Remineralization of artificial carious lesions using a novel fluoride incorporated bioactive glass dentifrice

Remineralizacja preparowanych in vitro ubytków próchnicowych środkiem zawierającym nowe bioaktywne szkło wzbogacone fluorkami

Abdulmajeed Mohammed Alhussain^{1,A–D,F}, Abdulhakeem Ali Alhaddad^{1,A–D,F}, Mahdi Mohammed Ghazwi^{1,A–D,F}, Imran Farooq^{2,A–F}

¹ Dental Intern, College of Dentistry, Imam Abdulrahman Bin Faisal University, Dammam, Saudi Arabia

² Department of Biomedical Dental Sciences, College of Dentistry, Imam Abdulrahman Bin Faisal University, Dammam, Saudi Arabia

A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation;

D – writing the article; E – critical revision of the article; F – final approval of the article

Dental and Medical Problems, ISSN 1644-387X (print), ISSN 2300-9020 (online)

Dent Med Probl. 2018;55(4):379–382

Address for correspondence

Imran Farooq

E-mail: drimranfarooq@gmail.com

Funding sources

None declared

Conflict of interest

None declared

Received on June 13, 2018

Reviewed on September 17, 2018

Accepted on October 4, 2018

Published online on November 23, 2018

Cite as

Alhussain A, Alhaddad A, Ghazwi M, Farooq I. Remineralization of artificial carious lesions using novel fluoride-incorporated bioactive glass dentifrice [published online as ahead of print on November 23, 2018]. *Dent Med Probl.* 2018;55(4):379–382. doi:10.17219/dmp/97311

DOI

10.17219/dmp/97311

Copyright

© 2018 by Wrocław Medical University
and Polish Dental Society

This is an article distributed under the terms of the
Creative Commons Attribution Non-Commercial License
(<http://creativecommons.org/licenses/by-nc-nd/4.0/>)

Abstract

Background. Remineralization potential of dentifrices with novel compositions that can restore minerals back into incipient carious lesions has not been extensively studied so far.

Objectives. The aim of this study was to assess the efficacy of a dentifrice based on novel fluoride incorporated bioactive glass in remineralizing artificial carious lesions in human enamel, and compare it with a standard fluoride-containing dentifrice.

Material and methods. Twenty-four human extracted teeth were sectioned at the cemento-enamel junction to obtain enamel blocks. These blocks ($n = 24$) were randomly divided into 3 groups, with each group containing 8 specimens: group 1 (negative control group; distilled water), group 2 (positive control group; fluoride toothpaste) and group 3 (test group; BioMin™ F toothpaste). Artificial carious lesions were produced in the enamel surfaces by exposing them to a demineralization solution (6% citric acid, pH 2.2) for 96 h. After demineralization, the specimens were brushed with manual toothbrushes in a toothbrush simulation machine (each sample received 800 strokes). For brushing the specimens from group 1, 20 mL of distilled water was used, for group 2 – 20 mL of slurry of toothpaste mixed with artificial saliva, and for group 3 – 20 mL of slurry of toothpaste (BioMin F) mixed with artificial saliva. The micro-hardness data (VHN – Vickers hardness number) was collected at baseline (sound enamel), post-demineralization and post-remineralization.

Results. The biggest difference between the post-remineralization and post-demineralization values was observed in group 3 (mean VHN = 118.73), followed by group 2 (mean VHN = 60.54) and group 1 (mean VHN = 47.44). All the groups revealed significant differences ($p < 0.05$) when the post-demineralization and post-remineralization values were compared to baseline values within each group.

Conclusions. The BioMin F group outperformed the other 2 groups in terms of remineralizing the demineralized enamel structure.

Key words: enamel, remineralization, fluoride bioactive glass

Słowa kluczowe: szkliwo, remineralizacja, bioaktywne szkło wzbogacone fluorkami

Introduction

Dental caries initiates with demineralization of the tooth structure caused by the acids produced by the cariogenic bacteria residing in dental plaque, after they ferment dietary carbohydrates.¹ In the past, dental caries was considered as an irreversible infectious bacterial disease, but recently, it has been established that caries is a complex multifactorial disease, which occurs when the equilibrium between demineralization and remineralization is disturbed.² A decrease in pH can cause a net loss of minerals from the tooth surface, also known as demineralization, whereas the process of restoring lost mineral ions to the tooth structure (which usually occurs at high pH) and strengthening the lattice network is known as remineralization.³

