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The Effect of Toothbrushing Using Microcurrent Toothbrush on the Fluoride Concentration on the Enamel Surface*

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ABSTRACT

The purpose of this study is to verify the effect of toothbrushing using a microcurrent toothbrush on the fluoride concentration on the enamel surface of teeth. Sixty samples were produced with sound cattle teeth and divided into control and experimental groups (ordinary and microcurrent toothbrush groups) to perform the brushing for four minutes using an automatic toothbrushing machine in a 2% NaF solution for each type of toothbrush. The specimen measured the microhardness of enamel surface after the pH cycling treatment, and the fluoride ratio of the enamel surface was identified by scanning electron microscope observation and EDS analysis. The microhardness of the enamel surface of the microcurrent toothbrush group was higher after brushing (279.64) than before brushing (267.74) (p=0.008). In scanning electron microscopic observation, it was confirmed that the enamel surface after brushing with a microcurrent toothbrush appeared a slight irregular spherical crystals, forming fluoroapatite. According to the EDS analysis of the scanning electron microscopy, the fluoride content of the enamel surface was 1.76% in the control group, while 22.29% in the microcurrent toothbrush group. In sum, it has been found that toothbrushing using microcurrent toothbrushes is effective in preventing tooth decay by increasing the formation of fluoroapatite in the enamel.

1. Introduction

Against dental caries, high fluoride concentration on the enamel surface increases the resistance to the progress of desorption. In other words, fluorine ions are more fully coupled than hydrogen ions or other ions, creating highly acid-resistant apatite crystals. The fluoride concentration of the teeth is

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relatively high at 500 to 4,000 ppm at the enamel surface with 50㎛, but in the depths of the enamel, it decreases to 50~1,200 ppm, and the fluoride concentration of the ivory is between 200 and 1,500 ppm, which is the middle level between the depths and the surface layer of the enamel (WHO, 1994).

Fluoride-replated enamel crystals are much more resistant to acids produced in the process of caries, and even in parts where fluoride is missing, fluorine ion and other ions form the layer and become more resistant (Lippert, 2017), oral health managers are constantly interested in effective fluoride layer, which can increase the fluoride concentration of the enamel (Jang & Lee, 2006; Creeth et al., 2016). It is reported that fluoride application has different levels of fluoride absorbed into the enamel depending on the method of use, so it has different effects of preventing tooth decay (Rošin-Grget et al., 2002).

In general, expert fluoride application uses high concentrations of fluoride solutions or gels, or varnish, and are mainly used in solution surface application where sodium fluoride and fluoride are used (Seppä et al., 1994), and tray gel application using 1.23% acid fluorophosphate (Ripa, 1991). Recently, iontophoresis using a 2% sodium fluoride solution has been clinically applied (Lee et al., 2008; Kim et al., 2008; Chang et al., 2011). It was reported that iontophoresis is mainly used in conservative treatment for pulp or pain caused by an ulcer of oral mucosa and hyperesophoria of ivory with microcurrent and lidocaine anesthesia (Carlo et al., 1982), fluoride iontophoresis is the most effective method of fluoride application for the re-calcification of the early desorption legion of the enamel (Simon et al., 1995). However, fluoride iontophoresis is not only subject to dental visits, but also to the treatment of specialists using a special device for it.

Toothbrush treatment is the most basic method among various oral hygiene management methods, which is essential for preventing periodontal diseases and tooth decay. In general, self-fluoride application is typically done with fluoride toothpaste or fluoride solution. They contained 0.15% (1,500 ppm) or less of Sodium fluoride (NaF), sodium monofluorophosphate (SMFP), stannous fluoride (SnF2), and amino fluoride.

Fluoride toothpaste is known to have a lower concentration compared to fluoride solutions used in the expert fluoride application method, but it is known to have a higher effect of preventing caries because it is used more than twice a day (Oral Health School Compilation Committee, 2016). The systematic review of the Cochrane Collaboration (2010) stated that fluoride toothpaste had about 24% prevention effect, expert fluoride application had 28% prevention effect, and fluoride varnish application method had 43% prevention effect. Based on the above information, it can be inferred that toothbrushing with a microcurrent toothbrush to further enhance the fluoride application effect of fluoride toothpaste will increase the fluoride concentration of the enamel and ultimately increase the prevention effect of dental caries. In this study, the effect of toothbrushing using a normal toothbrush and a toothbrush with microcurrent of a similar level of body current was compared and analyzed for the effect on the fluoride concentration of the enamel. In order to determine the effect of oral hygiene management using a microcurrent toothbrush on fluoride concentration of enamel, measurement of the surface microhardness and field emission scanning electron microscopy (SEM) observation were conducted. The specific research objective was, first, to analyze the change in microhardness of the enamel after brushing using a microcurrent toothbrush, and second, after brushing using a microcurrent toothbrush, to observe scanning electron microscope findings of the enamel surface and to identify the fluoride ratio in the enamel through EDS analysis.

