Research Article

# Influence of 9.3 µm $CO_2$ and Er:YAG laser preparations on marginal adaptation of adhesive mixed Class V composite restorations with one component universal adhesive

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**Abstract: Purpose:** To examine the marginal adaptation in enamel and dentin of mixed Class V saucer shaped restorations where cavities were prepared by two different lasers. **Methods:** A handpiece-integrated Er:YAG laser @ 4.5 W, 300 mJ, 15 Hz (LiteTouch III) and a novel $CO_2$ laser @ 12.95 W, 19.3 mJ, 671 Hz (Solea 9.3 µm). Diamond bur preparation with a 25 µm diamond bur (Intensiv) in a red contra angle at high speed under water spray cooling served as the control. Eight cavities per group were readied and restored under simulation of dentin fluid with a one bottle universal adhesive (One Coat 7 Universal) and a nanohybrid resin composite (Everglow), applied in two layers. For every preparation technique, the adhesive system was applied in the selective-etch and the self-etch mode, resulting in six experimental groups. Marginal analysis was performed immediately after polishing and after simultaneous thermal (5-50°C, 2 minutes each) and mechanical (max. 49 N; 200,000 cycles) loading by using a SEM (×200 magnification). **Results:** Significant differences were found for all groups - except groups 2 and 5 - between initial and terminal results and between the groups as well ($P< 0.05$, 2-way ANOVA with Fisher's post-hoc test). The bur prepared group with selective-etch technique showed the best overall results after loading, followed by Er:YAG prepared self-etch group and $CO_2$-prepared selective-etch group. (*Am J Dent* 2021;34:31-38).

**Clinical significance:** By using a universal one-component adhesive system, marginal adaptation in enamel and in dentin depended on the preparation method and on the adhesive's application technique as well. When using lasers, Er:YAG in self-etch mode and $CO_2$ 9.3 µm in selective-etch mode total marginal adaptation showed results which were comparable to conventional bur preparation with selective-etch technique.

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## Introduction

Lasers are accepted by practitioners and patients as the routine technique for increased number of indications as they may provide an operation field with low microbiological contamination, reduction of bleeding, rapid and eventless wound healing as well as time savings. Lasers may also be used for hard tissue preparation, thus avoiding some drawbacks of rotary instruments such as the noise and vibration which may cause discomfort, pain and subsequently dental fear to patients, or the creation of a smear layer which may negatively influence adhesion.[1-8]

Laser ablation in dental hard tissues can be obtained with wavelengths strongly absorbed either in water (organic matrix of enamel and dentin), in hydroxyapatite of the mineral matrix $[Ca_5(PO_4)_3(OH)]$ or in both.[9] For more than a decade, this goal has best been achieved by an Er:YAG laser, because its wavelength is highly absorbed in water and moderately absorbed in hydroxyapatite. Recently, a novel 9.3 µm $CO_2$ laser has been introduced to the market, which, besides working on soft tissues, is claimed to be indicated for cavity preparations as well. In contrast to an Er:YAG, this laser's wavelength is highly absorbed in hydroxyapatite and moderately absorbed in water.[3,5,9,10]

Currently, cavity preparation is usually followed by a resin composite restoration, which is bonded to the tooth structure with an adhesive system.[11] One component universal adhesives are becoming more popular as they simplify and accelerate bonding procedures.[12,13] Compared to multi-step etch-and-rinse systems, one component universal adhesives are less technique sensitive and indicated for a wide variety of restorative procedures and adhesion strategies.[13-19]

Even if some studies[20,21] reported microtensile and microleakage results, very little is known about the quality of marginal adaptation in enamel and dentin of resin composite restorations placed in Er:YAG laser prepared cavities and bonded with a universal adhesive system. To the authors' knowledge, no single study has been published on the dentin/enamel marginal adaptation of resin composite restorations placed after preparation with a 9.3 µm $CO_2$ laser.

This study evaluated and compared the surface effects of an Er:YAG laser and a novel 9.3 µm $CO_2$ laser on enamel and dentin micromorphology, as well as the quality of marginal adaptation of direct adhesive composite restorations in laser-treated mixed Class V cavities. The underlying null hypothesis stated that there were no micromorphological differences on the enamel and dentin surfaces as well as no significant differences in marginal adaption between mechanical bur, Er:YAG and 9.3 µm $CO_2$ laser in both self-etch and selective enamel etch technique.

