

Research Article

Effect of Fluoride Varnish on Enamel Demineralization around Orthodontics Brackets *In vitro*

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- Prevention

Abstract

Purpose: The purpose of this *in vitro* study is to evaluate the efficacy of fluoride varnish in preventing enamel demineralization lesions adjacent to orthodontic brackets.

Methods: Brackets were bonded to 60 extracted human premolars with traditional composite resin and resin modified glass ionomer cement (Both without fluoride) and 15 teeth were randomly assigned to four equal test groups. Demineralization of enamel was evaluated in longitudinal buccolingual tooth sections using polarized light microscopy.

Results: ANOVA ($P < 0.05$) indicated significant differences in depth and area of demineralized enamel in all the groups. Those teeth treated with fluoride varnish exhibited 50% less demineralization than the control teeth in both the composite and the resin modified glass ionomer cement groups.

Conclusion: Fluoride varnishes should be considered for use as a preventive adjunct to reduce enamel demineralization adjacent to orthodontic brackets, particularly in patients who exhibit poor compliance with oral hygiene and home fluoride use.

INTRODUCTION

Tooth enamel is the hardest and most highly mineralized substance of the body [1], and with dentin cementum, and dental pulp is one of the four major tissues, which make up the tooth. It is the normally visible dental tissue of a tooth and must be supported by underlying dentin. Ninety-six percent of enamel consists of mineral; with water and organic material composing the rest of the structure.

Enamel's primary mineral is hydroxyapatite, which is a crystalline calcium phosphate [2,3]. The large amount of minerals in enamel accounts not only for its strength but also for its brittleness.

Demineralization is the process of removing minerals, in the form of mineral ions, from dental enamel. A substantial number of mineral ions can be removed from the hydroxyapatite lattice work without destroying its structural integrity.

Remineralization is the process of restoring minerals - again, in the form of mineral ions - to the hydroxyapatite's lattice work structure. Both remineralization and demineralization occur on the surface of the tooth.

Caries occurs only when dental plaque accumulates over the pellicle and is exposed to dietary fermentable carbohydrates. When microorganisms in the plaque lower its pH to a critical

point, the mineral phase of enamel (hydroxyapatite) begins to dissolve and diffuse outward into the acidic plaque that is under saturated with hydroxyapatite. The remineralization process is greatly enhanced by low levels of fluoride in saliva or plaque. First stage of caries involves dissolution of the enamel surface. As demineralization proceeds into the subsurface, mineral loss in the subsurface becomes greater than at the surface. White spot lesions contain subsurface demineralization that alters the color of the enamel. Clinical evidence indicates that fluoride is most effective in inhibiting the beginning of caries and less effective in inhibiting the progression of a lesion. Successful preventive treatments involve strengthening the enamel surface with fluoride and blocking the diffusion of mineral ions from the enamel surface with sealants. Fluoride varnishes also leave deposits of calcium fluoride on the enamel surface that contributes to the formation of fluorapatite in the enamel [4].

BACKGROUND

Enamel demineralization remains common negative sequelae of orthodontic treatment in the absence of proper oral hygiene. Orthodontic treatment with fixed appliances predisposes patients to large accumulations of bacterial plaque and, therefore, to demineralized enamel "white spot" lesions. Considering the mechanical difficulties of removing plaque with orthodontic brackets in place, compliance with proper oral hygiene is critical.

The incidence of enamel demineralization and caries during orthodontic care is increasing. Gorelick et al., (1982) found white spot lesions for nearly 50% of patients who underwent orthodontic treatment [5]. These white spot lesions are due to demineralization of the enamel by organic acids produced by cariogenic bacteria [6]. The presence of white spot lesions after removal of orthodontic appliances is a discouraging finding to a specialty whose goal is to improve facial and dental esthetics. Although orthodontists have long recognized this problem and have attempted to prevent it, demineralization continues to be a problem. Smales reported that plaque more readily accumulates on resin bonded materials than on enamel [4]. Gwinnett and Ceen found a significant increase in plaque on the gingival side of bonded brackets [7]. In addition, Ogaard, et al., (1998) reported that these lesions could develop in 4 weeks time or the average time between orthodontic visits [8].

