Application of Direct Laminate Veneer for Enamel Protection during Orthodontic Treatment: An In Vivo Evaluation

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The aims of this study were to examine the enamel protective effect of a direct laminate veneer against acidic conditions, measure the amount of fluoride released from the veneer and which was taken up by the enamel, as well as evaluate the usefulness of the veneer for orthodontic treatment. First, a veneer was applied to rabbit incisors. Then, apart from using profile measurement microscope and electron probe microanalyzer to measure the roughness and fluoride concentration in the enamel of veneer-covered tooth surface, those of the uncovered adjacent tooth surface were measured too. It was found that the veneer protected the enamel surface from acidic conditions. Furthermore, fluoride ions were taken up by both the uncovered adjacent enamel area as well as by the covered area. These findings suggested that covering the enamel surface with a direct laminate veneer before bracket bonding might be a valuable means of tooth protection and caries prevention.

Key words: Direct laminate veneer, Caries prevention, Orthodontics

INTRODUCTION

Since the introduction of multi-bracket systems in orthodontic treatment, significant progress has been made in the successful bonding of brackets to teeth\(^1\), thereby replacing the former system of cementing preformed bands. However, in order to achieve sufficient bond force to the tooth surface, acid etching is required when resin adhesive materials are used – and acid etching can lead to enamel dissolution or defects\(^2\)\(^-\)\(^8\). One of the most frequently encountered problems is localized decalcification of the enamel around the bonded bracket, commonly referred to as a ‘white spot’\(^9\). In a study by Basdra et al.\(^10\), nearly 50% of orthodontic patients exhibited clinically visible white spot lesions during a treatment that lasted approximately two years. But in general, prevalence of smooth lesions during treatment has increased up to 50%\(^11\)\(^,\)\(^12\). This may be caused by inadequate patient oral hygiene and poor diet control, as well as by an increase in susceptibility to caries due to dissolution of the highly calcified prismless enamel by acid etching, or it could be attributed to reduced self-cleaning activity in the area around the bracket base.

Recently, glass ionomer cement (GIC)\(^13\)\(^-\)\(^18\) has been used as an adhesive material for the purpose of enamel protection because no etching is required for application. However, it is nevertheless difficult for patients to maintain an adequate level of oral hygiene throughout the bracketing period. Indeed, since the direct bonding method was introduced, we have seen many cases of caries and decalcification.

Previously, Miwa et al.\(^19\) reported on the application of a direct laminate veneer (veneer) as a pre-treatment for the bracket bonding procedure. Direct laminate veneer consisted of resin and glass ionomer cement; no acid etching or other invasive treatments to the enamel were required (Fig. 1). In addition, this veneer exhibited sufficient bond strength during the treatment period.

However, there have been no in vivo evaluations of the enamel protective effect of the veneer against acidic conditions, or of the fluoride released from the veneer and which is taken up by the enamel. The

![Fig. 1 Direct laminate veneer was structured by GIC and resin. GIC was painted as a thin layer on the enamel surface. Resin was then applied to the GIC layer using a transparent matrix formed from the labial surface of the tooth prior to the procedure.](image-url)
aims of this study, therefore, were to examine the enamel protective effect of the veneer against acidic conditions \textit{in vivo}, measure the amount of fluoride released from the veneer and which was taken up by the enamel, as well as evaluate the usefulness of the veneer in orthodontic treatment.

**MATERIALS AND METHODS**

**Preparation of direct laminate veneer**
Twenty-one male Japanese albino rabbits (2.0-2.5 kg) were used in this study. Table 1 shows the materials used for the experimental application of the direct laminate veneer. The surfaces of the rabbit incisors were polished for 10 seconds using a polishing agent (Prophypaste RDA120, Clean Chemical Sweden AB), washed with distilled water, and dried. Before bonding, the entire buccal surface was covered with masking tape in which a hole (1.5 mm in diameter) was punched to standardize the veneer area.

A glass ionomer cement (Fuji Ortho LC, GC, Tokyo, Japan), diluted by doubling the liquid ratio compared with the powder-liquid ratio recommended by the manufacturer, was painted as a thin layer on the labial surface using a small brush, and hardened by light radiation for 20 seconds. A resin (Gradia GC, Tokyo, Japan) was then applied to cover the GIC layer and hardened by light radiation for 40 seconds. The resultant thickness of GIC and resin layer was 150 \textmu m each.