## Materials and Methods

For this study, 48 intact, caries-free human molars, which were stored immediately after extraction in 0.1% thymol solution were cleaned (scaler, ultrasound) and brushed with a rotary brush with toothpaste (Signal[a] RDA 50) in a handpiece. They were randomly assigned to six experimental groups of

Table 1. Ablation parameters: Situation A preparation parameters, situation B finishing/conditioning of cavities and filling procedures: Light-curing 1 (after application of adhesive system), light-curing 2 (after application of each composite layer).

| | Group 1 | Group 2 | Group 3 | Group 4 | Group 5 | Group 6 |
|---|---|---|---|---|---|---|
| Preparation tool | Handpiece | | $CO_2$ laser | | Er:YAG laser | |
| Wavelength | - | | 9.3 µm | | 2.94 µm | |
| Tip/spot characterization (name, size) | Intensiv Suisse Football (white ring, 25 µm) | | 1.25 mm | | | |
| 1.3 × 14 mm | | | | | | |
| Working distance | In contact | | 4-15 mm (ca. 10 mm) | | 1-2 mm | |
| Spray amount | 50 % | | A + B: 100 % | | A: 5<br>B: 3 | |
| Pulse energy | - | | A: 19.3 mJ<br>B: 6.11 mJ | | A: 300 mJ<br>B: 50 mJ | |
| Pulse frequency | - | | 671 Hz | | 15 Hz | |
| Power | - | | A: 50 % -> 1.95 W<br>B: 20 % -> 4.1 W | | A: 4.5 W<br>B: 0.75 W | |
| Finishing/conditioning | $H_3PO_4$ | | 9.3 µm | 9.3 µm + $H_3PO_4$ | 2.94 µm | 2.94 µm + $H_3PO_4$ |
| Etching | Selective-etch ($H_3PO_4$ 15 sec. on enamel) | Self-etch | Self-etch ($H_3PO_4$ 15 sec. on enamel) | Selective-etch | Self-etch | Selective-etch ($H_3PO_4$ 15 sec. on enamel) |
| Bonding | One Coat 7 Universal (20 sec.) | | | | | |
| Light-curing 1 | 1 × 20 sec. (Valo 1,000 mW/cm$^2$) | | | | | |
| Filling | 2 layers Brilliant EverGlow | | | | | |
| Light-curing 2 | 2 × 20 sec. (Valo 1,000 mW/cm$^2$) | | | | | |

similar size (Table 1). To prepare the teeth for the experiments, their apices were sealed with an adhesive-system (OptiBond FL[b]) and the roots were fixed in the center of custom-made specimen holders using a cold polymerizing resin (Technovit 4071[c] resin cold curing). For the simulation of dentin fluid, horse serum (donor horse serum[d]) was 1:3 diluted with phosphate buffered saline[d] (PBS). To feed the diluted horse serum into the pulpal chamber, a cylindrical hole was drilled at the side surface of the tooth at the CEJ and a metal tube (needle Terumo[e] with a diameter of 1.2 mm) was inserted and glued with an adhesive system (OptiBond FL). The tube was connected to a flexible silicone hose to enable a preparation and filling of the teeth under the simulation of the dentin fluid flow. On each tooth, one saucer-shaped Class V cavity was prepared at the dentin-enamel junction under up to ×20 stereomicroscope magnification with a 2.94 µm Er:YAG laser (LiteTouch III[f]), a 9.3 µm $CO_2$ laser (Solea 9.3µm[g]) and a mechanical 25 µm diamond bur[h] (Table 1). The groups prepared with burs served as the positive control groups.

Prepared teeth were air-dried before bonding. Adhesive procedures for all groups used a one component universal adhesive (One Coat 7 Universal[i]), following the manufacturer's instructions either for the selective-etch or the self-etch protocol. In the latter case, phosphoric acid was applied on enamel for 15 seconds, rinsed with water for 20 seconds and air-dried for 2 seconds. In both adhesive protocols, the universal adhesive was slightly rubbed into the cavity surface for 20 seconds, softly air dried for 5 seconds with oil-free compressed air and light cured for 20 seconds (Valo[j] cordless; standard power 1,000 mW/cm$^2$). Subsequently, the cavities were filled with a nanohybrid resin composite (Coltène Brilliant EverGlow,[i] Shade A2/B2,) in two layers and light-cured for 20 seconds (Table 1). For finishing and polishing, flexible discs (SofLex[k]) were used and for concave areas, rubber polishing tips (Brownie and Greenie[l]) were used under up to ×20 stereo microscope magnification. Impressions with a polyvinylsiloxane (addition-type) impression material (President[i] light body) were taken after brush-cleaning the surface with toothpaste (Signal, RDA 50).[22,23]

The restored teeth were then subjected to repeated thermal and mechanical stresses in a chewing machine, under constant simulation of dentin fluid flow (mechanical stress ×200,000 with maximum 49 N and thermal stress between 5°C and 50°C).[22,23]

After loading, replicas were taken again following the above described procedures.

For the evaluation of marginal adaptation, replicas before and after aging were poured out with an epoxy resin (EpoFix[m]), gold sputtered and subjected to a quantitative marginal analysis in a scanning electron microscope under ×200 magnification (Zeiss Gemini - Sigma 300 VP[n]).

