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Laser Application in Remineralisation of Enamel after Interproximal Reduction: An In-vitro Scanning Electron Microscopic Study

Dentistry Section

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ABSTRACT

Introduction: Interproximal Reduction (IPR) or reproximation of enamel is of prime importance in orthodontics for correcting arch length-tooth size discrepancies. Despite its widespread application, IPR has been associated with adverse effects on the enamel surface.

Aim: The aim of this study was to investigate whether laser application, when combined with fluoride application, enhances the remineralisation potential of enamel.

Materials and Methods: This in-vitro study was conducted at Saveetha Institute of Medical and Technical Sciences (SIMATS), Chennai, Tamil Nadu, India, from December 2021 to July 2022. It involved 54 extracted teeth, divided into three groups of 18 samples each. Reproximation of 0.25 mm was performed on the proximal enamel surface of each tooth. Group 1 served as the control, group 2 was subjected to Fluor Protector, and group 3 received low-level laser therapy (Er,Cr:YSGG) after Fluor Protector application. Fluor Protector (Ivoclar Vivadent) was applied for seven days. Following the seven-day fluoride administration, a laser treatment using 0.75 W of power and 8.5 J/cm^2 of energy was applied for 20 seconds. Microhardness testing was conducted on the samples using a Vickers Hardness Tester. All specimens underwent surface topographic analysis with Scanning Electron Microscopy (SEM) and were evaluated for mineral content (% weight) using SEM-Energy Dispersive X-Ray analysis (EDX). Paired t-tests were performed to compare the pre- and post-microhardness values, while one-way Analysis Of Variance (ANOVA) test and Tukey's Post-hoc test were used to compare the microhardness values between the groups.

Results: The mean microhardness values recorded for group 1, group 2, and group 3 were 209.4±18.4 N/mm^2, 215.16±21.0 N/mm^2, and 233±18.05 N/mm^2, respectively. ANOVA test revealed a significant difference in microhardness values between group 1 and group 2 (p-value=0.004), as well as between group 1 and group 3 (p-value=0.001). The microhardness value was highest for group 3, followed by group 2 post-intervention. SEM analysis showed that laser-treated enamel surfaces were smoother, with well-coalesced enamel rods. The porous structure of enamel was lost due to fluoride deposition in group 2 and group 3, resulting in a smooth surface.

Conclusion: The combined application of fluoride and laser therapy demonstrated synergistic effects in remineralising the slenderised enamel. This simple, non-invasive technique may benefit patients undergoing IPR procedures by reducing the occurrence of dental caries.

Keywords: Fluoride, Low-level laser therapy, Reproximation, Surface topography

INTRODUCTION

IPR is a minimally invasive procedure that involves removing less than 0.5 mm of external enamel from the mesial and distal surfaces of the teeth. Also known as slenderisation or reproximation, IPR is an orthodontic treatment method used to reduce crowding [1]. This procedure is commonly performed to create additional arch length in patients with mild to moderate crowding, ensuring compliance with Bolton's ratio and avoiding extraction in borderline cases or when extraction therapy is undesirable. Despite its significant application in orthodontics, some authors have reported on the complications of IPR. Potential complications of enamel removal include hypersensitivity, irreversible damage to the dental pulp, increased plaque accumulation, and periodontal disorders [2-4].

Contrary to these findings, a clinical study that subjected enamel to IPR followed by polishing [5] claimed that IPR did not have a negative impact on the health of the treated teeth. This conclusion was supported by Jarjoura et al., who demonstrated that fluoridation after IPR resulted in only minor enamel changes [6]. There are three primary methods for reducing interproximal enamel: air-rotor stripping, diamond-coated discs, and abrasive metal strips [7]. However, it is important to note that employing improper IPR methods could result in a rough enamel surface, promoting plaque growth and creating a favourable environment for caries initiation and progression [7]. Consequently, despite the lack of evidence suggesting an increased risk of caries on treated teeth, the potential for abraded enamel to be more susceptible to caries formation has hindered the widespread acceptance of IPR.

