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Clinical Evaluation and Microleakage of Fissure Sealant: Effect of Laser Etching vs Acid Etching

Lazer ve Asit ile Pürüzlendirmenin Fissür Örtücünün Mikrosızıntısına Etkisi ve Klinik Değerlendirilmesi

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ABSTRACT

Aim: The aim of this study was to evaluate the clinical success and the sealing ability of a resin based fissure sealant that applied enamel surface which was etched with conventional phosphoric acid and Er:YAG laser at different energy levels.
Material and Methods: The study consisted of two parts; the evaluation of retention, marginal discoloration, marginal integrity and new caries formation for the clinical success after the treatment at 3, 6, 12 months; the evaluation of microleakage and penetration depth to fissures using dye penetration test. The in vivo part of the study was the surface preparation processes, 72 teeth in each group; It consists of 32% phosphoric Acid, Er:YAG laser- MSP (Medium Short Pulse) and Er:YAG laser- QSP (Quantum Square Pulse). A total of 45 impacted human third molar teeth were used for in vitro part. Data were analyzed using SPSS 21.
Results: The relationship between the success rates of 12-month clinical follow-up of fissure sealants and age, gender, and localization was assessed and there was no statistically significant correlation. When all criteria were considered, the most successful group was QSP. All the groups demonstrated microleakage regardless of the surface preparation techniques. However, there was no statistically significant difference between the groups.
Conclusion: It is considered to determine the optimal energy level of laser will increase the clinical success.

Keywords: Enamel, Fissure sealants, Er-YAG laser, Surface preparation, Microleakage

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ÖZ

Amaç: Bu çalışmanın amacı geleneksel olarak kullanılan asit ve farklı enerji seviyelerinde Er:YAG lazer ile pürüzlendirilmiş mine yüzeylerine uygulanan rezin esaslı bir fissür örtücünün klinik başarısını ve örtücülük kabiliyetini değerlendirmektir.
Gereç ve Yöntemler: Çalışma iki bölümden oluşmaktadır; tedaviden sonra klinik başarının 3., 6. ve 12. aylarda retansiyon, marjinal renklenme, marjinal bütünlük ve yeni çürük oluşumu açısından değerlendirilmesi; boya penetrasyon testi kullanılarak mikrosızıntının ve fissürlere penetrasyon derinliğinin değerlendirilmesi. Çalışmanın in vivo kısmı her grupta 72 diş olmak üzere yüzey hazırlığı işlemleri; %32 fosforik Asit, Er:YAG laser- MSP (Medium Short Pulse) ve Er:YAG laser- QSP (Quantum Square Pulse olmak üzere üç gruptan oluşmaktadır. İn vitro kısım için toplam 45 adet gömülü üçüncü molar diş kullanılmıştır. Veriler SPSS 21 kullanılarak analiz edilmiştir.
Bulgular: Fissür örtücülerin 12 aylık klinik takibindeki başarı oranları ile yaş, cinsiyet ve lokalizasyon arasındaki ilişki değerlendirilmiş ve istatistiksel olarak anlamlı bir ilişki bulunamamıştır. Tüm sonuçlar göz önünde bulundurulduğunda en başarılı grup QSP olmuştur. Tüm gruplar yüzey hazırlama tekniklerinden bağımsız olarak mikrosızıntı göstermiştir. Ancak gruplar arasında istatistiksel olarak anlamlı bir fark bulunmamıştır.
Sonuç: Lazerin optimal enerji seviyesinin belirlenmesinin klinik başarıyı artıracağı düşünülmektedir.