Finally, the masking tape was removed and the top edge of the veneer was polished using silicon points for finishing. All procedures for animal care were approved by the Animal Management Committee of Aichi-Gakuin University. Fig. 2 shows a schematic illustration and picture of the direct laminate veneer on rabbit incisors.

**Evaluation of the enamel protective effect of direct laminate veneer from acid conditions**
The animals were divided into two groups — acidic and neutral. At every 24 hours for four weeks, rabbits of the acidic group received 50 \textmu l of 65% phosphoric acid solution (Super Bond Etching Agent, Sun Medical, Shiga, Japan) dripped into the oral cavity, whereas the neutral group received 50 \textmu l of distilled water. The animals were sacrificed after four weeks, and the incisors were extracted.

1. Examination of the cross-section of incisors with direct laminate veneer
To examine the cross-section of incisors with veneer, the incisors were dried and embedded in epoxy resin (SCANDIPLEX, SCANDIA, Hagen, Germany). The incisors were cut buccolingually with a microtome (LEICA SP1600, Leica, Tokyo, Japan), and sections containing the veneer were hence prepared. The surface of each cross-section was polished with a polisher (ECOMET-3, BUEHLER, Illinois, USA) and dried in a desiccator for 24 hours under vacuum. After drying, specimens were examined under a profile measurement microscope (VF-7500, KEYENCE, Tokyo, Japan) at \times 250 magnification. Samples consisted of both a neutral group (n=4) and an acidic group (n=4).

2. Quantitative analysis of tooth surface roughness
Veneer was removed from the enamel surface before examination. Ten arbitrary points were selected for tooth surface roughness measurement. Then, both the veneer-covered tooth surface (VTS) and uncovered adjacent tooth surface (UTS) were measured by profile measurement microscope (VF-7500, KEYENCE, Tokyo, Japan). As controls, incisors of the untreated group were also measured. Samples consisted of a neutral group (n=10), an acidic group (n=10), and an untreated group (n=10).

<table>
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<th>Table 1 Materials used in this study.</th>
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<tr>
<td>Material</td>
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<tr>
<td>Light-cured composite resin</td>
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<td>Glass ionomer cement</td>
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<td>20% Polyacrylic acid</td>
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<td>65% Phosphoric acid</td>
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Fig. 2 Schematic illustration (a) and picture (b) of direct laminate veneer on rabbit incisor.
3. Measurement of lesion depth
To determine lesion depth around the veneer, depths were measured as the distance from the demineralized lesion around the veneer to the underlying sound enamel using the profile measurement microscope at 10 points arbitrarily selected along the junction stretch between the area covered by the veneer and the area not covered by the veneer (Fig. 3). Samples consisted of a neutral group (n=10) and an acidic group (n=10).

Measurement of fluoride concentration from direct laminate veneer
Four animals were kept for two weeks in order to measure the concentration of fluoride uptake by enamel from the veneer. Fluoride concentration in enamel was measured using an electron probe microanalyzer (EPMA-1600, Shimadzu, Kyoto, Japan). To prepare for line analysis, incisors with veneer were dried and embedded in epoxy resin (SCANDIPLEX, SCANDIA, Hagen, Germany). They were then cut buccolingually with a microtome (LEICA SP1600, Leica, Tokyo, Japan), and sections containing the veneer were hence prepared. The surface of each cross-section was polished with a polisher (ECOMET-3, BUEHLER, Illinois, USA) and dried in a desiccator for 24 hours under vacuum. After drying, specimens were coated with evaporated carbon. Using the uncovered adjacent tooth surface (A), boundary area (B), and veneer-covered tooth surface (C), line analysis was performed perpendicular to the cross-section at depths of 5, 10, and 20 μm from the enamel surface (Fig. 4). Observed fluoride intensity (cps) was recorded as the fluoride concentration at the measurement site. Electron microscope settings were 15 kV (accelerating voltage) and 0.05 μm (probe current) with a probe diameter of 1 μm. Samples consisted of an experimental group (n=4) and an untreated group (n=4).

Statistical analysis
All data obtained were analyzed using Student's t-test.

RESULTS
Examination of the cross-section of veneer and incisors
In the neutral group, no enamel loss was observed at UTS and the surface was smooth (Fig. 5(a)). In the acidic group, UTS was decalcified and enamel loss...
was observed. The loss reached the dentin, and a gap was observed between VTS and UTS (Fig. 5(b)). In both groups, sealing of the enamel by the GIC layer was adequate, and no percolation was observed between the enamel and veneer (Figs. 5(a) and (b)).

