Comparison of Fluoride Uptake into Dental Enamel from two Types of Sodium Fluoride Varnishes (In vitro)

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ABSTRACT

Background and Aim: Different fluoride products such as gels, mouthwashes, toothpastes and varnishes have been used to increase tooth resistance against dental caries. Among these, fluoride varnishes gained more importance due to their advantages. The present in-vitro study aimed to measure the amount of fluoride uptake into enamel from Sultan and AriaDent varnishes.

Materials and Methods: In this experimental in-vitro trial, 40 fresh premolars were selected and sectioned into two mesial and distal halves. The samples were divided into 2 groups (n=20) and were exposed to either AriaDent or Sultan varnish products for one hour. Samples were stored in artificial saliva at 37°C for 24 hours and then were etched with 1 M acid perchloric for 30 seconds. After etching, the window surfaces were washed by 0.2 M Potassium hydroxide (KOH). Sampling was done using acid etch enamel biopsy technique and fluoride and calcium contents were measured by potentiometer and spectrophotometer devices. Data were analyzed by T-Test.

Results: Enamel fluoride uptake in the specimens exposed to AriaDent and Sultan varnishes was 3.27±1.89 ppm and 3.41±1.34 ppm, respectively. The calcium content of the enamel specimens exposed to varnish was 82.02±52.84 ppm for samples exposed to AriaDent and 71.11±52.68 ppm for those treated with Sultan. No significant difference was found between two varnishes regarding the fluoride uptake into enamel (p=0.36) or calcium content. (p=0.34)

Conclusion: It seems that local fluoride therapy with both varnishes (AriaDent and Sultan as the standard varnish) is equally effective.
Introduction
Addition of fluoride to drinking water is believed to be the most efficient and cost-effective method of fluoride intake. Due to variation in fluoride content of water in different areas and insufficiency of this ion in many regions, also lack of accurate and repeated measurement of fluoride content of water, it seems that other local means of fluoride exposure must be provided such as toothpastes, mouthwashes and gels in dental office or at home besides fluoride in drinking water.  

Fluoride varnishes show the highest anti-caries effect in children and it seems that using them twice or more per year is effective and beneficial in caries prevention. When fluoride varnish is used, tooth is covered with an adhesive biofilm for a short time; therefore, uptake of fluoride ions into tooth structure is increased. Release of fluoride ion after applying varnish is gradual over two days which provides safety for this caries preventive method. Local application of fluoride varnishes can increase amount of fluoride uptake into dentin and enamel. Increase in precipitation of fluoride in surface of dental hard tissues may differ based on concentration and composition of varnish. Fluoride depositions constantly dissolve in saliva which leads to significant reduction in fluoride amount one week after local usage of fluoride. After using fluoride-containing products, fluoride content of saliva is increased in specific time intervals based on concentration and type of product. Increase in fluoride amount for 0.04 ppm in adjacent dental hard tissues can decrease risk of caries. After applying varnish on tooth surfaces, fluoride content of saliva remains elevated for 32 hours. The present in-vitro study aimed to measure the amount of fluoride uptake into dental enamel from AriaDent and Sultan varnishes with cooling water. Each half was inspected with stereomicroscope (ZSM-1001, 10×40) to make sure there is no defect on enamel surface. Windows method and Acid etch enamel biopsy method were used in this experiment. First, circles with diameter of 6 mm were cut out of a piece of adhesive paper using a punch machine and then, using a precise engineering ruler, each circle was cut to two semi-circles. After being washed with deionized distilled water, prepared half teeth were stored in at room temperature until they were totally dried. Then, each semi-circle was pasted to the one-third cervical area of mesial and distal halves in the center of surface in buccolingual direction, in a way that in both halves, stickers were of equal distance from occlusal surfaces. Each of these adhesive pieces were burnished carefully onto teeth surfaces until they became fully adapted with tooth surface. These stickers outlined the area of tooth under biopsy. Then, the remaining surfaces of half teeth were carefully coated with acid resistant nail polish. This was done twice for each half tooth and each tooth was dried in at room temperature. When specimens were dried, adhesive semi-circles were removed so on all teeth surfaces windows of enamel with known area were obtained. Then each half tooth was washed with deionized water and dried at room temperature.