Anahtar Kelimeler: Mine, Fissür örtücü, Er-YAG lazer, Yüzey hazırlığı, Mikrosızıntı

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INTRODUCTION

Dental caries is one of the most common chronic diseases in childhood.[1] The prevention of caries is an important prerequisite for a healthy adult dentition.[2] Consequently, substantial importance is placed on the early diagnosis and treatment, preserving the tooth structure as much as possible and preventive programs that aim to reduce the rate of caries progression in pediatric dentistry.[3,4]

Pits and fissures on the occlusal surfaces of permanent molar teeth increase the risk of caries formation, by accumulating more microorganisms and debris in the grooves. Therewithal, the pits and fissures are the most susceptible to caries development and are the most affected tooth surface.[3,5]

The teeth are often at highest risk to caries during the eruption process, as the enamel is not fully matured, parents are often unaware of the newly erupting teeth, and cleaning difficulties can arise. It may also be a challenge for children to brush all surfaces of the new erupting teeth, effectively, as molars exhibit a complex morphology, with fissures covering 12% of the tooth surface.[2,6]

Thus, sealants play a crucial role, among the caries prevention practices. Sealant application is considered the most effective approach to prevent the formation of caries on fissures and aims to transform fissures that are ideal for accumulation of food remnants and bacterial colonization, into smooth surfaces that can be cleaned easily.[7,8]

Various materials are used as sealants.[9-11] The most critical factors affecting the clinical success of the sealants are the resistance to microleakage associated with the quality of the adhesion between the enamel surface and the sealant material, and consequently, the long-term retention.[10,12]

The method of enamel surface preparation is directly related to the adhesion.[13] Phosphoric acid is conventionally used for conditioning of enamel prior to sealant. However, this method is time-consuming, has an unacceptable taste for children and requires technical precision. Therefore, new alternative approaches, which increase the surface intensity of enamel, have evolved.[14] The discovery of laser treatment in dentistry is an alternative to many conventional methods. In recent years, laser is increasingly used in dentistry as an enamel surface conditioning technique.[15]

It is noticed that the pulse duration of lasers is a significant factor for the etching.[16] The recently-developed quantum-square pulse (QSP) setting of Er:YAG laser, allows for high-efficiency pulses to follow each other at the optimum speed, with low energy. Hence, the absorption and scattering of the laser beam and, additionally, undesirable thermal effects on the dental tissues, are prevented.[17,18]

The etching of enamel surface with various techniques is aimed to increase the success of the clinical outcome. This two-part study evaluates the success of a resin-based sealant, applied to the enamel surface, which is etched with conventional acid and laser at two different energy levels. Firstly, it was evaluated the clinical success of the sealants after the treatment, at 3, 6 and 12 months in vivo. Secondly, the dye penetration test is performed in vitro, to observe the microleakage. In the current literature, no studies compare the Er:YAG laser in QSP and medium-short pulse (MSP) modes and conventional acid etching, and evaluate the clinical success of a resin-based sealant in vivo and the microleakage in vitro, simultaneously. The initial study hypothesis was that the use of Er:YAG laser in QSP mode for etching before the sealant, provided statistically significant clinical success and microleakage.

MATERIAL AND METHODS

This study evaluated the success of a resin-based sealant applied to an enamel surface etched with acid and laser at two different energy levels in vivo, as well as the sealing ability of the sealant in vitro. The materials and equipment used in the study are listed in Table 1 and Table 2. Ethical committee approval for the experiments was obtained (2016/002).

The parents of all children involved in the in vivo survey were informed about the trial, and both verbal and written consent was obtained before procedures. The patients were also advised about the collection of the teeth to be used in the in vitro experiments, and verbal and written consent for the teeth indicated for extraction was attained.

Table 1 The materials used in the current study.