The most effective method described in the literature for the removal of plaque is mechanical tooth brushing with toothpaste.⁴ In 1969 there was a revolution in tissue engineering, when Prof. Larry Hench introduced bioactive glass (BG) (sodium calcium phosphosilicate glass), as it was the first material that exhibited exceptional bone bonding ability.^{5,6} The use of BG in dentistry has been encouraged recently due to its compositional resemblance to bone and dental enamel.⁷ The other reason for its recent extensive use is that it forms hydroxylapatite (HAP), which repairs the structure of bone and enamel, and also makes them resilient.⁸ The traditional BG composition (Bioglass®) is deficient in fluoride.⁷ Fluoride is an essential ion that helps in the formation of fluorapatite (FAP), which is more resistant to caries and acid encounters as compared to HAP.⁹ Recently, a new toothpaste consisting of fluoride incorporated BG has been introduced, and claims have been made that it causes sustained release of calcium, phosphate and fluoride ions, thus resulting in enhanced remineralization of the tooth structure.¹⁰

Therefore, the aim of this study was to assess the efficacy of a dentifrice based on novel fluoride incorporated BG in remineralizing artificial carious lesions in human enamel, and compare it with a standard fluoride-containing dentifrice.

Materials and methods

Ethical approval (Ref. No. 2018001) was obtained from the Scientific Research Unit of the institution (College of Dentistry, Imam Abdulrahman Bin Faisal University, Dammam, Saudi Arabia) and all the ethical protocols were strictly followed.

Twenty-four extracted human teeth were obtained from the Department of Oral Surgery of the College of Dentistry at Imam Abdulrahman Bin Faisal University, Dammam, Saudi Arabia. Teeth which were free from caries, obvious white spot lesions, restorations,

or other defects were included in this study. All the teeth were sectioned at the cemento-enamel junction using a water-cooled diamond saw (Isomet® 5000 Linear Precision Saw; Buehler Ltd., Lake Bluff, USA) and their roots were discarded. The crowns of all the teeth were then embedded in self-cure acrylic resin in such a manner that the buccal/labial surface of tooth enamel was exposed. The enamel surfaces to be treated were then polished with 600-grit wet silicon-carbide paper. An enamel window of approx. 4 × 4 mm was created on the exposed enamel surface by a nail varnish to ensure that the analysis would be performed only in that particular area.

Artificial saliva preparation and grouping of specimens

Artificial saliva (AS) was prepared by mixing NaCl (0.400 g), KCl (0.400 g), NaH₂PO₄ · H₂O (0.690 g), CaCl₂ · H₂O (0.795 g), and Na₂S · 9H₂O (0.005 g) in 1000 mL of deionized water, as proposed by Fusayama et al.¹¹ The pH of freshly prepared AS was 5.5, which was adjusted by adding 1 M of sodium hydroxide (NaOH) until neutral pH of 7.0 was achieved. The enamel blocks (n = 24) were randomly divided into 3 groups, with each group containing 8 specimens: group 1 (negative control group; distilled water), group 2 (positive control group; fluoride toothpaste) and group 3 (test group; BioMin™ F toothpaste (BioMin Technologies Ltd., London, UK)). The details of active and other ingredients of the tested toothpastes are shown in Table 1.

Artificial carious lesions

Artificial carious lesions were produced in the enamel surfaces by exposing them to a demineralization solution (6% citric acid, pH = 2.2) for 96 h, as described earlier by Wang et al.¹² The demineralizing solution was changed every day during this 96-hour period. The specimens were then washed with distilled water and air-dried for a day.

Remineralization procedure

The specimens were brushed with manual toothbrushes from the same manufacturer (TRISA AG, Triengen, Switzerland). For brushing specimens from group 1, 20 mL of distilled water was used, for group 2 – 20 mL of slurry of toothpaste (Colgate®; Colgate-Palmolive Arabia Ltd., Dammam, Saudi Arabia) mixed with AS in a ratio of 1:3, and for group 3 – 20 mL of slurry of toothpaste (BioMin F) mixed with AS. The brushing experiments were carried out inside a toothbrush simulation machine (toothbrush simulator, model ZM-3.8; SD Mechatronik GmbH, Feldkirchen-

Table 1. Composition of the tested toothpastes

Toothpaste	Manufacturer	Composition	Active ingredient
Colgate	Colgate-Palmolive Arabia Ltd., Dammam, Saudi Arabia	dicalcium phosphate dihydrate, sodium lauryl sulfate, sodium saccharin, tetrasodium pyrophosphate, cellulose gum, glycerin, water, flavor	sodium monofluorophosphate 1.1% w/w fluoride content: 1450 ppm F
BioMin F	BioMin Technologies Ltd., London, UK	fluoro calcium silicate, sodium lauryl sulfate, titanium dioxide, acesulfame potassium, carbomer, polyethylene glycol 400 (PEG 400), glycerin, silica, flavor, fluoride <600 ppm	BioMin F (fluoro calcium phosphosilicate) fluoride content: <600 ppm F

Westerham, Germany), under a continuous loading of 250 g, with 100 strokes/min for 4 days (each sample received 800 strokes, which is equivalent to 2 weeks of in vivo brushing). After every brushing cycle, all the specimens were thoroughly washed with distilled water. After the 4th day of brushing, the specimens were washed with distilled water and left to air-dry, prior to the surface micro-hardness analysis.

