PENETRATION DEPTH AND ENAMEL MICROHARDNESS OF RESIN INFILTRANT AND TRADITIONAL TECHNIQUES FOR TREATMENT OF ARTIFICIAL ENAMEL LESIONS

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ABSTRACT

INTRODUCTION: Enamel white spot subsurface lesions compromise esthetics and precede cavitation; therefore, they must be halted by effective materials.

OBJECTIVE: This study was done to evaluate the penetration depth and enamel microhardness of artificial enamel lesions treated with Icon, CPP-ACP, Exite F.

MATERIALS AND METHODS: A sample of 49 sound premolars, indicated for orthodontic extraction were used to form the study group. The teeth were coated with a nail varnish, leaving a window of 4 mm \times 4 mm, on buccal surfaces of sound, intact enamel. Teeth were immersed in demineralizing solution for 4 days to produce artificial enamel lesions. Seven of the demineralized teeth were treated with low viscosity resin (Icon Infiltrant, DMG, Hamburg, Germany), Seven with CPP-ACP (Tooth Mousse from G. C. Corporation, Japan) and seven with Exite F adhesive. Specimens were observed with a stereomicroscope and the penetration depth of the materials were measured. Enamel microhardness test were determined also on specimens using calibrated Vickers indenter.

RESULTS: The maximum penetration depth was found in group I (Icon) followed by group II (CPP-ACP) finally group III (Exite F) and there was significant difference between them p=0.001 and the maximum microhardness was found in group I (Icon) followed by group II (CPP-ACP) followed by group III (Exite F) finally group IV(control) and there was significant difference between them p=0.009.

CONCLUSIONS: Resin infiltration significantly has the highest penetration depth in comparison to Cpp-acp and Exite F, resin infiltration also significantly increase microhardness of demineralized enamel in comparison to Cpp-acp and Exite F.

KEYWORDS: resin infiltration, CPP-ACP, Exite F ,white spot, penetration depth, microhardeness

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INTRODUCTION

Dental caries is a world-wide chronic disease, easily detectable, and reversible at an early stage, but irreversible and destructive after cavitation. White-spot lesions are the earliest macroscopic evidence of enamel caries (1). Typically, the enamel surface layer stays intact during subsurface demineralization, but, without treatment, will eventually collapse into a full cavity (2). White spot lesions occur when the pathogenic bacteria have breached the enamel layer and organic acids produced by the bacteria have leached out a certain amount of calcium and phosphate ions that may or may not be replaced naturally by the remineralization process. This loss of mineralized layer creates porosities that change the refractive index (RI) of enamel that is usually translucent (3).

Several techniques have been proposed to treatment of the white spot lesions. The common treatment strategy for white spot lesions comprises restorative procedures, improvement of remineralization using CCP-ACP containing or fluoride containing products, microabrasion, argon-laser irradiation. Recently, 'resin infiltration technique' was introduced with the development of highlyflowable resin material (4,5).

Caries infiltration involves the low-viscosity light curing resins (Icon) composed of triethylene glycol dimethacrylate (TEGDMA) which completely fills pores within the tooth, replacing lost tooth structure and stopping caries progression (6,7). It penetrates into the lesion by capillary forces and creates a diffusion barrier inside the lesion and not only on the lesion surface (8,9). Therefore, success of caries infiltration technique, depends on the efficacy of this low viscosity resin or caries infiltrant to penetrate up to the depth of the white spot lesion and not just mask the lesion. Also the use of 15% hydrochloric acid for etching the surface layer is effective and postulated to be beneficial for a deeper infiltration of the resin into the body of the lesion (10), and the use of solvents such as ethanol, acetone and water in resin infiltrates show lower surface tension and viscosities compared with materials without solvents.

The concept of CPP-ACP as a remineralising agent was first postulated in 1998, in which ACP is stabilised by CPP, and these nanocomplexes act as a calcium and phosphate reservoir. These nanocomplexes have been shown to prevent demineralisation and promote remineralisation of enamel subsurface lesions in animal and in situ caries models.(11)

Exite F is a light-curing, fluoride releasing, singlecomponent total-etch adhesive. It is used in the placement of direct restorations (composites, compomers) as well as in the adhesive luting of indirect restorations (all-ceramics, composites) involving light-curing and it could be used in the arresting of demineralized enamel.

Since there is no ideal method until now to treat white spot lesions therefore, this study aimed to evaluate the penetration depth and enamel microhardeness of incipient enamel lesions treated with resin infiltrant, CPP-ACP, Exite F adhesive.

