The Tensile Bond Strength of Fissure Sealant to Enamel Treated by Nd–YAG Laser Irradiation with TiO$_2$ Suspension

Tetsu Matsunaga,¹ Tohru Hayakawa,² and Kensuke Matsune¹

Departments of ¹Pedodontics, ²Dental Biomaterials, Nihon University School of Dentistry at Matsudo, Matsudo, Chiba 271-8587, Japan

Correspondence to:
Kensuke Matsune
E-mail: kensuke.matsune@nihon-u.ac.jp

Abstract
In pediatric dentistry, treatment is performed mainly for oral health care and prevention of caries. Fissure sealants are often used in clinical practice for prevention of dental caries. The prevention of caries in occlusal fissures is of practical importance, since pits and fissures are more susceptible to caries than any area. It is possible to debride pits and fissures using laser irradiation in order to remove the fissure contents without altering the shape and structure of the fissures. Although the adhesive strength of fissure sealant to enamel may grow weak when a laser is used, there have been no reports regarding the adhesive strength of enamel to fissure sealant after Nd–YAG laser irradiation with 1% TiO$_2$ suspension. Therefore, the authors investigated the tensile bond strength of a commercial fissure sealant applied after Nd–YAG laser irradiation in contact mode with a fiber diameter of 400 μm, 5 pps, laser tip output power of 300 mJ, and with the use of 1% TiO$_2$ suspension. It was revealed that lasers could react to an enamel surface without any special treatment like application of a black pigment to the surface of the tooth when Nd–YAG laser irradiation was performed. Also, the tensile bond strength of TeethmateFi® to the enamel was lower after laser irradiation with 1% TiO$_2$ suspension, whereas that of Fuji III LC® was stable. From the SEM images of the interfaces between the fissure sealant and enamel, it was found that the enamel surface was affected to a depth of approximately 20–30 μm by laser irradiation. It was revealed that when the fissure sealant was applied with laser irradiation, the laser should be precisely irradiated and the use of a glass-ionomer fissure sealant was more effective.

Keywords:
Nd–YAG Laser, TiO$_2$, Tensile adhesive strength

Introduction
In pediatric dentistry, the focus of treatment has shifted from the management of caries to child oral health care and caries prevention (¹, ²). Among the preventive treatment measures for caries, fissure sealant application is effective in the prevention of pits and fissures in young permanent teeth (³). The tooth substance of young permanent teeth is immature and particularly susceptible to caries because of complicated fissures. In this case, the sealant will be effective. However, it is difficult to remove impacted debris and remaining microorganisms from the deepest area of the pits and fissures. Meanwhile, the microorganisms with inductive factors of caries in enamel fissures can be killed by Nd–YAG lasers. Furthermore, acid resistance of enamel can be obtained, so that the Nd–YAG laser’s effect of providing additional acid resistance to enamel has attracted attention as one of the new caries treatments and one of the methods for preventing caries (⁴–⁶). As the Nd–YAG laser penetrates enamel, it is necessary to apply black pigment to the surface of the tooth and prepare the tip of the laser to make the laser react with the enamel (⁷). Shibuya et al. (⁸) reported that it was possible to make the laser react to the enamel surface without specially treating
either the surface of the enamel or the tip of the laser, by applying TiO₂ suspension. However, there have been no reports about Nd–YAG laser irradiation and the tensile bond strength of resin and glass ionomer sealants when TiO₂ suspension is applied.

In this study, the tensile-bond strengths of a commercial resin fissure sealant and glass-ionomer fissure sealant to the enamel surfaces treated by Nd–YAG laser with 1% TiO₂ suspension, were determined and compared.

Materials and Methods
Fifty enamel surface specimens derived from 80 human third molars, which were extracted due to pericoronitis and preserved in 10% formalin solution, were used in this study. TeethmateF1® (Kuraray Medical Inc., Osaka, Japan) and Fuji III LC® (GC, Tokyo, Japan) were used as the resin fissure sealant and glass-ionomer fissure sealant, respectively. According to the manufacturers’ instructions, the enamel surface was etched by 40% phosphoric acid for 40 sec in TeethmateF1® and with a conditioner 10% polyacrylic acid for 10 sec in Fuji III LC®. Both sealants were photo-cured for 20 sec by Curing Light XL3000 (3M Dental Products, MN, USA).