To counteract the adverse effects of IPR, clinicians have utilised remineralisation techniques in the past and continue to do so. The remineralisation effects of fluoride varnish and casein phosphopeptide-amorphous calcium phosphate-nanocomplexes (CPP-ACP) on white spot lesions have been previously demonstrated and are the preferred methods for remineralisation after IPR [8-10]. In addition to established methods such as fluoride application, CPP-ACP, and the use of sound polishing to prevent caries formation, lasers have recently been investigated as an alternative for caries prevention and enhancing enamel's resistance to acids [11,12]. Laser application has been reported to improve fluoride uptake for remineralisation [13]. Studies evaluating the effect of laser irradiation followed by fluoride application on remineralisation of deciduous teeth have been conducted [14-17]. However, to the best of the authors' knowledge, no studies have been conducted to date to assess its efficacy in permanent teeth, particularly after IPR. Therefore, the present study was undertaken to validate a novel laser treatment method aimed at enhancing the remineralising effect after fluoride application following completion of IPR.

MATERIALS AND METHODS

The present in-vitro study was conducted at Saveetha Institute of Medical and Technical Sciences (SIMATS), Chennai, Tamil Nadu, India, during the period of December 2021 to July 2022. Ethical approval was obtained from the Institutional Ethical Committee (HEC/SDC/ORTHO-2003/21/017).

Inclusion and Exclusion criteria: A total of 54 healthy mandibular incisor teeth were extracted for orthodontic or periodontal reasons. These teeth were free of restorations, cracks, and attrition, and were included as study samples. Any teeth presenting interproximal and/or cervical caries, restorations, or excessive wear were excluded. The teeth were then mounted in an acrylic block measuring 1.5”×0.5” [Table/Fig-1]. Subsequently, the teeth were stored in distilled water for up to one month.

Sample size calculation: The sample size selection was calculated based on a reference study that compared Carbon Dioxide (CO_2) and diode laser for enamel remineralisation in conjunction with fluoride-containing components in primary teeth [18]. G*Power software version 3.0 was used for the sample calculation, and the power of the study was 99%.

[Table/Fig-1]: Tooth samples embedded in acrylic.

Study Procedure

The 54 samples were randomly divided into three groups, with 18 samples in each group. Enamel reproximation was performed on both the mesial and distal surfaces of each tooth [Table/Fig-2]. Interproximal enamel reduction of 0.25 mm was carried out using a J316SF bur 0.3 mm (Strauss and Co.) under water-cooling. The bur was replaced with a new one after every 20 reduction procedures. All procedures were performed by the same skilled clinician. The identical IPR protocol was followed in all three groups:

Group 1: Specimens were stored in artificial saliva without being subjected to laser or Fluor Protector.

Group 2: Fluor Protector only was used.

Group 3: Fluor Protector + Er, Cr: YSGG laser.

[Table/Fig-2]: Sample subjected to proximal reduction.

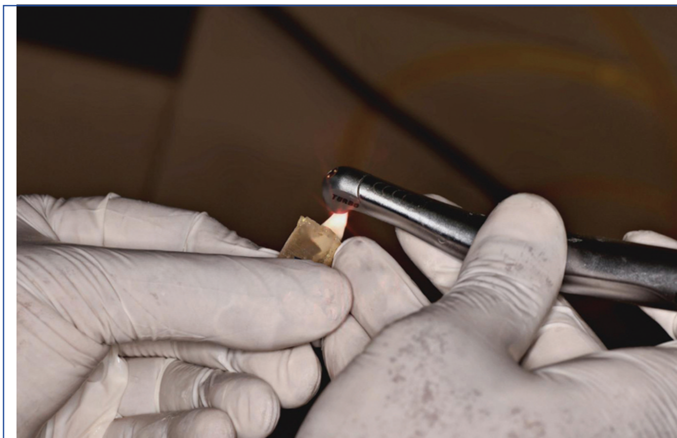
Fluoride varnish (Fluor Protector) was applied to the interproximal surfaces of group 2 [Table/Fig-3]. The varnish was applied to previously cleansed and dried tooth surfaces following the manufacturer’s instructions. Before being placed in artificial saliva, the varnish was allowed to dry for a minute. The remineralisation procedure was repeated twice daily for seven days, following standard protocols [11]. The samples were kept in artificial saliva between the remineralisation cycles.

[Table/Fig-3]: Samples subjected to application of Fluor Protector varnish.

Microhardness estimation pre-intervention: A Vickers hardness tester (Matsuzawa MMT7, Matsuzawa SEIKI Co., Ltd., Tokyo, Japan) with a 200 g load was used to create indentations on the enamel surface and measure the surface hardness of each sample. After allowing the loaded diamond to settle and rest on the enamel surface for 15 seconds, the Vickers hardness number was recorded. This process was carried out for all the samples before the intervention, and the microhardness data were documented.

