

Evaluating the comparative efficacy of Bioactive glass containing Toothpaste, Amine fluoride-containing toothpaste and CPP-ACPF on remineralization of Artificial Incipient Enamel Lesion

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ABSTRACT:

Objectives: The purpose of this in-vitro study was to evaluate the comparative efficacy of Bioactive glass containing toothpaste (BioMin) and Amine fluoride-containing toothpaste (Amflor), and CPP-ACPF (GC Tooth mousse plus) on remineralization of artificial incipient enamel lesion and evaluated using Vickers micro-hardness test. **Materials and Methods:** A total of fifty freshly extracted premolar teeth were collected and decoronated at 1mm below the CEJ. An adhesive tape of size 4x4 mm is placed on the labial surface of each specimen. Following this, the specimens were mounted in acrylic resin moulds such that only the adhesive tape was exposed, and on the removal of the tape, a window of 4x4 mm is created. Then the specimens were randomly divided into five groups, among which specimens of four groups were subjected to demineralization for four days at 37⁰c using an incubator. Group A (sound enamel), Group B (BioMin), Group C (CPP-ACPF), Group D (Amflor), Group E (demineralized lesion). The specimens of groups B,C,D were subjected to pH cycling for 10 days. Following which micro-hardness, all the specimens were evaluated using Vickers Micro-hardness Test. **Statistical analysis:** The analysis was done using One Way ANOVA and inter-group comparison was done using a post hoc test. **Results:** Results showed statistically significant differences (p<0.001) between groups B,C,D, and group E. Among the interventional groups even though there was no significant difference, group B (BioMin) showed higher micro-hardness values when compared to group C (CPP-ACPF) and group D (Amf). **Conclusion:** Under the limitation of this study, it can be concluded that Bioactive glass-based toothpaste (BioMin) is a novel technology that can be used for the remineralization of early enamel caries.

KEYWORDS: Remineralization, fluoride, enamel lesions, bioglass tooth paste.

INTRODUCTION:

Active research is going on to explore the scope for the remineralizing incipient carious lesion.¹ Initial caries can remineralize with the use of remineralization agents like fluoride, CCP-ACP, Bioglass.²

CCP-ACP toothpaste (GC Tooth Mousse plus) cream is being widely used as a remineralization agent. The CPP-ACP helps in the localization of amorphous calcium phosphate (ACP) at the tooth surface which helps to maintain a state of supersaturation. With the increased availability of calcium and phosphate ions, remineralization of the incipient lesion is promoted.

Bioactive glass is a unique material that enhances the remineralization process. The main component is amorphous calcium sodium phosphosilicate. This helps to release calcium, sodium, and phosphate ions in an aqueous stage which brings about

the process of remineralization.²

Amine Fluoride has greater remineralization potential due to its antiplaque effect and its ability to release fluoride ions.³

The pH cycling system has been used in the past in many studies to assess alteration in mineral changes in initial lesions.⁴ Even though there are other methods to assess remineralization, surface micro-hardness is the simplest, quick, and economical and requires a tiny area of specimen for testing.⁵

Hence this in-vitro study aims to evaluate the comparative efficacy of Bioactive glass containing toothpaste (BioMin) and Amine fluoride-containing toothpaste (Amflor), and CPP-ACPF (GC Tooth mousse plus) on remineralization of artificial incipient enamel lesion and evaluated using Vickers Micro-hardness Test.

METHODOLOGY:

Sample collection:

The study was conducted in Bapuji dental college, India for one year. Ethical clearance was obtained from the Institutional ethical committee (BDC/505/2014-2015). Fifty extracted human premolars which are extracted for orthodontic reasons were taken for the study. The teeth were free from any defects or cracks in the enamel. The teeth were cleaned of visible blood and gross debris and were maintained in a hydrated state during storage.

Sample preparation:

De-coronation of the teeth was done 1 mm below the CEJ using a diamond disc. The sample window preparation was made (4 mm × 4mm) using adhesive tape and coating of nail varnish applied to protect from acid attack. After drying, teeth were mounted on an acrylic block and the adhesive tape was removed using explorer resulting in a rectangular area on the enamel surface. Following which surface polishing was done in a sequential manner using 600 and 1200 grit sandpaper.

Randomization of Groups:

A total of fifty enamel samples were randomly divided into 5 groups. Group A - Sound enamel (No treatment- Positive control), Group B - Demineralized and treated with BioMintoothpaste, Group C - Demineralized and treated with Amflor toothpaste, Group D - Demineralized and treated with GC tooth mousse plus, Group E - Negative control group (demineralized but not treated with any solution).

Toothpaste solution preparation:

The supernatant solution was prepared in 1:3 dilutions (12g of each dentifrice in 36ml distilled water). Before starting the pH cycle, the suspension was stirred thoroughly every time.

Demineralizing and remineralizing phase:

The demineralizing solution and the remineralizing solution were prepared using the same method as per previous studies.⁶ To induce artificial caries formation each sample was then immersed in 10ml of demineralizing solution for 4 days in an incubator at a constant temperature of 37°C. All the samples were exposed to pH cycling to mimic the process of demineralization and remineralization that occurs in the oral cavity. The protocol for the pH cycle was similar to previous studies.^{6,7} The Surface Micro-hardness was evaluated using a Vickers hardness testing machine. The indentations produced by the load were captured digitally and micro-hardness values were calculated.