After checking the usual normality assumptions of data using the Kolmogorov and Smirnov and the Shapiro-Wilk tests, group differences were tested using a repeated measures ANOVA followed by Fisher's LSD post-hoc tests for pairwise mean comparisons.

## Results

The first part of this study was an analysis of dental hard tissue micromorphology (Figs. 1-3, A-D). While preparing the cavities, the first differences became clear regarding the time for ablation and the macromorphology of the dental surfaces. Ablation and finishing were faster with 9.3 µm $CO_2$ laser (00:01:13) followed by conventional bur (00:01:39) and Er:YAG laser Light-Touch (00:04:25). The relatively long period of time with the Er:YAG laser was due to the finishing of the deep and rough ablation patterns with less powerful laser settings. The whole surface of these cavities appeared whitish

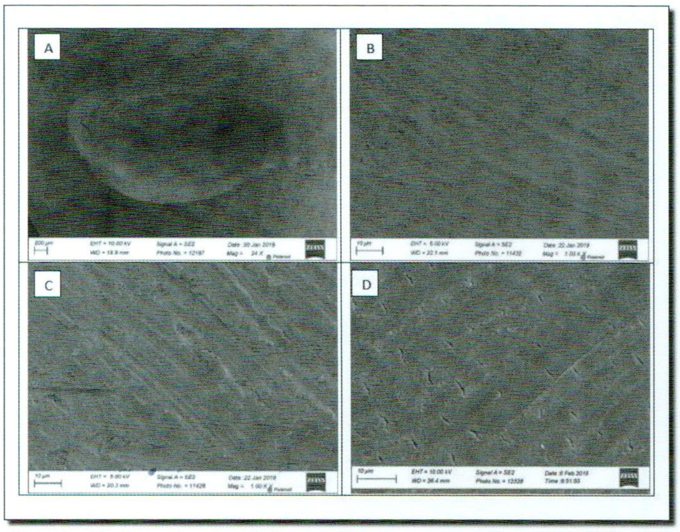

Fig. 1. Cavity micromorphology after bur preparation (FE-SEM): **A.** Overview (×24), **C.** Enamel (×1,000); **B/D.** In dentin (×1,000 and ×5,000).

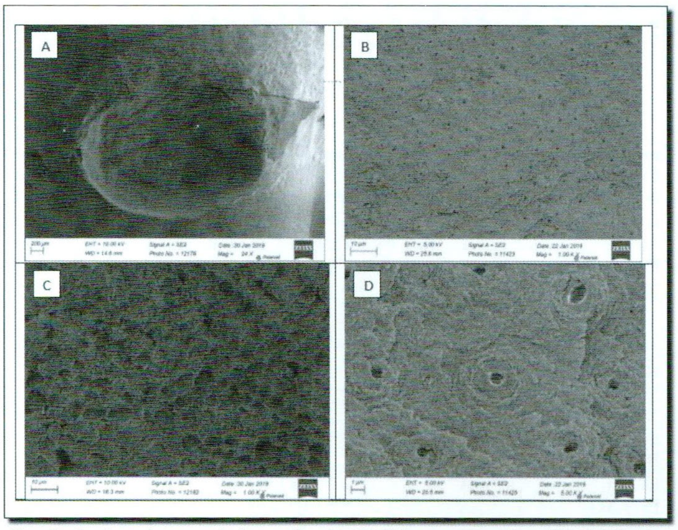

Fig. 3. Cavity micromorphology after Er:YAG-laser preparation (FE-SEM): **A.** Overview (×24), **C.** Enamel (×1,000) and **B/D.** In dentin (×1,000 and ×5,000).

opaque in comparison to the other two types. Figures 1-3, especially bur prepared cavities (Fig. 1) showed very clear and sharp margins. But also, $CO_2$ laser ablation left a well-defined cavity (Fig. 2). They differ from the irregular outline of Er:YAG prepared cavities (Fig. 3). This micromorphological surface was quite rough and showed honeycomb patterns in enamel in contrast to the other devices used in this study (Figs. 1C, 2C, 3C). The bur prepared cavities show quite a homogenous and smooth surface with flat grinding facets; $CO_2$ laser preparations are slightly more uneven in dentin due to melted "drops", which leave a hilly surface, whereas the enamel surface is quite smooth and glazed (Fig. 2C). Dentin tubuli were partly occluded by the melting drops, whereas they were widely open after Er:YAG laser preparation and completely covered by smear layer after bur ablation (Figs. 1 B,D, 2 B,D, 3 B,D).

The second part of this study was an analysis of quality of marginal adaptation regarding percentages of continuous margin of adhesive Class V composite restorations before and after aging.

