Cigarette smoke affects bonding to dentin

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This *in vitro* study evaluated the microtensile bond strength (μTBS) of composite resin bonded to dentin that had been contaminated by cigarette smoke. Ten extracted unerupted human third molars were used: Six molars were prepared for μTBS testing, while the other four molars were assigned to pre- and post-etching scanning electronic microscopy (SEM) analysis. The 20 specimens obtained from the 10 coronal portions were distributed into two experimental groups so that each tooth served as its own control. Group 1 underwent a daily toothbrushing simulation and exposure to a smoking simulation chamber, while Group 2 received only a daily simulated toothbrushing. Student's t-test demonstrated that Group 1 samples demonstrated significantly lower bond strength (49.58 MPa) than Group 2 samples (58.48 MPa). Pre and post-etching SEM analysis revealed the presence of contaminants on the dentinal surfaces of the Group 1 specimens. It was concluded that contamination by cigarette smoke decreases the bond strength between dentin and composite resin.

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Dentin is a more heterogeneous, humid, and organic substrate than enamel, which makes bonding to it more difficult.[1] Bonding restorative materials to dental substrates requires contact between the tooth and the adhesive system.[1,2] Studies involving different contaminant agents have indicated a decreased bond strength between contaminated dentin and composite resin.[3-9] In addition, the condition of the substrate can also affect bonding; as Tay and Pashley noted in 2004, pathologically altered dentin (such as noncarious sclerotic cervical dentin) demonstrates decreased bonding values when compared to normal dentin.[10]

According to a 2002 report, smokers consititute approximately 33% of the world's adult population, indicating that tobacco consumption has reached global epidemic proportions.[11] Cigarette smoke consists of two phases: the vapor phase and the particulate phase. The *vapor phase* is defined as the portion of cigarette smoke that would pass through a Cambridge glass fiber filter. The *particulate phase* (also known as *tar*) is the portion that is trapped on the glass fiber filter and consists of particles that range in diameter from 0.1 μm to <1.0 μm.[12]

Dentin exposure can result due to noncarious cervical lesions, dental erosion lesions, fractured teeth, and carious lesions.[13-21] In such situations, cigarette smoke is a potential contaminant.

The connection between tooth staining and tobacco consumption is well-established: The impregnation of cigarette smoke contaminants causes smokers' teeth to turn a yellow (or even black) color, and the staining level is positively influenced by the number of cigarettes consumed.[22-24] Nevertheless, there is no evidence in the literature indicating how exposure to cigarette smoke affects adhesive bonding. To test the hypothesis that exposure to cigarette smoke impairs bonding to dentin, this *in vitro* study evaluated the microtensile bond strength (μTBS) of a composite resin bonded to dentin that had been contaminated by cigarette smoke.

Materials and methods

Specimen preparation

Ten extracted human third molars were stored in distilled water at 4°C and used within three months of extraction. Using a diamond saw (Isomet 1000, Buehler Ltd.), the root portion of each tooth was removed by a perpendicular section at the cementoenamel junction. The dentinal surface of the remaining coronal portions were wet-ground using 600 grit sandpaper under copious water cooling until any pulp horn projections were eliminated. The substrate used for bonding was deep dentin via apical access.[25]

A longitudinal section was performed (in a mesiodistal direction) on each coronal portion, producing 20 halves from the 10 coronal portions. To isolate the dentinal surface, each half was embedded in epoxy resin, using a bipartite metallic matrix. After the epoxy resin set, each specimen was labeled and wet-ground (using 1200 grit sandpaper under copious water cooling) to prevent any epoxy resin from overlapping on the dentinal surface. The specimens were distributed into two experimental groups (n = 10), with half of each tooth assigned to each experimental group, so that each specimen served as its own control.