Surface micro-hardness analysis

Eight specimens from each group were used to evaluate the changes in the surface micro-hardness values. The Vickers surface hardness was measured using a digital micro-hardness tester (FM-ARS 9000; Future-Tech Corp, Kawasaki, Japan). Five indents were made on the polished surface of each specimen using a Vickers diamond indenter under a load of 100 g, applied for 10 s. An average of 5 indents was used for the analysis. The micro-hardness data (VHN – Vickers hardness number) was collected at baseline (sound enamel), post-demineralization and post-remineralization.

Statistical analysis

The results were analyzed statistically using the Kruskal–Wallis test, which was applied to compare the 3 groups. For comparison within each group of the post-demineralization and post-remineralization vs baseline values, the Wilcoxon signed-rank test was used. A *p*-value ≤0.05 was considered statistically significant.

Results

The surface micro-hardness analysis was performed by means of a Vickers tester (Fig. 1). The biggest difference between the post-remineralization and post-demineralization values was observed in group 3 (mean VHN = 118.73), followed by group 2 (mean VHN = 60.54) and group 1 (mean VHN = 47.44) (Table 2). For all the groups, 800 strokes were not enough to restore the structure completely back to the baseline values; still, the BioMin F group outperformed the other 2 groups in terms of remineralizing the demineralized enamel structure. All the groups revealed significant differences (*p* < 0.05) when the post-demineralization and post-remineralization values were compared to baseline values within each group (Table 2).

Fig. 1. Vickers indentation performed to assess the micro-hardness of the enamel surface

Table 2. Mean micro-hardness values (VHN – Vickers hardness number) for the 3 tested groups at baseline, post-demineralization and post-remineralization

Analysis time	Group 1 (distilled water)	Group 2 (fluoride toothpaste)	Group 3 (BioMin F toothpaste)	<i>p</i> -value
Baseline	569.98 ±29.41	482.04 ±66.13	497.56 ±71.28	0.015
Post-demin	122.75 ±56.03 ^a	209.50 ±151.22 ^a	164.93 ±171.20 ^a	0.456
Post-remin	170.19 ±73.69 ^a	270.04 ±136.67 ^a	283.66 ±161.55 ^a	0.184
Difference (post-remin – post-demin)	47.44	60.54	118.73	–
<i>p</i> -value	0.002	0.005	0.002	–

Data presented as mean ± standard deviation (SD); post-demin – post-demineralization; post-remin – post-remineralization; ^a significant difference of mean of the post-demin and post-remin values as compared to the baseline value within each group.

Discussion

In this study, the BioMin F toothpaste showed more remineralizing capability as compared to the fluoride-based toothpaste and distilled water, according to the surface micro-hardness analysis. The better remineralization potential of BioMin F could be ascribed to the difference in its composition with regard to a standard fluoride toothpaste. Conventionally, BG is composed of calcium sodium phosphosilicate and contains no fluoride.⁷ The presence of fluoride in toothpastes ensures enhanced remineralization and helps in the prevention of caries, as shown by the previous literature.^{13,14} The BioMin F toothpaste contains high-phosphate BG with fluoride within its BG composition.¹⁰ When BG is placed in the oral cavity, ionic exchange reactions take place and the glass begins to dissolve, releasing calcium (Ca^{2+}) and phosphate (PO_4^{3-}) ions,¹⁵ resulting in the formation of FAP, which is more acid-resistant and is quite desirable for various dental applications. On the other hand, fluoride from a regular toothpaste can be washed quickly by the salivary flow and the amount of FAP thus formed is also questionable.¹⁶ The high phosphate content of a BG toothpaste is also useful, as it helps to maintain the network connectivity of the glass and ensures the formation of FAP, as shown previously by Brauer et al.¹⁷

The literature lacks studies which have analyzed and compared the remineralization potential of BG toothpastes and fluoride toothpastes. A recent in vitro study reported more tubule occlusion on dentin specimens achieved by BioMin F as compared to Novamin™ (Sensodyne Repair®, Group Pharmaceuticals Ltd., Mumbai, India) and a standard fluoride toothpaste.¹⁸ Our results also demonstrated a better performance of BioMin F toothpaste, but in terms of enamel remineralization. To achieve the maximum benefit of fluoride, it should be deposited and released slowly.¹⁹ Farooq et al. previously developed the composition of fluoride-containing BG that formed apatite in Tris buffer solution within 6 h, which was much faster than in the case of 45S5 (Bioglass; 24 h).²⁰ Therefore, the superior remineralization potential of BioMin F could be attributed to the presence of fluoride, in addition to BG, in its composition.