MATERIALS AND METHODS

Forty nine extracted human permanent premolars, indicated for orthodontic extraction and periodontal reasons, were selected for this study. Teeth with cracks, caries or restorations were excluded. Calculus and stains were removed with hand scaler, rubber cup & polishing paste. The teeth were debrided and stored in normal saline at room temperature from the day of extraction until used. Composition and manufacturer of the materials used in the study are shown in table 1.

 Table 1: Composition and manufacturer of the materials used in the study.

Material	Composition	Manufacturer	
ICON (resin infiltrant)	1 syrige Icon-Etch 0.45ml, syringe Icon-Dry 0.45ml, 1 syrige Icon-Infiltrant 0.45ml	DMG, Hamburg, Germany.	
	Hydrochloric Acid Pyrogenic Silicic Acid Surface-active Substances		
CPP- ACP	CPP-ACP10%, Glycerol, D- sorbitol,	GC Corp., Tokyo, Japan.	
(GC Tooth mousse)	Sodium carboxyl methyl cellulose(CMC-Na),		
	Propylene glycol, silicon dioxide, Titanium		
	dioxide, Xylitol, flavoring,		
	Guar Gum,		
	Phosphoric acid, Sodium saccharin,		
	Zinc oxide,		
	Magnesium oxide,		
	Ethyl 4-hydroxybenzoate,		
	Propyl 4-hydroxybenzoate		
Excite F Adhesive	Phosphoric acid acrylate	IvoclarVivadent, Liechtenstein.	
	HEMA		
	Dimethacrylate		
	Silicone dioxide		
	Initiators		
	Stabilizers		
	Potassium fluoride in an alcohol solution		



Artificial white spot lesion

All the teeth were coated with a nail varnish, leaving a 4 mm \times 4 mm window on the middle of buccal surface. Root apices were sealed with cyanoacrylate to avoid penetration of demineralizion through the root canals. All the teeth were immersed in a demineralizing solution (2.2 mM calcium chloride, 2.2 mM monopotassium phosphate, 0.05 mM acetic acid having pH adjusted to 4.4 and 1M potassium hydroxide) for 4 days to create artificial white spot lesions (12).

For penetration depth investigation twenty one premolars with artificial white spot lesions were divided randomly into 3 groups with 7 specimens according to the material used in each group as follows:

group I: white spot lesions treated with Icon.

group II: white spot lesions treated with (CPP-ACP).

group III: white spot lesions treated with Exite F adhesive.

For microhardeness investigation twenty eight premolars with artificial white spot lesions were divided randomly into 4 groups with 7 specimens according to the material used in each group as follows:

group I: white spot lesions treated with Icon.

group II: white spot lesions treated with (CPP-ACP).

group III: white spot lesions treated with Exite F adhesive. **group IV:** white spot lesions left untreated (control group).

Material application method

Icon treatment : The surface of the lesion was etched using (Icon etchTM) 15% hydrochloric acid gel for 2 minutes. The etching gel thoroughly washed for 30 seconds using a water spray. Following etching the lesion desiccated by applying (Icon-DryTM) ethanol for 30 seconds and air dried according to the manufacturer's instructions. Low viscosity resin (Icon) was applied on the lesion surface using a microbrush and allowed to penetrate for 3 min. The excess material were removed using a cotton roll and the surface was light cured for 40 seconds using light cure device (Demetron, Dan-bury, CT, USA).

CPP-ACP treatment: The lesion was treated with CPP-ACP for 5 minutes and the excess was removed with gauze and the teeth samples were stored in artificial saliva (0.65 g/lpotassium chloride, 0.058 g/l magnesium chloride, 0.165 g/l calcium chloride, 0.804 g/l dipotassium hydrogen phosphate, 0.365 g/l potassium dihydrogen phosphate, 2 g/l sodium carboxymethyl cellulose and distilled water) until the next day. This remineralization procedure was repeated for 14 days as recommended by the manufacturer at pH = 7.2 and the artificial saliva changed every day (13).

Exite F treatment: The white spot lesion was treated with Exite F total etch for 20 seconds and then removed with water spray, then excess moisture was removed and the Exite F adhesive was applied for 10 seconds and light cured for 20 seconds according to the manufacturer's instructions.