Exposure to cigarette smoke

Using a smoking simulation device (Fig. 1), Group 1 specimens were exposed to 30 Marlboro cigarettes a day for 17 days. The specimens were inserted in this smoking simulation chamber and aligned so that their dentinal surfaces did not contact the bottom of the chamber. A total of 510 cigarettes were "smoked" by the end of the testing period. Each cigarette was consumed in approximately two minutes. At the end of each day of cigarette smoke exposure, simulated toothbrushing was performed on specimens in both groups.

Toothbrushing simulation

All specimens were subjected to a daily toothbrushing simulation, using a device created especially for this study (Fig. 2 and 3). The specimens were put into receptacles with their dentinal surfaces facing the heads of the electric toothbrushes (Oral-B Advance Power 400, Procter & Gamble). A toothpaste/water solution was prepared daily and poured on the dentinal surfaces of each specimen; at that point, the electric toothbrushes were turned on and the specimens were "brushed" for 30 seconds.[26,27]

μTBS testing

At the end of the 17-day period, a μTBS test was conducted on six specimens from Group 1 and their counterparts from Group 2. Using a three-step etch-and-rinse adhesive system (Scotchbond Multi-Purpose, 3M ESPE), the specimens were subjected to a bonding treatment in accordance with the manufacturer's instructions. Using a composite resin (Filtek Z250, 3M ESPE), the specimen's dentinal surface was built up to a height of 6 mm. The specimens were stored in distilled water (37°C) for 24 hours, then subjected to μTBS testing.

Fig. 1. *Top left*: Lateral view of the smoking simulator device. *Top right:* Anterior view of the smoking simulator device. A hose connected to a pressure compressor on the back of the device simulates smoking. *Bottom left*: The chamber interior where the specimens were inserted. *Bottom right:* A cigarette is positioned to simulate smoking.

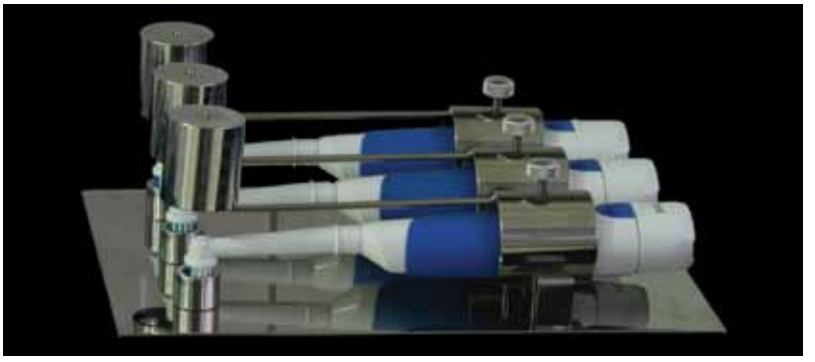

Fig. 2. A lateral view of the toothbrushing simulation device.

Each specimen was longitudinally sectioned in both directions to obtain rectangular sticks with a cross-sectional area of approximately 0.45 mm^2. The sticks were fixed to Geraldelli's jig with cyanoacrylate glue.[28] To isolate the dentin-composite interface for μTBS testing, the glue was applied to both extremities of each stick and at the dentin-enamel junction. Using

Fig. 3. The specimen and receptacle positioned for simulated toothbrushing.

Table. Mean μTBS values (in MPa); summary statistics by group, $p = 0.001$.

Group	Number of specimens	Mean μTBS (SD)
1	83	49.58 (17.41)
2	77	58.48 (15.92)

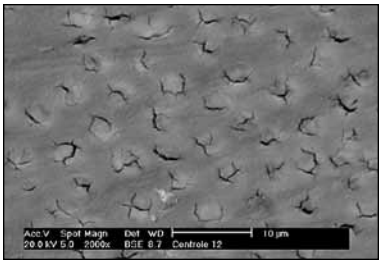

Fig. 4. The dentinal surface of a specimen from Group 2, after wet grinding (using 1200 grit sandpaper) and toothbrushing simulation (magnification 2,000x).

a universal testing machine (Instron 4444, Instron Corp.), the sticks were stressed at a crosshead speed of 0.5 mm/minute until failure. The μTBS values were expressed in MPa by dividing the imposed force at the time of fracture (in N) by the bonded area (in mm^2).