Er,Cr:YSGG laser irradiation: In group 3, fluoride was applied following the standard protocol used in group 2. After fluoride application, the samples were irradiated with an Erbium, chromium-doped yttrium, scandium, gallium, and garnet (Er,Cr:YSGG) laser (Biolase, Waterlase) for 30 seconds on each surface. The operating tip (device tip-sapphire with a tip length of 9 mm and a diameter of 600 µm) was inserted into the handpiece according to the manufacturer’s recommendations. The operator wore wavelength-specific protective eyeglasses. The laser tip was inspected for any scratches or contamination.

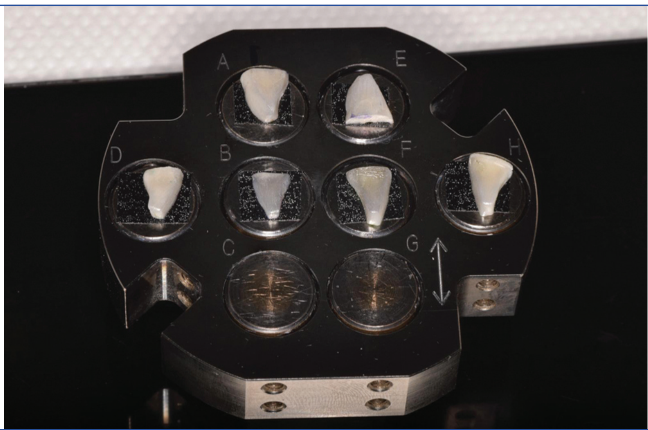
The device was calibrated to operate at a power of 0.75 W, an energy density of 8.5 J/cm, an energy per pulse of 12.5 mJ, a pulse width of 140 µs in H mode, and a frequency of 20 Hz without water cooling [19]. To prevent drying from affecting the viability of the samples, teeth were retrieved from artificial saliva, allowed to air dry for three seconds, and then immediately irradiated with the laser for 30 seconds on each surface [20]. The operator wore wavelength-specific protective glasses, and the laser’s point was kept parallel to the tooth’s surface at a standard distance of approximately 1 mm [Table/Fig-4].



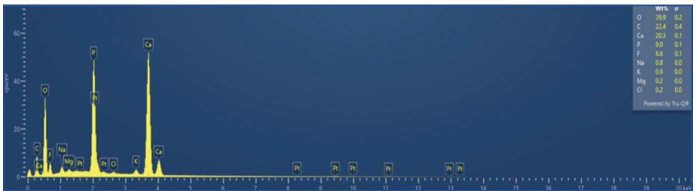
[Table/Fig-4]: Samples subjected to Er,Cr: Ysgg laser application.

Microhardness estimation post-intervention: The microhardness test was repeated for all the samples after the remineralisation process, and the values were recorded. Three indentations were made on each sample, and the average was calculated as the final value.

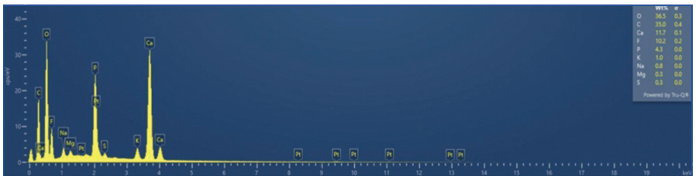
Scanning Electron Microscopy (SEM) and EDX: All specimens underwent surface topographic analysis using SEM and were also evaluated for mineral content (% weight) using SEM-EDX. The samples were sectioned, with the root portion kept separate. The crowns were then sectioned sagittally, splitting them into mesial and distal proximal surfaces. The samples were subsequently sputter coated with platinum [Table/Fig-5]. Pictures were taken at 1000× and 2000× magnification for all three groups [21]. Elemental analysis was performed for group 2 and group 3 to determine the amount of fluoride deposition [Table/Fig-6-8]. This analysis was not conducted for the control group since the samples were not subjected to fluoride application.



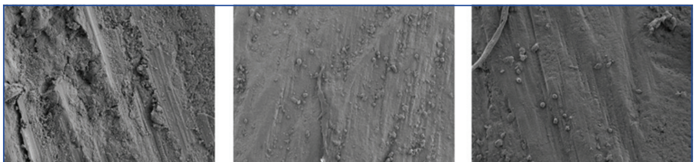
[Table/Fig-5]: Sectioned samples sputter coated with Platinum.



