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SEM-Evaluation of Enamel Surfaces After Orthodontic Debonding: A 6 and 12-Month Follow-Up *In Vivo* Study

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Summary: The purpose of this *in vivo* study was to compare the morphology of the enamel surfaces before bracket bonding and 6 and 12 months after debonding. Replicas of thirty-two maxillary second premolars of 16 volunteers were made before bracket bonding (T0), after debonding (T1), 6 months (T2), and 12 months (T3) later. Scanning electron microscope (SEM) images of the labial enamel surfaces were taken at T0, T1, T2, and T3 at increasing magnifications and analyzed according to the enamel damage index EDI. Data evaluation by using Friedman test followed by Wilcoxon signed ranks test with Bonferroni adjustment did not reveal statistically significant differences in the mean EDI at T0, T2, and T3, whereas the mean EDI at T1 was significantly higher than at T0, T2, and T3 ($p < 0.05$). The debonding procedure tested in this study produces no clinically relevant enamel damage. These alterations are reversible indeed, as a progressive restoration to pretreatment condition is evident after 6 months already and even more after 12 months. SCANNING 37:322–326, 2015. © 2015 Wiley Periodicals, Inc.

Key words: debonding, SEM, replicas, orthodontics

Introduction

At the end of orthodontic treatment with fixed appliances, brackets and any residual adhesive must be removed. Ideally, the debonding should restore the enamel surface as closely as possible to its pretreatment condition (; Schuler and Van Waes, 2003; Ozer et al., 2010). Several factors are involved in this procedure, the most important of which are the tools used for bracket removal and the instrumentation for resin removal (95; Brosh et al., 2004; Hosein et al., 2004; Kitahara-Céia et al., 2008).

No universally approved protocol has been established for the clean-up of adhesive resin after bracket removal. To date no instrument can achieve complete resin removal without affecting the enamel surface; scratches and grooves often appear on the enamel surface after the debonding procedure (Eliades et al., 2004; Ryf et al., 2012).

Most studies evaluating the effect of debonding have been conducted *in vitro*; they could not be highly relevant from the standpoint of both scientific and clinical evidence since major intraoral factors, such as saliva, oral hygiene, temperature, and pH changes are ignored (Alessandri et al., 2011; Zachrisson and Büyükyilmaz, 2012).

Furthermore the literature lacks of data about the persistence of the damage caused by debonding over time (Schuler and Van Waes, 2003).

For these reasons, starting from the results of a previous study about the effectiveness of a single debonding technique (Alessandri et al., 2011), an *in vivo* research was designed in order to compare the morphology of the enamel surfaces at the initial stage, immediately after debonding and 6 and 12 months after debonding. The null hypothesis was that no differences in the enamel surface morphology were detectable among the different stages.

Materials and Methods

Sixteen volunteer undergraduate students (5 women, 11 men; mean age, 23.8 ± 1.57 years) were included in the study and each of them signed informed consent. Brackets have been bonded on each second maxillary

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TABLE I Distribution of enamel damage index (EDI) grades before bonding (T0), after debonding and clean-up (T1), 6 months (T2), and 12 months (T3) later

	n	Grade 0[a]	Grade 1[b]	Grade 2[c]	Grade 3[d]	Mean	SD[e]	Median	Interquartile Range	Mean Rank
T0	32	27	5	0	0	0.2	0.369	0	0–0	2.2
T1	32	9	20	3	0	0.8	0.592	1	0–1	3.5
T2	32	28	4	0	0	0.1	0.336	0	0–0	2.2
T3	32	30	2	0	0	0.1	0.246	0	0–0	2.1

[a]Grade 0 = smooth surface without scratches, perikymata may be visible; [b]Grade 1 = acceptable surface, fine scattered scratches are present; [c]Grade 2 = rough surface, numerous coarses scratches and slight grooves are visible; [d]Grade 3 = surface with coarse scratches, wide grooves and enamel damage visible to the naked eye; [e]Standard Deviation.

bicuspid of the students and a total of 32 premolars were examined. Inclusion criteria were the absence of cavities on the labial surface and the lack of exposure to chemical agents (i.e., etching or bleaching agents)

The selected bicuspids were cleaned with nonfluoride pumice, then rinsed and dried. Replicas of the buccal surfaces were created from impressions of the teeth taken by employing polyvinyl siloxane (President Plus Light-Body, Coltene, Altstätten, Switzerland) and cast in epoxy resin (Epofix, Struers, Ballerup, Denmark) (T0) (Monaco et al., 2006; Campos et al., 2014).