All specimens that demonstrated both dentin and resin cohesive failures were eliminated from the research. The failure mode of each stick was analyzed under a 25x magnification microscope after debonding. The failure modes were classified into four types: A (adhesive failure), B (resin cohesive failure), C (dentin cohesive failure), and D (mixed failure—that is, adhesive failure with some dentin or resin cohesive involvement).[29]

To detect equality or difference between the tested groups, Student's t-test was applied to the μTBS mean values. After 17 days of toothbrushing simulation and exposure to cigarette smoke, the dentinal surfaces of the specimens in Group 1 were visibly contaminated. In order to better understand the contamination by cigarette smoke, pre- and post-etching scanning electronic microscopy (SEM) analysis (Philips XL-30, FEI Company) was performed on the remaining four specimens in Group 1 and their counterparts in Group 2.

Results

The mean μTBS values of the tested groups are presented in the table. Statistical analysis (Student's t-test) revealed significant difference between the mean bond strengths of the tested groups ($p = 0.001$). Group 1 specimens (49.58 MPa) demonstrated lower bond strength values than Group 2 specimens (58.48 MPa).

Failure mode analysis

The failure mode of the debonded sticks, as determined by means of stereomicroscopy (magnification 25x) rather than a statistical test, revealed only adhesive and mixed failures. No cohesive failures were found. Group 1 specimens exhibited 72 (86.7%) adhesive failures and 11 (13.2%) mixed failures, whereas Group 2 specimens demonstrated 63 (81.8%) adhesive failures and 14 (18.1%) mixed failures. The lower bond strength of the Group 1 specimens is in accordance with this group's higher incidence of adhesive failures.

SEM analysis

Figures 4–7 show pre- and post-etching SEM analysis of the dentinal surfaces in specimens from each group.

Discussion

The results of the present study show that cigarette smoke contamination decreases the μTBS values of dentin, confirming the hypothesis that dentinal exposure to cigarette smoke impairs bonding.

Dentin bonding systems are sensitive to contamination by an excess of water, saliva, and plasma, due to hydroxapatite's capacity for macromolecule adsorption.[30] Blood contamination reduces dentin bond strength because of blood's high protein content (6–7%). Blood, in combination with macromolecules (that is, fibrinogen and platelets), can form a film and prevent the adhesive system from infiltrating into the underlying dentin.[3] Some plasma macromolecules (as platelets) can range in diameter from 0.5–5 μm, while cigarette smoke particles can range in diameter from 0.1 μm to <1.0 μm.[12,30] Based on these factors, dentin hydroxyapatite may have adsorbed the cigarette smoke particles, preventing contact between dentin and the adhesive system and decreasing dentin μTBS values.

Considering the results of this study, patients who smoke should be excluded from clinical trials involving noncarious Class V adhesive restorations in which the exposed dentin stands as the main bondable substrate.[31]

μTBS values are influenced by the dimension and geometry of the interfacial area tested.[32] For this study, bonding was performed in deep dentin, and rectangular sticks (with a cross-sectional area of approximately 0.45 mm^2) were obtained using a nontrimming technique.[33] It is difficult to make

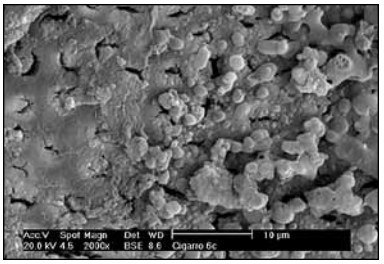

Fig. 5. The dentinal surface of a specimen from Group 1, after wet grinding (using 1200 grit sandpaper), toothbrushing simulation, and exposure to cigarette smoke (magnification 2,000x).