[Table/Fig-6]: EDX analysis of a sample from group 2 showing 6.6 wt% of Fluoride.



[Table/Fig-7]: EDX analysis of a sample from group 2 showing 10.2 wt% of Fluoride.



[Table/Fig-8]: Surface topographic analysis of samples from group 1, group 2 and group 3, respectively (from left to right).

STATISTICAL ANALYSIS

The Statistical Package for the Social Sciences (SPSS), software version 23.0 by SPSS Inc., Chicago, IL, USA, was utilised for statistical analysis. The normality of the data was assessed using the Shapiro-Wilk test. Since the data was found to be parametric with a p-value >0.05, paired t-tests were conducted to compare the pre and post microhardness values. Furthermore, one-way ANOVA test and Tukey's post-hoc tests were performed to compare the microhardness values among the groups, with a significance level set at p=0.05.

RESULTS

Microhardness: The control group (group 1 stored in artificial saliva) had a mean microhardness value of 209.4±18.4 N/mm^2. For group 2 (application of Fluor Protector only), the mean hardness values were 215.16±21.0 N/mm^2. In group 3 (Fluor Protector + Er, Cr:YSGG), the mean hardness value was 233±18.05 N/mm^2. Paired t-tests revealed significant differences in the microhardness values before and after fluoride application in group 2 (p-value=0.008) and group 3 (p-value <0.001).

ANOVA and Tukey's post-hoc tests for multiple comparisons showed significant differences in the microhardness values between group 1 and group 2 (p-value=0.004), as well as between group 3 and group 1 (p-value <0.001). However, there were no significant differences between group 2 and group 3, according to the statistical tests [Table/Fig-9-11]. ANOVA test and Tukey's post-hoc tests were performed to analyse significant differences in the microhardness values between the groups at T0 (post IPR) and T1 (post remineralisation).

Component	Wt%
Water, xylitol and thickener	97
Potassium fluoride (1450 ppm of fluoride)	0.45
Calcium glycerophosphate	>1.0
D-panthenol	>1.0
Additives, flavouring agents, stabilisers	>2.0

[Table/Fig-9]: Composition of Fluor Protector gel.

Group	T0 (post IPR) N/mm^2	T1 (Post remineralisation) N/mm^2	Mean difference	p-value
Group 1 (Control-not subjected to Fluor Protector)	201.3±37.5	209.4±18.4	8.1	0.1
Group 2	182.16±29.2	215.16±21.0	33.1	0.008
Group 3	192.±34.1	233±18.05	41.04	<0.001

[Table/Fig-10]: Dependent t-test performed to compare the microhardness values pre and post remineralisation treatment showing significant p-values.

Time period	Group		p-value
T0	Group 1	Group 3	0.1
		Group 2	0.249
	Group 2	Group 1	0.5
		Group 3	0.1
	Group 3	Group 1	0.1
		Group 2	0.249
T1	Group 1	Group 3	<0.001*
		Group 2	0.004*
	Group 2	Group 1	0.004*
		Group 3	0.249
	Group 3	Group 1	<0.001*
		Group 2	0.249

[Table/Fig-11]: ANOVA test and Tukey's post-hoc tests performed to analyse the significant differences in the microhardness values between the groups at T0 and T1.

SEM/EDX: SEM analysis showed that the laser-treated enamel surfaces were smoother, with well-coalesced layers of enamel rods. The porous enamel structure was lost due to fluoride deposition in group 2 and group 3, resulting in a smooth surface. In contrast, a rough, coarse, and porous enamel surface was observed in group 1. The highest accumulation of 10.2% atomic percent of fluoride was found in the group treated with Fluor Protector after laser treatment, whereas it was 6.6% in group 2, indicating increased fluoride absorption in group 3.

DISCUSSION

Low-level laser therapy has been shown to have various benefits in orthodontics, such as increasing the rate of tooth movement and increasing the pain threshold. It has also been found to be beneficial in inducing enamel remineralisation. The aim of this study was to analyse whether the addition of Er, Cr: YSGG laser, along with fluoride application, would have an additive beneficial effect on the remineralisation of reduced enamel surfaces during interproximal enamel reduction (IPR).

