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The effect of temperature change on fluoride uptake from a mouthrinse by enamel specimens

Serdar Baglar[1]
Adil Nalcaci[2]
Mustafa Tastekin[3]

ABSTRACT

Objective: The aim of this study was to examine the effect of temperature on fluoride uptake by enamel specimens from a 0.05% NaF-fluoridated mouthrinse (Oral-B Advantage; Oral-B Laboratories, Newbridge, UK).

Methods: Enamel specimens were prepared from extracted human maxillary central incisors. A fluoride-specific ion electrode was used to measure the uptake from a 2 ppm fluoride solution containing 50.0 mL of distilled water, total ion strength adjustment buffer, and fluoridated rinse at 3 different temperatures (room temperature, 25°C; human body temperature, 37°C; hyper-fever temperature, 43°C). One-way analysis of variance and least significant difference were used to assess intragroup and intergroup differences (P<.05).

Results: The study found that both the amount and the rate of fluoride uptake increased significantly with increase in temperature. This effect was particularly noticeable at 43°C.

Conclusions: The temperature of the NaF mouthrinse may easily and safely be increased beyond room temperature by placing a container of the NaF mouthrinse in a bowl of hot water, allowing greater fluoride penetration into the enamel from the mouthrinse when used at home as a routine prophylactic agent. (Eur J Dent 2012;6:361-369)

Key words: Fluoride uptake; temperature; enamel; mouthrinse

[1] Department of Restorative Dentistry, Faculty of Dentistry, Kirikkale University, Kirikkale, TURKIYE
[2] Department of Restorative Dentistry, Faculty of Dentistry, Ankara University, Ankara, TURKIYE
[3] Department of Chemistry, Faculty of Science, Ankara University, Ankara, TURKIYE

Corresponding author: Dr. Serdar Baglar
Department of Restorative Dentistry, Faculty of Dentistry, Kırıkkale University, Kurtulus Mh. Merkez, 71400 Kırıkkale, TURKIYE
Tel: +90 318 2244927
Fax: +90 318 2250685
Email: serdarbaglar78@gmail.com

INTRODUCTION

Fluoride penetration in the enamel occurs through the replacement of the relatively weak hydroxyl ions in the enamel mineral structure by the much more active fluoride ions, thereby improving the chemical stability of the enamel structure and making it more resistant to acids.[1-4] The presence of fluoride in the oral cavity, especially in the saliva, has also been shown to have beneficial effects.[5-8] For these reasons, a number of fluoride products with different contents and uptake levels

have been developed to provide supplemental fluoride in various forms. After dentifrices, fluoride mouthrinses represent one of the most useful fluoride products.[1,2,8-12] The fluoride mouthrinses commonly used today are neutral pH, 0.05% NaF, stable chemical compounds[13] that act topically to decrease enamel decalcification and increase remineralization.[14,15]

The most important function of fluoridated agents is to increase the enamel resistance to acid by decreasing solubility through the incorporation of fluoride into the enamel apatite structure. Fluorapatite formation is based on a chemical equilibrium that allows the penetration of fluoride into the enamel. As with all chemical reactions, this process is strongly influenced by concentration and temperature.[16] In a study examining the effects of heat on tin and fluoride uptake from topical SnF_2 solutions by enamel samples prepared from extracted bovine teeth, Putt et al[17] noted an increase in both tin and fluoride uptake with increase in temperature. Barrancos[18] also found that higher temperatures enhanced fluoride uptake by the enamel from topical acidulated phosphate fluoride (APF) solutions. In study conducted using powdered enamel over a wide temperature range, Stearn and Berndt[19] found APF solutions to be correlated with an increase in fluoride content and CaF_2 formation. Stookey and Stahlman20 suggested that thermal changes resulting from friction were responsible for the enhanced fluoride uptake provided by fluoride-impregnated prophylactic cups.

In order to better understand the effects of temperature on fluoride uptake, this study compared the rates and amounts of fluoride uptake by the enamel from a fluoridated mouthrinse at 3 different temperatures.

