

# Effects of Low-volume Mouth Rinsing after Toothbrushing with Newly Developed Fluoride Dentifrice on Salivary Fluoride Concentrations

Taeko Osawa, Wenqun Song, Kazuaki Kawamura, Hirohisa Arakawa

*Department of Oral Health, Graduate School of Kanagawa Dental College, Yokosuka, Japan*

**Objective:** To investigate the effects of low-rinsing volume after toothbrushing with fluoride dentifrice (paste and gel dentifrice, 950 ppm F, new products with cationated cellulose and original products) on salivary fluoride retention.

**Methods:** Subjects comprised seven adults with a mean age of 40.8 years. Experimental groups were 1: New paste, rinse once with 15 ml of water, 2: New paste, rinse twice with 25 ml of water, 3: New paste, rinse four times with 30 ml of water, 4: Original paste, rinse once with 15 ml of water, 5: New gel, rinse once with 25 ml of water, 6: New gel, no rinsing, 7: Original gel, rinse once with 25 ml of water. They brushed their teeth for 3 min 30 s with 0.5 g of each dentifrice, after which six samples of unstimulated whole saliva were collected from immediately after brushing (0 min) to after 150 min.

**Results:** A statistical test for the AUC in group 1-4, fluoride retention was significantly higher in group 1 and 4 ( $p < 0.01$ ). In group 5-7, fluoride retention was highest in group 6 ( $p < 0.001$ ). As a result of multiple regression analysis with the AUC concerning the factors influencing salivary fluoride concentration, volume of water, baseline salivary fluoride and the amount of fluoride remaining in the mouth were selected as explanatory variables.

**Conclusion:** For both the new and original products, larger rinsing volume and more number of rinses significantly reduced salivary fluoride retention after toothbrushing with fluoride dentifrice, yield to reduction of their cationated cellulose effects.

**Keywords:** fluoride dentifrice, paste and gel dentifrice, low-volume rinsing, salivary fluoride retention

## Introduction

Fluoride-containing dentifrice first came into use as a home-care product in the 1940s, and in the 21st century, is used by an estimated 1.5 billion people worldwide (1). It is regarded as having made the greatest contribution to the global-scale decline in caries since the 1970s (2). Many studies have shown, however, that the caries-suppressing effects of fluoride denti-

frice are greatly affected by post-brushing methods of mouth rinsing, including the number of rinses and the volume of water used. In a three-year study of post-brushing methods of mouth rinsing and the incidence of caries in children who brushed with fluoride dentifrice, children who were not in the habit of using a cup but wet their toothbrushes, scooped up water with their hands, or placed their mouth under the tap to rinse it had significantly fewer caries than those who rinsed their mouths thoroughly using a cup for rinsing (3). A study that compared post-brushing mouth-rinsing habits in adults also found that those who had more caries rinsed their mouths significantly more often, used more water, and had significantly lower post-brushing salivary fluoride concentrations (4).

Preventing the loss of fluoride from the mouth by refraining from spitting out during brushing, rinsing with water only a few times after brushing, and other means of supplying as much flu-

Corresponding author **Wenqun Song**

Department of Oral Health, Graduate School of Kanagawa Dental College, Yokosuka, 82 Inaoka-cho, Yokosuka, Kanagawa [238-8580], Japan. Tel: +81-46-822-8862, Fax: +81-46-822-8862, E-mail: song@kdu.ac.jp

Received September, 5, 2013, Revised December, 11, 2013, Accepted December, 14, 2013

oride as possible to the oral environment have been suggested as ways of improving the caries-preventing effect of fluoride dentifrice (5). Most Japanese are in the habit of brushing their teeth at least twice a day (6), but they use only a small amount of dentifrice (7-9) and rinse their mouths several times with large amounts of water after brushing (9-11), which tends to reduce the caries-inhibiting effect of fluoride dentifrice. Dr. Shimoido of our department (12) has proposed a method of toothbrushing for adults that consists of selecting a dentifrice with a fluoride concentration of close to 1,000 ppm, using at least 0.5 g of this dentifrice, refraining as far as possible from spitting out during brushing, and rinsing the mouth out at the conclusion of brushing with less than 25 ml of water for under 4 s once or twice. In this study, we compared the salivary fluoride concentration after brushing with either a newly formulated NaF dentifrice containing hydroxyethyl cellulose dimethyl diallyl ammonium chloride cationated cellulose or the original product, in order to investigate the effect of low-volume mouth rinsing on salivary fluoride retention.