20 pairs of half teeth were placed for one hour in constant magnetic stirring machine (DELTA-HM-101) with speed of 70-80 rpm (the first 20 samples were placed in AriaDent varnish and the second 20 samples in Sultan varnish). Then, experimental halves were washed and dried and kept in artificial saliva at 37ºC for 24 hours. In this stage, all samples went under Acid etch enamel biopsy, in order to be measured for their fluoride and calcium content.

40 plastic containers itemized for sampling were washed with deionized water and dried and were coded with someone blind to the process. In each container, 1 M of perchloric acid (HClO4) was poured for 30 seconds. Each experimental half was gripped with a plier and was immersed in designated containers and biopsy from each window was performed for 30 seconds. Tip of the plier was coated with acid resistant nail polish in order to prevent ion interaction and loss of balance in solution. Samples were stirred in acidic
solutions with slight moves while being biopsied to prevent the returning of released fluoride ions into enamel structure. One biopsy from each sample was obtained. Enamel surface of window area was washed with 2 cc of 2% potassium hydroxide (KOH) solution 0/2 M. all these stages were done inside plastic containers and in each container 3 cc of biopsy solution was obtained. Then, the window on tooth surface was dried immediately with a small cotton ball and this cotton ball was placed inside the plastic container. The Container’s lid was tightened after biopsy and solution was taken to laboratory of colorup company for measurement of fluoride and calcium content. Potentiometer was used for measuring the fluoride content in of solutions. For calcium measurement, Atomic Absorption Spectrophotometry method was applied with use of Flame Atomic Absorption Spectrophotometer machine (Perkin-Elmer/-603) with sensitivity of 0.01 ppm. In order to eliminate any probable error, standard solutions with precisely measured concentrations of calcium ion (standard solutions with 1-15 ppm calcium concentration) were used and a light absorption curve was drawn based on concentration of the solution and concentration of the sample was determined by placing the amount of absorption in the curve. \(^{13,14}\) Presence of phosphate compounds in biopsy solutions obtained from dental enamel is one factor causing error in measurement of calcium. These compounds form stable combinations with calcium and prevent proper calcium evaporation process. To eliminate the effect of this interfering factor, 0.1 M EDTA (BDH Co.) was used for releasing calcium. The Amount of calcium in different samples was measured in ppm. Each measurement was repeated 3 times and the mean value was reported.

The Mean of indices in two groups of Sultan and AriaDent varnishes were statistically analyzed by T-Test.

**Results**

Based on the findings of the present study, the amount of fluoride ion in AriaDent varnish samples was 3.27±1.89 ppm and in samples of Sultan varnish was 3.41±1.34 ppm. Although the amount of fluoride ion in samples of Sultan varnish was estimated to be little higher than AriaDent varnish samples, this difference was not statistically significant. (p=0.36)(Diagram 1)

The Amount of calcium in AriaDent samples was 82.02±52.84 ppm and in Sultan samples was 71.11±52.68 ppm. Although these amounts are reported higher for AriaDent varnish, the difference between two varnishes regarding calcium amount is not statistically significant. (p=0.34)

**Discussion**

Based on the results of this study, the amount of fluoride ion in samples of AriaDent and Sultan varnishes is similar in range. Most other related studies were performed on other fluoride-containing products including gels, toothpastes and mouthwashes and few studies exist on fluoride varnishes. Pai et al. showed that some local fluoride-containing products such as APF gel can cause more fluoride uptake in comparison with NaF product. \(^{15}\) In the mentioned study, experimental methods similar to our study were adopted. Also, Schemerhorn et al. Reported that fluoride varnish containing tri-calcium phosphate (TCP) causes less fluoride deposition in sound and demineralized enamel comparing to amorphous calcium phosphate (ACP)-containing varnish. \(^{16}\) Maas et al. reported that Bifluoride and Duo-fluoride varnishes showed the highest amount of fluoride release in remineralization and demineralization solutions. \(^{17}\) Considering that neither
of the mentioned varnishes were discussed in the present study, results of these two studies cannot be compared.