MATERIALS AND METHODS

The chemical substances and equipment used in this study are listed in Tables 1 and 2, respectively.

Preparation of the Specimens

A total of 30 extracted human maxillary central incisors were used in the study. All the teeth were visually examined to ensure that they were free from apparent caries, macroscopic cracks, abrasion, and excessive staining on the lingual and buccal surfaces. Prior to use, the teeth were cleaned by gentle brushing with a fluoride-free toothpaste and stored in distilled water containing 0.1% thymol at 4°C. The crowns were separated from the roots 2 mm above the cemento-enamel junction and sectioned mesiodistally; then, the labial surfaces were sectioned into 3 slabs by using a diamond saw (Isomet; Buehler, Lake Bluff, IL, USA). The inner surfaces of each section were abraded

Table 1. Chemical materials used in the experiment.

MATERIAL	MANUFACTURER
TISAB	Ank. Univ. Department of Chemistry, Faculty of Science, Analytical Chemistry Laboratory
Ultra Distilled Water	Ank. Univ. Department of Chemistry, Faculty of Science, Analytical Chemistry Laboratory
Mouthrinse (0.05% NaF)	Oral-B Advantage, Oral-B Laboratories, Newbridge, UK.
Acetic Acid (99.5%)	Merck (100056) Darmstadt, Germany
NaCl (99.5%)	Merck (106404) Darmstadt, Germany
NaOH (High purity)	Merck (106462) Darmstadt, Germany
Standard fluoride solution (100 ppm)	Orion Research (940907) Boston, M.A., U.S.A.

Table 2. Equipment used in the experiment.

EQUIPMENT	MANUFACTURER
Combined fluoride ion selective electrode	Orion Model 96-09, Orion Research, Boston, MA, U.S.A
Glass electrode	Orion Model 91 Series, Ag/AgCl pH electrode, Boston, M.A, U.S.A
Automatic pipette 100 µL – 1000 µL	Brand, ± 5 µL, measured with accuracy, Germany
Automatic pipette 0.5 µL – 5 µL	Brand,± 0.02 µL, measured with accuracy , Germany
Potentiometer	Consort C863 Multi - parameter analyser, Parklaan, Belgium
Magnetic Stirrer	Chiltron hotplate, magnetic stirrer HS31 Scientific Wendurer, England
Circulating water bath pump	Nüve BM 102 Industrial Supplies and Trading Company Ankara, Turkey
Analytical Balance	Gec Avery, Model, VA304, 0,1 mg measured with accuracy, UK

using wet 360- and 600-grit silicon carbide paper to remove any remaining dentin, and the slabs were examined under a stereomicroscope (Leica MZ12; Leica AG, CH-9435 Heerbrugg, Switzerland) to confirm that they consisted of enamel alone. All specimens were stored in distilled water until the experiment.

Specimens were randomly divided into 3 groups (n = 30) for treatment at different temperatures, namely, room temperature, 25°C; human body temperature, 37°C; and hyper-fever temperature, 43°C. Each group was further divided into 3 subgroups (n = 10) to demonstrate the reproducibility of the measurements in each temperature group, and the average mass of each subgroup was calibrated (0.7412 ± 0.0043 g) using an analytical balance to ensure standardization (Table 3). The slabs were stored in distilled water in polyethylene tubes that had been previously washed with distilled water, rinsed with bidistilled water, and dried in a sterilizer oven on blotting paper.

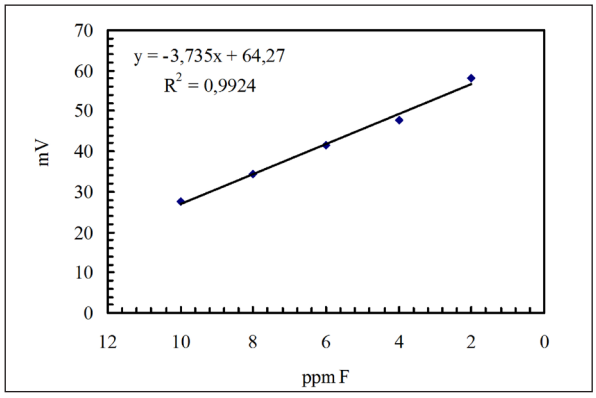

Figure 1. Calibration graph used to determine fluoride content of mouthrinse.