2. Materials and Methods

2.1 Specimen preparation

Among incisors of cattle, those without surface loss, pigmentation, or desorption were refrigerated at 4˚C before the experiment, and 50 sound teeth were finally used as the research subjects.

The subjects of the experiment were classified into a control group and two toothbrushing groups. The sound teeth were not treated with fluoride, cut in half, and used as the control group (A), while the other half were assigned as the experimental group. Toothbrushing groups, which were the experimental group, were classified into a group (B) using an ordinary toothbrush (LG Household and Health Care, Korea) and a group (C) using an ultra-thin microcurrent toothbrush (GI, Korea).

The experimental conditions for each group are as shown in Table 1. The size of the samples studied was referenced by the study of Jang (2004) and Kim (2016), and 40 samples were calculated using G*power 3.17, with the significance level (α) of 0.05, effect size 0.20, and verification power (1-β) of 0.80, but 60 samples for the final analysis were made considering that samples with exposed dentin while brushing would be excluded.

Table 1. Experimental conditions by experiments and control

Group	Sort	N	Experimental conditions
Control group	A	20	Without any fluoride treatment.
Brushing group	B	20	Toothbrushing in 2% NaF solution for 4 minutes using a regular toothbrush
	C	20	Toothbrushing in 2% NaF solution for 4 minutes using a microcurrent toothbrush

After brushing the teeth, the target teeth were cut only the enamel part using dental cutting bur, carborundum paper, and caliper, and the specimen was produced with an area of about 4 × 4mm. Also, the samples from the control group (group A) teeth without fluoride application were also produced in the same way.

2.2 Toothbrushing and pH-cycling

For the toothbrushing groups, brushing was conducted for 4 minutes in 2% NaF solution using an automatic toothbrushing machine with each type of toothbrushes (ordinary toothbrush and micro-current toothbrush).

The pH-cycling process was carried out for five days for the control samples and the samples with toothbrushing (see Fig. 1).

The manufacturing process of the desorption solution required for the pH-cycling process is as follows. The tertiary distilled water was added to the 50cc 1.0M lactic acid to 500cc, and the 50% NaOH solution was added to set it to pH 5.0. Then, 1.5g HAP (tribase calcium phosphate) and 20% HCl solution were added together, dissolved for 30 minutes maintaining pH 5.0 in the

stirrer and then filtered. Add 200ml of 1% carbopol stock solution and 50cc 1.0M lactic acid to the filtered solution, add distilled water to make it a little less than 1000cc, and add 50% NaOH solution to make pH 5.0. After that, a 1L desorbed solution was completed by adding tertiary distilled water to make it 1000cc.

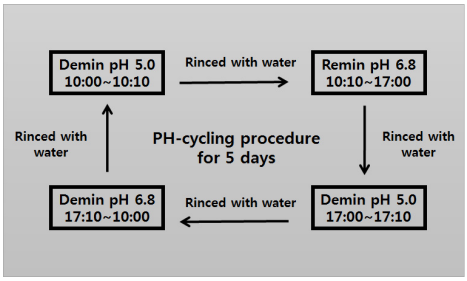


Fig. 1. pH-cycling process

The method of manufacture of 1L of desorbed solution (Lee et al., 2012) is as follows. The tertiary distilled water was added to 50cc 1.0M lactic acid to 500cc, and the 50% NaOH solution was added to set at pH 5.0. 1.5g HAP (tribase calcium phosphate) and 20% HCl solution were added together and dissolved in the stirrer maintaining pH 5.0 for 30 minutes, and then filtered in the funnel. One percent carbopol stock solution 200ml and 50cc 1.0M lactic acid were added to the filtered solution, and tertiary distilled water was added to make it a little less than 1000cc. The remineralized solution is made as follows. 0.002g ascorbic acid, 0.03g glucose, 0.058g NaCl, 0.17g CaCl2, 0.16g NH4Cl, 1.27g KCl, 0.16g NaSCN, 0.33g KH2PO4, 0.2g urea, and 0.34g Na2HPO4 were measured and put in 1,000cc of tertiary distilled water, set it at pH 6.8 using 50% NaOH solution and 20% HCl solution, and dissolved it in a constant temperature water tank (37℃) for 20 hours.