## Subject and Methods

### 1. Subjects and materials

This study was approved by the Ethics Committee of Kanagawa Dental University (no. 165). Subjects were 7 adults (3 males, 4 females) of mean age  $40.8 \pm 11.2$  years.

The products used in these experiments were Check-Up Standard<sup>®</sup> paste and Check-Up Gel<sup>®</sup> (new products created by blending the original products with cationated cellulose, and original products alone; 950 ppmF as NaF, Lion Dental Products Tokyo).

### 2. Methods

Table 1 shows the combinations of dentifrices and mouth-rinsing methods used. Experiment (1): rinsing once with 15 ml of water after brushing with the new paste; experiment (2): rinsing twice with 25 ml water after brushing with the new paste; experi-

ment (3): rinsing four times with 30 ml of water after brushing with the new paste; experiment (4): rinsing once with 15 ml of water after brushing with the original paste; experiment (5): rinsing once with 25 ml of water after brushing with the new gel; experiment (6): brushing with the new gel without rinsing; and experiment (7): rinsing once with 25 ml of water after brushing with the original gel.

All experiments were performed on weekday mornings. Subjects first brushed their teeth with non-fluoride dentifrice and rinsed their mouths thoroughly with distilled water, after which unstimulated whole saliva was collected for 5 min at rest (to measure the baseline salivary fluoride concentration). They then brushed their teeth with 0.5 g of the appropriate dentifrice for each experiment for 3 min 30 s, and after spitting out once, rinsed their mouths with the volume of distilled water stipulated for that experiment. Residual suspended solids on the toothbrush, substances spat out after brushing, and substances spat out during rinsing were collected as far as possible. Whole saliva was collected six times for 5 min, immediately after the end of the experiment (0 min) and 15, 30, 60, 120 and 150 min later. Subjects engaged in deskwork until the end of saliva collection, during which time they were forbidden to eat or drink. The samples were diluted as required, equal volumes of TISAB II were added, and fluoride concentration was measured with a fluoride ion electrode. The amount of fluoride remaining in the mouth was calculated from the difference between the amount of fluoride in the dentifrice used and the amount collected outside the mouth.

During the experiment, subjects were instructed to brush their teeth with a non-fluoride dentifrice from after their evening meal two nights before the experiment until the experiment itself. A washout period of at least two days was left between measurement days. Statistical tests used are described in the Results section.

## Results

Salivary fluoride concentration after using fluoride dentifrice

**Table 1.** Combinations of dentifrices and rinsing methods

Group	Dentifrices	Rinsing volume	Frequency of rinsing	Reasons of various rinsing methods
①	New paste	15 ml	1	New rinsing method of new dentifrice
②	New paste	25 ml	2	Former rinsing method of new dentifrice
③	New paste	30 ml	4	Large amount of rinsing method of new dentifrice
④	Original paste	15 ml	1	New rinsing method of original dentifrice
⑤	New gel	25 ml	1	Former rinsing method of new dentifrice
⑥	New gel	0	0	New rinsing method of new dentifrice
⑦	Original gel	25 ml	1	Former rinsing method of original dentifrice