Fluoride Measurement

A fluoride-specific ion electrode (Orion model 96-09; Orion Research, Boston, MA, USA) was used to measure the fluoride concentrations. The total ion strength adjustment buffer (TISAB) was used as a stabilizer.[7,20]

Preparation of Calibration Solution from Standard Fluoride Solution

A standard 100 ppm fluoride solution (Orion Research 940907, Boston, MA., USA) was used to prepare the calibration solutions.[7,21] In order to establish calibration graphs, 50 mL of the calibration solutions were prepared using 25 mL of TISAB solution and 25 mL of 2, 4, 6, and 8 ppm fluoride solutions prepared from a standard fluoride solution of 100 ppm fluoride and distilled water. The actual fluoride content of the mouthrinse was determined using the calibration graph constructed from the calibration solutions (Figure 1).

Calibration of the Mouthrinse Fluoride Solution

Using the standard solution calibration graph (R^2 = 0.9924), the fluoride concentration of the mouthrinse was determined to be 235 ppm, which is similar to the measurement reported by the manufacturer (230 ppm). On the basis of this calibration graph, the rinse volumes were calculated for solutions of 0.5, 1.0, 1.5, 2.0, and 2.5 ppm fluoride. Calibration graphs for all the temperatures were constructed for determining the fluoride uptake by the enamel from the mouthrinse solution.

Table 4 shows the fluoride ppm and the corresponding mouthrinse volumes for 50 mL of the calibration solutions.

Table 3. Subgroups of enamel specimens and their masses.

25°C Enamel Specimens I. Subgroup	37°C Enamel Specimens I. Subgroup	43°C Enamel Specimens I. Subgroup
1a, 2a, 3a, 4a, 5a,	1b, 2b, 3b, 4b, 5b,	1c, 2c, 3c, 4c, 5c,
6a, 7a, 8a, 9a, 10a	6b, 7b, 8b, 9b, 10b	6c, 7c, 8c, 9c, 10c
Σ= 0.7355 g	Σ= 0.7424 g	Σ= 0.7386 g
II. Subgroup	II. Subgroup	II. Subgroup
11a, 12a, 13a, 14a, 15a,	11b, 12b, 13b, 14b, 15b,	11c, 12c, 13c, 14c, 15c,
16a, 17a, 18a, 19a, 20a	16b, 17b, 18b, 19b, 20b	16c, 17c, 18c, 19c, 20c
Σ= 0.7461 g	Σ= 0.7456 g	Σ= 0.7461 g
III. Subgroup	III. Subgroup	III. Subgroup
21a, 22a, 23a, 24a, 25a,	21b, 22b, 23b, 24b, 25b,	21c, 22c, 23c, 24c, 25c,
26a, 27a, 28a, 29a, 30a	26b, 27b, 28b, 29b, 30b	26c, 27c, 28c, 29c, 30c
Σ= 0.7386 g	Σ= 0.7371 g	Σ= 0.7411 g

Separate solutions were prepared for each temperature at all ppm values. The mV values were recorded from the potentiometer screen after the readings stabilized. Calibration graphs for 25°C, 37°C, and 43°C are shown in Figures 2, 3, and 4, respectively.