The manufacturing method of 1L of the remineralization solution (Lee et al., 2012) is as follows. In the third distilled water, 0.002g ascorbic acid, 0.03g glucose, 0.058g NaCl, 0.17g CaCl2, 0.16g NH4Cl, 1.27g KCl, 0.16g NaSCN, 0.33g KH2PO4, 0.2g Eura, 0.34g Na2HPO4 were measured and added. Using 50% NaOH solution and 20% HCl solution, the pH was set to 6.8, and a constant temperature water tank was set to 37°C and dissolved for 20 hours.

2.3 Measurement of the vickers hardness number

Using a Digital Microhardness Tester (MXT-α7, Matsuzawa Co., LTD, Japan), all specimens are divided into 6 parts per specimen, measuring the Vickers Hardness Number (VHN) with a weight of 200 grams before brushing, and measuring 3 locations per part for average. The samples used in the first experiment were cleaned with distilled water and soaked in distilled water for 7 days before measuring the microhardness in the same way as above. In the second experiment, toothbrushing was performed for each type of toothbrush, cleaned with distilled water, treated for five days during pH-cycling process, and measured the microhardness in the same way.

2.4 Morphological examination

Using SEM (Sigma 500, Carl Zeiss, LTD, Germany), fluoride concentration on the enamel surface by each type of toothbrush (microcurrent toothbrush and ordinary toothbrush) was observed. The enamel surfaces of each experimental and control specimen were observed at 500 times, 4,000 times, and 10,000 times after the platinum sheath in the ion sputter. In addition, the atomic content was measured by taking a dot mapping image of elements P, F, and Ca on the tooth surface using EDS (10,000 magnification).

2.5 Statistical analysis

For the collected data, the Wilcoxon-Signed Ranks test was performed on the measured enamel surface microhardness before and after brushing using the SPSS 23.0 program. After the significance was tested by Kruskal-wallis test for each type of toothbrush, the follow-up test was performed by the Scheffe test for multiple comparisons.

3. Results and Discussion

3.1 Microhardness of the enamel surface

Table 2 is the result of oral hygiene control using microcurrent toothbrush and surface microhardness measurement for comparison and analysis with fluoride ion concentration absorbed in the enamel after brushing with an ordinary toothbrush. There was no significance between the microhardness of the control group (266.80 points) and the ordinary brushing group (264.09 points) and the microcurrent brushing group (267.74 points) before brushing (baseline), securing the homogeneity (p=0.839). After four minutes of brushing in a 2% NaF solution of each type of toothbrush, it was soaked with distilled water for seven days and measured its microhardness. While the ordinary toothbrush group was reduced to 257.73 points, but the microcurrent toothbrush group was slightly higher at 269.71 points (p=0.058).

Table 2. Comparison of microhardness of the enamel after toothbrushing by types of toothbrush

Group	N	Baseline	Exp. I†	p-value*
		Mean±SD	Mean±SD	
Control group	18	266.80±31.53	255.69±24.11	0.677
Ordinary toothbrush group	18	264.09±17.01	257.73±14.87	0.107
Microcurrent toothbrush group	18	267.74±13.87	269.71±14.21	0.742
p-value**		0.839	0.058	

* by Wlicoxon Signed Ranks Test at α=0.01
** by Kruskal Wallis Test at α=0.01
† Group that measured the microhardness after brushing with 2% NaF solution for 4 minutes, then immersed in distilled water for 7 days

Table 3 is the result of measuring the microhardness after brushing for 4 minutes in 2% NaF solution of each type of toothbrush, washing it with distilled water, and then processing it for 5 days through pH-cycling. The microhardness of the ordinary toothbrush group was reduced to 245.04 points after brushing and pH-cycling treatment compared to before the brushing (264.09 points), but the microhardness of the microcurrent toothbrush group was increased to 279.64 points after brushing and pH-cycling treatment compared to before the brushing (267.74 points), resulting in a significant difference (p=0.008). The microhardness by each type of toothbrush was higher in the microcurrent toothbrush group than in the ordinary toothbrush group (p=0.022).