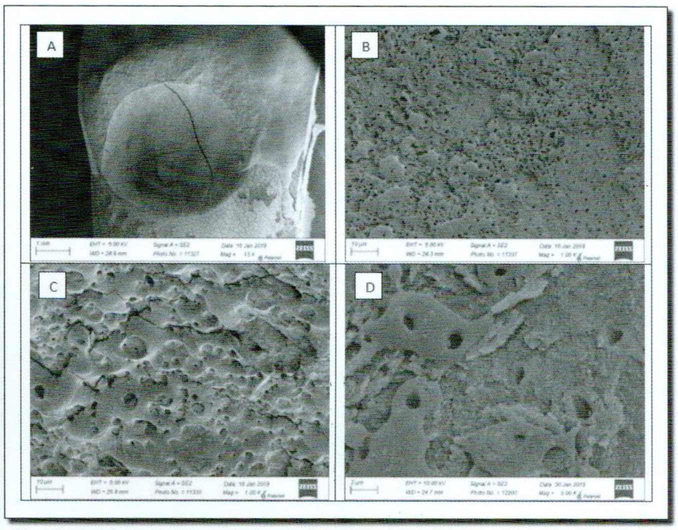

Fig. 2. Cavity micromorphology after $CO_2$-laser preparation (FE-SEM): **A.** Overview (×13), **C.** enamel (×1,000); **B/D.** in dentin (×1,000 and ×5,000).

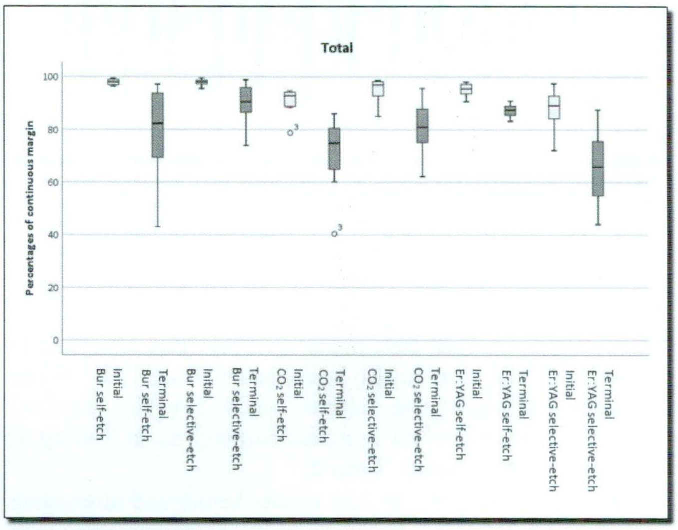

Fig. 4. Box-whisker-plots showing percentages of continuous margin in overall margins at baseline and aged for all experimental groups; herein represents the total length of a boxplot the range of the data, the outline of the box the first and third quartile and the line within the box the median value. Outliers are marked with small circles and are no longer in the range of the 1.5× interquartile-distance.

With the exception of group 5 (Er:YAG laser preparations in combination with self-etch adhesive restorations) and group 2 (bur with selective enamel-etch) all teeth showed a significant decrease in percentages of continuous overall margins between baseline and aged values. Regarding enamel separately, there was no significant reduction of continuous margin, only in group 5; in dentin there was no significant decrease for Er:YAG and $CO_2$ self-etch as well as bur selective-etch. Comparing aged values of enamel and dentin of the same experimental group, significant differences between percentages of continuous margins were evident in group 3 ($CO_2$ self) and group 6 (Er:YAG selective-etch).

Group 2 showed the best overall results regarding the entire margins followed by group 5 (Fig. 4). Dividing the aged results of self-etch and selective-etch samples, it was clear that, within the selective-etch technique, the bur performed significantly better than $CO_2$, which was better than Er:YAG laser prepara-

Table 2. Significant differences between the aged values of experimental groups in overall total margins.

| Self-etch | | Selective enamel-etch | |
|---|---|---|---|
| Bur | AB | Bur | A |
| $CO_2$ | B | $CO_2$ | A |
| Er:YAG | A | Er:YAG | B |

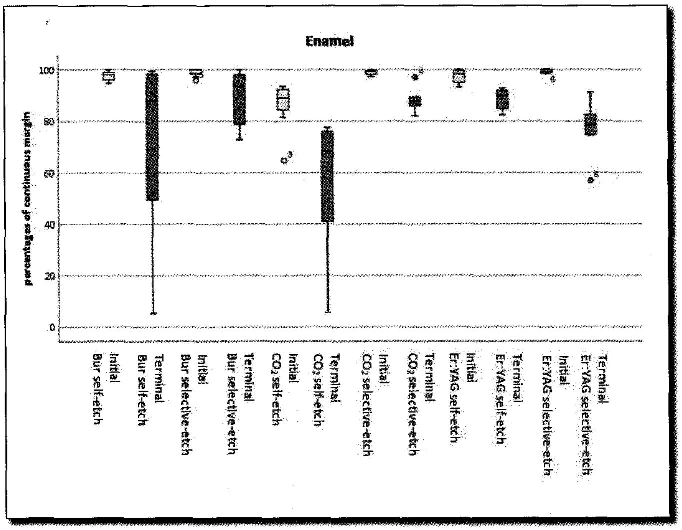

Fig. 5. Box-whisker-plots showing percentages of continuous margin in enamel at baseline and aged for all experimental groups; herein represents the total length of a boxplot the range of the data, the outline of the box the first and third quartile and the line within the box the median value. Outliers are marked with small circles and are no longer in the range of the 1.5× interquartile-distance.

