



Remineralization Effect of Bioactive glass Toothpaste Versus Sodium Fluoride Toothpaste on White Spot Lesions Around Orthodontic Brackets: An In Vitro Study

Vahid Mollabashi¹, Maryam Heydarpour^{2*}, Abbas Farmani³, Kimia Saadat⁴, Maryam Farhadian⁵

¹ Associate Professor, Department of Orthodontics, Dental Research Center, School of Dentistry, Hamadan University of Medical Sciences, Hamadan, Iran

² Postgraduate Student, Department of Orthodontics, School of Dentistry, Hamadan University of Medical Sciences, Hamadan, Iran

³ Assistant Professor, Dental Research Center and Dental Implant Research Center, Hamadan University of Medical Sciences, Hamadan, Iran

⁴ Dentist, School of Dentistry, Hamadan University of Medical Sciences, Hamadan, Iran

⁵ Associate Professor, Department of Biostatistics, School of Public Health and Research Center for Health Sciences, Hamadan University of Medical Sciences, Hamadan, Iran

***Corresponding author:** Maryam Heydarpour, Department of Orthodontics, School of Dentistry, Shahid Fahmideh Street, Hamadan City, Hamadan Province, Iran.

Email: maryamheydarpour97@gmail.com

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Abstract

Aim: Formation of white spot lesions (WSL) subsequent to orthodontic treatment is a cosmetic concern. This research assessed the effects of bioactive-glass toothpaste on remineralization of orthodontic white spot lesions.

Methods: Orthodontic brackets were bonded to seventy extracted premolars. Then they were submerged in a demineralization solution (pH=4.52) at 37°C for 96 hours, which led to the formation of artificial caries lesions on enamel. Occlusal brackets were assigned to the treatment group, whereas gingival brackets were assigned to the control group. Specimens were allocated to one of two groups: group 1) toothpaste containing sodium fluoride (1,450 ppm), group 2) toothpaste containing bioactive glass (5%). The samples underwent a 14-day remineralization/demineralization cycle consisting of a twice-daily 30-minute submersion in a demineralizing solution (pH=4.52, 37°C) followed by a five-minute toothpaste treatment for the occlusal of the brackets. Each sample was analyzed using a polarized microscope, and AutoCAD 2007 was used to quantify the mineralization depth and area. Using SPSS version 23, the independent and paired t-tests were used to assess data statistically.

Results: In terms of both remineralization area and depth, the study's findings revealed a statistically significant difference between the experimental and control groups ($p < 0.001$). Area and depth differences between the two remineralization methods in the treatment group were also statistically significant ($p < 0.001$).

Conclusion: Both sodium fluoride and bioactive-glass toothpaste are useful for remineralizing the WSL, while bioactive-glass toothpaste seems to work more effectively than sodium fluoride toothpaste.

Keywords: Bioactive glass, Orthodontics, Remineralization, Sodium fluoride.

1. Background

White spot lesions (WSL) are an adverse result of orthodontic treatments, caused by poor oral hygiene (1). Fixed orthodontic appliances prevent total oral hygiene, and reduce natural oral self-cleaning process causing accumulation of plaque and decreasing oral pH (2). Consequently, enamel

demineralization occurs resulting in WSL, which is explained as subsurface enamel porosity and is evident on the tooth surface because of its milky white opacity (3). WSL implies preliminary formation of enamel caries (4) which usually forms after four weeks of bonding (5). Studies indicated

different prevalence rates of WSLs in orthodontic patients as a result of various measurement techniques or criteria (5), which vary from 15-85% (6). The primary purpose of orthodontic therapy for patients is to produce an aesthetically attractive smile. Unfortunately, the presence of a WSL distracts from this objective (7). Consequently, this often occurring lesion makes a serious challenge for orthodontists (8). Studies have shown that remineralization may be the next significant advancement in the early treatment of caries lesions (9). Researchers and doctors are beginning to see the non-invasive remineralization therapy as a therapeutic option for WSL (6). Fluoride compounds are one of the first therapies for WSL, although additional remineralizing substances, such as casein phosphopeptide-amorphous calcium phosphate (10) and biomimetic hydroxyapatite (11), have also been suggested (11). However, fluoride's important role in remineralization cannot be understated, even at low quantities (12). Additionally, it has been shown that plaque calcium and phosphate levels are inversely related to enamel demineralization (12,13). In order to hasten the remineralization process, attempts are being undertaken to boost the concentrations of calcium, fluoride, and phosphate ions (8).