Clinically applied fluoridated varnishes work by adhering to the enamel surfaces and forming a layer of Calcium Fluoride (CaF_2) [22]. This CaF_2 layer acts as a physical barrier against acidic environments created by sugars and also acts as a reservoir of calcium and fluoride ions at lower pH levels [22,23]. This enhances the enamel's potential for remineralisation. Laser treatment has been investigated as an alternative for caries prevention and has been shown to enhance enamel's resistance to acids when used in conjunction with fluoride application. Since IPR in orthodontic treatment causes enamel loss, the study aimed to compare the effectiveness of laser treatment combined with fluoride treatment on the degree of remineralisation.

The results of the study showed that the use of Er, Cr: YSGG laser irradiation along with fluoride application significantly increased the enamel microhardness of permanent teeth compared to the group that only underwent fluoride application. Additionally, the increased fluoride content in group 3 supports the theory of the formation of a calcium fluoride (CaF_2) layer, resulting in the formation of a more permeable enamel. It was challenging to compare the findings of this study with previous studies since none of them used fluoride in conjunction with lasers on reduced interproximal enamel surfaces. However, the findings of this study are consistent with earlier research [13,15,18,24,25] that concluded that combining Er, Cr: YSGG and CPP-ACP significantly increased microhardness, regardless of the sequence of laser application (laser first or CCP-ACP first).

Anaraki SN et al. and Kaur T compared the resistance of deciduous teeth to an acidic environment using two different types of lasers. The surface microhardness values were significantly higher for the CO2 laser compared to the Er, Cr:YSGG laser [26,27]. SEM analysis showed that the CO2 laser-radiated enamel surfaces had a rough, fractured appearance, which could serve as plaque-retentive zones. In contrast, the Er, Cr:YSGG group displayed a smooth, shiny enamel surface without any fractures, indicating resistance to acidic dissolution [26,27].

The effectiveness of lasers in enhancing remineralisation is linked to their wavelength, which is in line with the absorption peak of carbonated hydroxyapatite, the main component of tooth enamel (85%). The energy generated by the Er, Cr:YSGG laser is absorbed and converted to heat without causing injury to underlying tissues. This leads to ultrastructural and chemical changes in the irradiated enamel, resulting in enhanced resistance to acid dissolution [28,29]. As suggested by Chin-Ying SH et al., this effect is not only due to laser-induced enamel alterations but also to an increase in fluoride uptake in the irradiated enamel [30]. These two mechanisms are believed to contribute to the potential remineralisation effects.

Other mechanisms that can improve remineralisation include the formation of numerous spherical precipitates on tooth surfaces when laser-fluoride application is used. These precipitates morphologically resemble calcium-fluoride-like deposits and act as a reservoir to replenish fluoride, allowing for higher fluoride uptake [31]. Another mechanism is the uptake of fluoride in the form of firmly bound fluoride [30].

Limitation(s)

The present study had several limitations. Firstly, it was conducted as an in-vitro study with a limited sample size. Therefore, the results may not fully reflect the outcomes in clinical settings. Further in-vivo/in-vitro studies with larger sample sizes are necessary to accurately quantify the Er, Cr:YSGG parameters that yield the best results using various assessment methods.

Additionally, it is important to establish criteria for determining the optimal laser administration sequence. Further investigation is needed to determine whether the laser should be used before, after, or concurrently with fluoride treatment. Each sequence may have different clinical significance and outcomes, which should be explored in future studies.

CONCLUSION(S)

Low-level laser therapy has become increasingly important in orthodontics, particularly in accelerating tooth movement. Another emerging area of interest is the effect of laser therapy on enamel remineralisation. The results of the present study concluded that the combination of laser therapy and fluoride application had an increased potential for remineralisation. This simple and non-invasive procedure could be beneficial for patients undergoing interproximal enamel reduction (IPR), as it can cause significant enamel loss and increase the risk of caries formation. Long-term studies are needed to compare the efficacy of single versus multiple surface treatment applications. Further investigations are also crucial to compare the effectiveness of Er, Cr:YSGG laser therapy in preventing demineralisation. Additionally, studies should be conducted to evaluate different combinations of low-level laser therapy and various remineralising agents.

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Manufacturer's name: Fluor Protector, Ivoclar Vivadent, Bengaluru, Karnataka, India.