Quantitative tooth surface analysis
Fig. 6 shows the average roughness and standard deviations for all groups. The tooth surface roughness of UTS in the acidic group \( (1.11 \pm 0.47 \mu m) \) was significantly greater than all the other categories: neutral group UTS \( (0.45 \pm 0.10 \mu m) \); neutral group VTS \( (0.43 \pm 0.10 \mu m) \); acidic group VTS \( (0.43 \pm 0.10 \mu m) \); and untreated group \( (0.45 \pm 0.11 \mu m) \) \( (P < 0.001) \). On the other hand, there were no significant differences between the untreated group and the neutral groups (i.e., both UTS and VTS), as well as with acidic group VTS.

Measurement of lesion depth
The mean lesion depth around the veneer was 110.9 ± 2.2 \( \mu m \) in acidic group — which was significantly larger than that in neutral group \( (2.1 \pm 0.5 \mu m) \) \( (p < 0.001) \) (Fig. 7).

Measurement of fluoride concentration
Figs. 8(a) and (b) show the typical fluoride intensity profiles of untreated and veneer-covered enamel.

In the enamel of the untreated group without veneer, fluoride intensity was weak regardless of the depth from the enamel surface, and no large changes were observed among the varying depths. On the other hand, in the enamel of the experimental group, fluoride intensity sequentially increased from the enamel not covered with veneer to the enamel directly beneath the veneer. Moreover, as a function of enamel depth, fluoride intensity decreased in each region (i.e., A, B, or C) as enamel depth from the surface increased (i.e., 5, 10, and 20 \( \mu m \) from enamel surface).

Fluoride intensity and comparison by enamel depth are shown in Fig. 9. The fluoride intensity of position C decreased steadily from the surface towards the inner part, and significant differences were observed between 5 \( \mu m \) \( (35.8 \pm 6.3 \text{ cps}) \) and 10 \( \mu m \) \( (23.8 \pm 0.5 \text{ cps}) \) \( (P < 0.01) \), and between 10 \( \mu m \) \( (23.8 \pm 0.5 \text{ cps}) \) and 20 \( \mu m \) \( (16.5 \pm 1.4 \text{ cps}) \) \( (P < 0.05) \). At position B, significant differences were observed between 10 \( \mu m \) \( (15.3 \pm 3.0 \text{ cps}) \) and 20 \( \mu m \) \( (13.5 \pm 2.5 \text{ cps}) \) \( (P < 0.01) \). However, no significant differences were observed among the results at position A. As for the results between the untreated group and the other positions, significant differences were observed.

DISCUSSION

Structure and function of direct laminate veneer
The use of a perforated laminate to bond to teeth without enamel etching or the unnecessary removal of enamel. The perforated laminate consists of a resin surface layer and a GIC base layer. GIC bonds to the enamel20-24), and its bonding strength is so high that a perforated laminate can be applied directly to the enamel without etching.

Concerning the bonding of GIC and resin, Farah et al.25) investigated the strength between resin-modified glass ionomer cement and light-cured composite resin. They reported that the materials showed a strong mutual adherence by chemical bonding, and that the bonding strength was adequate for clinical use. In an in vitro experiment using a ther-
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Fig. 8 (a) Typical fluoride intensity profiles of untreated enamel.

Fig. 8 (b) Typical fluoride intensity profiles of veneer-covered enamel, where A: Uncovered adjacent to tooth surface; B: Boundary area; and C: Veneer-covered tooth surface.

Fig. 9 Comparison of fluoride intensity. A: Uncovered adjacent tooth surface; B: Boundary area; C: Veneer-covered tooth surface. *** indicates P<0.001, ** indicates P<0.01, * indicates P<0.05 (as compared with Untreated group). ## indicates P<0.01, # indicates P<0.05.

In this experiment, we applied veneer to rabbit incisors and dripped phosphoric acid into the oral cavity every 24 hours for four weeks to examine whether it would protect the enamel in vivo. As a result, the tooth surface not covered with veneer was roughened, and enamel and dentin loss were likewise observed on the tooth surface not covered with veneer. As shown in Fig. 7, lesion depth was 110.9 ± 2.2 µm. These data seemed to indicate that, since the mouth is an acidic environment, it is inevitable that tooth surfaces are not only demineralized and roughened, but that some tooth material is also lost. However, no enamel loss was observed in the enamel surface covered with veneer, and no percolation was observed between the enamel and veneer. The GIC formed an adhesive bond with enamel20-24), and it appeared to be quite effective in sealing enamels.