tions. Within the self-etch mode, Er:YAG was significantly better than the bur, which was better than $CO_2$. When comparing self-etch with selective enamel-etching, Er:YAG laser preparations showed significant differences in contrast to $CO_2$ and bur preparations (Table 2).

In enamel margins, the best results before and after aging were seen for the bur-prepared cavity in combination with selective enamel-etching, whereas the worst results were observed for the $CO_2$-laser prepared group with self-etch adhesive (Fig. 5). Within the aged values of the self-etch samples, group 5 (Er:YAG) showed the best results, followed by the bur and $CO_2$-laser (for $P< 0.05$), whereas after application of the selective-enamel-etching, the bur was significantly different from Er:YAG and $CO_2$ was not significantly different from either. Differences between the two application forms of the adhesive systems were significant for the bur and $CO_2$ laser, but not for Er:YAG group (Table 3).

In dentin, group 2 showed also the best aged values, whereas group 6 revealed the worst results before and after aging (Fig. 6). In the self-etch mode, the three devices were equal, but in selective-etch, the bur was significantly better than $CO_2$, which was significantly better than Er:YAG. Aged values for each preparation device in self-etch or selective enamel-etching were significantly different for Er:YAG and $CO_2$ laser, but not for the bur group (Table 4).

Comparing values of all three devices together for self-etch and selective enamel-etching, there were significant differences in enamel and dentin, but when analyzing the complete marginal length (enamel and dentin together) there was no difference. Influence of the device used for preparation was significantly different between bur preparations and lasers, but not between the two lasers.

Table 3. Significant differences between the aged values of experimental groups in enamel.

| Self-etch | | Selective enamel-etch | |
|---|---|---|---|
| Bur | B | Bur | A |
| $CO_2$ | C | $CO_2$ | AB |
| Er:YAG | A | Er:YAG | B |

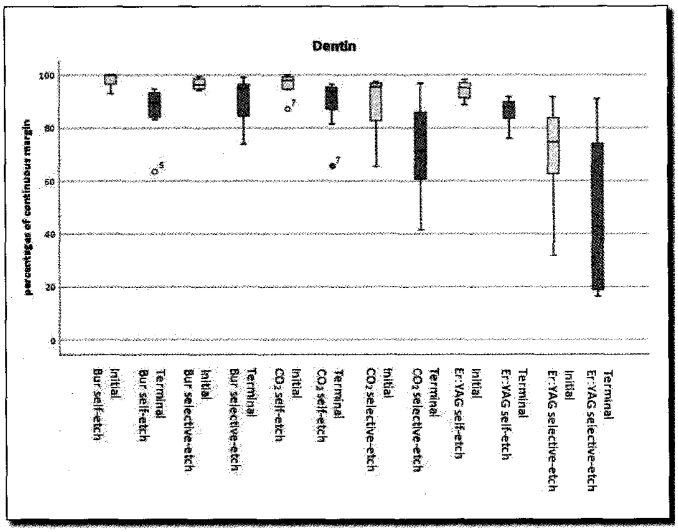

Fig. 6. Box-whisker-plots showing percentages of continuous margin in dentin at baseline and aged for all experimental groups; herein represents the total length of a boxplot the range of the data, the outline of the box the first and third quartile and the line within the box the median value. Outliers are marked with small circles and are no longer in the range of the 1.5× interquartile-distance.

Table 4. Significant differences between the aged values of experimental groups in dentin.

| Self-etch | | Selective enamel-etch | |
|---|---|---|---|
| Bur | A | Bur | A |
| $CO_2$ | A | $CO_2$ | B |
| Er:YAG | A | Er:YAG | C |

The choice of the cavity preparation device (laser or bur) was statistically more influential than the bonding technique.

## Discussion

Marginal quality of adhesive restorations depends on several factors such as the choice of the restorative material, cavity preparation tools, its parameters, and the operator experience.[22,24-27]

In this study the focus was on the comparison of the influence of cavity preparation by using an Er:YAG and a novel 9.3 mm $CO_2$ laser to a control group, which was prepared by a conventional bur and two different application techniques of a universal one component self-etching adhesive system. This choice was based on the fact that the comparisons between different lasers and burs based on microleakage or bond strengths have already been published, but to the authors' knowledge, no information is available on the quality of marginal adaptation of composite restorations combined with a one component universal self-etching adhesive system with cavities prepared with the novel 9.3 µm $CO_2$ laser or an Er:YAG laser.