Orthodontists have long attempted to reduce demineralization with limited success. Research efforts in cariology have demonstrated the beneficial influence of topical fluoride in decreasing lesion size and reducing formation rates of lesions [9-11]. Efforts to minimize the formation of white spot lesions should include meticulous oral hygiene and topical fluoride application [12,13]. Because patients' compliance can be a limiting factor, the orthodontist should investigate methods to achieve these goals with less than ideal patient cooperation. The beneficial effects of dentifrices and/or home use of fluoride solutions have been confirmed. Todd, et al., reported that the application of fluoride varnish (Duraflor) promoted 50% less enamel demineralization [14]. This was corroborated by Ogaard, et al., and recently, Vivaldi-Rodrigues, et al., observed a 44% reduction in the incidence of white spot lesions with tri-monthly application of fluoride varnish after 12 months of corrective orthodontic treatment [15,16]. Teeth that had fluoride varnish, applied around composite resin-bonded brackets showed a 35% reduction in demineralized lesion depth [17]. Advantages of fluoride varnish over other topical fluoride regimens include providing fluoride protection of enamel despite patient noncompliance and delivering the fluoride in a sustained manner over a longer period of time. The longer contact time with enamel enables fluoride varnishes to incorporate significantly more fluoride in enamel when compared with acidulated phosphate fluoride (APF)-gel and amine fluoride applications [18].

Brudevold et al., observed that the efficiency of topical fluoride application was directly related to the exposure period to enamel [19-24]. A longer exposure period permanently increased the amount of fluoride retained in the enamel, enhanced the formation of fluoridated hydroxyapatite, and reduced the acid solubility of enamel.

Seppa reported that fluoride varnish is not inactivated by dental plaque and does not require any previous prophylaxis [25]. Koch et al., reported that the fluoride uptake from Duraflor varnish is increased significantly by its application to dry surfaces compared with wetted surfaces. It is unclear whether the increased fluoride uptake is due to enhanced uptake by dry enamel or because the fluoride varnish adheres better to a dry surface thus remaining on the enamel surface longer with subsequent fluoride uptake. De Bruyn and Arends recommended

normal tooth brushing and drying of the tooth surface before application of the varnish [21]. The varnish sets upon contact with saliva and remains on the tooth until is removed by brushing.

The application of fluoride varnish is a preventive protocol that does not require patient compliance. Prolonged contact time with fluoride varnish permits significantly more incorporation of fluoride than other cooperation-based fluoride applications [26-31]. Peterson et al., observed that a tri-monthly application of fluoride varnish resulted in a dramatic reduction in caries incidence and the application of a fluoride varnish can be easily adapted to current orthodontic bonding techniques.

Composite resins are commonly used to bond orthodontic brackets to teeth. Research has demonstrated that plaque more readily accumulates on composite resin adhesive than on enamel [30]. Fluoride-releasing bonding agents have the potential to minimize demineralization around orthodontic brackets [32]. The critical factors for success of these materials are adequate bond strength for orthodontic appliances and sustained fluoride release. Although the composite resin has adequate bond strength, fluoride release over time is controversial [25,33-35]. An initial burst of fluoride has been associated with minimizing demineralization, but detectable fluoride levels taper off after 96 hours [36]. Glass ionomer cements on the other hand have been shown to consistently release fluoride over time [32,37]. They also have the ability to take up and re-release fluoride after application of a topical fluoride source [37-39]. Glass ionomer cements have been shown to inhibit secondary caries as well as reverse demineralized enamel lesions [40]. Although the property of fluoride releasing would appear to make glass ionomer cements an ideal bonding agent for orthodontic brackets, the adequacy of bond strength for successful clinical bonding is questionable, and weaker than resins [38-40].

Hybrid glass ionomer cements have been developed that combine the desirable properties of composite resin bond strength and glass ionomer fluoride release.