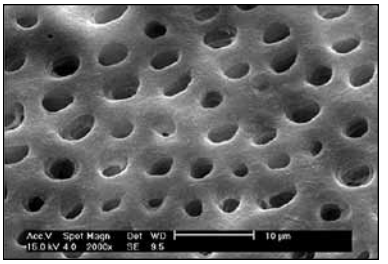

Fig. 6. The dentinal surface of a specimen from Group 2, after wet grinding (using 1200 grit sandpaper), toothbrushing simulation, and 15 seconds of etching with 35% phosphoric acid (magnification 2,000x).

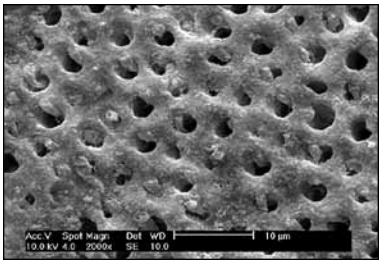

Fig. 7. The dentinal surface of a specimen from Group 1, after wet grinding (using 1200 grit sandpaper), toothbrushing simulation, cigarette smoke exposure, and 15 seconds of etching with 35% phosphoric acid (magnification 2,000x).

realistic comparisons between μTBS values from different studies due to the variations in interfacial geometry and dentin depth.[32,34,35] The failure mode analysis showed a higher prevalence of adhesive failures than mixed failures; in addition, Group 1 exhibited more adhesive failures than Group 2, indicating a weaker hybrid layer for dentin contaminated by cigarette smoke.

In order to expose dentinal substrate to cigarette smoke, a device was constructed to simulate smoking and its effect on the oral cavity. It is possible that the presence of saliva, oral soft tissues, and sclerotic dentin could modify the contamination pattern of cigarette smoke.[10,36] This was an *in vitro* study that utilized a short smoking regime (510 cigarettes during 17 days, or slightly more than one pack of cigarettes per day). Future studies should consider increasing the exposure to cigarette smoke and providing qualitative assessments regarding contamination level and bond strength.

Because toothbrushing could affect and modify the cigarette smoke contamination pattern, a toothbrushing simulation device was constructed to create a more accurate clinical simulation. Each specimen was "brushed" for 30 seconds a day, simulating brushing three times a day using a pressure of 200 g.[26,37] After simulated toothbrushing and exposure to cigarette smoke, it was visually verified that specimens in Group 1 had taken on a black color. SEM analysis showed that the darkening process probably occurred due to contamination by the cigarette smoke particulate phase (tar) (Fig. 5).[12] Post-etching SEM revealed the presence of cigarette smoke contaminants that appear to have been modified by acid-etching; this particulate phase partially fills and blocks the dentinal tubules lumens. Acid-etching apparently could not remove the modified smear layer completely; as a result, the surface remained contaminated (Fig. 7).

This study utilized a three-step etch-and-rinse adhesive system. Acid-etching could not remove the cigarette smoke contaminants from the dentinal surface, which suggests that future studies should involve conservative approaches for smear layer removal, such as sandblasting and smear layer modification by means of a self-etch adhesive system.

Student's t-test was applied to each specimen. There appears to be a trend to conduct statistical analysis by using the μTBS mean value of sticks from the same tooth as the experimental unit.[38,39] Because intratooth variability is greater than intertooth variability, the specimen alone cannot be considered independent.[39,40] In the present study, 83 specimens were subjected to cigarette smoke and 77 were not; in addition, dentin depth was standardized and each tooth served as its own control.[40]

Conclusion

The results of this study indicate that contamination by cigarette smoke decreases the bond strength between dentin and composite resin. Little is known of cigarette smoke's influence on adhesive restorations in daily practice. The results of this *in vitro* study alone cannot assess the clinical effectiveness of adhesive restorations containing composite resin in the restored dentin of patients who smoke. However, patients should be alerted that bonding to dentin that has been contaminated by cigarette smoke may result in restoration loss and microleakage.

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Disclaimer

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