

Assessment and comparison of microleakage of a fluoride-releasing sealant after acid etching and Er: YAG laser treatment – An *in vitro* study

R. VIJAYARAGHAVAN, V. ARUN PRASAD RAO¹, N. VENUGOPAL REDDY¹, R. KRISHNAKUMAR¹, D. K. SUGUMARAN¹, G. MOHAN¹

Abstract

Aim: The aim of the present study was to estimate and compare the microleakage of a pit and fissure sealant after etching and Er:YAG laser treatment. **Materials and Methods:** Twenty non-cariou premolars extracted for orthodontic reasons were equally divided into two groups. Samples in Group- I were treated with 37% phosphoric acid. Samples in Group II were irradiated with Er: YAG laser at 400 mJ at 4 Hz. Sealant was placed and light cured. Then, the samples were subjected to thermocycling. The samples were then immersed in 1% methylene blue. The samples were sectioned and examined under stereomicroscope at $\times 10$ magnification. **Results:** Acid etched samples showed significantly less microleakage when compared to laser etching and it was statistically significant ($P < 0.01$).

Keywords: Er:YAG laser, microleakage, sealants

Introduction

The major concern of modern dentistry, mainly for the last decade, has become focused on reducing patients' risk for caries, stimulating preventive measures, and preserving tooth structure, indicating, as often as possible, non-invasive conservative techniques instead of proceeding with an invasive healing treatment.^[1] Sealing occlusal pit and fissures in teeth is a common and highly effective caries preventive method.^[2] Pit and fissure sealants have been widely employed since the 1970s and were considered an efficient preventive method, as they can block the development of caries lesions on occlusal surfaces.^[3]

The preventive benefit of this treatment relies upon the ability of the material to promote an appropriate sealing of pits, fissures or eventual enamel defects, and remain completely intact and bonded to enamel surface.^[1] The marginal sealing ability of sealing materials is extremely

important for successful treatment. Lack of sealing allows the occurrence of marginal leakage, i.e. passage of bacteria, fluids, molecules and ions through the tooth-material interface, which can prompt caries lesion progression underneath the restoration.^[4]

In 1955, Buonocore described phosphoric acid as a simple means for increasing the adhesion of acrylic filling materials to enamel surfaces.^[5] Effective adhesion to enamel has been achieved with relative ease and has repeatedly proven to be a durable and reliable clinical procedure for routine applications in modern adhesive restorative dentistry.^[6]

Nevertheless, a disadvantage attributed to acid conditioning is that demineralization of enamel surface makes it more permeable and prone to long-term acid attack and caries, especially if the demineralized substrate was not completely filled by resin monomers. In order to overcome this limitation, studies have investigated alternative procedures for treatment of enamel surface, such as the Er:YAG laser irradiation.^[7]

Laser energy is greatly absorbed by the dental enamel, promoting superficial modifications, which has treatment significance.^[5] Tooth conditioning by lasers has been suggested as a means of preparing dental surfaces for adhesive procedures.^[8]

Maria Cristina Borsatto *et al.*,^[1] reported that preparing pits and fissures exclusively by Er:YAG laser did not result in optimal penetration of sealant into etched enamel. The laser irradiation alone was not able to produce a high-quality, dye penetration-resistant interface.^[1]

J Moshonov *et al.*,^[2] observed no significant difference between acid etching and laser etching.^[2] Tatjana Dostalova *et al.*,^[5] reported that Er:YAG laser etching can apparently replace acid

Department of Pedodontics and Preventive Dentistry, Vivekanandha Dental College for Women, Elayampalayam, Tiruchengode, Namakkal District, 'Rajah Muthiah Dental College and Hospital, Annamalai University, Chidambaram, Tamil Nadu, India

Correspondence: Dr. R. Vijayaraghavan, Department of Pedodontics and Preventive Dentistry, Vivekanandha Dental College for Women, Elayampalayam, Tiruchengode, Namakkal District, Tamil Nadu, India. E-mail: drvijay.ragavan@gmail.com

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etching with similar effect on enamel and without negative influence of phosphoric acid.^[5] Jung-Ho Kim *et al.*,^[9] concluded that Er:YAG laser treated enamel were more resistant to acid attack than phosphoric acid-etched enamels.^[9]

The literature available regarding the quality of pit and fissure sealants after Er:YAG laser preparation was rare and not conclusive.^[1,2,5] In this context, the purpose of the present study was to assess and compare the degree of marginal leakage of a pit and fissure sealant after acid etching and Er:YAG laser treatment.

