The influence of caries-affected dentin on bonding effect of deciduous teeth

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The influence of caries-affected dentin on bonding effect of deciduous teeth

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Abstract

Aim

The aim of this study was to investigate the influence of caries-affected dentin on bonding effect of deciduous teeth, in turn to provide guidance for caries removal strategy of deciduous tooth caries before bonding.

Methods

Extracted second deciduous molars with one adjacent surface carious were collected. The half tooth with caries adjacent surface were grouped into the caries-affected group (Group test), while the caries-free half were grouped into the control group (Group control). For the Group test, carious tissue was removed with slow speed ball drill under the indication of caries indicator. For the Group control, at the other side of the occlusal surface sound dentin was prepared to the similar deep level. Single bond Universal was applied according to the manufacture's instructions. The defective part was filled with Shofu Beautifil II Namer, a commonly used resin composite in pediatric dentistry. Vertical sticks were obtained and subjected to tensile stress test. The morphology of the bonding surfaces were observed by scanning electron microscope and transmission electron microscopy.

Results

Electron microscope observation showed that there was no significant difference in morphology between caries-affected dentin and normal dentin. The minerals and smear layer were removed after acid etching, and the collagen network of dentin tubules and intertubular dentin was exposed. The surface of caries-affected dentin was relatively rough and partially decomposed. TEM images of the caries-affected deciduous tooth dentin showed that the hydroxyapatite was sparsely distributed in strips. In normal dentin, hydroxyapatite was thicker and denser. The acidic environment produced by the bacteria demineralized the dentin part, which corresponded to the surface morphology of the caries-affected dentin as seen by SEM. There was no significant difference in the micro-tensile strengths between Group test and Group control ($P > 0.05$).

Conclusion

In vitro, caries-affected deciduous tooth dentin could achieve similar bonding strength to normal tooth dentin when Single Bond Universal adhesive was applied. The results of this study may provide theoretical basis for partial caries removal strategy of deciduous tooth caries treatment.

Keywords

Caries-affected dentin; micro-tensile strength; deciduous tooth; Single bond Universal adhesive

1 Introduction

Dental caries is the main content of clinical work for general pediatric dentists. In clinical work, dental caries must be treated with removal of carious tissues before restoration. Resin filling of deciduous teeth has always been the main method for repairing tooth defects of deciduous teeth[1-3]. Dentin is the main structure of deciduous teeth and the most common bonding interface. For carious teeth, dentin that has been demineralized without bacterial invasion, was named caries affects caries-affected dentin (CAD)[4, 5]. This part is the significant part of clinical bonding. With the development of preservation dentistry, on the pulp wall pediatric dentists generally remove only the part of dentin that bacteria invaded.

Caries removal techniques used in pediatric dentistry include bur drilling, chemomechanical method and laser. Different methods have their own advantages and disadvantages. The use of burs is popular worldwide but usually causes discomfort and pain. Carisolv is a chemomechanical caries removal system, including sodium hypochlorite (0.95%) and 3 amino acids (lysine, leucine, and glutamic acid) in a gel preparation. After treatment with Carisolv, the softened carious dentin can thus be removed. These methods are likely to retain part of the caries-effected dentin [6-9] . Under the requirements of preservation and comfortable dentistry, it is necessary to retain part of the hardened caries-affected dentin on the pulp wall of cavity, especially in the treatment of children, which can reduce the use of anesthetics and reduce the pain and discomfort.

Previous studies [10-14] have compared the effects of artificial demineralized dentin and normal dentin on bonding strength. However, whether the caries-affected dentin really affects the bonding effect or not is unknown. In the present study, we investigated the influence of caries-affected dentin on bonding effect based on extracted carious deciduous teeth.

2 Materials and methods

2.1 Sample Preparation

After approval from the Institutional Ethics Board, 12 second deciduous molars dentin with one adjacent surface carious were collected from the Department of

Stomatology of the Stomatology Hospital of Zhejiang University Medical College in 2018. The teeth were stored in 0.1% sodium azide solution and used within 3 months.

The enamel of the tooth was removed with high-speed diamond bur and dentin was exposed. The half tooth with caries adjacent surface were grouped into the caries-affected group (Group test), while the caries-free half were grouped into the normal control group (Group control). The caries cavity was dripped with the caries detector dye and rinsed. Then a slow speed ball drill with the spray of water was used to remove the stained dentin. Next, caries detector dye was used again, and the stained dentin was removed. Repeat this operation until the dentin cannot be stained (Group test), and then at the other side of the occlusal surface sound dentin was prepared to the similar deep level and used as a normal dentin group (Group control).