One limitation of our study is its in vitro nature. Actual in vivo conditions could be different and may offer more dynamic challenges to the tested materials. To the best of the authors' knowledge, this study is the first to compare the remineralization potential of BioMin F and fluoride in a head-to-head trial. This study could prove useful and serve as a basis for future quantitative and clinical studies, which can be performed to analyze the effects of these dentifrices under more vigorous in vivo conditions.

Conclusions

The BioMin F group outperformed the other 2 groups in terms of remineralizing the demineralized enamel structure. Future in vivo studies are recommended to evaluate its clinical efficacy.

References

1. Featherstone JD. Dental caries: A dynamic process. *Aust Dent J*. 2008;53:286–291.
2. Kawasaki K, Ruben J, Tsuda H, Huysmans MC, Takagi O. Relationship between mineral distributions in dentine lesions and subsequent remineralization in vitro. *Caries Res*. 2000;34:395–403.
3. González-Cabezas C. The chemistry of caries: Remineralization and demineralization events with direct clinical relevance. *Dent Clin North Am*. 2010;54:469–478.
4. Collins WJN, Walsh TF, Figures KH. *A Handbook for Dental Hygienists*. 4th ed. Oxford, UK: Butterworth-Heinemann. 1998:272–273.
5. Jones JR. Review of bioactive glass: From Hench to hybrids. *Acta Biomater*. 2013;9:4457–4486.
6. Krishnan V, Lakshmi T. Bioglass: A novel biocompatible innovation. *J Adv Pharm Technol Res*. 2013;4:78–83.
7. Farooq I, Imran Z, Farooq U, Leghari A, Ali H. Bioactive glass: A material for the future. *World J Dent*. 2012;3:199–201.
8. Pradeep K, Rao PK. Remineralizing agents in the non-invasive treatment of early carious lesions. *Int J Dent Case Rep*. 2011;1:73–84.
9. Mohammed RN, Lynch RJ, Anderson P. Effects of fluoride concentration on enamel demineralization kinetics in vitro. *J Dent*. 2014;42:613–618.
10. <https://www.biomin.co.uk/products/biointm-f-toothpaste>. Accessed May 10, 2018.
11. Fusayama T, Katayori T, Nomoto S. Corrosion of gold and amalgam placed in contact with each other. *J Dent Res*. 1963;42:1183–1197.
12. Wang Y, Mei L, Gong L, et al. Remineralization of early enamel caries lesions using different bioactive elements containing toothpastes: An in vitro study. *Technol Health Care*. 2016;24:701–711.
13. Somaraj V, Shenoy RP, Shenoy Panchmal G, Kumar V, Jodalli PS, Sonde L. Effect of herbal and fluoride mouth rinses on *Streptococcus mutans* and dental caries among 12–15-year-old school children: A randomized controlled trial. *Int J Dent*. 2017;2017:5654373.
14. Clark MB, Slayton RL; Section on Oral Health. Fluoride use in caries prevention in the primary care setting. *Pediatrics*. 2014;134:626–633.
15. Khalid MD, Khurshid Z, Zafar MS, Farooq I, Khan RS, Najmi A. Bioactive glasses and their applications in dentistry. *J Pak Dent Assoc*. 2017;26:32–38.
16. Aasenden R, Brudevold F, Richardson B. Clearance of fluoride from the mouth after topical treatment or the use of a fluoride mouthrinse. *Arch Oral Biol*. 1968;13:625–636.
17. Brauer DS, Karpukhina N, O'Donnell MD, Law RV, Hill RG. Fluoride-containing bioactive glasses: Effect of glass design and structure on degradation, pH and apatite formation in simulated body fluid. *Acta Biomater*. 2010;6:3275–3282.
18. Shaikh K, Pereira R, Gillam DG, Phad S. Comparative evaluation of desensitizing dentifrices containing BioMin®, Novamin® and fluoride on dentinal tubule occlusion before and after a citric acid challenge – a scanning electron microscope in-vitro study. *J Odontol*. 2018;2:1000105.
19. ten Cate JM. Current concepts on the theories of the mechanism of action of fluoride. *Acta Odontol Scand*. 1999;57:325–329.
20. Farooq I, Tylkowski M, Müller S, Janicki T, Brauer DS, Hill RG. Influence of sodium content on the properties of bioactive glasses for use in air abrasion. *Biomed Mater*. 2013;8:065008.