The enamel was then etched with 35% orthophosphoric acid gel (Scotchbond, 3M Unitek, Monrovia, California) for 30 seconds, rinsed with water and carefully dried until the etched surfaces became white frosty (Al Shamsi et al., 2007). Subsequently, a bracket (Victory Series; 3M Unitek, Monrovia, California) was bonded (Transbond XT Light Cure adhesive primer and Transbond XT adhesive resin, 3M Unitek, Monrovia, California) on the center of clinical crown of each selected bicuspid following the manufacturer's instructions. A dental probe was used to remove the excess resin from the periphery of the bracket base. Resin polymerization was obtained by using a diode unit light (Ortholux, 3M Unitek) for 10 seconds from the mesial side and 10 seconds from the distal side.

The brackets were removed 7 days after bonding, to be sure that polymerization was complete in terms of bond strength efficacy (Eliades et al., 2004).

The bracket debonding was performed by gently squeezing the mesial and distal wings with bracket removal pliers (Ormco Corporation, Glendora, California). A 12-blade tungsten carbide bur (H247; Komet, Lemgo, Germany) driven on a low speed hand piece at 20,000 rpm, without water cooling, was used to remove the gross adhesive remnant from the enamel surface. Subsequently, finishing the residual resin with medium, fine, and ultrafine discs (Sof-Lex; 3M Dental, St Paul, Minnesota) driven on a low speed hand piece at 10,000 rpm was carried out (Oliver and Griffiths, '92). Proper removal of the composite resin was checked on the basis of visual inspection of the enamel surfaces under a dental operating light. The same operator (G.A.) accomplished all clinical steps. At that time (T1), another impression of the buccal bicuspid surfaces was taken and replicated in epoxy resin. Patients were not allowed to get professional dental hygiene for the following 12 months. Six months (T2) and 12 months (T3) later, new impressions of the buccal surfaces of the bicuspids were taken and epoxy resin replicas were obtained.

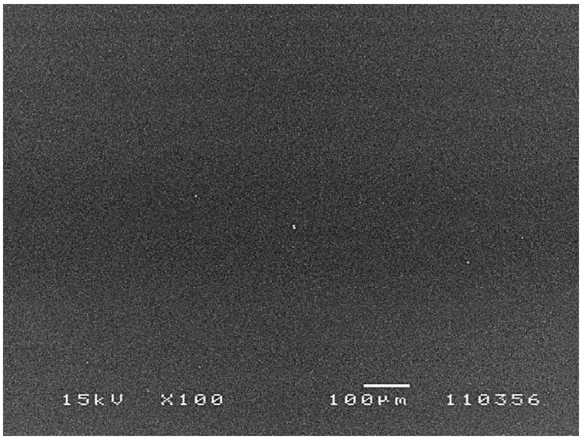

Fig. 1. SEM microghraph (100×) of an enamel surface with an EDI grade of 0.

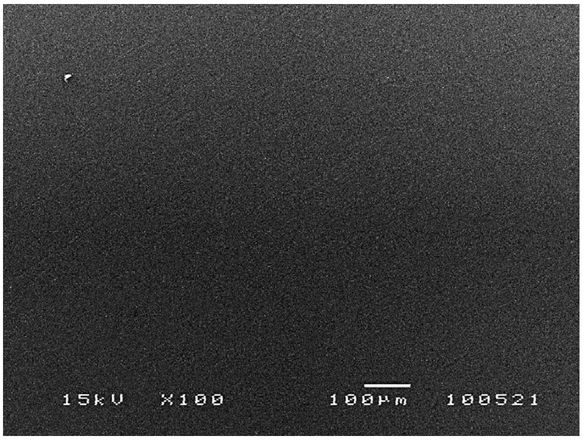

Fig. 2. SEM microghraph (100×) of an enamel surface with an EDI grade of 1.