In another study by Mahmudian et al. on Iranian fluoride-containing products, it was showed that two 0.2% sodium fluoride mouthwashes and one 1.23% APF gel manufactured in Iran caused fluoride uptake similar to foreign brands and in some cases, this uptake level was even higher for Iranian products. The Results of the mentioned study are in line with the present study regarding the similarity between fluoride uptake from Iranian products and foreign standard ones which can be due to conforming of Iranian manufacturing units of established standards in this regard.

On the other hand, another study showed that fluoride uptake into enamel after usage of foreign stannous fluoride gel was higher compared to that of 0.05% sodium fluoride made in Iran. Also Biria et al. compared the amount of fluoride uptake into enamel of permanent teeth after usage of Sina and Sultan fluoride gels and reported inadequate fluoride uptake of Sina gel in to dental enamel. In another study, Navabi et al. reported that Duraflor varnish causes higher fluoride uptake compared to APF fluoride gel. In Biria et al. study fluoride uptake in two depths of biopsy was evaluated but in Navabi et al. study and our study, fluoride uptake was measured only in one depth of biopsy. The Similarity of findings of recent studies regarding significant fluoride uptake after using Sultan varnish shows the established characteristics of this ADA approved product. On the other hand, Eronat et al. found no significant difference in fluoride uptake of varnishes and gels after considering neutral NaF and APF local gels and Duraphat and F-Protector fluoride varnishes. In a study by Patil and Anegundi it was found that dental enamel treated with Amine fluoride had the highest amount of fluoride uptake.

The Probable reason for dissimilarity in results of different studies can be due to evaluation of various types of varnishes and gels produced by different manufacturers.

Various amounts of fluoride uptake from several products have been reported. In the present study, in spite of no statistical significance, Sultan varnish showed higher fluoride uptake in comparison to AriaDent varnish.

The Amount of fluoride ion in different fluoride products including toothpastes, gels or varnishes can fluctuate due to various factors. The First factor in this regard is high or low amount of fluoride ion added to the formulation of a product which needs more evaluation and consideration by their manufacturers. Since the amount of fluoride in of local fluoride products directly correlates with the well-being of individuals in the society, the related authorities should be more attentive to their formulation to produce a reliable and beneficial product so that the probable effects of fluoride deficiency can be avoided. The Second factor in this regard is lack of uniform supply of fluoride in different produced series of products or in each related produced series. Also, the presence and type of abrasives in a product can be effective in deactivation of fluoride ion and reduction of its anti-caries properties.

In experiments that measure amount of fluoride ion, only free fluoride ion is measured and if it is in form of a salt along with abrasives (for example calcium fluoride salt next to calcium carbonate abrasive in toothpaste) measurement is not possible; hence the presence or absence of inappropriate abrasives and their amount in formulation can be another effective factor influencing the amount of fluoride ion in various products. Since metallic ions like Ca and Al etc. can discontinue the activity of fluoride ion by surrounding or forming a compound with it, the presence of unwanted metallic ions in the composition of some products such as colors and flavor additives can inhibit the activity of fluoride ion and finally reduce the uptake of fluoride.

**Conclusion**

Since there is no statistical significant difference between AriaDent and Sultan varnishes regarding the amount of fluoride uptake into enamel, AriaDent varnish can be considered suitable for local fluoride therapy.

**Conflict of interests**

Authors report no conflict of interest related to this study.

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