An Nd–YAG laser device (CONTAC LASE, S.L.T. product) was used in this study and the irradiation was performed in contact mode for 35 sec or 1 sec with a fiber diameter of 400 μm (not processed silica), 5 pps and laser tip output power of 300 mJ using 1% TiO₂ suspension.

SEM Observation of the enamel surface before sealant application
After the teeth preserved in 10% formalin solution were washed under a stream of water, the buccal enamel surface of the third molar teeth was ground with a sequence of 800-grit and 1,000-grit silicon carbide papers. Six specimens were divided into six experimental groups: only polished, phosphoric acid etched, polyacrylic acid etched, laser irradiation, and phosphoric acid etched and polyacrylic acid etched after laser irradiation groups. Two specimens of the non-irradiated were prepared as follows: the polished enamel surface was etched with 40% phosphoric acid aqueous solution or 10% polyacrylic acid aqueous solution. The other two specimens of the laser-irradiated were prepared as follows: after 1% TiO₂ suspension was applied onto the polished buccal enamel surface, Nd–YAG laser irradiation was performed; the laser-irradiated buccal enamel surface was then etched with 40% phosphoric acid aqueous solution or 10% polyacrylic acid aqueous solution.

Six specimens were sputter-coated with a platinum alloy using an Ion Coater (JFC–1600, Japan Electrics, Tokyo, Japan). Specimens were then mounted onto aluminum stubs, and sputter-coated with a platinum-palladium alloy using Ion Coater (JEF–1600, Japan Electrics). Six specimens were examined at numerous magnifications and tilt angles with a field-emission scanning electron microscope (FE-SEM, JSM-63400F, Japan Electronics) at 5 kV.

Tensile bond strength of the fissure sealant
The buccal enamel surfaces of the third molar teeth were polished with 1000-grit silicone carbide paper and 40 specimens were obtained. In order to identify the adhesive area, a 0.3-mm-thick masking tape with a circular hole (diameter=2.5 mm) was attached to the polished enamel surfaces. They were then prepared accordingly to be included in the non-laser or laser-irradiated groups. The enamel surface was etched and dried by air blowing. The fissure sealant was applied to the defined enamel surfaces, and after the fissure sealant hardened, a sandblast-treated stainless steel bar was fixed with super-bond C&B (Sun Medical) on the hardened fissure sealant surface. After immersion in water at 37 °C for 1 day, the tensile bond strengths were measured at a tensile speed of 1 mm/min by an Instron Universal Testing Machine (TCM–500CR, Shinko Engineering) (Fig. 1).

The adhesive test was conducted on 40 defined enamel surfaces in five specimens. The surfaces had been treated as follows: with no laser irradiation, laser irradiation for 35 sec over the entire surface with 1% TiO₂ suspension, no laser irradiation in the pseudo-fissure created in the center (a hole (4πr²×
1/2 + πr²×h) with 0.5 mm diameter and 0.5 mm depth was made by jet carbide bur) and laser irradiation for 1 sec in the pseudo-fissure with 1% TiO₂ suspension (Fig. 2). The laser irradiation was conducted in such a manner so that the tip of fiber was adjusted until it just touched and made contact with the enamel surface perpendicularly. It was moved 400 μm at each laser irradiation under a stereomicroscope, SZX-ZB12 (Olympus). Results of the tensile bond strength are shown as the mean value ± standard deviation (S.D.). Statistical analysis was performed by one-way ANOVA and Turkey tests.

**Observation of the adhesive interface between fissure sealant and polished enamel**

Silicon ring molds with an inner diameter of 3.2 mm and thickness of 2 mm were temporarily bonded onto the etched enamel surface or laser-irradiated and etched enamel surface. TeethmateF1® or Fuji III LC® was applied to the etched enamel surface inside the silicone ring molds, and then photo-cured for 20 sec by Curing Light XL3000 (3M Dental Products, MN, USA). Thereafter, the silicone ring mold was removed and the bonded specimen immersed in water at 37 °C. After 1 day, the bonded specimens were cross-sectioned parallel to the tooth axis and the cross-sectioned surface was polished by using 1000–grit, 1200–grit and 1500–grit water-resistant silicone carbide papers and 6 μm, 3 μm, 1 μm and 0.25 μm diamond paste (Stratek) sequentially. Following that procedure, argon–ion etching was performed for 30 sec using a small-sized ECR-type ion shower apparatus (EIS-200ER, Elionix), operating at 1 kV, 15 mA (9), and sputter-coating was done with a platinum–palladium alloy. Four specimens were examined at numerous magnifications and tilt angles in a FE-SEM.