The testing protocol was based on the one proposed by Bader & Krejci.[22] Protocols for tooth sample preparation and analyses were used, but the laser-devices and their parameters, as well as the materials and their application protocols were different from the study of Bader & Krejci.[22]

Great importance was given to the preparation of the tooth samples, especially to the simulation of dentin fluid flow to simulate as close as possible the clinical conditions. This enabled the ablation of the dental hard tissue and the adhesive procedures under the conditions close to those of vital teeth. When using extracted human teeth without flooding the pulpal chamber and the dentin tubuli with simulated dentin fluid, teeth are relatively dry, which may modify the interaction between lasers with wavelength absorbed in water as well as the interaction with adhesive systems, as studies showed reduced ablation effectiveness of Er:YAG laser systems and impaired bond strength due to excessive dehydration. As an example, since the thermomechanical laser ablation vaporizes water in the dental tissues, hydrophilic monomers may no longer easily diffuse into the dentin, which might affect the dentin-composite-interface and thus the quality of adhesive restorations. Dentin fluid may promote rehydration of the irradiated tissues and thus allow better diffusion of the hydrophilic monomers.[23,28,29]

In this study, percentages of continuous margins were recorded, based on SEM images of the restorative margins before and after loading, which is considered more relevant and closer to the clinical reality than the previously mentioned evaluation methods of bond strength or microleakage measurements.

As the identical evaluation methodology was used in the present study, the obtained percentages of continuous margins may be directly compared to previous studies.[22,30] This comparison showed that the results of this study with Er:YAG laser preparation with high pulse energy and finishing with low pulse energy in combination with one component self-etching universal adhesives had the same trend.

The quality of adhesion to lased dental surfaces is controversially discussed in the literature.[31] Analysis of bond-strength and of microleakage reported better,[32,33] comparable[34] and worse results[29,35-37] of lased surfaces than for bur preparations. In view of the results of the present study, the controversy of laser application in dentistry and especially in dental hard tissue ablation is confirmed and the importance of the laser type in combination with adhesion strategies (total-/self-etch) is highlighted. At the same time, even better results for marginal adaptation with Er:YAG laser than with the mechanical diamond bur in the self-etch group were reported. Laser treatments may provide more conservative and minimally or even non-invasive dental treatments in less time and less painful treatments.[4,5]

The differences between the initial results and the results after thermomechanical loading allow certain predictions on the long-term performance of the restorative systems tested, which is especially valuable for the clinicians. Within this study there was no significant decrease in the quality of the total marginal adaptation for Er:YAG laser prepared cavities in combination with the self-etch application of the one component universal adhesive system (group 5), as well as in the bur prepared cavities with selective enamel-etch technique (group 2). These procedures may be considered promising for clinical applications and should be tested in a clinical study. However, by evaluating marginal adaptation on enamel and dentin separately, it was noticed that in enamel margins the above-mentioned Er:YAG laser group performed well. The bur-selective-enamel-etching had a slightly significant difference.

Both above-mentioned groups performed well before and after loading. In enamel, margins of the Er:YAG laser group (group 5) did not show any differences between the values before and after loading, whereas this difference was slightly significant for the bur preparations (group 2) ($P= 0.0455$). This indicates that Er:YAG laser-cavity preparation followed by the application of a self-etching universal adhesive seems to offer the most stable adhesion between enamel surfaces and the adhesive system, followed by bur preparation and selective enamel etching. On margins located in dentin, no significant differences were found between the results before and after loading for groups 3 and 5. However, the relatively good adaptation on dentin in the $CO_2$ group might have been promoted by the poor marginal adaptation in enamel, which might have reduced the effects of the composite shrinkage stress at the dentin adhesive interface to a certain extent by reducing the C-factor and thus making an additional free surface available.[38]

Comparing the results of marginal adaptation in enamel and dentin after loading within the same experimental group, significant differences between percentages of continuous margins were evident in group 3 ($CO_2$ self-etch 56.7% in enamel and 89.47% in dentin) and in group 6 as well (Er:YAG selective-etch 47.2% in dentin and 77.45% in enamel). Based on the present results, these two procedures may not be advisable for clinical use.