Table 3. Comparison of the microhardness after toothbrushing by type of toothbrush by pH-cycling treatment

Group	N	Baseline	Exp. II†	p-value*
		Mean±SD	Mean±SD	
Control group	18	266.80±31.53	256.75±27.02[a]	0.301
Ordinary toothbrush group	18	264.09±17.01	245.04±33.55[a,b]	0.389
Microcurrent toothbrush group	18	267.74±13.87	279.64±13.36[b]	**0.008**
p-value**		0.839	**0.022**	

* by Wlicoxon Signed Ranks Test at α=0.01
** by Kruskal Wallis Test at α=0.01
[a,b] Means followed by different letters are statistically significantly different by Scheffe test at α=0.01
† Group that measured the microhardness after brushing with 2% NaF solution for 4 minutes, then immersed in distilled water for 5 days

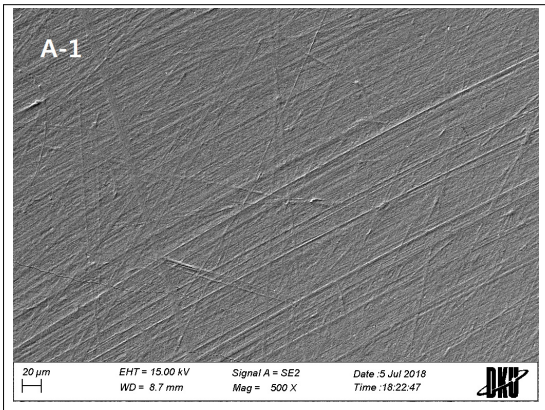
3.2 SEM observations

As a result of observation with a scanning electron microscope on the enamel surface, and the untreated control group turned out to be even and smooth (A-1 and A-2).

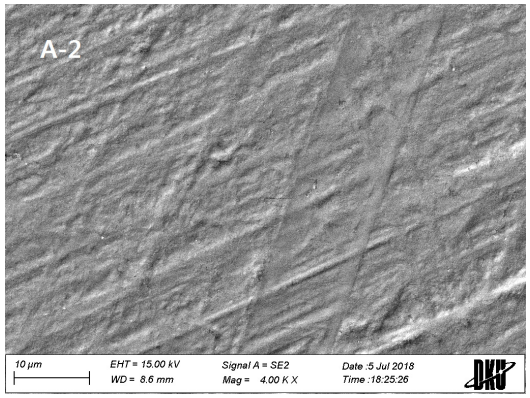
The scanning electron microscopic findings of the enamel surface observed after a pH-cycling treatment for five days after brushing for 4 minutes in 2% NaF solution are believed to be that irregular spherical crystals formed, which seems like fluoroapatite, in the ordinary toothbrush group (B-1 and B-2) and the microcurrent toothbrush group (C-1 and C-2).

However, due to the short period of this experiment, it was not enough to understand the exact process of the change, and given the prior studies, it was suggested that the test period needs to be extended to at least 28 days in order to confirm the formation of distinct fluoroapatite on the enamel surface.

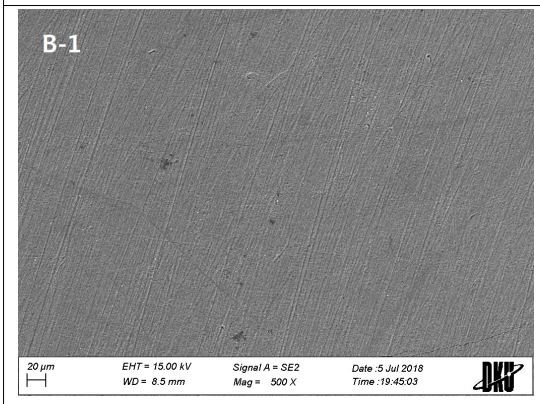
As a result of observation by scanning electron microscope on the enamel surface that were brushed in 2% NaF solution for 4 minutes, washed with distilled water, soaked in distilled water for 7 days, while the control group was unchanged, the microcurrent toothbrush group (C) showed slightly irregular crystals of calcium fluoride and crystals of globular shapes with angles, forming a new foundation that appeared to be fluoroapatite (see fig. 3 and 4). Therefore, toothbrushing using microcurrent toothbrush is found to be effective in forming fluoroapatite in the enamel, so it is believed to be useful for preventing tooth decay