**Results**

**SEM observation of the enamel surface before sealant application**

When the buccal enamel surface was etched with 40% phosphoric acid, typical etching patterns corresponding to the alignment of the enamel prisms were observed (Fig. 3B). In contrast, when the enamel surface was etched by the 10% polyacrylic acid aqueous solution, only a blank outline of the enamel prism was observed (Fig. 3C). The degree of decal-

![Fig. 1. Diagram showing an adhesive specimen.](image)

![Fig. 2. Diagram showing polished enamel surface treated for adhesives (cross-section).](image)

![Fig. 3. SEM images of the enamel surfaces before sealant application (×1500). A: Polished enamel surface, B: 40% phosphoric acid, C: 10% polyacryl acid.](image)
cification of the buccal enamel was strongly dependent on the type acid utilized compared with no application of acid (Fig. 3A).

When the Nd–YAG laser was applied to the polished enamel surface, a glass and lava–like structure showing an irregular pattern with large and small sized particles, was observed on the surface (Fig. 4). This glass and lava–like structure remained unchanged on the laser–irradiated enamel surface, even though 40% phosphoric acid was applied. However, since the non–irradiated enamel surface, surrounding the laser–irradiated enamel, was etched by 40% phosphoric acid, a typical etching pattern was observed (Fig. 5A and Fig. 5B). Similar to the result obtained after phosphoric acid etching, even though 10% polyacrylic acid etching was applied, the glass and lava–like structure was retained (Fig. 6A and Fig. 6B).

![Fig. 4. SEM image of the enamel surface treated by laser irradiation (×500).](image)

![Fig. 5. SEM images of the enamel surfaces with 40% phosphoric acid after laser irradiation. A: ×500, B: ×1500.](image)

![Fig. 6. SEM images of the enamel surfaces with 10% polyacrylic acid after laser irradiation. A: ×500, B: ×1500.](image)

**The tensile bond strength of fissure sealant**

Table 1 shows the mean tensile bond strengths of TeethmateF1® and Fuji III LC® to the laser non–irradiated and irradiated enamel surfaces without and with pseudo–fissure. In TeethmateF1®, the application of Nd–YAG laser to the polished enamel surface resulted in a significant decrease in the mean bond strength. A significant difference in the mean bond strength between the laser–irradiated group and non–irradiated group was observed. In contrast to the result obtained with TeethmateF1®, a specific decrease in the mean bond strength was not observed when Fuji III LC® was applied to the laser–irradiated enamel surface. In the non–irradiated surface, the strength of Fuji III LC® was significantly lower than that of TeethmateF1®. Otherwise, the strengths of TeethmateF1® and Fuji III LC® were similar in laser irradiated surfaces.

<table>
<thead>
<tr>
<th></th>
<th>TeethmateF1 (Mpa)</th>
<th>Fuji III LC (Mpa)</th>
</tr>
</thead>
<tbody>
<tr>
<td>non-laser</td>
<td>11.7±3.1 (n=5)</td>
<td>5.7±2.2 (n=5)</td>
</tr>
<tr>
<td>laser</td>
<td>4.1±2.2 (n=5)</td>
<td>5.8±1.3 (n=5)</td>
</tr>
<tr>
<td>non-laser (pseudo-fissure)</td>
<td>13.6±5.3 (n=5)</td>
<td>9.1±2.2 (n=5)</td>
</tr>
<tr>
<td>laser (pseudo-fissure)</td>
<td>8.1±2.7 (n=5)</td>
<td>9.7±1.6 (n=5)</td>
</tr>
</tbody>
</table>

* : p<0.05

![Table 1. Tensile bond strength of sealant to enamel treated by non-laser or laser irradiation.](image)
The mean tensile bond strengths of TeethmateF1® and Fuji III LC® to the laser non-irradiated and irradiated enamel surfaces with pseudo fissure. In TeethmateF1®, the irradiation of Nd–YAG laser to the polished enamel surface with pseudo fissure resulted in an insignificant decrease in the mean bond strength compared with non-irradiated. In contrast to the result obtained from TeethmateF1®, a decrease in the mean bond strength was not observed when the Fuji III LC® was applied to the laser-irradiated enamel surface. However, the strength of the non-irradiated enamel surface of Fuji III LC® was significantly lower than that of TeethmateF1®.