Product	Properties	Composition	Manufacturer
Clinpro™ Sealant	Resin based fissure sealant.	Bis-GMA, TEGDMA, EDMAB [Ethyl 4- (dimethylamino) benzoate] Diphenyidodonium hexafluorophosphate, BHT (Butyl hydroxytoluene), CQ (Camphorquinone), 1-7% Reinforced inorganic filler (Silane-treated amorphous silica), TBATFB (Tetrabutylammonium Tetrafluoroborate), TiO2 (Titanium Dioxide), Rose bengal sodium.	3M ESPE, St. Paul, MN, USA.
Scotchbond™ Universal Etchant	32% phosphoric acid gel.	Water, phosphoric acid, synthetic amorphous silica, fumed, crystalline free, polyethylene glycol, aluminum oxide.	3M ESPE, St. Paul, MN, USA.
Microscopy Cerstistain® Fuchsin (C.I. 42510)	Basic fuchsin.	C.I. Basic Red 9.	Merck KgaA, Damnstadt, Germany.

Table 2 The equipment used in the current study.

Equipment	Properties	Instruments	Manufacturer
Light-Walker AT	Er:YAG Laser System	Fiber tip suitable for contact handpiece (Code: 71766, Cylindrical, sapphire, Diameter: 1.3 mm, Length: 8 mm, Maximum energy: 600 mJ) Protective laser glasses.	Fotona, Slovenia.
Isomet 1000 Precision Saw	Slow-speed sectioning device	Double-sided cutting diamond fine blade (Isomet Diamond Wafering Blades, Buehler GmbH Dusseldorf, Germany).	Buehler Lake Bluff, IL, USA.
Kavo DiagnoDent Pen 2190	Laser fluorescence divice	Sapphire tips.	Biberach, Germany.

The study consists of 3 groups: Acid ething, MSP and QSP (Table 3). The group treatments performed.

Table 3 The study groups.

Groups	Procedure
GRUP1: Acid	%32 phosphoric acid etching (20 second)
GRUP 2: QSP (Quantum Square Pulse)	Er:YAG laser QSP setting (15 second)
GRUP 3: MSP (Medium Short Pulse)	Er:YAG laser MSP setting (15 second)

In vivo Study

The in vivo investigation was conducted on 54 patients (26 male, 28 female), aged between 7-11 (±1,26) years, who had no systemic disorder. Children who were referred to our clinic and examined by a single investigator, and presented no occlusal problem, bruxism, clamping, non-carious first permanent molar teeth that had completed the eruption, and had no developmental defects or restorations, were included in the study. There were 3 groups in the trial, with 72 teeth in each group. The teeth were primarily examined clinically and radiologically, and also with a laser fluorescence device (KaVo DIAGNO Dent Pen 2190, Biberach, Germany).

In order to evaluate the clinical efficacy of the all preparation techniques belonging to the same patient, at most two study groups were used at the same time in each patient. Each tooth group was considered equal in each study group, by applying a split-mouth design. All dental treatments were completed before sealant, and all children were given oral hygiene motivation.

Clinical Procedure

The plaque and debris on the tooth surfaces were cleaned with brush/water before the sealant was applied. During the treatment, roll cotton and rubber dam were used to prevent moisture, and the surfaces dried with an air-water spray. The occlusal surfaces were etched with acid for 20s and Er:YAG laser at two different energy levels (120mJ, 10Hz, 1.2 W ;60%water, 40% air) for 15s (Fig.1), then washed and air-dried. Next, ClinproTM (3M ESPE, St. Paul, MN, USA) sealant was applied, in accordance with the manufacturer's instructions. Early occlusal contact points were examined, and, if necessary, excesses were removed with finishing burs, followed by polishing, and, thus, the restoration completed.

Figure 1: The etching of the occlusal surfaces with Er:YAG laser.

Clinical Evaluation

The clinical success of the sealants, according to the modified United States Public Health Services (USPHS) criteria, including retention, marginal integrity, marginal discolouration and secondary caries formation, was evaluated at 3, 6 and 12 months (Table 4). 19 The clinical investigation for the success of the sealant was assessed by a different, calibrated and blinded investigator. The sealants denoted 'unsuccessful' were repaired or renewed for ethical reasons, during this period.

Table 4 The modified USPHS criteria.