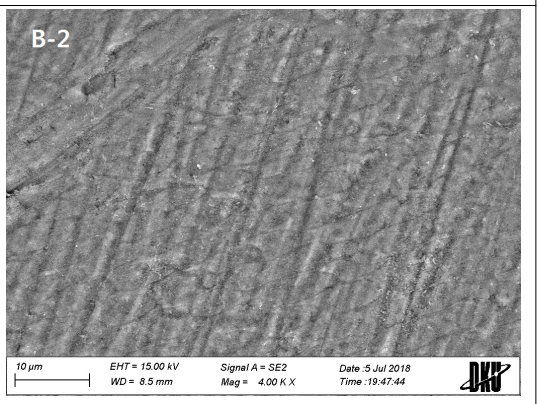
A-1. Control group (×500)



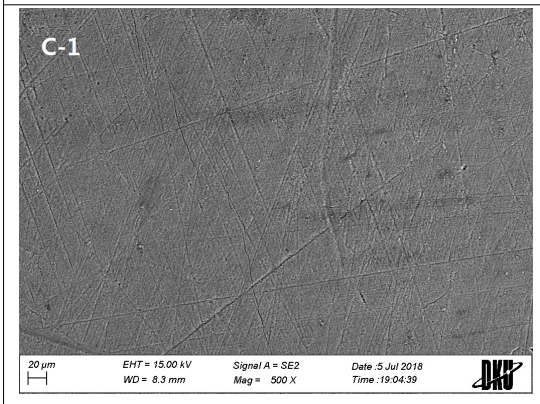
A-2. Control group (×4,000)



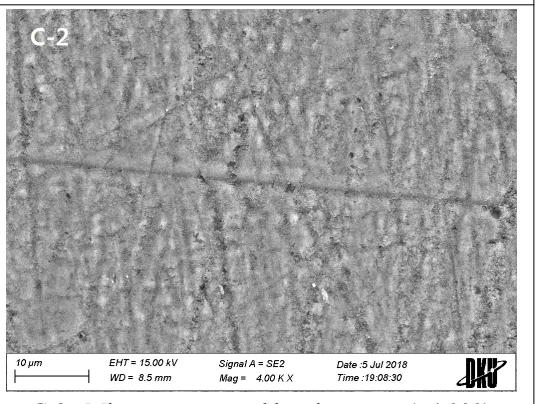
B-1. Ordinary toothbrush group (×500)



B-2. Ordinary toothbrush group (×4,000)

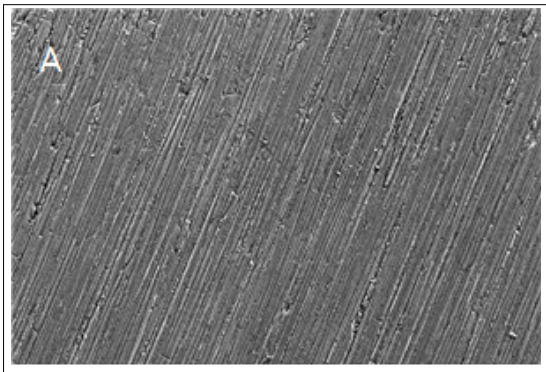


C-1. Microcurrent toothbrush group (×500)

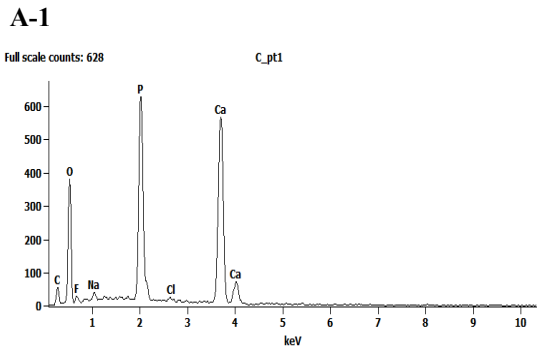


C-2. Microcurrent toothbrush group (×4,000)

Fig. 2. Results of observation through scanning electron microscope on the enamel surface after brushing by type of toothbrush