Determination of Fluoride Uptake by the Enamel from the Mouthrinse Solution

Circulation water bath temperatures were adjusted for all temperatures and subgroups. Test solutions of 0.426 mL of the fluoride mouthrinse were freshly prepared for each subgroup. Different pipettes were used for TISAB, distilled water, and the mouthrinse. Magnetic stirrers and small magnets were used to maintain the stability and integrity of the solutions. New magnets and containers were used for each experiment. All experiments were performed using freshly prepared solutions.[22] Temperature sensors, thermometers, and fluoride-specific electrodes were held in place in the solutions by using fixed holders. The surfaces of the electrodes and the temperature equipment were washed with distilled water and dried between groups. The fluoride content of each solution was measured using a potentiometer and recorded prior to the addition of the enamel specimens at 15-s intervals up to 1 min, 1-min intervals from 1 to 10 min, 5-min intervals from 10 to 50 min, and 10-min intervals from 50 to 120 min. The fluoride uptake by the enamel specimens was calculated in mV by subtracting the measured concentration from the previously measured concentration before the addition of the specimens. The same procedures were repeated for all the temperatures and subgroups.

Fluoride-ion activity potential was measured in mV by using a potentiometer. The calibration graphs were constructed with mV values on the y-axis and the ppm F^- values on the x-axis; the ppm F^- values were calculated using the equation y = mx + n.

Statistical Analysis

The uptake of fluoride by the enamel specimens from the mouthrinse was measured and recorded from 15 s to 120 min. Graphs of the fluoride uptake for each temperature group were drawn,

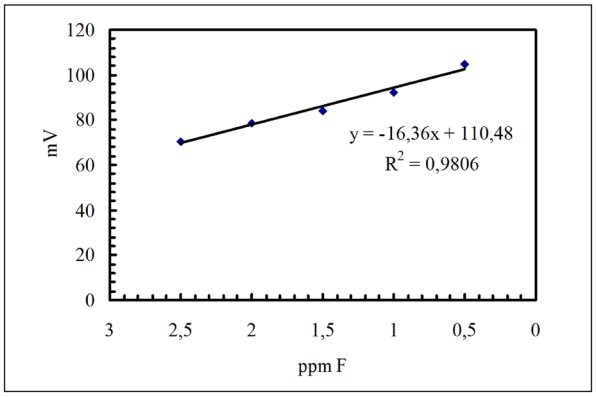

Figure 2. Calibration graph (25°C).

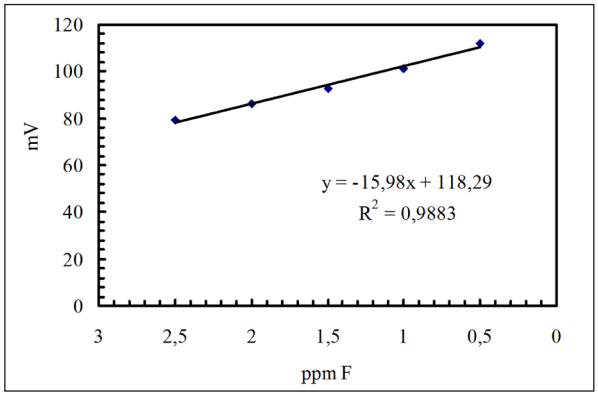

Figure 3. Calibration graph (37°C).

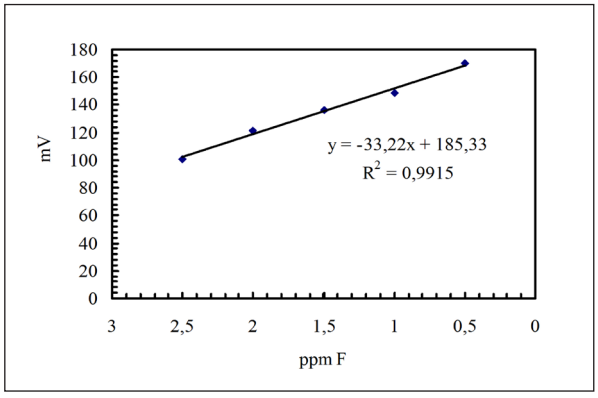

Figure 4. Calibration graph (43°C).

Table 4. 50mL calibration solutions (fluoride ppm and mouthrinse volume).

F ppm	mL mouthrinse
0,5	0.106
1	0.213
1,5	0.319
2	0.426
2,5	0.532

and intragroup and intergroup comparisons were assessed using one-way analysis of variance and the least significant difference, with the level of significance set at P<.05.