Properties of hybrid glass ionomer cements appear to improve on some of the disadvantage of composite resins and traditional glass ionomer cements [41]. Teeth with RMGI-bonded (resin modified glass ionomer cement) brackets demonstrated a 50% reduction in lesion depth whether or not fluoride varnish was applied. RMGI adhesives have been demonstrated to sustain fluoride release long after initial application but they only protect a limited area immediately adjacent to the orthodontic brackets [39-41].

MATERIALS AND METHODS

Sixty extracted premolars for orthodontic reasons were collected, immediately stored in a 0.1% thymol solution. Each tooth was subsequently disinfected with a 0.12% chlorhexidine solution and the residual calculus, bone, and soft tissue were removed [41,42].

The enamel surface of all teeth was then cleaned with a mixture of pumice and distilled water using a prophylaxis cup on a slow speed hand piece. All teeth were rinsed with distilled water and designated at random into four equal groups of 15 teeth (Table 1).

Table 1: Groups to which teeth were assigned.

Group	N	Condition	Bonding adhesive
1	15	Control, no fluoride	Composite resin (Ideal adhesion system, Accu Bond)
2	15	Fluoridated varnish	Composite resin (Ideal adhesion system, Accu Bond)
3	15	Control, no fluoride	RMGI Cement (Ideal Plus, Glass ionomer ortho band cement)
4	15	Fluoridated varnish	RMGI Cement (Ideal Plus, Glass ionomer ortho band cement)

Abbreviations: RMGI Cement: Resin Modified Glass Ionomer Cement

A window, the size of an orthodontic bracket base, was cut out from a piece of adhesive tape. The tape was placed with the window centered on the facial surface on a tooth to limit acid etching of the entire enamel surface. The enamel, exposed by the window in the tape was etched for 30 seconds with 35% phosphoric acid gel, rinsed with distilled water for the same amount of time, and thoroughly dried with compressed air.

Stainless steel orthodontic brackets (GAC, International Inc. Bohemia, NY, ROT OMNI type) were bonded to all 30 teeth in group 1 and 2 using a chemically cured composite bonding resin following the manufacturer's instructions and 30 teeth in group 3 and 4 using RMGI cement (resin modified glass ionomer cement). Twenty minutes after bonding, the adhesive tape was removed from each tooth and any excessive adhesive residue from the tape was discarded.

A box was drawn with a graphite pencil around each bracket with the perimeter of the box 2 mm from the bracket margin. All teeth were then painted with a thin coat of acid-resistant (non-fluoridated) varnish on all surfaces except the 2mm area between the brackets and the box that was drawn around the bracket.

The 30 teeth with bonded brackets were randomly distributed into two equal groups. The teeth in group 1 received no further treatment while those in group 2 were dried and the exposed enamel within the lines of the box was painted with a thin layer of Vanish (OMNI Vanish 5% Sodium Fluoride, White Varnish 3M ESP) fluoride varnish. The other 30 teeth bonded with RMGI cement were randomly distributed into two equal groups. The teeth in group 3 received no further treatment while those in group 4 were dried and the exposed enamel within the lines of the box was painted with a thin layer of Fluoride Varnish (OMNI Vanish 5% Sodium Fluoride, White Varnish 3M ESP).

After allowing the varnish to dry for 5 min, all teeth in each group were stored in separated beakers of a 200ml artificial saliva solution (Caphosol: Artificial Saliva, CYTOGEN Corporation) at neutral pH. After 12h of cycling in artificial saliva an artificial caries solution was introduced. Twice daily, with a 6-h interval, all teeth were immersed for 1h in beakers containing 200ml of artificial caries solution (Caphosol with 0.01 M of lactic acid at pH 4.4). Both solutions were stored in an incubator at a constant temperature of 37°C and artificial caries solution was changed every 3 days during the experimental period of 35 days [6,13].

After 1h of exposure to the caries solution challenge, all teeth in each group were removed, rinsed with deionized water, and brushed for 5 seconds on each surface using a Colgate Classic soft-

bristled toothbrush, without dentifrice, to stimulate mechanical wear of the varnish material. As a result, each tooth in all groups was "brushed" twice daily. The teeth were cycled between the artificial saliva and caries challenge following this protocol for 35 days. Fluoride varnish was reapplied only to the teeth in groups 2 and 4 on day 15.