Retention	Marginal Integrity	Marginal Discoloration	Secondary Caries Formation
ALPHA: Sealant was intact.	**ALPHA**: Closely adapted, no visual evidence of a crevice along the margin.	**ALPHA**: There was no discoloration.	**ALPHA**: Absence of caries.
BRAVO: Sealant was partially intact.	**BRAVO**: Visible crevice along the margin into which the explorer penetrate or catch.	**BRAVO**: There was a superficial discoloration of the sealant but can be removed by polishing.	
CHARLIE: Sealant was totally lost.	**CHARLIE**: Visible evidence of a crevice along the margin into which the explorer penetrate or catch.	**CHARLIE**: There was a deep coloration on the edges of the sealant and it can not be removed by polishing.	**CHARLIE**: Presence of caries.

In vitro Study

A total of 45 impacted human third molar teeth were examined in vitro. The teeth were cleaned of organic debris and stored in distilled water at room temperature, and then randomly divided into 3 groups, for the microleakage to fissures. The etching of enamel surfaces and sealant application were performed as per the in vivo part. Samples were subjected to 10,000 thermal cycles between 5 and 55°C, with a dwell time of 30s and were kept in distilled water until the test.20 After thermocycling, the root apices of the teeth were covered with wax. All tooth surfaces, except the occlusal surfaces, were isolated with two layers of nail varnish, leaving a 1mm window around the sealants. Subsequently, the samples were immersed in 0.5% fuchsin staining solution (Merck KgaA, Damnstadt, Germany) for 24h. All the samples were then washed in plain water to remove excess dye. After drying, the samples were mounted in acrylic resin blocks. The crown portions were sectioned buccolingually into three pieces (Fig.2). The sectioned samples were examined under a stereomicroscope (Leica Microsystems Ltd, Heerbrugg, Swiss) at ×40 magnification. The degree of the microleakage was scored by using the criteria listed in Table5.

Figure 2: The sectioned tooth samples for the evaluation of microleakage.

Table 5 The scores of microleakage.

Score	
0	no dye penetration
1	dye penetration restricted to occlusal half of the tooth/sealant interface
2	dye penetration restricted to gingival half of the tooth/sealant interface
3	dye penetration up to the depth of the groove and beneath the sealant

Statistical Analysis

Data were analyzed using SPSS21 (SPSS, Chicago, USA). The single sample Shapiro-Wilk test was applied, to examine whether each factor had a normal distribution. Friedman's test for dependent multiple comparisons and two related sample tests for binary comparisons were used. For the microleakage measurements, Post-hoc Tukey and one-way ANOVA were conducted, to determine statistically significant differences between the groups. The relationships between the success rates at 12-month clinical follow-up of the sealants with age, gender and localization were assessed by chi-square and Fisher's exact tests. Statistical significance was set at $p<0.05$.

RESULTS

The initial hypothesis was rejected based on the results of the study. The in vivo part of this study involved 54 children aged between 7-11 years. A total of 216 permanent first molar teeth were fissure-sealed under three different surface conditions and evaluated in a split-mouth design. Three patients were excluded at the 3-month follow-up because they didn't come to the control appointments. Patients were assessed according to the modified USPHS criteria at 3-,6- and 12-month appointments.

There were no statistically significant correlations between the clinical success rate and these variables. The most frequent failure was the impairment of marginal integrity. When all criteria were considered, the most successful group was QSP. Failure cases in all follow-up periods are given in Table6.

Table 6 The rates and percentages of modified USPHS criteria of all groups in 12 month follow-up period.