RESULTS

The fluoride absorption by the enamel specimens from the mouthrinse was found to increase with increase in temperature (Table 5).

Enamel Fluoride Concentrations at 25ºC

The changes in the mV and ppm values of the fluoride solution at 25°C are shown in Figures 5 and 6, respectively. At 25°C, most of the reduction in the solution's fluoride concentration occurred in the first 16 min, during which the concentration decreased at a rate of approximately 20% to 1.60 ppm. At 50 min, the mean fluoride concentration had decreased to 1.56 ppm (23%) and at 120 min, the mean fluoride concentration had decreased to 1.52 ppm (25%). Differences in the rate of change in fluoride concentrations were significant up 70 min (P<.05) but they were not significant from 70 min to 120 min (P>.05).

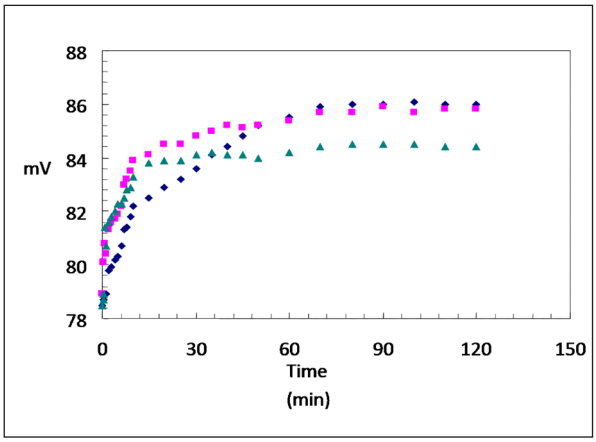

Figure 5. Changes (mV) in fluoride solution 25°C (♦:I.subgroup; ■:II.subgroup; ▲:III.subgroup).

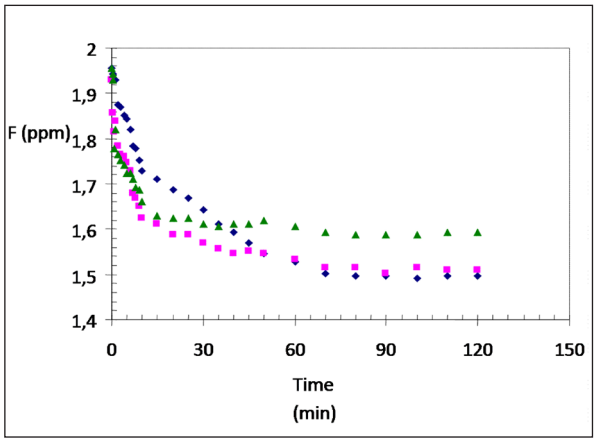

Figure 6. Changes in fluoride uptake (ppm) by enamel from solution at 25°C (♦: I. subgroup ; ■: II. subgroup; ▲: III. Subgroup).

Table 5. Enamel fluoride uptake from mouth rinse, by temperature/time.