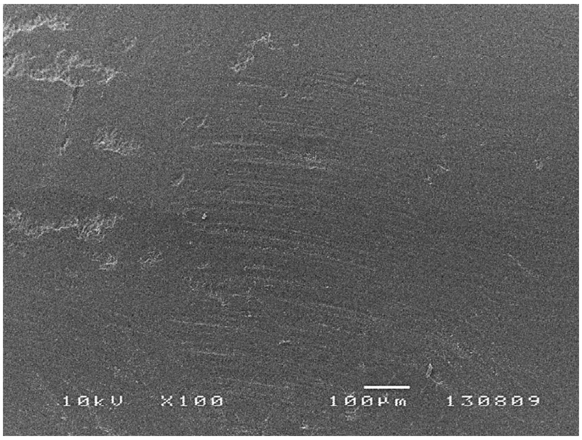

Fig. 3. SEM microghraph (100×) of an enamel surface with an EDI grade of 2.

The dental replicas were mounted on aluminum stubs, sputter-coated with a 300 Å layer of gold and palladium (Sputter Coater SC7620; Polaron, East Grinstead, United Kingdom) and analyzed with a high vacuum scanning electron microscope (JSM-5200; JEOL, Tokyo, Japan) capturing secondary electron images at 10–15 kV, at a working distance of 20 mm. The replica technique represents a high precision method to inspect tooth morphology, as demonstrated by its employment not only in orthodontics research but also in many dentistry fields (Monaco et al., 2006; Baroni et al., 2014; Campos et al., 2014)

The enamel surfaces of the second maxillary bicuspids at T0, T1, T2, and T3 were evaluated following the enamel damage index (EDI) (Schuler and van Waes, 2003), adapted from the surface roughness index by . This index comprises of 4 grades: 0, smooth surface without scratches, and perikymata might be visible; 1, acceptable surface, with fine scattered scratches; 2, rough surface with numerous coarse scratches or slight grooves visible; and grade 3, surface with coarse scratches, wide grooves and enamel damage visible to the naked eye.

Microphotographs of the epoxy resin replicas were taken at increasing magnifications (35 and 100 times) to determine the EDI grade. The 35× magnification allowed examination of the labial surface of the tooth on the whole; the center of tooth clinical crown area was then inspected in detail at 100× to assess the quality of the surface morphology. A systematic method (Marchionni et al., 2010) providing records on predetermined points on the surface of sample has been adopted for SEM images observation and interpretation, allowing 100% repeatability in finding the same observation fields.

EDI evaluations were performed by an expert and calibrated examiner (M.S.), trained in micromorphologic evaluations (Alessandri et al., 2011; Zanarini et al.,2013), who did not know to which phase (T0, T1, T2, or T3) the specimens belonged. The evaluations were repeated by the same examiner 1 week later. If any discrepancies arose, a third assessment was performed to determine the final score. The study design was approved by the Local Institutional Review Board.

Statistical Analysis

A power analysis was conducted to determine the correct sample size. By hypothesizing a medium effect size as relative magnitude of the experimental treatment along the measurements (T0, T1, T2, and T3) in the enamel damage index of 0.5 points, 10 specimens were required at least. A non parametric Friedman ANOVA test was used to determine any difference among the measurements ($p < 0.05$). Post-hoc pair-wise comparisons between measurements were calculated by using the Wilcoxon signed rank test with Bonferroni adjustment. Statistical significance was defined as $p < 0.05$.

Results

No discrepancies arose in the determination of the EDI grades between the first and second assessments by the examiner, so a third assessment was not required. All 32 teeth were examined 4 times (at T0, T1, T2, and T3). The distribution of the EDI grades is shown in Table I. Figures 1, 2, and 3 show representative examples of enamel surfaces exhibiting the different EDI grades observed. The non-parametric Friedman test revealed statistically significant differences among measurements ($p < 0.05$). Subsequently, post-hoc comparisons were carried out by using the Wilcoxon test: no statistically significant differences in the mean EDI at T0, T2, and T3 were found, whereas the mean EDI at T1 was significantly higher than at T0, T2, and T3 ($p < 0.0125$).