		Modified USPHS Criteria			
Groups	**Score**	**Retention** n(%)	**Marginal Integrity** n(%)	**Marginal Discoloration** n(%)	**Secondary Caries Formation** n(%)
1	Alfa	47 (69,12%)	40 (58,82%)	20 (29,42%)	63 (92,65%)
	Bravo	17 (25%)	24 (35,3%)	43 (63,23%)	
	Charlie	4 (5,88%)	4 (5,88%)	5 (7,35%)	5 (7,35%)
	*	*46,053*	*53,82*	*104,61*	*5,4*
2	Alfa	45 (78,95%)	46 (68,66%)	35 (52,24%)	66 (98,51%)
	Bravo	12 (21,05%)	18 (26,86%)	30 (44,78%)	
	Charlie	0	3 (4,48%)	2 (2,98%)	1 (1,49%)
	*	*27,558*	*48,504*	*64,789*	*9*
3	Alfa	36 (52,17%)	28 (40,58%)	19 (27,54%)	66 (95,65%)
	Bravo	24 (34,78%)	27 (39,13%)	38 (55,07%)	
	Charlie	9 (13,05%)	14 (20,29%)	12 (17,39%)	3 (4,35%)
	*	*81,742*	*103,332*	*107,328*	*9,923*

**: chi square value. p>0.05, statistically insignificant.*

In the 12-month follow-up period, the differences between Acid and QSP, and Acid and MSP were not statistically significant while there was a significant difference between QSP and MSP in retention, marginal integrity and marginal discolouration of the sealants. There was no significant difference between the three groups in the comparison of secondary caries formation.

The in vitro part was performed on 45 impacted human third molar teeth, and the microleakage was evaluated. The microleakage scores of all groups (Table7) were non-significant by Tukey's test. All the groups demonstrated microleakage, regardless of the surface preparation techniques. When the groups were compared for the extent of microleakage, based on the degree of dye penetration between the sealant and tooth substance interface, the lowest values were obtained in Acid group, but the difference was not significant.

Table 7 The rates and percentages of dye penetration test.

	Score			
Groups	**0**	**1**	**2**	**3**
1	3 (20%)	3 (20%)	5 (33,3%)	4 (26,7%)
2	7 (46,7%)	4 (26,7%)	2 (13,3%)	2 (13,3%)
3	7 (46,7%)	5 (33,3%)	2 (13,3%)	1 (6,7%)

p>0.05, statistically insignificant. Kruskal-Wallis test.

DISCUSSION

The present study was conducted to evaluate the clinical success and microleakage of a fissure sealant by three different etching techniques. Due to their nature, pits and fissures are prone to dental plaque accumulation, and cannot benefit from the remineralisation effect of fluoride as much as the smooth surfaces.[4] Therefore, the sealing of chewing surfaces is required. Sealants are a somewhat effective treatment for the first dental visit in pediatric dentistry. In addition, this preventive technique is cost-effective, without loss of dental tissue.[21,22] Nonetheless, sealants may not stay whole, and thus, should be examined at regular intervals and repaired, if necessary.[23,24]

Adhesion between the sealant and dental tissue and long-term retention are the most significant factors assessing the success.[10] Therefore, the tooth surfaces must be prepared prior to the treatment.[25] Different fissure modification methods are used to prime tooth surfaces.

The use of acid is an accepted and standard method. Although 32-37% phosphoric acid for 20s, is routinely used as an enamel conditioner, it has some disadvantages. Consequently, alternative techniques that further increase the surface energy of enamel have been sought.[14]

The laser vaporizes water trapped in the hydroxyapatite matrix on the surface, causing microablation. Thus, the enamel surface becomes rough and irregular, similar to acid etching. It changes the calcium/phosphorus and carbonate/phosphate ratios of enamel and provides less soluble contents in the acidic medium. Besides, it can be absorbed only in the superficial parts of the dental hard tissues, thereby preventing thermal damage to deep tissues.[26,27]

There are several advantages of the laser over conventional methods, such as the faster and more controlled preparation. Particularly, the use of laser etching for children whose isolation is difficult is favourable. In the literature, conflicting results are reported. Some studies state that the enamel etching with the Er:YAG laser exhibits weaker adhesion than conventional methods, while others describe it as similar or superior to that of traditional methods.[28-30]