Penetration depth investigation:

The penetration depth of the materials into the lesions were determined by immersing the Specimens in methylene blue dye for 24 h at 37°C in an incubator and sectioned into two halves along the bucco lingual plane with a diamond disc mounted on a low speed handpiece. The specimens, were then observed under the stereomicroscope at $\times 80$ magnification for determining the depth of penetration of the materials in microns. The measurements were made on three different locations and the average value was calculated (14).

Enamel microhardeness investigation:

The roots of the teeth discarded. Then, the buccal portion of specimens were embedded in acrylic resin with outer buccal surface exposed. The surface microhardness of the specimens determined by using microhardness tester with a Vickers elongated diamond pyramid indenter. A certain constant load of 50 g applied to the treated surface for 10 s (15). Three indentations placed on the center of the buccal surface and the average value considered. The diagonal length of the indentation measured by built in scaled microscope and Vickers values converted to microhardness values.

The results were analyzed by one-way analysis of variance (kruskal-wallis). Multiple comparisons between groups were performed by (post hoc) test (16).

RESULTS

Results of the penetration depth are shown in (Table 2) (Fig. 1).

Table 2: Comparison between the different groups according to penetration depth.

	Group I "ICON"	Group II "CPP-ACP"	Group III "Excite F"		
Min.	17.68	14.99	11.00		
Max	31.32	24.39	19.46		
Mean	26.32	19.27	15.47		
S.D.	4.61	3.21	3.60		
K	16.33				
P	0.001*				
P1		0.003*	0.001*		
P2			0.0312*		

P1 comparison between group I (icon) and both group II (CPP-ACP) and III (Exite F). Statistically significant at $P \le 0.05$.



Figure 1: Bar chart showing comparison between the mean values of different groups according to penetration depth.



Figure 2: Max. and min. depth of penetration of caries infiltrant.

The highest results was obtained in group I resin infiltrant (Icon) ranged from (17.68-31.32) µm (Fig. 2) with a mean of (26.32±4.61) followed by group II tooth mousse (CPP-ACP) ranged from (14.99-24.39) µm (Fig. 3) with a mean of (19.27 ± 3.21) , finally the penetration depth in group III (Exite F) adhesive ranged from (11.00-19.46) µm (Fig. 4) with a mean of (15.47 ± 3.60) and was found to be statistically significant than the other groups (p=0.001). The results of the microhardeness are shown in (Table 3) (Fig. 5).



Figure 3: Max. and min. depth of penetration of tooth mousse.



Figure 4: Max. and min. depth of penetration of Exite Fadhesive.

Table 3: Comparison between the different groups according to microhardness.

	Group I "ICON"	Group II "CPP-ACP"	Group III "Excite F"	Group IV "Control"	
Min.	174.0	165.5	159,3	137.6	
Max	208.0	182.6	184.6	160.0	
Mean	188.61	176.03	167.26	149.03	
S.D.	10.25	6.44	8.31	8.65	
K P	18.11 0.009*				
P1		0.048*	0.013*	0.002*	
P2			0.038*	0.009*	
P3				0.045*	

P1 comparison between group I (icon) and other groups. P2 comparison between group II (CPP-ACP) and both group III (Exite F) and IV (control group). P3 comparison between group III (Exite F) and IV (control group). *Statistically significant at $P \leq 0.05$.

The highest results found in group I resin infiltrant (Icon) ranged from (174.0-208.0) kg/mm² with a mean of (188.61±10.25), followed by group II tooth mousse (CPP-ACP) the enamel microhardness ranged from (165.5-182.6) kg/mm² with a mean of (176.03 ± 6.44) , followed by group III (Exite F) adhesive ranged from (159.3-184.6) kg/mm² with a mean of (167.26±8.31), finally in group IV (control) ranged from (137.6-160.0) kg/mm² with a mean of (149.03±8.65) and was found to be statistically significant than the other groups (p=0.009).



Figure 5: Bar chart showing comparison between the mean values of the different groups according to microhardness.

DISCUSSION

One of the most recent methods for conservative therapy of incipient caries is represented by local application of sealing agents with role of infiltrating hard dental tissues (17-18). The results of the present study regarding penetration depth showed that the Icon group revealed the highest value followed by CPP-ACP group followed by Exite F group. These findings may be attributed to the Icon infiltrant resin compostion which exhibit a very low viscosity, low contact angles to the enamel and high surface tension. These properties are important for complete depth of penetration of the resin infiltrant into the body of the enamel lesions (19) and also the use of hydrochloric acid gel (Icon etch) erodes the surface layer more effectively than 37% phosphoric acid used with Exite F (20). Paris et al. (21) indicated that pit and fissure caries lesions treated with an infiltrant showed significantly higher penetration depth than treatment with sealant.