In TeethmateF1®, the creation of pseudo-fissure resulted in insignificant increases in the mean bond strength in non-laser and laser irradiation groups. In contrast to the result obtained from TeethmateF1®, the increases in the mean bond strength were significantly observed when Fuji III LC® was applied to the non-laser and laser irradiated enamel surfaces with pseudo-fissure.

**Observation of adhesive interface between fissure sealant and polished enamel**

Fig. 7A and Fig. 7B show the SEM images of the adhesive interface between TeethmateF1® and the non-laser-irradiated or laser-irradiated enamel surfaces, respectively. When TeethmateF1® was applied to the non-laser irradiated or laser irradiated enamel surface, the adhesive interface of TeethmateF1® and enamel were closely bound. The SEM images after laser irradiation showed cracks up to approximately 30 μm on the enamel surface layer immediately below the interface. Fig. 8A and Fig. 8B show the SEM images of the adhesive interface between Fuji III LC® and the non-laser-irradiated or laser-irradiated enamel. Both figures show a space between the enamel surface and Fuji III LC®, which might be due to the shrinkage of the sealant. Meanwhile, many images showed that Fuji III LC® was detached from the enamel on the interface between them, and a part of the Fuji III LC® remained closely attached to the enamel after the internal fracture. Many parts where Fuji III LC® was detached from the enamel were observed on the interface after laser irradiation, and an image where parts of the enamel were detached, together with the Fuji III LC®, also was observed. The depth of the detached enamel was approximately 20 μm at the maximum.

**Discussion**

Lasers have been used effectively for the prevention and treatment of caries and excision of soft tissue in pediatric dentistry (2, 10-14). The advantages of the Nd–YAG laser, for example, its acid resistance effect, have also been reported (4-6, 15, 16). The tip of the fiber had to be prepared either by sharpening the tip with a file and attaching carbide or incorporating carbide in the tip of the fiber (17-19). If the fissures are irradiated by Nd–YAG laser before sealant application, the deposits in the fissures of the enamel can be removed, so it is indeed
effective for cleaning before sealant application (20). According to a report on preventive fissure sealant application, irradiating of an Nd–YAG laser to fissures before applying the fissure sealant can effectively remove tooth deposits (21). Furthermore, the Nd–YAG laser immediately raised the temperature of the surface of the tooth to over 1000 °C, destroying bacteria, and since the laser is precisely irradiated on the surface coated with a black pigment, the heat of the laser does not affect the dental pulp (21). Recently, it was reported that the laser irradiation could be localized to the surface of the enamel by using a TiO₂ suspension without any special treatment (7). Therefore, an additional investigation was conducted concerning the bonding of fissure sealant after the laser has been made to react with the enamel using 1% TiO₂ suspension.

Based on a report by Hasegawa et al. (22), the enamel surface demonstrates a lava–like structure under laser irradiation conditions of more than 15 pps, 83 mJ and 1.25 W and the acid resistance of the enamel is improved. Kito et al. (23) have reported on the low-output effect on the enamel surface, saying that even low outputs ranging from 100 mJ to 300 mJ can affect the surface layer of the enamel. Therefore, contact irradiation was performed with a fiber diameter of 400 µm, 5 pps and laser tip output power of 300 mJ when using a 1% TiO₂ suspension in this study.