On day 35, all teeth were removed from the artificial saliva solution, dried thoroughly; and photos were taken and the brackets were removed. Buccolingual longitudinal sections of approximately 400µm were made of each tooth using a high-speed diamond saw (Isomet 2000 Precision saw Buehler; Buehler Ltd., Lake Bluff, IL). The teeth were then rinsed with de-ionized water and stored in separate containers (with labels identifying the teeth from each group) of de-ionized water.

Upon examination, each tooth section was dried with absorbent paper and placed on a histological slide for evaluation under polarized light microscopy using an Olympus BX50 microscope with a 3CC Pro Series digital camera attached to it. Photomicrographs were made at 20X magnification with maximum illumination. Image Pro-Plus software was used to analyze the digital photomicrographs. This software projects a reference line on a computer screen representing 500µm. This line was traced onto tracing acetate to create a template for measurement of each enlarged digital photomicrograph. Three vertical lines were drawn on the acetate at 250µm intervals, perpendicular to the horizontal reference line; thereby, separating the area below the 500µm line into three equal sections for measurements (L_1 , L_2 and L_3). The tracing acetate template was centered over the enamel lesion on the computer screen with the horizontal reference line superimposed on the image of the enamel surface and the center vertical line (L_2) bisecting the length of the lesion. Within the area below the reference line, three depth measurements (in µm) were taken at each of the perpendicular lines (L_1 , L_2 , and L_3) for each lesion.

RESULTS AND DISCUSSION

Differences in mean maximum lesions depth between the 4 groups were analyzed using two way ANOVA to determine the impact of fluoride varnish and bonding material, pair wise t-tests with Bonferroni correction were used to compare individual groups.

Demineralization lesions were recorded for all samples in each group and were measured in mm depth.

Two way ANOVA

Two-way ANOVA was conducted on Composite/RMGI Cements with fluoride varnish and without (Cement vs. Fluoride Varnish) to test the effect of different materials with fluoride varnish on demineralization lesions.

Fluoride varnish with RMGI cement or Composite was significant ($p=0.000$) among all other groups. RMGI cement revealed less demineralization than Composite ($p=0.005$). The Cement type itself did not significantly affect demineralization when used with fluoride Varnish.

All pair wise comparison was made (Table 2).

Discussion

This study found that: (1) the application of fluoride varnish

within 1mm of the brackets base did reduce enamel lesion depth when brackets were cemented with the composite and RMGI cement, (2) the lesion depth in teeth cemented with RMGI cement and composite cement without fluoride varnish were not significantly different (Figure 1,2).

The literature was found to be limited in studying the relationship between enamel demineralization around orthodontic brackets, fluoride varnish, composite cement and RMGI cement in reducing the demineralization.

The hypothesis of this study was that fluoride varnish prevents enamel demineralization adjacent to metallic orthodontic brackets, whether the bracket is cemented with Composite or RMGI cement. The statistical significant of the results indicates that the 2 groups with fluoride varnish demonstrated less demineralization than the other groups without fluoride varnish.

Table 2: Pair wise comparison of groups.

Groups	Mean (mm)	SD (mm)	p-value comparison of gr 1	p-value comparison of gr 2	p-value comparison of gr 3	p-value comparison of gr 4
1 Composite, no fluoride	0.899	0.3630	--	0.006	1.000	0.000
2 Composite, fluoride	0.529	0.3031	0.006	--	0.237	0.053
3 RMGI, no fluoride	0.752	0.2765	1.000	0.237	--	0.000
4 RMGI, fluoride	0.241	0.1895	0.000	0.053	0.000	--