There are several ways to increase the phosphorous and calcium ions near the dental plaque. Recent research has shown the effectiveness of calcium sodium phosphosilicate bioactive glass, popularly known as NovaMin, in the treatment of WSL (4,14–16). NovaMin starts to exchange H⁺ ions with sodium (Na) particles when it is placed in an aqueous environment, which results in the release of phosphate and calcium from calcium sodium phosphosilicate particles. The sodium ions interaction with the hydrogen cations in pH is what causes the short spike. A calcium phosphate coating is created on the surface of the tooth thanks to NovaMin and saliva, and the sediments progressively change into hydroxyl-carbonate apatite. Chemically and structurally, this substance resembles biological hydroxyapatite (8). Additionally, the temporary pH increase brought by NovaMin helps to avoid demineralization (8). Due to NovaMin's ability to produce calcium and phosphorous ions in the creation of fluorapatite, a mixture of fluoride and NovaMin has also been proposed as synergic in the remineralization process (4,17). Toothpaste is commonly applied every day by the patients with fixed orthodontic appliances and at the same time, bioactive-glass has a proven function to improve remineralization. This study evaluated the outcomes of bioactive-

glass toothpaste on the remineralization of WSL around orthodontic brackets.

2. Methods

Intervention

This study was approved by the research ethics committee of Hamadan University of Medical Sciences (ID: IR.UMSHA REC1397.786). Seventy healthy premolars that were extracted for orthodontic therapy were immersed in saline. The surface plaques were rubbed for 15 seconds with a rubber cap and pumice powder. The point of contact between the bracket and the buccal surface of the tooth was then identified. The tooth surface was etched for fifteen seconds and then rinsed with water (30 seconds). Brackets (Dentaurem, Germany) were bonded (Relicane bond, India) to enamel and cured (Detroit Dental Manufacturing Co., USA). One operator bonded all the brackets. The teeth were fixed to a block of epoxy resin with their buccal sides exposed. Each tooth was immersed for four days in a demineralization solution (containing 2 mmol/L calcium chloride, 2 mmol/L trisodium phosphate, 75 mmol/L buffer acetate) with a pH of 4.6. They were then extracted from the demineralized solution, washed with deionized water, and divided into two groups (35 per group). The occlusal portion of the bracket was allocated to the treatment group, while the gingival portion was assigned to the control group. The teeth were then treated with fluoride and bioactive-glass according to the following protocol for 14 days: The teeth were soaked in demineralizing solution (pH=4.52, 37°C) for 30 minutes twice daily, followed by five minutes toothpaste treatment with an electronic toothbrush (Oral-B, Vitality Cross Action, USA). Group 1's occlusal brackets received 1450 ppm sodium fluoride paste, whereas group 2 received 5% bioactive glass paste (soaked in 1:3 slurries with Deionized water). In order to imitate oral conditions, samples were immersed in separate containers containing synthetic saliva (pH=7.00, 37°C) with the specified formula (NaCl 0.4, KCl 0.4, CaCl₂.2H₂O 0.795, NaHPO₄.2H₂O 0.78, Na₂S.2H₂O 0.005, urea 1.0, H₂O 1.0 g/L). After each treatment, the saliva solution was discarded and replaced. Each block was equipped with its own toothbrush to avoid cross-contamination. After two weeks, the first and second groups' premolars were removed from the artificial saliva, and their brackets were separated. Afterwards, a uniaxial cutting machine sliced them vertically (18). Each sample was sandpapered to a thickness of 50 m and examined using a polarized microscope (Dewinter

Technologies, Italy) at a magnification of 40X. The AutoCAD 2007 program was used to estimate the mineralization depth and area of all premolars on the treatment and control sides based on an image of each sample. Three samples from the first group and two samples from the second group were shattered and destroyed by the cutting equipment. During enamel sandpapering, three samples from the first group and four samples from the second group were damaged. All specimens that were destroyed were eliminated from the research, leaving 29 specimens in each group.

Statistical Analysis

The data were analyzed using SPSS software version 23.0 (SPSS Inc., Chicago, IL, USA). Using Shapiro Wilks's test, the data's normality was assessed. Student's t-test and paired sample t-test were used to compare the groups. The confidence interval was established at 95%, and $p < 0.05$ was considered statistically significant. For intra and inter-examiner reliability, the measurements of area and depth of remineralization in both methods were repeated twice by two observers.

3. Results

Two treatment approaches were used to remineralize the surface. The teeth were sectioned and studied using a polarized microscope to measure the depth and extent of lesion remineralization. Figure 1A to D shows photographs of specimens of the treatment and control groups.

Shapiro Wilks test was used to verify data's normality. In terms of intra-examiner (ICC= 0.99) and inter-examiner (0.98) agreement for both area and depth of remineralization, the measurement's

reliability was excellent. Repeated measurements have a proven capacity to boost accuracy while lowering examiner- or method- error (19).

