lack of stability which may compromise their efficacies.

Collaborators

AF FABRUCCINI performed the experiment, analyzed the data and wrote the paper. LL ALVAREZ performed the experiment. M MALTZ analyzed the data. LN HASHIZUME conceived, designed the experiment, analyzed the data and wrote the paper.

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