REFERENCES

[1] Pindoria J, Fleming PS, Sharma PK. Inter-proximal enamel reduction in contemporary orthodontics. Br Dent J. 2016;221(12):757-63.
[2] Vicente A, Ruiz AJO, Paz BMG, López JG, Bravo-González LA. Efficacy of fluoride varnishes for preventing enamel demineralization after interproximal enamel reduction. Qualitative and quantitative evaluation [Internet]. PLOS ONE. 2017;12:e0176389. Available from: http://dx.doi.org/10.1371/journal.pone.0176389.
[3] Zachrisson BU, Nyøygaard L, Mobarak K. Dental health assessed more than 10 years after interproximal enamel reduction of mandibular anterior teeth. Am J Orthod Dentofacial Orthop. 2007;131(2):162-69.
[4] Zachrisson BU, Minster L, Øgaard B, Birkhed D. Dental health assessed after interproximal enamel reduction: Caries risk in posterior teeth [Internet]. American Journal of Orthodontics and Dentofacial Orthopedics. 2011;139(1):90-98. Available from: http://dx.doi.org/10.1016/j.ajodo.2010.09.002.
[5] Sheridan JJ, Ledoux PM. Air-rotor stripping and proximal sealants. An SEM evaluation. J Clin Orthod. 1989;23(12):790-94.
[6] Jarjoura K, Gagnon G, Nieberg L. Caries risk after interproximal enamel reduction [Internet]. Am J Orthod Dentofacial Orthop. 2006;130(1):26-30. Available from: http://dx.doi.org/10.1016/j.ajodo.2004.08.024.
[7] Lapenaite E, Lopatiene K. Interproximal enamel reduction as a part of orthodontic treatment. Stomatologija. 2014;16(1):19-24.
[8] Bayrak S, Tuloglu N, Bicer H, Sen Tunc E. Effect of fluoride varnish containing CPP-ACP on preventing enamel erosion. Scanning. 2017;2017:1897825.
[9] Zawaideh FI, Owais AI, Mushtaha S. Effect of CPP-ACP or a Potassium Nitrate Sodium Fluoride Dentifrice on Enamel Erosion Prevention [Internet]. J Clin Pediatr Dent. 2017;41(2):135-40. Available from: http://dx.doi.org/10.17796/1053-4628-41.2.135.
[10] Rajendran R, Kunjusankaran RN, Sandhya R, Anilkumar A, Santhosh R, Patil SR. Comparative evaluation of remineralizing potential of a paste containing bioactive glass and a topical cream containing casein phosphopeptide-amorphous calcium phosphate: An in-vitro study. Pesquisa Brasileira Em Odontopediatria e Clínica Integrada. 2019;19(1):01-10. Doi: 10.4034/pboci.2019.191.61.
[11] Bharti M, Nagar P, Aminah M, Singh P. Effect of laser and fluoride application for remineralization of the carious lesion: A polarized microscopic study. Int J Contemp Med Res. 2017;4(2):489-92.