Temperature	Time / ppm													
°C	15s	30s	45s	1m	2m	3m	4m	5m	6m	7m	8m	9m	10m	15m
	2	2	2	2	1,967	1,9364	1,967	1,9303	1,9181	1,9181	1,8936	1,8753	1,8444	1,8395
25	1,857	1,857	1,8386	1,8142	1,7836	1,7836	1,7592	1,7531	1,7286	1,6797	1,6797	1,6553	1,6347	1,6264
	1,9364	1,9303	1,8203	1,7775	1,7653	1,7531	1,7408	1,7225	1,7225	1,7103	1,6919	1,6858	1,6614	1,6309
	1,9561	1,9249	1,8998	1,7934	1,7058	1,6245	1,6244	1,5932	1,5619	1,5431	1,5118	1,4931	1,4757	1,4538
37	1,9624	1,9437	1,8748	1,831	1,7309	1,649	1,6245	1,5931	1,5557	1,5182	1,4806	1,4493	1,4276	1,4074
	1,9437	1,8999	1,8686	1,7935	1,7372	1,6308	1,587	1,5433	1,5119	1,4744	1,4306	1,3868	1,3354	1,2854
	1,6626	1,5181	1,3645	1,2532	1,1358	1,0545	0,9552	0,9341	0,8769	0,8438	0,8378	0,8197	0,8107	0,7987
43	1,5993	1,4729	1,2863	1,1448	1,0394	0,9551	0,8739	0,8047	0,7444	0,6993	0,6722	0,6512	0,6421	0,6301
	1,6896	1,5542	1,4548	1,3314	1,208	1,1026	0,9913	0,8739	0,7715	0,6873	0,6331	0,6241	0,615	0,603
oC	20m	25m	30m	35m	40m	45m	50m	60m	70m	80m	90m	100m	110m	120m
	1,8017	1,8123	1,7918	1,7652	1,7487	1,7118	1,7101	1,6856	1,6608	1,6575	1,6575	1,6571	1,657	1,657
25	1,6264	1,5962	1,5736	1,5642	1,5546	1,5527	1,5427	1,5218	1,5074	1,5074	1,5007	1,5074	1,5035	1,5035
	1,6247	1,6247	1,6125	1,6064	1,6124	1,6125	1.610	1,6063	1,5941	1,588	1,588	1,588	1,594	1,594
	1,4326	1,4219	1,3854	1,3548	1,3365	1,2847	1,2687	1,2133	1,1675	1,1345	1,078	1,0321	0,9989	0,9982
37	1,3762	1,3012	1,2738	1,2437	1,1987	1,1439	1.249	1,0623	0,9964	0,9265	0,8867	0,8127	0,7964	0,7887
	1,1997	1,1128	1,0768	1,0325	0,9978	0,9765	0,9565	0,9417	0,9265	0,8953	0,8417	0,8123	0,8053	0,8067
	0,7806	0,7716	0,7595	0,7505	0,7415	0,7415	0.740	0,7384	0,7233	0,7083	0,7143	0,7143	0,7173	0,7203
43	0,615	0,606	0,5999	0,594	0,591	0,591	0,594	0,597	0,597	0,5999	0,597	0,597	0,5999	0,5999
	0,5879	0,5699	0,5639	0,5548	0,5488	0,5428	0,5388	0,5338	0,5338	0,5428	0,5368	0,5397	0,5398	0,5398

Enamel Fluoride Concentrations at 37ºC

Changes in the mV and ppm values of the fluoride solution at 37°C are shown in Figures 7 and 8, respectively. At 37ºC, most of the reduction in the solution's fluoride concentration occurred in the first 12 min, during which the fluoride concentration of the solution decreased at a rate of approximately 30% to 1.4 ppm. At 50 min, the mean fluoride concentration had decreased to 1.00 ppm (50%) and at 120 min, the mean fluoride concentration had decreased to 0.7 ppm (65%). Differences in the rate of change in fluoride concentrations were significant up to 60 min (P<.05) but they were not significant from 60 min to 120 min (P>.05).

Enamel Fluoride Concentrations at 43ºC

Changes in the mV and ppm values of the fluoride solution at 43°C are shown in Figures 9 and 10, respectively. At 43°C, most of the reduction in the solution's fluoride concentration occurred in the first 5 min, during which the fluoride concentration of the solution decreased at a rate of approximately 65% to 0.7 ppm. At 50 min, the mean fluoride concentration had decreased to 0.60 ppm (70%), and no further changes in concentration were observed. Differences in the rate of change in fluoride concentrations were significant up 50 min (P<.05) but they were not significant from 50 min to 120 min (P>.05).

A Comparison of the Fluoride Uptake by the Enamel Specimens at Different Temperatures

Regardless of temperature, fluoride uptake occurred most rapidly during the first few minutes, after which it decreased until the enamel reached saturation. Moreover, the speed and amount of fluoride uptake increased with temperature, with majority of the fluoride uptake at 25°C occurring during the first 16 min, compared to the first 12 min at 37°C and the first 5 min at 43°C. Significant increase occurred in the rate of uptake until 70 min at 25°C, 60 min at 37°C, and 50 min at 43°C (P<.05), after which the changes were not statistically significant.