Materials and Methods

Twenty sound, non-carious young premolars, which had been extracted for orthodontic reasons, were selected and stored in sterile saline. The teeth were randomly divided into two groups, with ten premolars in each group. In Group I, the occlusal surfaces were etched with 37% phosphoric acid gel for 30 s, rinsed with air–water spray for 20 s, and gently air dried.^[2] In Group II, laser irradiation of the enamel occlusal surface without contact and in scanning mode was done using an Er:YAG laser (Fidelis, Fotona) with 400 mJ per pulse and 4 pulses per second, at 12-mm working distance with air–water spray,^[3] followed by placement of sealant material (Helioseal F, Ivoclar Vivadent, Pineview Drive Amherst, NY USA), and light cured using a conventional light source for 20 s. The teeth were then stored in saline at room temperature in plastic containers. Then, the samples were subjected to thermocycling at 750 cycles in baths at 55°C and 6°C, for 10 s in each bath under electronic control.^[2]

The root apices were sealed with acrylic resin. All the samples were then covered with three layers of nail varnish, except for the 1-mm window at the resin–sealant interface, and immersed in 1% methylene blue solution for 24 h. After being immersed in the 1% methylene blue solution, all samples were embedded in chemically activated acrylic resin and sectioned in the microtome. Slices were then examined under stereomicroscope at 10X magnification connected to a digital video camera (Samsung, China) lens macro connected to a computer. The images were recorded in a CD-R (700 MB-Moserbaer, India) and analyzed for marginal leakage. Microleakage was measured by the degree of dye penetration. [Table 1]^[3,10] [Figures 1-5].

The data were tabulated and subjected to statistical analysis using Kruskal-Wallis test and Chi-Square test.

Results

Microleakage scores for Group I and Group II are shown in Table 2. Group II showed more number of specimens with high scores [Table 3]. The mean and standard deviation for Group I and Group II are shown in Table 3, and the value was statistically significant [Table 4].



Figure 1: Score 0



Figure 2: Score 1

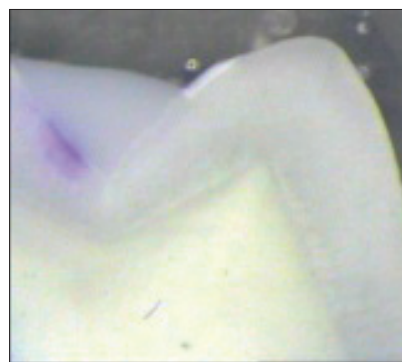


Figure 3: Score 3

Table 1: Dye penetration scores

Score	Penetration of the dye
0	No microleakage
1	Dye penetration restricted to the occlusal third of one of the sealants' wall (buccal or lingual)
2	Dye penetration restricted to the occlusal third of both the sealants' walls
3	Dye penetration restricted to the medium third of one of the sealants' walls
4	Dye penetration restricted to the medium third of both the sealants' walls
5	Dye penetration restricted to the pulpal third of one of the sealants' walls
6	Dye penetration restricted to the pulpal third of both the sealants' walls
7	Total dye penetration along the cavity walls including the pulpal wall