The lack of consensus on this issue is attributed to the use of different laser systems or settings.[31] Especially, the laser pulse duration is recognised to be a significant factor.[16] It has become possible to control the pulse duration and wavelength of the laser beam with the development of variable-square pulse technology. Less energy is lost by heat, due to the relatively higher energy of the shorter than longer pulse modes and, thus, the thermal effect that occurs in the tissue is minimized with the efficient and safe ablation.[19] It has also been mentioned that the enamel surface quality required for high bond strength can be obtained using the recently-developed QSP setting of the Er:YAG laser.[17,18] Our study supported this claim, in that QSP was found to be the most successful group in consideration of the clinical success rates.

The fissure-sealed teeth should be examined clinically and radiographically, at regular intervals.[32,33] It is documented that the retention failure of the sealants is at the highest rate immediately after application and in the first months.[2,34] There is a consensus that the success rate of sealants will increase with the regular controls.[3,35]

Kumar et al.[36] investigated the retention rates of a fissure sealant placed with 37% phosphoric acid and Er,Cr:YSGG laser etching at 3, 6, 9 and 12 months clinical follow-up. However, the difference between the groups was statistically non-significant. In another study, Durmus et al.[37] applied 35% phosphoric acid, Er:YAG laser and laser combined with acid, prior to sealant and evaluated the retention and new caries formation ratios at 3, 6, 9, 12 and 18 months. Based on the results, it was concluded that laser combined with acid etching exhibits superior retention ratios. Likewise, in our study, the laser groups presented improved clinical success rates, as similar to the previous research.[36,37]

The long-term success and retention of sealants depend on their resistance to microleakage.[5,10] Dye penetration is one of the most commonly used and reliable method for this purpose, detecting even small amounts of leakage.[38] Hence, it was implemented in the current study. The samples were also subjected to thermal cycling 10,000 times, at temperatures ranging from 5-55 °C, to simulate thermal changes in the mouth.[20]

Güçlü et al.[39] evaluated the microleakage of a sealant applied to the teeth that were conditioned with acid, Er:YAG laser or combined laser and acid. Microleakage assessments were realised using 0.5% fuchsin dye. There was no statistically significant difference between the groups, but lower microleakage values were seen in the group treated with the combined laser and acid. Similarly, there was no significant difference between the groups regarding microleakage, in the present study. In another report that evaluated the microleakage of the sealant applied to the enamel surface etched with the Er:YAG laser, the laser pre-treatment was deemed an alternative method of sealant application.[40]

One of the major limitations in prospective studies, as in our research, is the decrease in the recall rate of the patients when the follow-up period is prolonged. Therefore, we believe that further studies, in which more patients are included with longer follow-up periods, are needed.

This study assessed the methods intended for increasing the retention that affects the clinical success of sealants used commonly in pediatric dentistry. The most important factor in determining the retention of sealants before their application is the conditioning of the enamel surface. For this purpose, determination of the optimal energy level of the laser is considered to determine the clinical success. Within the limitations of this study, QSP exhibited superior results for the clinical success while Acid afforded the better microleakage outcomes. However, no statistical difference was observed among the groups.