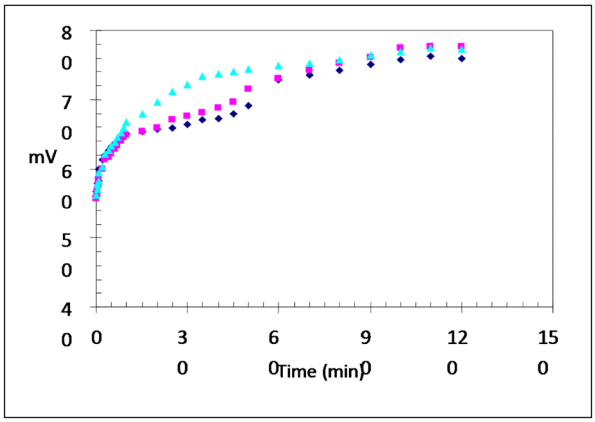

Figure 7. Changes (mV) in fluoride solution 37°C (♦:I.subgroup; ■:II.subgroup; ▲ :III.subgroup).

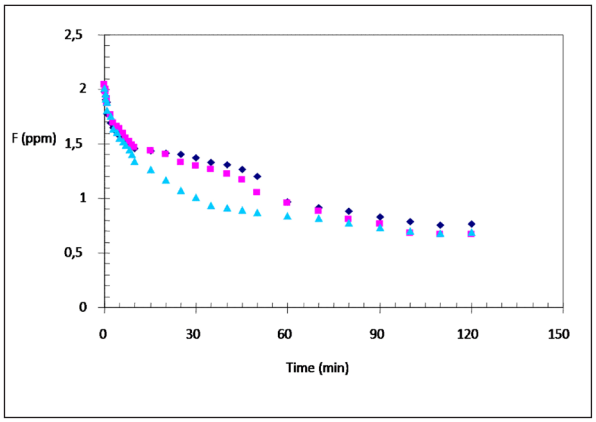

Figure 8. Changes in fluoride uptake (ppm) by enamel from solution at 37°C (♦: I. subgroup ; ■: II. subgroup; ▲: III. Subgroup).

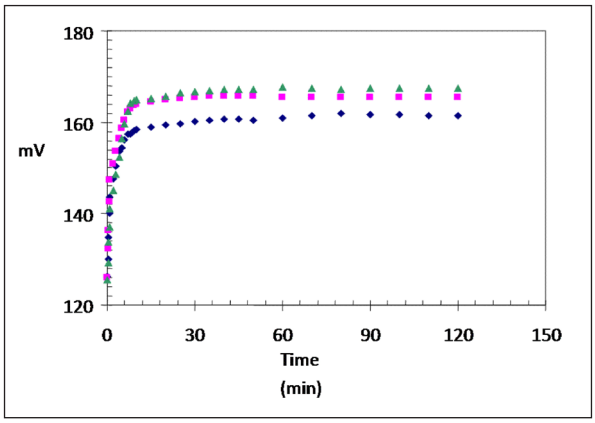

Figure 9. Changes (mV) in fluoride solution at 43°C (♦:I.subgroup; ■:II.subgroup; ▲ :III.subgroup).

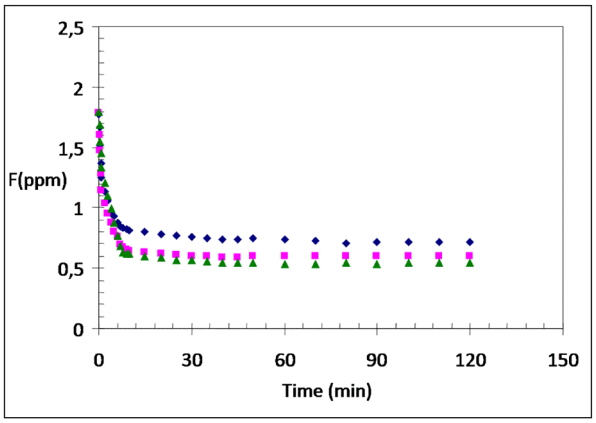

Figure 10. Changes in fluoride uptake (ppm) by enamel from solution at 43°C (♦: I. subgroup ; ■: II. subgroup; ▲: III. Subgroup).