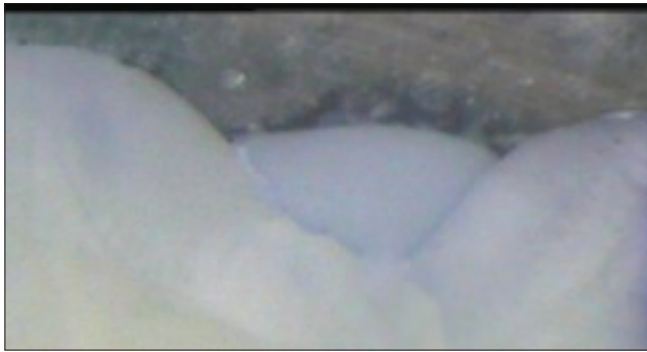


Figure 4: Score 6

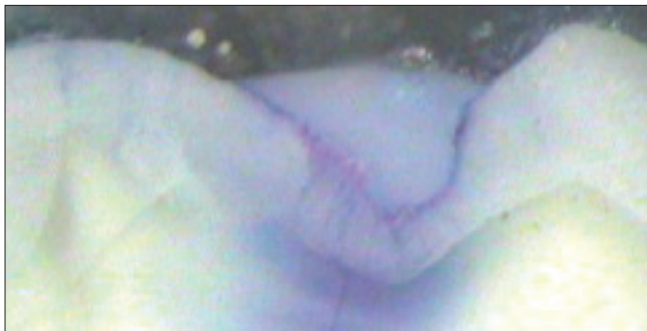


Figure 5: Score 7

Discussion

The marginal sealing is important for sealant success because penetration of bacteria beneath the sealant might allow caries onset and/or progression. It has also been advocated that if complete caries removal was precluded or missed, the sealing ability of the filling material seems to be more important than its cariostatic properties.^[4]

An *in vitro* model was chosen in the present study to standardize the model, and allowed thermocycling to simulate stress caused by thermal variation.^[11] Thermocycling is a method used widely in dental research, particularly when testing the performance of adhesive materials. It aims at thermally stressing the adhesive joint at the tooth–restoration interface by subjecting the restored teeth to extreme temperatures compatible with temperatures encountered intraorally.^[12] The most widely concentration used at present for enamel is 37% phosphoric acid. Acid etching of enamel appeared to improve retention by selectively eroding certain hydroxyapatite formations and facilitated penetration with development of resin tags.^[13] Lasers with a wide range of characteristics are available today and are being used in several fields of dentistry. Laser energy is absorbed by the dental enamel, promoting superficial modifications, which may have clinical significance.^[2]

Short, high-energy pulses allowed effective tissue removal with almost no temperature elevation to the surrounding

Table 2: Dye penetration scores obtained for all sections in each group

Group	Dye penetration scores								Total	% of samples showing microleakage
	0	1	2	3	4	5	6	7		
I	3	2	0	1	0	0	0	4	7	70
II	2	2	0	0	0	0	1	5	8	80

Table 3: Mean and standard deviation for Group I and Group II

Group	Mean	SD
Group I	3.3	3.3
Group II	4.3	3.3

Table 4: Chi-Square test

Scores	Group I	Group II
Low	2	2
Medium	1	0
High	4	6
Total	7	8

Calculated Chi-Square value $\chi^2 = 13.27$, P value = 0.0041 ($P < 0.01$) (significant), Low: Score 1 + score 2, Medium: Score 3 + score 4, High: Score 5 + score 6 + score 7

tissue.^[3] Water irrigation seemed to effectively prevent the thermal damage. Water irrigation was particularly important in reducing thermal effects. The spray allowed cleaning of the ablation site, supplied an increased efficiency for the ablation rate, and promoted the ablation process (photoacoustic effect).^[14]

In the present study, 70% of the samples in Group I showed microleakage [Table 2]. Similar results were obtained in the previous investigations.^[15,16] Microleakage could be expected in all the restorative materials. The most likely explanation for this could be the difference in the thermal expansion co-efficient of sealant and the enamel.^[17]

In the present study, the samples prepared using 37% phosphoric acid gel obtained a mean microleakage score of 3.3 [Table 3]. The existing literature had shown values ranging between 1.429 and 3.3.^[18,19] The difference in findings might be due to the difference in the materials used. More fluid resins might penetrate fissures more deeply and spread more rapidly over the surface.^[20]

In the present study, 80% of the samples in Group II showed microleakage [Table 2]. M. N. Youssef *et al.*,^[3] investigated the effects of Er:YAG laser irradiation on the microleakage of pit and fissure sealants. The authors observed microleakage in all the samples that were prepared by laser. The difference in the finding could be due to difference in the type of dye material used. The authors used 50% silver nitrate, which has a relatively smaller particle size when compared with 1 % methylene blue.^[3] In a previous investigation, 63 % of the samples prepared with Er:YAG laser showed microleakage.^[21] This could be due to the difference in the laser parameters used. The authors used 1000 mJ/pulse at 10 Hz for laser

irradiation.^[21] J. Moshonov *et al.*,^[1,2] did not observe microleakage in any of the samples. The authors used Er:YAG laser at 800 mJ/pulse and 12 pulses per second.^[2]