Intergroup Comparison

In the treatment group, there was a statistically significant difference between the two procedures for the depth (p -value < 0.001) and area of remineralization (p -value < 0.001), as shown in Tables 1 and 3. However, there was no statistically significant difference between the two procedures in the control group for the depth (p -value=0.057) or area of remineralization (p -value=0.080) as shown in Tables 1 and 2.

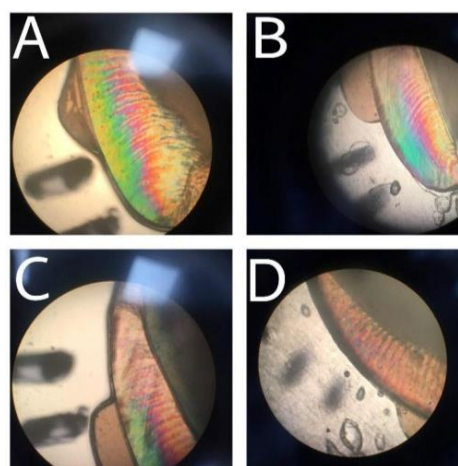


Figure 1. A tooth enamel 2 weeks after using fluoride toothpaste. B, tooth enamel 2 weeks after using bioactive glass toothpaste. C, The image of gingival part of bracket as a control in fluoride group. D, The image of gingival part of the bracket as a control in bioactive glass group

Table 1. Intra- and inter-group comparison of the area of remineralization

Method	Treatment group Mean \pm SD	Control group mean \pm SD	Difference mean \pm SE	Intragroup comparison P-value*
I	959251.75 \pm 33.89	109213.86 \pm 98.20	850037.89 \pm 669.57	<0.001
II	1763797.82 \pm 45.75	164337.13 \pm 13.48	1599460.69 \pm 85.15	<0.001
Mean difference \pm SE	-804546.06 \pm 10.12	-55123.27 \pm 30.68		
Intergroup comparison P-value**	<0.001	0.080		

*paired t-test

**t-test

Table 2. Intra-group and intergroup comparison the depth of remineralization

Method	Treatment group Mean \pm SD	Control group mean \pm SD	Difference mean \pm SE	Intragroup comparison P-value*
I	879.24 \pm 18.97	205.00 \pm 17.35	674.24 \pm 41.49	<0.001
II	1228.17 \pm 25.65	311.86 \pm 23.33	916.31 \pm 64.23	<0.001
meandifference \pm SE	-348.93 \pm 58.40	-106.86 \pm 54.87		
Intergroup comparison	<0.001	0.057		

P-value**

*paired t-test

**t-test

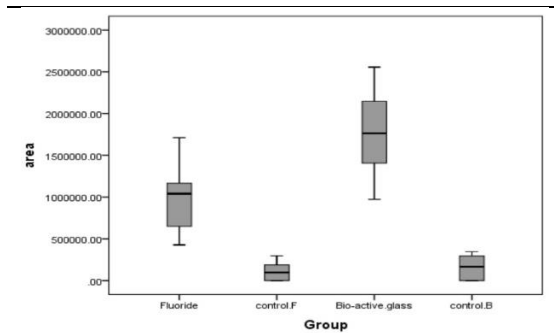


Figure 2. Box plot for remineralization area

Intra-group Comparison

The results also revealed significant differences in each of the two approaches for the remineralization area (p -value < 0.001) and depth (p -value < 0.001), as shown in Tables 1 and 2, respectively.

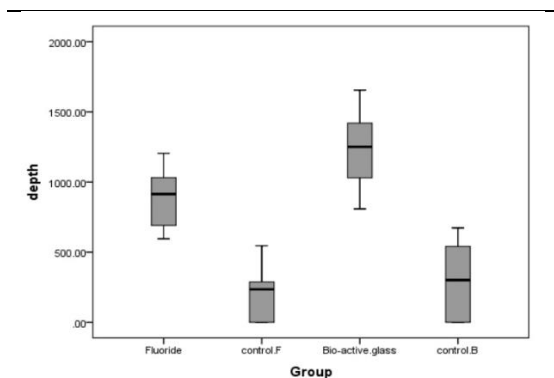


Figure 3. Box plot for remineralization depth

4. Discussion

In this research, the remineralization area and depth were significantly different between the treatment and control groups for each of the two procedures, showing that both approaches increased the WSL surrounding the orthodontic brackets. The study results showed significant differences in the area and depth of remineralization between the treatment groups in both methods, indicating that bioactive-glass toothpaste has a higher rate of remineralization than fluoride toothpaste.