Close examination showed that the applied adhesion strategy with phosphoric acid had a positive impact on adhesion in enamel for the bur and the $CO_2$ group as well. This type of acid seems to be more effective in creating microroughness than the weaker acid monomers present in the universal adhesive. Especially for the $CO_2$ lased enamel surfaces, this might be essential as this wavelength apparently modifies the chemistry and morphology of the enamel's surface due to the high absorption of its emission wavelength in hydroxyapatite. Heating of enamel due to the laser irradiation leads to a loss of the carbonate phase from the enamel crystals and further to a reduction in acid dissolution of enamel.[39] This "hardening" and increase in acid resistance might be the reason for the lower performance of the adhesive in self-etch mode in enamel in this group. This fact has also been previously described. Rechmann et al[29] demonstrated by bond strength tests with 9.3 µm $CO_2$ laser that, contrary to the etch-and-rinse systems (OptiBond Solo Plus and Peak Universal Bond), the self-etch system Peak SE (self-etching primer and Universal Bond), which was applied after preparation on rinsed and left damp surfaces, performed significantly worse than the non-lased control group (barely half of the control values). In addition, Scotchbond Universal did not perform well in the control group (half of the strength of PEAK SE in control) and had no further decrease

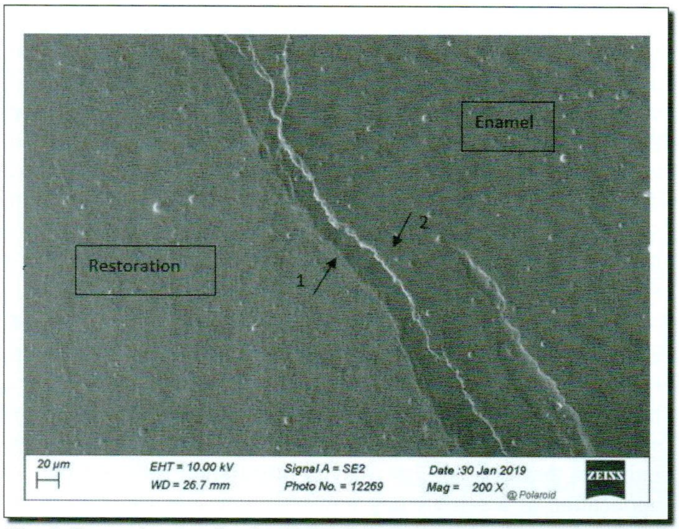

Fig. 7. Restoration margin after Er:YAG-laser preparation (×200; FE-SEM), continuous restoration margin (1) and enamel fracture (2).

after $CO_2$ laser ablation. The weaker values might be due to the fact that less acidic self-etching bonding agents are not as powerful in transforming the laser-treated molten enamel surface into a strong bonding surface. Rechmann et al[29,40] showed with scanning electron microscopy that phosphoric acid was capable of breaking up molten surfaces, creating additional pores for micro-retention, while the weaker acidic monomers of the self-etch system obviously were not able to favor those transformations.

Er:YAG prepared cavities with self-etching technique performed the best compared to the two other preparation techniques. The enamel values of continuous margins for the Er:YAG group were even better for self-etch technique than for selective enamel etching, although not statistically significant. When phosphoric acid was applied to Er:YAG lased enamel, the continuity of the margins in enamel after loading decreased from 88.5% with the self-etch approach to an average of 77.5%. The main reasons for this decrease were enamel fractures along the restoration margins, which accounted for approximatively 10% of the openings and were exclusively present in this group (Fig. 7). This could be explained by the fact that lasing weakens the target tissues to a certain degree. Unlike phosphoric acid, the mechanism of tissue removal by laser is not demineralization. This ablation process causes water and dental organic component vaporization, promoting micro-explosions that cause the resulting destruction of inorganic substances resulting in microscopic surface irregularities, enlarging the surface into which the adhesive system may penetrate leading to a strong and durable enamel-adhesive interface. However, if these porosities are too pronounced and associated with microcracks, they may weaken the enamel, which results in enamel fractures.[22,34,41,44,45] In such a case, adhesion to lased and etched surfaces may become so strong that the loading forces are deviated to the dental tissue producing fractures in enamel because of weaker crystal bond due to the subsurface damages induced by the laser. Summarizing, this means that the percentage of pure marginal openings in this group was about 87% after aging (88.5% baseline).

Furthermore, the better performance of the Er:YAG laser in combination with a self-etching universal adhesive compared to bur preparations might be attributed to the absence of debris, as laser prepared surfaces are free of smear layer. In comparison with acid etching, laser treatment is less technique-sensitive and may thus give better control over the area that needs to be precisely pretreated for adhesion.[22,34,42,43]

Similar to enamel, Er:YAG laser preparation of dentin leads to a smear layer-free, rough surface with open dentin tubuli, which may be ideal for bonding, as smear layer in this context may reduce the surface energy and decrease the reactivity of the dentin to the bonding systems.[28,44-46] Depending on the laser parameters: fluence, frequency, wavelength; and the tissue's optical and thermal characteristics, lasers with wavelengths between 8-11 µm can induce alterations in the physical and chemical composition in enamel and also in dentin. The Er:YAG laser produces changes in the composition and conformation of the organic matrix (collagen), $OH^-$ radical and the water present.[47]

Some studies[36,37] explained the inferior adhesion values in dentin after laser application with selective ablation of organic tissues, leading to less collagen left to be exposed and consequently to be hybridized. These findings are not in agreement with those of the present study where laser-prepared cavities showed similar results to conventionally prepared ones when applying a self-etching adhesive system. This might be explained by the fact that, contrary to most published studies, the laser-prepared cavities in this study were finished after preparation with high pulse energies with less powerful laser settings, which removed the subsurface damages induced by high energy pulses so that fewer negative effects were present on the surface of dentin, so that already weak acids can expose collagen fibers of underlying dentin and thus build a stable adhesive-tooth interaction.