In the present study, the samples prepared by Er:YAG laser obtained a mean microleakage score of 4.3 [Table 3]. The previous investigations conducted had microleakage scores ranging between 0.76 and 3.51.^[21,22] The difference in the findings might be due to difference in the microleakage evaluation method. The authors calculated the microleakage by measuring the length of penetration of dye material between the sealant–enamel interface. Maria Cristina Borsatto *et al.*,^[22] obtained a mean microleakage score of 3.51 after Er:YAG laser preparation. In their investigation, microleakage was calculated as percentage after measuring the length of the resin–sealant interface.^[22]

In the present study specimens prepared with 37% phosphoric acid gel showed less microleakage scores than the laser-irradiated specimens, and the difference was statistically significant.[Table 4] Similar results were obtained in the previous investigation on Er:YAG laser.^[1,21,22]

A suitable explanation for such performance would be that due to its thermally induced microexplosive ablation process, the Er:YAG laser does not provide a selective dissolution of the mineral phase. Hence, it doesn't create an even, uniform etching pattern similar to that obtained with etchant solutions. Instead, laser ablation yields a random fragmentation and removal of dental substance with a real cleavage of the enamel prism pathway.^[6]

Acid etching provides suitable substrate for adhesion, because it removes the smear layer and creates a uniform microretentive pattern, due to the selective dissolution and removal of hydroxyapatite crystals.^[17]

The morphological recesses clearly differ from the well-arranged microporosities characteristic of acid etching. In addition, the Er:YAG laser beam does not have continuous emission and, therefore, does not provide a homogeneous etching of tooth surface, leaving non-lased areas between the pulses. Consequently, it is likely that such irregular microstructure leads to bonding failures and undermined marginal sealing. Er:YAG laser irradiation of pits and fissures does not eliminate the need for etching.^[15]

Disruption as a result of microexplosions weakened the enamel and gave rise to a more heterogeneous surface than that obtained by acid etching. Acid etching typically produced a repeating surface pattern, with cracks and fissures no deeper than 12 μm that were readily filled with resin. In contrast to acid-etch treatment, laser etching produced extensive surface fissuring and less regular and less homogeneous surface patterns arising from the union of different craters.^[13]

No gap was found in acid-etched enamel specimens, and resin tags were visible. On the other hand, laser irradiated samples, whether acid etched or not, presented with gaps and no mechanical interlocking.^[7]

One of the potential disadvantages of enamel acid etching is that the acid causes demineralization of the most superficial layer. As a result, this surface becomes more susceptible to long-term acid attack and caries, especially when resin impregnation is defective because of air bubbles or saliva contamination. Such effects are particularly important, given that plaque tends to accumulate at interfacial surface. The physiochemical changes caused by laser etching can be expected to decrease long-term susceptibility to acid attack and caries. This reduction may be related to changes in Ca:P ratio, reduced carbonates, and pyrophosphate formation, together with reduced water and organic component contents. It has also been suggested that laser etching might create re-mineralization/micro-spaces that trap free ions.^[13]

Mozammal Hossain *et al.*, reported that application of Er:YAG with and without water spray was effective for caries prevention.^[23] Cebellos *et al.*, reported 56% reduction in primary enamel surface lesion depth when compared with the acid-etched group.^[24]

Moisture contamination of the enamel may lead to reduced penetration of the sealant and, therefore, microleakage of the bacteria at the margins may increase the chances of caries developing. Tooth isolation may be achieved by the use of cotton rolls or rubber dam. Both techniques require skill and precision, and are time consuming. The preparation of tooth surface by laser does not require isolation of the tooth, and thus, can save the dentist this step prior to applying the sealant material on the enamel surface.^[2]

Microleakage strictly refers to ingress of oral bacteria, which have an approximate diameter of 0–5 μm .^[17] The size of the methylene blue molecule is 1.2 nm².^[25] Therefore, dye leakage tends to be a severe test for microleakage.^[18]

Conclusion

In the present investigation, laser and conventional etching were compared only with respect to marginal leakage. Different environmental conditions and different ecologies in various mouths may influence the microleakage of the sealant placed using either technique. Adhesive interface, micromorphology, and the alterations in substrate compounds, under different laser parameters should be investigated to achieve optimum irradiation conditions for pit and fissure sealant placement. Moreover, further *in vivo* researches on the longevity of the restorations are required.

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