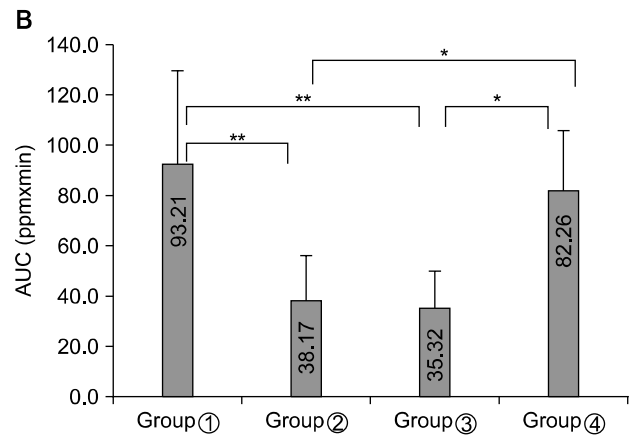
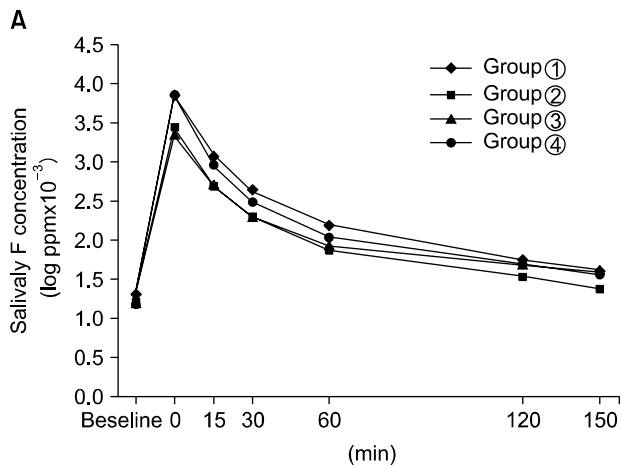
was greatest immediately after brushing and then declined steeply in all experiments, with a mean concentration after 150 min of 0.04 ppm. We also calculated the areas under the curve (AUCs) for salivary fluoride concentration up to 150 min after use. The salivary fluoride concentration results are shown in Figures 1A and 1B for the pastes and Figures 2A and 2B for gels.

One-way analysis of variance (ANOVA) followed by multiple comparison testing of AUCs (Scheffé's multiple comparison test) for each experiment showed that for the paste dentifrice shown in Figure 1B there were significant differences between experiments (1) and (2), (1) and (3), (4) and (2), and (4) and (3). In particular, fluoride retention was lowest in experiment (3), which used high-volume rinsing four times with 30 ml of water. Salivary fluoride concentrations immediately after experiments (1) and (4) were 7.4 ppm and 7.2 ppm, respectively, with the concentration 0.2 ppm higher after the new dentifrice was

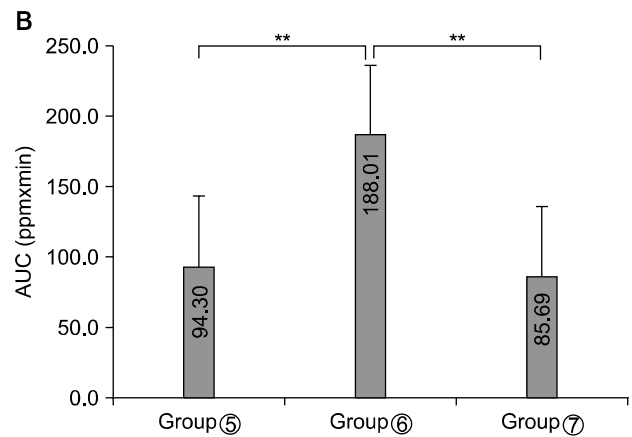
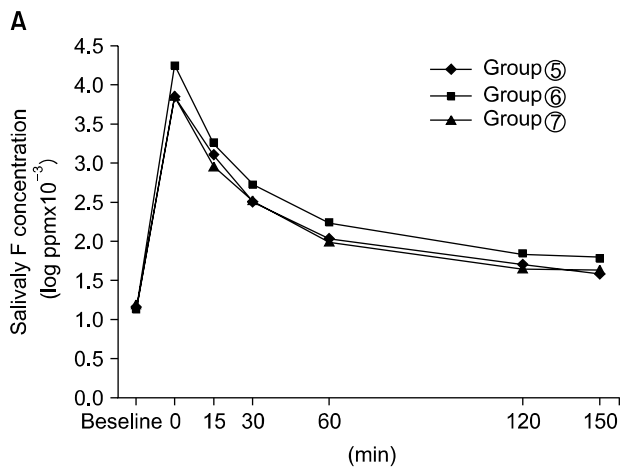
used, but although it subsequently also remained high there were no significant differences in AUCs.