DISCUSSION

The present study examined the effect of temperature on fluoride uptake by enamel specimens from a mouthrinse. In order to ensure comparability between groups, utmost care was taken in the preparation of the samples to provide standardization of volume and mass. Numerous studies have relied on fluoride-specific ion electrodes for measuring the fluoride uptake.[1,7,23-28] Our study also utilized a fluoride-specific ion electrode as a practical, economical, and accurate measuring tool.

Fluoride mouthrinses may be categorized by NaF concentrations as either low concentration (230 ppm, 0.05%) or high concentration (900 ppm, 0.2%).[29] In addition to NaF formulations, SnF2, amine fluoride, and ammonium fluoride mouthrinses are also available.[5,30-32] Mouthrinses routinely available on the market tend to be low-concentration, neutral-pH, 0.05% NaF solutions.[12,33,34] In this study, a 0.05% NaF mouthrinse solution was chosen because it possessed a stable structure, almost neutral pH, and high solubility; did not cause enamel discoloration (as with SnF_2); and was readily available on the market.

In line with many earlier studies, our study found that majority of the fluoride uptake by the enamel specimens occurred during the first minutes following application and continued over time. Moreover, our study showed that increasing the temperature of the fluoride solution resulted in an increase in the fluoride uptake by the enamel specimens as well as a decrease in the time needed to reach saturation. Fluoride absorption was found to cease at around 60 min; however, measurements continued until 120 min to control for additional changes/recycling.

Mouthrinse has been reported to be one of the most effective methods of providing low but consistent fluoride levels in the oral cavity.[6,32,35,36] As with other fluoride preparations, fluoridated mouthwashes are designed to increase the level of fluoride released in the saliva and maintain it at a certain level.[37] Most importantly, fluoridated agents are able to increase the resistance and decrease the solubility of the enamel to acids through the incorporation of fluoride ions into the enamel apatite structure.

As with all chemical reactions, the degree and speed of penetration of fluoride into the enamel and the formation of fluorapatite are strongly dependent upon concentration and temperature.[16] However, very few studies have examined the effect of temperature on fluoride uptake by the enamel from a mouthrinse. Barrancos[18] reported that fluoride absorption increases with increase in room temperature, and Stookey and Stahlman[20] showed that uptake from a fluoride solution could be increased by raising the temperature of the storage container by scrubbing. Moreover, in a study examining the effects of heat (25°C, 45°C, 65°C, and 85°C) on tin and fluoride uptake from a topical SnF_2 (8%) solution by enamel samples prepared from extracted bovine teeth, Putt et al[17] reported increase in both fluoride and tin uptake in line with increase in temperature, with the most significant increase in fluoride content observed at 65°C and 85°C. Our study used a 0.05% NaF solution and examined temperatures up to only 43°C, the body's hyper-fever limit. Nevertheless, in line with these previous studies, significant increase in fluoride uptake was observed with increase in temperature.

CONCLUSIONS

Our study found that the amount and rate of fluoride uptake by tooth enamel increased with increase in temperature. The temperature of NaF mouthrinse may easily and safely be increased beyond room temperature by placing a container of NaF mouthrinse in a bowl of hot water, allowing greater fluoride penetration into the enamel when the mouthrinse is used at home as a routine prophylactic agent. It should be noted that fluoride absorption and release may change depending upon an individual's dental structure and eating habits. Other studies are required to examine the effects of temperature on fluoride uptake in the enamels of different populations from other mouthrinse derivatives and at different temperatures and application times.

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