Discussion

Damage to the enamel due to debonding has been, and still is, a concern to clinicians because it is reported to occur *in vitro* and *in vivo* (Al Shamsi et al., 2007; Pont et al., 2010; Zanarini et al., 2013). The outermost enamel layer is reported to contain more fluoride and have higher hardness than the deeper zone (Øgaard, 2001). Consequently, scratches and grooves caused by the removal of resin can lead to reduced resistance of the enamel to the organic acids, making teeth more susceptible to plaque decalcification ().

Even though there is lack of consensus about the safest, least aggressive technique to remove residual resin, the use of a tungsten carbide bur followed by discs is supported by several authors. suggested using a tungsten carbide bur to remove bulk and residual resin from teeth and to finish the underlying enamel using graded discs or ceramic wheels. Others (Rouleau et al., '82;) rated the tungsten carbide bur used at low speed as the "gold standard." In accordance with , proposed the use of a 12-bladed tungsten carbide bur followed by Sof-Lex discs to produce a smoother enamel surface. In a previous article Alessandri Bonetti et al. (2011) tested the effectiveness a high-speed 12-bladed tungsten carbide bur followed by finishing with graded medium, fine, and superfine Sof-Lex and observed that it did not restore the enamel surface to its pretreatment condition, even though there was no clinically relevant damage. The same instrumentation was adopted in the present study. On a larger study sample than the previous article, a comparison between the enamel surface appearance before bonding and at 6 and 12-month follow-up after debonding, was carried out to evaluate any changes occurred to the enamel over time. By classifying SEM images, taken at a magnification of 100×, through the EDI grading system (Pont et al., 2010; Alessandri et al., 2011), the damage severity was established.

Only 9 tooth surfaces out of 27 showing a grade of 0 at T0 were classified again as grade 0 at T1, whereas the remaining 20 obtained a grade of 1 at with T1. Among the 5 tooth surfaces exhibiting a grade of 1 at T0, 2 had the same grade at T2, and the remaining 3 received a grade of 2. No specimen was classified as grade 3. Similarly to the findings of other researches, it emerged that all rotating instruments that are effective for the removal of resin remnants determine some abrasion to the enamel layer (). The debonding technique adopted in this study determined some surface alterations, however they were not severe enough to affect the integrity of the enamel surface (Alessandri et al., 2011).

At T2 and T3 a significant improvement in surface quality was noted: 18 surfaces out of 20 with a grade of 1 at T1 were classified as grade 0 at T2; 1 specimen of the remaining 2 was classified as grade 0 at T3, whereas the other received a grade 1 also at T2 and T3. As for the 3 tooth surfaces with a grade of 2 at T2, 1 of them was even classified as grade 0 at T2, 1 improved 1 point in the EDI at T2 and another point at T3, thus becoming a grade 0, whereas the third one received a grade of 1 either at T2 or at T3. These findings are in contrast with those of Schuler and Van Waes (2003), who reported no sign of improvement after 12 months in the large majority of the analyzed teeth. One possible explanation for this difference could be the SEM magnification adopted in the latter study (10×) and the different clean-up technique. No other follow-up researches are available in literature.

The disappearance of scratches may be explained by the precipitation of minerals within saliva on the enamel surface allowing to consider saliva a valid healing aid, able to repair early enamel lesions caused by debonding. (; Alessandri Bonetti et al., 2013; Paganelli et al., 2015). Furthermore the tooth surface is subjected to physiological buccal wear with age (about 2 microns per year) (). Over time new scratches could appear from toothbrushing and other mechanical procedures, indeed.

Conclusions

In light of the findings of the present study the following conclusions can be drawn:

- The clean-up technique adopted produces superficial enamel damage (statistically significant difference between T0 and T1).
- The surface alterations produced by the clean-up technique adopted are reversible as a progressive restoration to pretreatment condition is already evident after 6 months, and even more after 12 months (no statistically significant differences among T0, T2, and T3).

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