It was thought that because GIC bonds chemically to enamel, near-edge sealing on the direct laminate veneer bonding side was excellent, and hence phosphoric acid was prevented from invading through the boundary between the veneer and enamel. Due to these findings, direct laminate veneer was considered to have prevented the loss and demineralization of enamel from the acidic mouth environment by having the tooth surface covered during the experimental period of four weeks.

These in vivo results are considered to support the finding by Miwa et al.19) concerning the sufficient physical properties of the veneer.
Release of fluoride from direct laminate veneer and acid resistance of the enamel

Despite advances in orthodontic materials and techniques, decay development around brackets during orthodontic treatment continues to be a problem. Therefore, prevention of these lesions during treatment is an important concern for both the orthodontist and patient.

O'Reilly et al.\textsuperscript{26} reported that fluoride regimes, such as combining a dentifrice (1100 ppm fluoride) and a mouth rinse (0.05% sodium fluoride), have been shown to reduce or prevent white spot lesions in orthodontic patients. Unfortunately, a compliance rate of only 13% was achieved with patients asked to decrease their caries risk with a daily fluoride mouth rinse\textsuperscript{27}. Therefore, in-office topical fluoride treatments have also been suggested to minimize the need for patient compliance\textsuperscript{30,32}. Nevertheless, demineralization lesions of significant depth (75 μm) can develop in just four weeks – a time shorter than many orthodontic appointment intervals of six to 10 weeks\textsuperscript{11}. To avoid these problems of compliance and treatment intervals, manufacturers have incorporated fluoride into orthodontic bonding cement to help prevent or reduce decay around the brackets.

In the present research, fluoride intensity was measured using EPMA and high values were indicated for both positions A and C in the experimental group as compared with the untreated group (Fig. 8). This was because fluoride was taken up by the enamel from the GIC that composed the direct laminate veneer. Moreover, it should be noted that a high value was not only indicated for position C (which was directly covered by veneer) but also for uncovered position A. This finding showed that fluoride released from the veneer was also taken up by the uncovered surrounding enamel. Gorton et al.\textsuperscript{30} reported that fluoride was taken up by the enamel surrounding the brackets, and demineralization was controlled when fluoride-releasing GIC was used as a bracket adhesive. Moreover, when brackets were bonded using GIC, Chadwick et al.\textsuperscript{31} believed that it contributed to the significant increase in fluoride concentration of the adjacent enamel. Based on the slightly higher fluoride intensity at position A compared with the untreated group (Fig. 9), it is thought that even at a depth of 20 μm, fluoride was taken up not only by the enamel directly beneath the veneer, but also by the surrounding enamel. Yamamoto et al.\textsuperscript{32} filled prepared cavities using fluoride-releasing restorative material in vivo, and after one month, they reported fluoride uptake in enamel up to a maximum depth of 47 μm using two-dimensional mapping by electron probe microanalysis (EPMA). It is thought that a similar result was obtained in this experiment.

As our results showed, fluoride ions were taken up by the adjacent enamel not covered by veneer, as well as by the covered enamel area. Thus, fluoride may also be delivered to contact points with adjacent teeth and the enamel below the gingival margin. In this connection, the use of direct laminate veneer is considered to be extremely advantageous for the prevention of caries in low, self-cleaning areas.

Forsten\textsuperscript{30} reported that GIC has a fluoride recharging capacity due to a reservoir effect. In this connection, fluoride release from the GIC veneer is expected to give additional protection to the teeth throughout treatment, and daily use of fluoridated toothpastes and/or rinses can reinforce this fluoride uptake-release feature. However, many aspects concerning the fluoride recharge of GIC, including optimal concentration, recharge intervals, and properties of GIC, remain to be evaluated. Hence, further studies on these subjects are still needed.

**CONCLUSION**

Protection of teeth from demineralization and caries is essential for obtaining optimal results in orthodontic treatment. Direct laminate veneer shows great potential in this aspect because it does not require enamel etching before application.

Besides, fluoride released from the veneer appeared to improve the acid resistance of not only the enamel covered with veneer but also the enamel not covered with veneer, such as the uncovered contact areas on adjacent teeth and areas under the gingival margin.

GIC – which composes direct laminate veneer – can be recharged by the administration of fluorides in the oral cavity. In this manner, continuous release of fluoride from the veneer is expected. Thus, the veneer may have a caries preventive effect by not only physically protecting the enamel, but also by strengthening the enamel with fluoride even when oral hygiene is markedly reduced during orthodontic treatment.

**REFERENCES**