Abbreviations: RMGI Cement: Resin Modified Glass Ionomer Cement

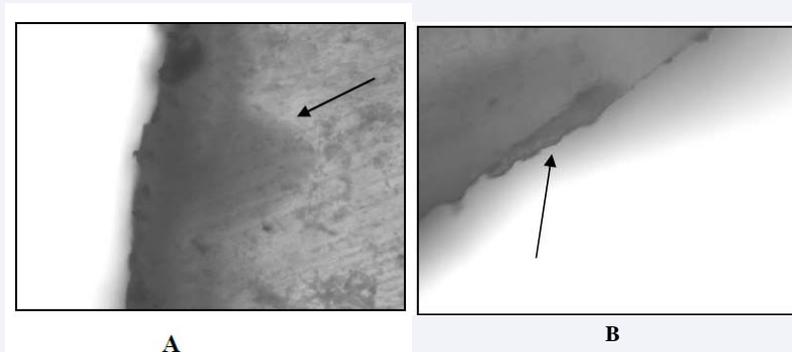


Figure 1 Polarized light photomicrograph of an enamel lesion adjacent to the site of an orthodontic bracket on a tooth (indicated by the arrows) A: with composite resin only and B: with composite resin and fluoride varnish.

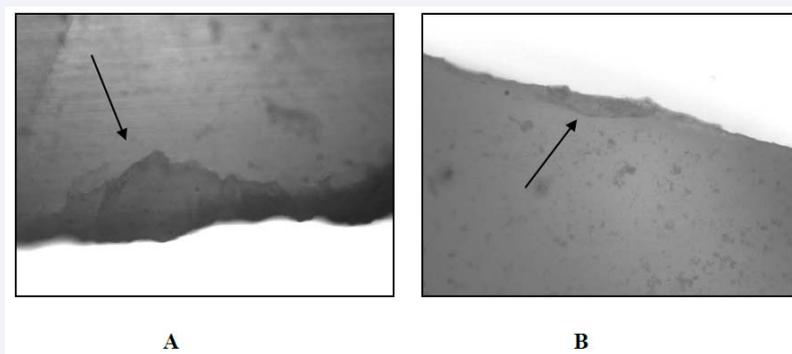


Figure 2 Polarized light photomicrograph of an enamel lesion adjacent to the site of an orthodontic bracket on a tooth (indicated by the arrows) A: with RMGI cement only and B: with RMGI cement and fluoride varnish.

The secondary aim of this study was to compare between Composite and RMGI cement for Ortho bracketing in preventing enamel demineralization. It has been shown in this *in vitro* study that there was no difference in the lesion depth between these 2 groups.

The result of this study showed that the mean demineralization depth of the lesion with RMGI cement and fluoride varnish was the least. It is speculated that the burst of fluoride released from the initial curing of the RMGI cement resulted in higher fluoride ion uptake in the enamel when compared with composite and the enamel will uptake more fluoride ions from the fluoride varnish to, this explains why the lesion depth is less in this group.

CONCLUSION

Orthodontics treatment attempts to provide as esthetic dental and facial treatment result for patients. Unfortunately, a distinct disadvantage of fixed orthodontic therapy is the development of decalcification, or white spots, adjacent to brackets during the course of treatment. This *in vitro* study evaluated the ability of a fluoride varnish to inhibit demineralization of enamel adjacent to orthodontic brackets when compared with non-fluoridated enamel.

Analyzing the measurements obtained from photomicrographs of enamel sections of human teeth, it may be concluded that fluoride varnish (Vanish) promotes a reduction of about 50% in the mean depth of enamel demineralization lesions adjacent to orthodontic brackets bonded with composite resin or RMGI cement.

Considering the results of this and many other studies that have demonstrated the efficacy of fluoride varnish in reducing the incidence and depth of enamel demineralization, orthodontist should consider its routine use in clinical practice and consider using RMGI cement, especially for patients exhibiting poor oral hygiene.

CLINICAL CONSIDERATIONS

Orthodontics should strive for 2 oral hygiene goals: (1) prevent the formation of enamel white spot lesions, and (2) if they occur, prevent their progression to cavitation. Fluoride varnish should be an adjunct in dealing with or attempting to prevent white spot lesion development. A fluoride varnish can be applied as a preventive measure after initial bonding. Further applications could be administered at subsequent appointments when patients exhibit poor oral hygiene or have dietary risks. Successive applications could also be made in areas that already have developed white spot lesions to inhibit their progression.

Patients are instructed to avoid eating for half an hour after application and to refrain from brushing the teeth the night of application. Repeated applications of fluoride varnish are needed to maintain its caries-preventive effect.

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