For the gel dentifrice shown in Figure 2B, there were significant differences in salivary fluoride concentration immediately after brushing and in the AUCs between experiments (5) and (6) and experiments (6) and (7). Similarly to the paste dentifrice, there were no significant differences when the same method was used with the new and original products in experiments (5) and (7).

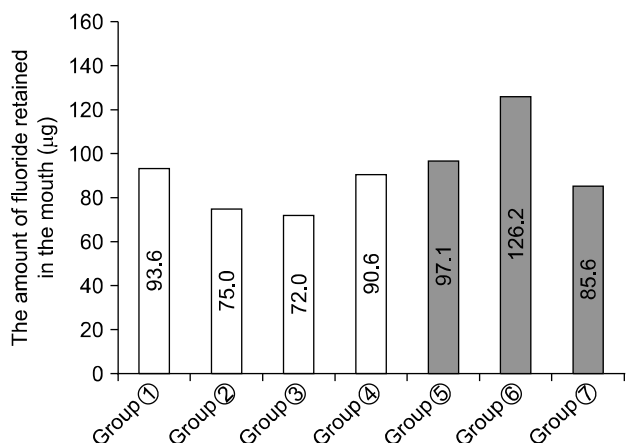
Figure 3 shows the amount of fluoride remaining in the mouth after brushing. The highest figure was 126.2 µg in experiment (6), in which gel dentifrice was used without rinsing. The highest value for paste dentifrice was 93.6 µg in experiment (1), in which rinsing was performed once with 15 ml of water, and the lowest value was 72.0 µg in experiment (3), when rinsing was performed four times using a total of 120 ml of water. One-way



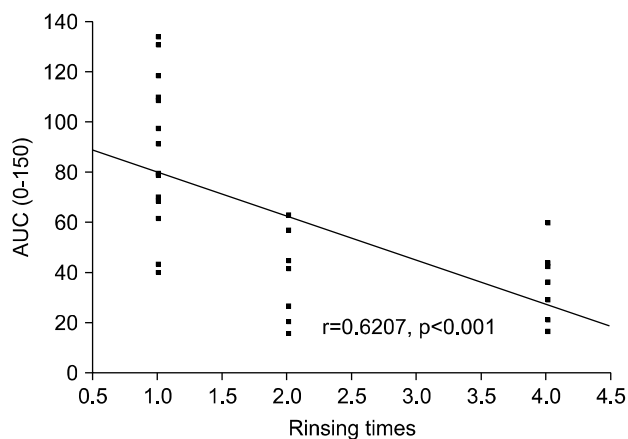
**Figure 1.** (A) Changes over time in salivary fluoride concentration after toothbrushing with paste dentifrice. (B) AUC of salivary fluoride concentrations after toothbrushing with paste fluoride dentifrice. \* $p < 0.05$ , \*\* $p < 0.01$ .



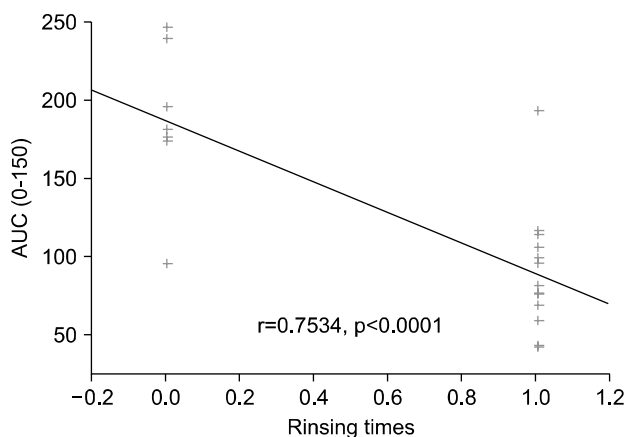
**Figure 2.** (A) Changes over time in salivary fluoride concentration after toothbrushing with gel dentifrice. (B) AUC of salivary fluoride concentrations after toothbrushing with gel fluoride dentifrice. \*\* $p < 0.01$ .