Moreover in the application of the Icon infiltrant the use of Icon-dry (which contains 99% ethanol) for 30 s. Addition of ethanol increases the penetration coefficient by decreasing the viscosity and contact angle, The mixtures of the Icon dry containing large amounts of HEMA, TEGDMA, and ethanol may be associated with high penetration coefficients (22). The findings of the present study come in agreement with Priya et al (14), in that the infiltrant successfully penetrated into the artificially created white spot lesion.

The results of CPP-ACP group regarding penetration depth is lower than Icon group and this may be explained as the CPP-ACP possesses concentration ratio of calcium and phosphate and fluoride similar to that of natural tooth. The effect of CPP-ACP localizes the amorphous calcium phosphate by CPP onto the tooth surface, which creates a hypersaturation state with respect to tooth mineral. This concentration gradient drives calcium phosphate into the tooth and this the process of remineralization and mineral deposition usually affected by several factors such as saliva composition and time of remineralization to give an effective depth of penetration into enamel lesion in contrast the method of resin infiltration missing to these factors (23).

Exite F showed the lowest penetration depth value than Icon and CPP-ACP and this may be explained as the Penetration into and the arrest of artificial enamel lesions by dental adhesives have been investigated in several laboratory studies. Although a complete infiltration of shallow lesions could be accomplished using these products, penetration into natural, deeper lesions could be achieved, at most, superficially. Since adhesives have been developed mainly for the purpose of adhesion, it was not surprising that these materials showed inferior penetration capability (24-25). This come in agreement with Paris et al (26), in that the penetration depth indicated that enamel surfaces treated with resin infiltrant showed significantly higher penetration depth than treatment with Exite adhesive.

Regarding enamel microhardness, the results of the present study showed that the Icon group revealed the highest value followed by CPP-ACP group followed by Exite F group. The high microhardness result of Icon infiltrant may be attributed to the ability of low-viscosity resin to fill the spaces between the remaining crystals of porous lesions and create a diffusion barrier not only at the surface, but also within the enamel lesion body.

Therefore, a resin-infiltrated layer should be able to strengthen the demineralized enamel structure and prevent further wear and cavitation. Some clinical studies have reported that micro-invasive caries treatment with resin infiltration was an effective and safe approach to arrest initial caries lesions and preserve demineralized enamel (27-28).

The findings of the present study agreed with those reported by Torres et al. (29) and Paris et al. (30) in that the microhardness of carious lesions was significantly increased with resin infiltration and the lesion progression reduced when compared with untreated artificial lesions after demineralization. Taher et al. (31) also indicated that enamel surfaces treated with an infiltrant showed significantly higher surface hardness than treatment with fissure sealant.

The microhardeness results for CPP-ACP group is lower than Icon group and this may be due to the dependence on remineralization of white spot lesion by the artificial saliva on the deposition of calcium ions (32-33). In contrast, the low-viscous resin could penetrate into deeper lesions of the white spot lesion immediately after treatment. That explains why the microhardeness immediately showed significant recovery for the Icon group, whereas there was improvement in microhardeness only after 2 weeks of remineralization in the CPP-ACP group in this study. In our study, the CPP-ACP did not have an effective result on the white spot lesion. This might be because the time of CPP-ACP treatment in the study. This is agreed with Pancu et al. (34) who found that caries infiltration leads to increase the hardness of infiltrating lesions compared to the fluoride group which depend in the treatment on the time of remineralization with artificial saliva

Exite F result regarding the enamel microhardness was lower than Icon and CPP-ACP groups but higher than the control group and this may be attributed to the very thick oxygen inhibition layer which forming the Exite F compared to the other materials, so Exite F give microhardness values higher than control group (demineralized enamel), although this established adhesive layer was thick enough to prevent further demineralization. It was very inhomogenous, and only partly polymerized resin layer due to the solvent (water), and it could not protect the lesions from further acidic influences (24).

CONCLUSIONS

Under the conditions of this study, these conclusions can be derived.

- 1. Resin infiltration significantly has the highest penetration depth in comparison to CPP-ACP and Exite F.
- 2. Resin infiltration significantly increased microhardness of demineralized enamel in comparison to CPP-ACP and Exite F.

CONFLICT OF INTEREST:

The authors declare that they have no conflicts of interest.

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