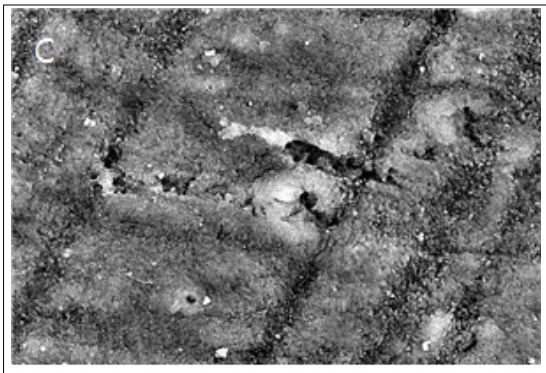


A. Control group (×10,000)

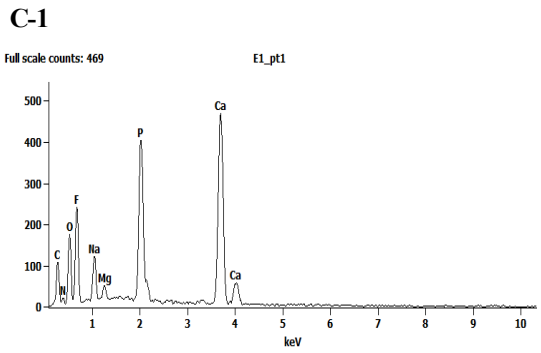


A-1. Atomic content of the enamel surface of the untreated control group(%)

Fig. 3. Results of observation through scanning electron microscope on the enamel surface of the control group



C. After microcurrent toothbrushing (×10,000)



C-1. The atomic content of the enamel surface after microcurrent toothbrushing (%)

Fig. 4. Observation results of the enamel surface of the microcurrent toothbrush group by scanning electron microscope

As a result of EDS analysis of the observation of the enamel surface by scanning electron microscope after four minutes of brushing in 2% NaF solution and pH-cycling treatment for five days, the untreated control group showed 1.76% of fluoride and the microcurrent toothbrush group showed 22.29% (see Table 4).

Table 4. Atomic content of the enamel surface by the observation with scanning electron microscope (Unit: %)

Group	Element					
	C	O	**F**	Na	P	Ca
Control group	2.55	38.03	**1.76**	0.80	18.75	37.61
The microcurrent toothbrush group	6.27	18.57	**22.29**	3.90	12.60	30.31

This study was an in vitro experiment that observed microhardness and scanning electron microscope findings seven days after specimen production. In order to measure the process of the change of fluoride concentration in the enamel after brushing teeth, it is suggested that pH-cycling process is carried out for at least 28 days, or that in-vivo experiments would make the research results more feasible. In addition, it is necessary to observe the depths of enamel produced by the process of desorption and remineralization through the observation of CLSM (confocal laser scanning microscope).

4. Conclusion

This study was attempted to determine the effect of toothbrushing using a microcurrent toothbrush on the production of fluoroapatite on the enamel surface.

60 samples were produced using sound cattle teeth, and the control group (A) and the experimental group were divided into the ordinary toothbrush group (B) and the microcurrent toothbrush group (C), and toothbrushing was performed for 4 minutes using an automatic toothbrushing machine in 2% NaF solution by each type of toothbrush. The microhardness of the enamel surface of the specimen were measured after p-H cycling, and the fluoride ratio of the enamel surface was identified by scanning electron microscope observation and EDS analysis. The main analysis results are as follows:

1) The enamel microhardness of the microcurrent toothbrush group was higher after brushing (279.64) than before brushing (267.74) ($p=0.008$). That of the control group and the ordinary toothbrush group was slightly lower after brushing, but not a significant difference ($p<0.05$).

2) Scanning electron microscopic observations showed that the enamel surface after brushing with a microcurrent toothbrush appeared slightly irregular globular crystals, forming fluoroapatite.

3) According to EDS analysis of scanning electron microscope, the fluoride content of the enamel surface was 1.76% in the control group while 22.29% in the microcurrent toothbrush group.

To sum up the above results, toothbrushing using a microcurrent toothbrush has been found to be effective in preventing tooth decay by increasing the formation of fluoroapatite in the enamel. If the oral hygiene by brushing teeth can increase the effect of preventing tooth decay in daily life, the use of microcurrent toothbrushes is expected to greatly contribute to the reduction of the national medical expenses and the promotion of the oral health of the people.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

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