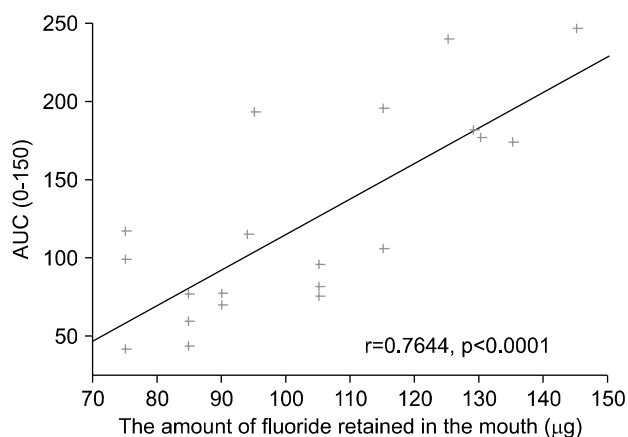
**Figure 3.** Amount of fluoride retained in the mouth after dentifrice use in each experimental group.



**Figure 5.** Correlation of rinsing times and salivary fluoride retention as AUC after toothbrushing with paste fluoride dentifrice.



**Figure 4.** Correlation of rinsing times and salivary fluoride retention as AUC after toothbrushing with gel fluoride dentifrice.



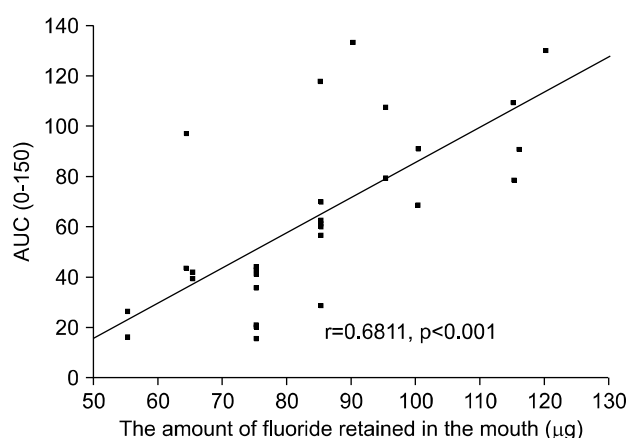
**Figure 6.** Correlation of the amount of fluoride retained in the mouth and fluoride retention as AUC after toothbrushing with gel fluoride dentifrice.

ANOVA followed by Scheffé’s multiple comparison test showed that there were significant differences in ANOVA ( $p < 0.05$ ), however, there were no significant differences in Scheffé’s test. In experiments (5), (6) and (7) with gel dentifrice, there were significant differences between experiments (5) and (6) and experiments (6) and (7) (ANOVA,  $p < 0.001$ ; Scheffé’s multiple comparison,  $p < 0.001$ ). The amount of fluoride remaining in the mouth after brushing clearly differed when different numbers of rinses and amounts of water were used.

We performed single regression analysis separately for paste and gel dentifrices with the AUCs for salivary fluoride concentration up to 150 min after brushing as Y and volume of water, number of rinses, and amount of fluoride remaining in the mouth in each experiment as X. This showed that the number of rinses and the volume of water used have a strongly negative correlation with AUCs, and that the amount of fluoride remain-

ing in the mouth exhibited a strongly positive correlation. These results are shown in Figures 4 and 6 for the gel dentifrices and Figures 5 and 7 for the paste dentifrices.

In order to investigate the effects of factors influencing salivary fluoride concentration, such as the number of rinses, volume of water, amount of fluoride remaining in the mouth, salivary fluoride concentration before the start of the experiment (baseline value) and volume of saliva secretion, we carried out stepwise multiple regression analysis with the AUC as the target variable (Y). Volume of water (X1), baseline salivary fluoride (X2) and the amount of fluoride remaining in the mouth (X3) were selected as explanatory variables, and the following regression formulae were used: for paste dentifrice,  $Y = -21.5 - 0.3X1 + 1668.8X2 + 0.9X3$  ( $p < 0.001$ ); for gel dentifrice,  $Y = -63.7 - 3483.4X2 + 2.3X3$  ( $p < 0.001$ ).