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REFERENCES

1. Lam A. Increase in Utilization Of Dental Sealants. J Contemp Dent Pract. 2008;9:81-7.
2. Subramaniam P, Konde S, Mandanna DK. Retention Of A Resin-Based Sealant And A Glass Ionomer Used As A Fissure Sealant: A Comparative Clinical Study. J Indian Soc Pedod Prev Dent. 2008;26:114-20.
3. Beauchamp J, Caufield PW, Crall JJ, Donly K, Feigal R, Gooch B, Ismail A, Kohn W, Siegal M, Simonsen R. Evidence-Based Clinical Recommendations For The Use Of Pit-And-Fissure Sealants: A Report Of The American Dental Association Council On Scientific Affairs. J Am Dent Assoc. 2008;139:257-68.
4. Fejerskov O. Changing Paradigms In Concepts On Dental Caries: Consequences For Oral Health Care. Caries Res. 2004;38:182-91.
5. Cehreli SB, Gungor HC, Karabulut E. Er,Cr:YSGG Laser Pretreatment Of Primary Teeth For Bonded Fissure Sealant Application: A Quantitative Microleakage Study. J Adhes Dent. 2006;8:381-6.
6. Khan AA. The Permanent First Molar As An İndicator For Predicting Caries Activity. Int Dental J. 1994;44:623-7.
7. Pardi V, Sinhoreti MA, Pereira AC, Ambrosano GM, Meneghim MDE C. In Vitro Evaluation Of Microleakage Of Different Materials Used As Pit-And-Fissure Sealants. Braz Dent J. 2006;17:49-52.
8. Griffin SO, Oong E, Kohn W, Vidakovic B, Gooch BF, Bader J et al. The Effectiveness Of Sealants in Managing Caries Lesions. J Dent Res. 2008;87:169-74.
9. Cohen L, Sheiham A. The Use Of Pit And Fissure Sealants in The General Dental Service İn Great Britain And Northern Ireland. Br Dent J. 1988;165:50-3.
10. Simonsen RJ. Pit And Fissure Sealant: Review Of The Literature. Pediatr Dent. 2002;24:393-414.
11. Simonsen RJ. Retention And Effectiveness Of Dental Sealant After 15 Years. J Am Dent Assoc. 1991;122:34-42.
12. Bhatia M, Patel A, Shirol D. Evaluation Of Two Resin Based Fissure Sealants: A Comparative Clinical Study. J Indian Soc Pedod Prev Dent. 2012;30:227-30.
13. Yazici AR, Karaman E, Baseren M, Tuncer D, Yazici E, Unluer S. Clinical Evaluation Of A Nanofilled Fissure Sealant Placed With Different Adhesive Systems: 24-Month Results. Oper Dent. 2009;34:642-7.
14. Garcia-Godoy F, Harris ON, Helm DM. Primary Preventive Dentistry. In Harris ON, Godoy-Garcia F (ed), 6th edn. New Jersey, Pearson, 2009; pp 285-318.
15. Karandish M. The Efficiency Of Laser Application On The Enamel Surface: A Systematic Review. J Lasers Med Sci. 2014;5:108-14.
16. Lukac M, Primc NM, Pirnat S. Quantum Square Pulse Er:YAG Lasers For Fast And Precise Hard Dental Tissue Preparation. J LAHA. 2012;1:14–21.
17. Gutknecht N, Lukac M, Marincek M, Perhavec T, Kazic M. A Novel Quantum Square Pulse (QSP) Mode Erbium Dental Laser. J Laser Health Acad. 2011;1:15–21.
18. Lukac M, Marko M, Ladislav G. Super VSP Er:YAG Pulses For Fast And Precise Cavity Preparation. J Oral Laser Appl. 2004;4:171–3.
19. Grgurević J, Grgurević L, Miletić I, Karlović Z, Krmek SJ, Anić I. In Vitro Study Of The Variable Square Pulse Er:YAG Laser Cutting Efficacy For Apicectomy. Lasers Surg Med. 2005;36:347-50.
20. Gale MS, Darvell BW. Thermal cycling procedures for laboratory testing of dental restorations. J Dent. 1999;27:89-99.
21. Lavonius E, Kerosuo E, Kervanto-Seppälä S, Halttunen N, Vilkuna T, Pietilä I. A 13-Year Follow-Up Of A Comprehensive Program Of Fissure Sealing And Resealing İn Varkaus, Finland. Acta Odontol Scand. 2002;60:174-9.
22. Matalon S, Slutzky H, Mazor DY, Weiss EI. Surface Antibacterial Properties Of Fissure Sealants. Pediatr Dent. 2003;25:43-8.
23. De Luca-Fraga LR, Pimenta LA. Clinical Evaluation Of Glassionomer/ Resin-Based Hybrid Materials Used As Pit And Fissure Sealants. Quintessence In, 2001;32:463-8.
24. Rethman J. Trends in Preventive Care: Caries risk assessment and indications for sealants. J Am Dent Assoc. 2000;131 Suppl:8s-12s.