Pulse energy (mJ), pulse frequency (Hz), power output (W), water flow rate (ml/s) and air pressure (bar) may be adapted for each device in order to avoid any adverse effects such as disintegrations, microcracks, leaflets, loosely bound particles and burned or melted spots, which may lead to increased microleakage and inferior marginal quality of adhesive restorations and efficient ablation.[22,48] Bader & Krejci[22] proposed higher energies for a fast ablation and finishing of the cavities with less powerful settings (less energy and water) to finish and smoothen the surfaces. However, when applying phosphoric acid to the enamel bevel, percentages of continuous margins decrease significantly for both lasers (groups 4 and 6) in comparison to the self-etch groups and both bur groups. Preparations with the Er:YAG laser showed significantly more open margins than the $CO_2$ laser group.

The negative impact in this study of the selective enamel etching on lased cavities, in enamel, but mainly in dentin, may be explained as follows: micromorphological surfaces of laser preparations are more uneven (Er:YAG > $CO_2$) than bur preparations with only flat grinding facets. For Er:YAG laser, peritubular dentin is deeply ablated and dentin tubuli free of smear layer. After $CO_2$ laser ablation, the surface is quite hilly due to melted drops which occlude partly open dentin tubuli. All the deep irregularities are filled with water when rinsing the phosphoric acid from the enamel bevel. It might be possible that drying the surfaces following the manufacturer's instructions for 2 seconds is insufficient for such surface patterns. It seems to be suitable for flat and fast drying bur-ablated surfaces.

After cavity preparation and before enamel-etch or self-etch of the whole cavity, surfaces were dried to the operator's estimation for suitable surfaces (5 seconds), that do not show visible water, but on the other hand, were not dehydrated/desiccated. With the 2 seconds drying after etching the enamel, the surfaces are not covered by much water anymore, but are still wet to an uncertain amount.

This persisting water in the micro-irregularities might dilute the bonding agent with its acids and monomers to an unknown and variable percentage, which could also explain the variances between the results. For that reason, the single components of the agent are not as effective as usual and the etched-like pattern is less prominent. In dentin, tubuli are not enlarged and collagen fibrils are less infiltrated, which leads to smaller, shorter, and less connected resin tags.[29,49]

Furthermore, it is possible that after the application of the bonding agent and its dissolution, evaporating the solvent and water in the bonding is more difficult. If the water and the solvent are not well evaporated it might create some bubbles within the hybrid/bonding layer, that weakens the interface and thus aging may have a greater impact.[50,51]

These might all be reasons for a weaker interface and for lower percentages of continuous margins.

Within the limitations of this in vitro study, the results led to the conclusion that a specific combination of laser type and adhesion settings significantly influenced the results of marginal adaptation, both in enamel and dentin. Er:YAG and 9.3 µm $CO_2$ laser preparations may be valuable alternatives to conventional bur preparations in combination with a one component universal adhesive system, under the conditions of a specific application technique of the adhesive.

Consequently the null hypothesis stating that there were no micromorphological differences on enamel and dentin surfaces as well as no significant differences in marginal adaptation between mechanical bur, Er:YAG and 9.3 µm $CO_2$ laser in both self-etch and selective enamel-etch application technique was rejected. Clinical studies are necessary to confirm these in vitro findings.

a. Colgate-Palmolive, New York, NY, USA.
b. KaVo Kerr, Orange, CA, USA.
c. Kulzer, Hanau, Germany.
d. Bioswisstec, Schaffhausen, Switzerland.
e. Terumo, Somerset, NJ, USA.
f. Light Instruments, Yokneam, Israel.
g. Convergentdental, Needham, MA, USA.
h. Intensiv, Montagnola, Switzerland.
i. Coltène/Whaledent, Altstätten, Switzerland.
j. Ultradent, Provo, UT, USA.
k. 3M, St. Paul, MN, USA.
l. Shofu, Kyoto, Japan.
m. Struers, Willich, Germany.
n. Karl Zeiss Microscopy, Cambridge, UK.

*Acknowledgements:* To Ms. Isaline Rossier, and Ms. Luciana Caseiro for their assistance in specimen preparation and observation in the scanning electron microscope and Dr. Ivonne Chaskélis for her advice on the laser.

*Disclosure statement:* The authors declared no conflict of interest. This research was supported by Coltène/Whaledent, who provided restorative materials and by Convergentdental in allocating their laser system.

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