**Figure 7.** Correlation of the amount of fluoride retained in the mouth and fluoride retention as AUC after toothbrushing with paste fluoride dentifrice.

## Discussion

In order to improve the caries-preventing effects of fluoride dentifrice, it is important to both increase the amount of fluoride supplied to the mouth and to reduce the amount flushed out of the mouth by post-brushing rinsing (5). Japanese, however, are in the habit of brushing with a small amount of dentifrice (7-9) and rinsing their mouths several times with large amounts of water after brushing (9-11). It is impracticable to dramatically reduce the number and volume of rinses, solely out of consideration for increasing fluoride retention in the mouth after brushing, as this leaves an unpleasant aftertaste. In our department, we have therefore investigated methods of increasing the time for which the salivary fluoride concentration is maintained at over 0.05 ppm, regarded as the lower limit for promoting remineralization, after brushing using the fluoride dentifrice (13).

Taking into account Japanese circumstances, our recommendations for adults comprise selecting a dentifrice with a fluoride concentration of close to 1,000 ppm, using at least 0.5 g of this dentifrice, refraining as far as possible from spitting out during brushing, and rinsing the mouth out at the conclusion of brushing with less than 25 ml of water for under 4 s once or twice, and we have worked to promote their widespread adoption (12). Fifteen years later, we investigated the use of a new fluoride dentifrice formulated with cationated cellulose (14), as well as the practicability of decreasing the amount of rinsing, with the aim of increasing fluoride retention on the surface of the teeth after brushing. We found that after brushing with a mildly flavored dentifrice, restricting rinsing to once for 4 s with 15 ml of water was acceptable to general consumers (15). In this study, we investigated the efficacy of low-volume rinsing after brushing with a new fluoride dentifrice. Cury et al. (16) stated that fluo-

ride concentration in whole saliva can be used as an indicator of fluoride bioavailability, while Rolla & Ekstrand (17) found that fluoride retention in the oral reservoir following fluoride dentifrice use was affected by the fluoride level in whole saliva. We therefore used fluoride retention in whole saliva as an indicator in our experiments.

From our experimental results, whether the new or original gel and paste dentifrices were used, increasing the number and volume of rinses significantly decreased salivary fluoride retention after fluoride dentifrice use, which may very well also decrease its caries-preventing effects. The effects of rinsing with a large volume of water (30 ml×4) in experiment (3) meant that fluoride retention in the mouth was very low. Fluoride retention was significantly higher in experiment (1) than in experiment (2), which used the previously proposed method of rinsing. Nevertheless, although the AUC in experiment (1) was 10 ppm×min higher than in experiment (4), which adopted the same rinsing method used in experiment (1) for the original dentifrice, this difference was not significant.

For gel dentifrices, Shimoido (12) proposed rinsing once for less than 4 s with less than 25 ml of water after tooth brushing. Experiments (5) to (7) using the gel type were therefore designed as shown in Table 1. We found that although the rinsing method was the same, the AUC was 9 ppm×min higher in experiment (5), using the cationated cellulose formulation, than in experiment (7), which used the original dentifrice, although this difference was not significant. Based on the high fluoride retention seen in experiment 6, in which no post-brush rinsing was performed, we should consider recommending the patients with high caries risk use gel dentifrice without rinsing for the second brushing when double brushing.

In this study, which used saliva as the test substance, we found no significant differences in salivary fluoride concentration as a result of the effects of the new dentifrice, which contained an improved coating material compared with the original product. Although a study by Kubota et al. (14) found that fluoride retention on the tooth surface after brushing with the new product was better than with the original product, increasing retainability on the surface does not appear to promote fluoride retention in the oral environment. Further studies into fluoride retention on the surface of human teeth after fluoride dentifrice use are necessary.