25. Celiberti P, Lussi A. Use Of A Self-Etching Adhesive On Previously Etched İntact Enamel And Its Effect On Sealant Microleakage And Tag Formation. J Dent. 2005;33:163–71.

26. Evans DJ, Matthews S, Pitts NB, Longbottom C, Nugent ZJ. A Clinical Evaluation Of An Er:YAG Laser For Dental Cavity Preparetion. Br Dent J. 18, 2000;8:677-9.

27. Simsek M, Yildiz E. Lasers In Pediatric Dentistry. Gaziantep Med J. 2014;20:113-9.

28. Dunn WJ, Davis JT, Bush AC. Shear Bond Strength And SEM Evaluation Of Composite Bonded To Er:YAG Laser-Prepared Dentin And Enamel. Dent Mater. 2005;21:616–24.

29. Gurgan S, Kiremitci A, Cakir FY, Gorucu J, Alpaslan T, Yazici E, Gutknecht N. Shear Bond Strength Of Composite Bonded To Er,Cr:YSGG Laser-Prepared Dentin. Photomed Laser Surg. 2008;26:495–500.

30. Marthaler TM. Changes in Dental Caries 1953-2003. Caries Res. 2004;38:173-81.

31. Firat E, Gurgan S, Gutknecht N. Microtensile Bond Strength Of An Etch And Rinse Adhesive To Enamel And Dentin After Er:YAG Laser Pretreatment With Different Pulse Duration. Lasers Med Sci. 2012;27:15-21.

32. Welbury R, Raadal M, Lygidakis NA. European Academy Of Paediatric Dentistry. EAPD Guidelines For The Use Of Pit And Fissure Sealants. Eur J Paediatr Dent. 2004;5:179-84.

33. Rushton VE, Horner K, Worthington HV. Factors İnfluencing The Frequency Of Bitewing Radiography in General Dental Practice. Community Dent Oral Epidemiol. 1996;24:272-6.

34. Dennison JB, Straffon LH, More FG. Evaluating Tooth Eruption On Sealant Efficacy. J Am Dent Assoc. 1990;121:610-4.

35. Feigal RJ, Donly KJ. The Use Of Pit And Fissure Sealants. Pediatr Dent. 2006;28:143-50.

36. Kumar G, Dhillon JK, Rehman F. A Comparative Evaluation Of Retention Of Pit And Fissure Sealants Placed With Conventional Acid Etching And Er,Cr:YSGG Laser Etching: A Randomised Controlled Trial. Laser Ther. 2016;25:291-8.

37. Durmus B, Giray F, Peker S, Kargul B. Clinical Evaluation Of A Fissure Sealant Placed By Acid Etching Or Er:YAG Laser Combined With Acid Etching. Oral Health Prev Dent. 2017;15:157-62.

38. Shortall AC. Microleakage, Marginal Adaptation And Composite Resin Restorations. Br Dent J. 1982;153:223-7.

39. Güçlü ZA, Dönmez N, Tüzüner T, Odabaş ME, Hurt AP, Coleman NJ. The Impact Of Er:YAG Laser Enamel Conditioning On The Microleakage Of A New Hydrophilic Sealant--Ultraseal XT Hydro. Lasers Med Sci. 2016;31:705-11.

40. Memarpour M, Kianimanesh N, Shayeghi B. Enamel Pretreatment With Er:YAG Laser: Effects On The Microleakage Of Fissure Sealant in Fluorosed Teeth. Restor Dent Endod. 2014;39:180-6.