Statistical Analysis:

All the samples were measured for surface micro-hardness and the mean for each group was determined. The statistical analysis was done using one-way ANOVA and Tukey's post hoc test.

RESULTS:

The mean values of all the groups are represented in graphical and tabular form. (Table 1)

- The mean value of group A (Sound enamel) has a significant difference when compared with mean values of group B (BioMin), C (Amflor), D (CPP-ACPF), E (Demineralized enamel) p-value <0.0001. (Table 2)
- The mean values of Group B (BioMin), Group C (Amflor), Group D (CPP-ACPF) did not show any significant difference when compared. (Table 2)
- The mean values of Group A (Sound enamel), Group B (BioMin), Group C (Amflor), Group D (CPP-ACPF) showed significant differences when compared to the mean value of Group E (Demineralized enamel) p-value <0.0001. (Table 2)

Box whisker's graph showing confidence intervals and standard deviation

Bar graph showing the comparison between mean surface micro-hardness values of the interventional group.

Table 1. Comparison of mean intervention surface micro-hardness values of enamel samples between interventional groups using one-way analysis of variance.

	Sound enamel	BIOMIN	AMFLOR	CPP_ACPF	Demineralized enamel
SMH (Mean±SD)	351.26±11.38	333.42±7.15	324.72±9.08	331.90±10.67	272.26±4.23
One-way ANOVA	117.70				

F value	
Probability value	<0.0001**

** statistically highly significant at $p < 0.01$

Table 2. Results of Tukey's HSD post hoc analysis using one-way Analysis of variance to find a significant difference in mean post-intervention surface micro-hardness of enamel samples between the interventional groups.

Variable	Interventional groups	Interventional groups	Probability value
Post Intervention Surface micro-hardness values (PISMH)	Sound enamel	BIOMIN	0.0001**
		AMFLOR	0.0001**
		CPP-ACPF	0.000*
		DL	0.000*
	BIOMIN	AMFLOR	0.33
		CPP-ACPF	1.00
		DL	<0.0001**
	AMFLOR	BIOMIN	0.3
		CPP-ACPF	0.7
		DL	<0.0001**
	CPP-ACPF	BIOMIN	1.0
		AMFLOR	0.7
Demineralized enamel		<0.0001**	

*statistically significant at $p < 0.05$, ** statistically highly significant at $p < 0.01$

DISCUSSION:

Dental caries is one of the most common diseases which destroys tooth substances which include enamel, dentine.^{8,9} Minimal invasive dentistry and prevention of carious lesions are the main objectives of dental health in the modern era.^{10,11} Early enamel lesions can remineralize at early stages but in the advanced phase can lead to pulpitis and tooth loss.¹³ The anti-cariogenic effect of fluoride due to its presence in saliva can enhance remineralization but additional agents like CCP-ACP are mostly recommended.^{14,15} Therefore in the present study, the remineralizing potential of newer agents was evaluated.

CCP-ACP toothpaste with 900ppm fluoride (GC Tooth Mousse plus) cream is being widely used as a remineralization agent. Various studies have proved its effectiveness in enamel remineralization.¹⁶⁻¹⁸

The bioactive glass has been incorporated in the toothpaste for utilizing its unique biomimetic properties for remineralization.^{19,20} This novel material stimulates the remineralization of incipient lesions by increasing the bioavailability of free calcium and phosphate ions. This brings about the formation of hydroxylapatite crystals.²¹⁻²⁴

Due to the limited literature available on the remineralization efficacy of BioMin (Bioactive glass), the present study was conducted to assess and compare the remineralizing effect of CPP-ACPF (GC Tooth Mousse plus), BIOMIN (Bioactive glass), and AMFLOR (Amine fluoride) on artificially induced enamel lesions using surface micro-hardness analysis.

These findings are similar to other studies where significant differences were observed for surface micro-hardness (SMH) of enamel samples after treating them with remineralizing agents like CPP-ACPF (GC Tooth Mousse plus), BIOMIN (Bioactive glass), and AMFLOR (Amine fluoride) when compared to surface micro-hardness of demineralized enamel lesion.²⁵⁻²⁸

In the present study, the surface micro-hardness values of Group B (BioMin), Group C (Amflor), and Group D (CPP-ACPF) showed higher values when compared with Group E (Demineralized lesion). When compared among the interventional groups, Group B showed higher values when compared with Group C and D. [Bharat](#) et al conducted a study that showed that Bioactive glass has better remineralizing potential when compared to CPP-ACPF. This is because bioactive glass is a biomimetic agent which brings about a remineralization process similar to the human body resulting in restoration of tooth structure.^{29,30} It also forms hydroxyl carbonate apatite (HCA) crystals directly, without the intermediary Amorphous calcium phosphate which increases its remineralizing potential.^{31,32}

In the present study, it was compared and there was no significant difference among all the three remineralizing agents which was in accordance with the study done by Narayana et al.²

In the present study, there was a significant difference between the sound enamel group and the interventional groups which are contrary to the study done by Shetty et al. in which there was no significant difference between the sound enamel group and the interventional groups.¹² Thus further studies are required to establish the role of Bioactive glass as a remineralizing agent.

CONCLUSION:

Under the limitations of this study, it can be concluded that Bioactive glass-based toothpaste (BioMin) is a novel technology that can be used for the remineralization of early enamel caries. The Vicker's micro-hardness values produced by BioMin were highest followed by CPP-ACPF and AMFLOR.

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