## Conclusion

We found that large-volume mouth rinsing (rinsing numerous times with a large amount of water) reduces the amount of fluoride retained in the mouth, and that fluoride retention in the mouth after brushing gradually decreases. This tendency is evi-

dent similarly for both paste and gel dentifrices. Increasing the number of rinses also increases the volume of water used. In light of the results of experiment (6), in which only one post-brushing spit and no rinsing are allowed, brushing actions would ideally be improved so as to avoid rinsing, but people are generally unwilling to accept the notion of not rinsing. If 25 ml is regarded as the volume of a single mouthful in adults, it may be more effective and practicable consciously to restrict the number of rinses to once or twice.

## Acknowledgements

We are indebted to the colleagues of this investigation for their cooperation.

## References

1. Rugg-Gunn A. Preventing the preventable- the enigma of dental caries. *Br Dent J* 2001;191(9):478-88.
2. Buischi YP, Burt BA, Ghandour I, et al. Report of a WHO expert committee on oral health status and fluoride use. Fluorides and oral health (WHO Technical report Series 846). Geneva: World Health Organization; 1994:26-9.
3. Chesters RK, Huntington E, Burchell CK, Stephen KW. Effect of oral care habits on caries in adolescents. *Caries Res* 1992; 26(4):299-304.
4. Sjögren K, Birkhed D. Factors related to fluoride retention after toothbrushing and possible connection to caries activity. *Caries Res* 1993;27(6):474-7.
5. Sjögren K, Birkhed D, Rangmar B. Effect of a modified tooth-paste technique on approximal caries in preschool children. *Caries Res* 1995;29(6):435-41.
6. Hanada N, Ando Y, Ueno M, et al. Comprehensive guide the survey of dental diseases (2005). Tokyo: Oral Health Association of Japan; 2005:38-9.
7. Arakawa H, Kuroha K, Kawamura K, Uematsu M, Yamaguchi K, Hirata Y, Iizuka Y. Study on amount of dentifrice used (I). *J Shonan College* 1995;6:11-7.
8. Kuroha K, Kato M, Komiyama M, Shimoido S, Iwase Y, Arakawa H, Iizuka Y. Study on amount of dentifrice used (II). *Kanagawa Shigaku* 1996;30(4):397-404.
9. Iwase Y, Hirata Y, Kawamura K, Kuroha K, Arakawa H, Iizuka Y. Influence on toothbrushing habits of soft-paste fluoride dentifrice. *J Dent Hlth* 1996;46:472-3.
10. Mihata T, Toda S, Arakawa H. The influence of dentifrice with or without foaming agent on effectiveness of mouth cleaning. A clinical trial. *J Dent Hlth* 2000;50:361-74.
11. Iwase Y. Oral sensations and fluoride retention in saliva after toothbrushing with a foam-type fluoride dentifrice. *Kanagawa Shigaku* 1999;34(1):27-42.
12. Shimoido S. Caries preventive potentialities of various fluoride products for home-use. A study through the fluoride concentration in saliva at the time of rinsing in the morning. *Kanagawa Shigaku* 1999;34(1):43-60.
13. Featherstone JDB, Zero DT. Clinical and biological aspects of dentifrices. Laboratory and human studies to elucidate the mechanism of action of fluoride-containing dentifrices. New York: Oxford University Press; 1992:41-51.
14. Kubota M, Fujikawa H, Uno D, Yamamoto K, Ujiie T. Studies on fluoride retention and remineralization by fluoride dentifrice combined with cationic cellulose. *J Dent Hlth* 2005;55(4):384.
15. Osawa T, Song W, Ishiguro A, Nakamukai M, Ishida N, Arakawa H. Feasibility investigation of low-volume rinsing with fluoride dentifrice. *Int J Clin Prev Dent* 2013;9(3):125-9.
16. Cury JA, Del Fiol FS, Tenuta LMA, Rosalen PL. Low-fluoride dentifrice and gastrointestinal fluoride absorption after meals. *J Dent Res* 2005;84(12):1133-7.
17. Rölla G, Ekstrand J. Fluoride in dentistry: fluoride in oral fluids and dental plaque 2. Copenhagen: Munksgaard; 1996:215-29.