SEM images revealed that if enamel was irradiated with a laser using 1% TiO₂ suspension, it is possible to make lasers react with the surface layer. From the SEM images, some parts of the markedly uneven enamel surface had evaporated and there were glass and lava–like structures and pores, which were in agreement with the findings reported by Kito et al. (23). Based on this fact, it was revealed that if 1% TiO₂ suspension was used, laser irradiation could cause the same reactions on the enamel surface without special treatment as previously reported. The glass and lava–like image was observed on the enamel irradiated by laser. This glass and lava–like sintered part has a stronger resistance to acid (24), and a greater prevention of caries can be obtained by applying fluoride or fluoride slow–release sealant materials. Regarding the effects of various pretreatments on the adhesive strength between enamel and commercial fissure sealant, Matsune et al. (25) reported that resin fissure sealants were more strongly affected by pretreatment than glass–ionomer fissure sealants. Ariyaratnam et al. (26) reported that although Nd–YAG laser roughened the surface of enamel, it did not provide a surface as retentive as a surface treated with conventional acid etching and the laser cannot be recommended as a viable alternative to acid etching in the case of resin fissure sealant. From the results of this experiment, although the adhesive strength of TeethmateF1® to enamel decreased by approximately 60% when all the constrained surfaces were irradiated by the laser, no differences were observed when Fuji III LC® was applied. From the above results, TeethmateF1® was strongly affected by the enamel treatment more than Fuji III LC® was. As for the tensile bond strength when pseudo–fissures were prepared, although the tensile bond strength of TeethmateF1® slightly increased, that of Fuji III LC® increased significantly. Furthermore, when lasers were irradiated exclusively on fissures, although the tensile bond strength of TeethmateF1® decreased by approximately 40%, that of Fuji III LC® showed almost no difference, and it was revealed that Fuji III LC® remained mostly unaffected by the surface of the enamel before being applied to it. The value of the tensile bond strength that decreased by approximately 40% coincides with that of the report by Suzuki et al. (27) ; therefore, it was clear that the adhesive strength between resin materials and enamel decreases if a laser is irradiated. It was also shown that if laser irradiation was limited to the fissure crevices, most of the enamel surface that needed etching was not affected by laser irradiation, so its effect is the same as that of a normal preventive sealant. However, from the results of tensile bond strength tests, TeethmateF1® was affected by laser irradiation and its tensile bond strength decreased most likely because of the size of the pseudo–fissures. However, Fuji III LC® was not affected by laser irradiation.
From the images of the interface between the enamel surface and fissure sealant, TeethmateF1® was more tightly bonded to the enamel than Fuji III LC®. It is said that bonding between the enamel surface and resin is mainly caused by the anchor effect mechanically supported by a resin tag formed by the resin monomer that diffuses into the enamel roughened by etching treatment (28). Another study demonstrated that laser etching had a similar or greater etching effect than acid etching (29). However, Isogai et al. (30) reported that the value of the shear test when a fissure sealant was applied after Er: YAG laser irradiation was near half of the value of the phosphoric acid treatment.

Meanwhile, a glass ionomer fissure sealant can directly adhere to the tooth due to its nature, so it was considered that it produced stronger adhesion, and it was revealed that part of the sealant remained on the adhesive interface. Large part of the glass ionomer fissure sealant in the interface was detached because air bubbles were produced during the procedure (25), its adhesion to enamel is weak compared to TeethmateF1® applied after treatment with 40% phosphoric acid and its penetration into enamel is weak (31). However, it was observed that when the laser was locally irradiated, the tensile bond strength of Fuji III LC® did not decrease and remained effective more than that of TeethmateF1®, when compared with the case of non-laser irradiation. It is reported that some cracks appeared on the enamel of the adhesive interface after laser irradiation, and in this study, cracks were observed in both TeethmateF1® and Fuji III LC®. In particular, when glass ionomer fissure sealant that has a strong adhesiveness to the tooth substance was applied, cracks and partial ruins were observed on the enamel at the interface. As reported, the compressive strength of the enamel after irradiation by a laser decreases by 15%, and it is considered that laser irradiation affected not only the enamel surface but also its inside. In addition, when the glass ionomer fissure sealant was applied, together with its strong adhesiveness to the tooth substance, part of the enamel was detached along with the sealant when the sealant was detached from the enamel.

According to the results of this research, it is suggested that it is possible to avert fissure sealant detachment, clean and sterilize the inside of a fissure and effectively uptake fluoride which is gradually discharged from the glass ionomer fissure sealant at the same time by precise laser irradiation on the fissure. However, excessive laser irradiation should be avoided because the enamel surface may become brittle. It has been reported that the retention rate of glass ionomer fissure sealant in fissures is lower than that of resin fissure sealant so far; however, no differences in the progression rate of caries is shown between resin and glass ionomer fissure sealants because of the re-calcification response by release of fluoride in glass ionomer (3).

From the results of this study, it is suggested that if fissure sealants are applied in combination with the Nd-YAG laser, the combination of glass ionomer fissure sealant is more effective than that of resin fissure sealant.

Acknowledgments
The authors wish to thank Prof. T. Maeda, Department of Pediatric Dentistry and Prof. N. Nishiyama, Department of Biomaterials, Prof. T. Ikemi and Prof. Y. Kozawa, Nihon University School of Dentistry at Matsudo, for their kind suggestions and advice.

References