[12] Asl-Aminabadi N, Najafpour E, Samiei M, Erfanparast L, Anoush S, Jamali Z, et al. Laser-Casein phosphopeptide effect on remineralization of early enamel lesions in primary teeth. J Clin Exp Dent. 2015;7(2):e261-67.
[13] Yilmaz N, Baltaci E, Baygin O, Tüzüner T, Ozkaya S, Canakci A. Effect of the usage of Er,Cr:YSGG laser with and without different remineralization agents on the enamel erosion of primary teeth. Lasers Med Sci. 2020;35(7):1607-20.
[14] De Sant'Anna GR, Paleari GSL, Duarte DA, Brugnera A Jr, Soares CP. Surface morphology of sound deciduous tooth enamel after application of a photo-absorbing cream and infrared low-level laser irradiation: An in-vitro scanning electron microscopy study. Photomed Laser Surg. 2007;25(6):500-07.
[15] Elwardani GE, Harhash TAH, Zaky AA. Effect of Er,Cr:YSGG on remineralization using CPP-ACPF (MI-Paste Plus) after enamel erosion caused by carbonated soft drink in primary teeth: In-vitro study. Open Access Maced J Med Sci. 2019;7(7):1184-92.
[16] Nair MJ, Rao A, Jayaprakash K, Natarajan S, Kumblekar V, Suprabha BS. Effectiveness of laser fluorescence-based device in detecting the extent of re-mineralisation in primary teeth compared to the conventional method: An study. Indian J Dent Res. 2022;33(3):292-96.
[17] González-Sotelo A, Rodríguez-Vilchis LE, Contreras-Bulnes R. Effect of Er:YAG and fluoride varnishes for preventing primary enamel demineralisation. Oral Health Prev Dent. 2019;17(4):317-21.
[18] Memarpour M, Soltanimehr E, Sattarahmady N. Efficacy of calcium- and fluoride-containing materials for the remineralization of primary teeth with early enamel lesion [Internet]. Microsc Res Tech. 2015;78(9):801-06. Available from: http://dx.doi.org/10.1002/jemt.22543.
[19] Ana PA, Tabchoury CPM, Cury JA, Zezell DM. Effect of Er,Cr:YSGG laser and professional fluoride application on enamel demineralization and on fluoride retention. Caries Res. 2012;46(5):441-51.
[20] De Benedetto MS, Morais CC, Novaes TF, de Almeida Rodrigues J, Braga MM, Mendes FM. Comparing the reliability of a new fluorescence camera with conventional laser fluorescence devices in detecting caries lesions in occlusal and smooth surfaces of primary teeth. Lasers Med Sci. 2011;26(2):157-62.
[21] Jayarajan J, Janardhanam P, Jayakumar P, Deepika. Efficacy of CPP-ACP and CPP-ACPF on enamel remineralization-An in-vitro study using scanning electron microscope and DIAGNOdent® [Internet]. Indian J Dent Res. 2011;22(1):77-82. Available from: http://dx.doi.org/10.4103/0970-9290.80001.
[22] Ganss C, Schlueter N, Klimek J. Retention of KOH-soluble fluoride on enamel and dentine under erosive conditions-A comparison of in-vitro and in situ results [Internet]. Arch Oral Biol. 2007;52(1):09-14. Available from: http://dx.doi.org/10.1016/j.archoralbio.2006.07.004.
[23] Sancakli HS, Sar Sancakli H, Austin RS, Al-Saqabi F, Moazzez R, Bartlett D. The influence of varnish and high fluoride on erosion and abrasion in a laboratory investigation [Internet]. Aus Dent J. 2015;60(1):38-42. Available from: http://dx.doi.org/10.1111/adj.12271.
[24] Tulumbaci F, Oba A. Efficacy of different remineralization agents on treating incipient enamel lesions of primary and permanent teeth [Internet]. J Conserv Dent. 2019;22(3):281-86. Available from: http://dx.doi.org/10.4103/jcd.jcd_509_18.
[25] Natarajan PV, Jain R. In-vitro comparative evaluation of enamel microhardness remineralised with Diode and Er,Cr:YSGG lasers along with CPP-ACP remineralising agent. J Clin Diagn Res. 2021;15(12):ZC29-ZC33. Doi: 10.7860/JCDR/2021/51546.15790.
[26] Anaraki SN, Serajzadeh M, Fekrazad R. Effects of laser-assisted fluoride therapy with a CO2 laser and Er,Cr:YSGG laser on enamel demineralization. Pediatr Dent. 2012;34(4):e92-96.
[27] Kaur T. SEM evaluation of enamel surface changes and enamel microhardness around orthodontic brackets after application of CO2 Laser, Er,Cr:YSGG laser and fluoride varnish: An in vivo study [Internet]. J Clin Diagn Res. 2017;11(9):ZC59-ZC63. Available from: http://dx.doi.org/10.7860/jcdr/2017/30292.10603.
[28] Jabin Z, Nasim I, Vishnu Priya V, Agarwal N. Quantitative analysis and effect of SDF, APF, NaF on demineralized human primary enamel using SEM, XRD, and FTIR. Int J Clin Pediatr Dent. 2021;14(4):537-41.
[29] Stern RH, Sognnaes RF. Laser inhibition of dental caries suggested by first tests in vivo. J Am Dent Assoc. 1972;85(5):1087-90.
[30] Esteves-Oliveira M, Zezell DM, Ana PA, Yekta SS, Lampert F, Eduardo CP. Dentine caries inhibition through CO(2) laser (10.6 μm) irradiation and fluoride application, in-vitro. Arch Oral Biol. 2011;56(6):533-39.
[31] Chin-Ying SH, Xiaoli G, Jisheng P, Wefel JS. Effects of CO2 laser on fluoride uptake in enamel. J Dent. 2004;32(2):161-67. Doi: 10.1016/j.jdent.2003.10.004.

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