Current *in vitro* research similar to ours indicated similar results. Consistent to our study, Taha et al. (20) systematically reviewed 11 laboratory-based studies to assess the effectiveness of bioactive-glass to promote enamel

remineralization. They found that bioactive glass, in addition to topical remineralization agents like fluoride and CPP-ACP, may improve enamel remineralization when employed in various compositions. The impact of topical NovaMin on tooth remineralization was compared with sodium fluoride in a research by Golpayegani et al. (15). According to the findings of this research, when compared to dentifrice that contained fluoride, NovaMin dentifrice seemed to have a stronger impact on the remineralization of carious-like lesions on permanent teeth.

Ggorgievska et al. (21) studied the remineralization abilities of different toothpaste compositions, including toothpaste containing bioactive-glass. Their study used energy dispersive X-ray and scanning electron microscopy (SEM) to show that the bioactive-glass toothpaste has a high efficiency in fostering enamel remineralization, similar to our study. However, in a recent systematic review by Khigmatgar et al. (14), their results indicated no significant clinical evidence to confirm the effectiveness of NovaMin in the remineralization of the WSL.

Consistent to our study, Gokce et al. (13) evaluated the effects of toothpastes with various compositions on remineralization of WSLs around orthodontic brackets. They concluded that the toothpaste containing NovaMin can effectively improve WSLs in comparison to the fluoride toothpaste and other types of toothpastes containing probiotics.

However, conflicting results were observed in a *in vitro* investigation conducted by Ballard et al. (22) on the Restore toothpastes (which contains bioactive-glass), MI Paste Plus, and previDent 5000. According to their findings, these products were not superior to the control group to resolve the WSL aesthetically. This difference could be contributed to measurement method of the enamel remineralization. They objectively measured the changes in the esthetic appearance of WSLs.

Our study's findings and those of Haghgo et al. (23), whose *in vitro* experiment examined the effectiveness of NovaMin and Nano-hydroxyapatite in remineralization of carious lesions of permanent teeth, differ in many important ways. They demonstrated that there was no statistically significant difference in the degree of surface micro hardness between NovaMin and Nano-hydroxyapatite dentifrices. Most *in vitro* studies

reported significant advantages to use NovaMin for remineralizations of WSLs(13,15,20,21); however, clinical studies on orthodontic patients (24) have not observed any clinically or statistically significant differences. This inconsistency could be because of the differences in the oral cavity environment in clinical studies including the effects of saliva, plaques, tooth movement, mastication and so forth, which could wash away the NovaMin toothpaste before it could have an effect on the lesions or because of the less invasive methods of analyzing WSL used by the clinical studies (DIAGNOdent, etc.).

Utilizing remineralizing agents is the major method for preventing the development of WSLs (2). Topical fluoride is often the primary treatment approach, followed by fluoride compositions of variable strengths (such as bonding materials, sealants, gels, mouth rinses, various types of toothpastes, and varnishes) (25,26). There are, however, alternatives to fluoride-based toothpastes, such casein phosphopeptide-amorphous calcium phosphate (CPP-ACP) and bioactive-glass toothpaste (27,28). On the other hand, antimicrobial pastes, mouthwash solutions, and varnishes containing chlorhexidine have been included into a number of therapy modalities in an effort to reduce biofilm. However, the ability of chlorhexidine to inhibit and manage caries via its antibacterial activity is controversial, since the data is still inconclusive, and therapy may be necessary for advanced lesions (5).

The demineralization and remineralization of enamel have been studied using a variety of direct and indirect techniques, including as abrasion, scraping, indentation, atomic force microscopy, and microhardness (29). In the current experiment, remineralization of enamel lesions was evaluated using a polarized microscope. Indentation and scratching tests can only evaluate tooth's surface, therefore they cannot accurately measure caries, remineralization, or demineralization. Microhardness is measured by cutting a specimen with a diamond blade (30). Most of microhardness is determined by experimental conditions in addition to demineralization and remineralization. However, it has been shown that other factors may affect microhardness as well. Since the arrangement of enamel crystals and their mineral content are closely related to microhardness, this physical criterion may be used to assess the degree to which teeth are mineralized (31). Demineralization and remineralization may be evaluated most effectively using a polarized microscope (32).

The contribution of fluoride to prevent tooth

decalcification is well documented (33–35). In our study, both types of toothpastes showed a considerable increase in remineralization of WSL around orthodontic brackets, and bioactive-glass toothpaste has a higher rate of remineralization than fluoride toothpaste. However, more evidence is needed to elucidate the effects of bioactive-glass on WSLs in orthodontic patients.

Conclusion

Both fluoride and bioactive-glass toothpastes were successful in remineralization of the WSLs; however, bioactive-glass toothpaste seems to have a stronger impact on remineralizing the WSL on permanent teeth.

Conflict of interest

The authors have no conflict of interest to declare.

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