After the dentinal surface was dried with an absorbent, the single bond universal adhesive (Single bond Universal SBU, USA) was applied on the dentin for 20 seconds, blow lightly for 5 seconds, light (VALO CORDLESS) cured further. Next, a composite resin (Shofu Beautifil II, Japan) was added in 1-mm-thick increments up to 5 mm thickness. Then, each increment was light cured for 20 seconds. The samples were rinsed in the water and further stored in 37 ° C saline. The two groups of the samples were embedded in the plaster to avoid the cracks during the cutting process. The obtained samples were segmented into about 1.0 mm × 1.0 mm sticks by using the diamond bur of saw shaped and water used as coolant. The actual cross-sectional area of each samples (S, mm^2) was calculated using vernier caliper records. A total of 24 dentin blocks were used in this study (in each group 12 dentin blocks were evaluated).

2.2 Analysis of micro-tensile bond strength (μTBS)

International Standards Organization (ISO) Technical Specification No. 11405 has been used in this study[15]. This specification provides guidance on substrate selection, storage, and handling as well as essential characteristics of different test methods for quality testing of the adhesive bond between restorative materials and tooth structure. It also presents some specific test methods for bond strength measurements.

Twelve specimens of caries-affected dentin and normal dentin were evaluated respectively. The sticks were placed on a paper towel to absorb the excess moisture. Individual sticks were bonded onto a test block of the micro-tensile testing machine (Bisco, Schaumburg, IL, USA). The block was composed of two test jaws with sticks testing surfaces, linkage and slide keys. We ensured that no tension was placed on the stick, and we placed a drop of glue on each jaw in the middle of the test stick mount area. All sticks were parallel to the direction of the linkage to prevent sideways vectors for tension. The test block was run at a speed of 0.8 mm / min until the sample was broken.

Then μTBS was calculated by dividing the load at failure (P) by the cross-sectional bonding area. And according to the formula: micro-tensile bond strength (μTBS) = P / S (MPa.)

2.3 Analysis of Scanning electron microscope

Three samples each of the caries-affected dentin group specimens and the normal dentin group specimens were selected, and the observation surface was treated with 37% phosphoric acid for 15 seconds to remove mineralization on the dentin surface, dehydrated, finally polished and sputtered with gold-palladium. Scanning electron microscopy (SEM, Nova Nano 450) was used to observe the two types of demineralized dentin. The most representative areas were photographed.

2.4 Sample Preparation for Transmission Electron Microscopy

The caries-affected and normal dentin specimens were fixed in Karnovsky's fixative and postfixed in 1% osmium tetroxide. After fixation, each specimen was rinsed three times with phosphate buffer saline (PBS). The specimens were dehydrated in an ascending series of ethanol (30−100%) and acetone. Then, they were subsequently embedded in epoxy resin. Ultrathin sections (70−90 nm) were prepared and examined by transmission electron microscopy (TEM, JEM-1230, JEOL, Tokyo, Japan) operated at 80 kV.

2.5 Statistical analysis

The statistical software SPSS l7.0 was used. For the analysis and comparison between the groups regarding the micro-tensile bond strength, paired t-test was used. $P < 0.05$ was considered statistically significant.

3 Result

3.1 Assessment of micro-tensile bond strength

Micro-tensile bond strengths for Group test and control were showed in Figure 1. The results showed that there was no statistical difference between Group test and Group control ($P > 0.05$).

3.2 Failure models

Figure 1 showed the distribution of failure modes for the different groups in primary teeth. Most of the samples showed cohesive failure. According to the Chi-square test (Fisher's exact test), there were no statistically significant differences in failure modes between the groups ($p = 0.244 > 0.05$).

3.3 Bonding morphology observation

SEM micrographs of the deciduous dentin after dentin preparation were shown in Figure 2. After removal of the caries with burs, the caries-affected dentine surface was rough (A-B). There was little obvious residual smear layer on the dentine surface with few dentinal tubules remaining occluded with smear plugs. This surface was irregular (original magnification 2000×). The control normal dentine surface presented open dentinal tubules distributed on a smooth surface almost free of a smear layer (C-D). The surface was generally free of smear layer and accompanied by open dentinal tubules. Some collagen fibrils were both visible at group test and group control (original magnification 2000×).

3.4 Transmission electron microscopic observation

Unstained TEM images of cross-section of the caries-affected deciduous tooth dentin and normal deciduous tooth dentin (5000×, 50000×) are shown in Figure 3. Unstained TEM images of cross-section of the interfaces between dentin (caries-affected deciduous tooth dentin and normal deciduous tooth dentin) and resin (5000×, 50000×)

were shown in Figure 4. Hydroxyapatite is sparsely distributed in strips in the caries-affected deciduous tooth dentin (50000×). In normal dentin, hydroxyapatite is thicker and denser. The acidic environment produced by the bacteria demineralizes the dentin part, which corresponds to the surface morphology of the caries-affected dentin as seen by SEM. There was no significant difference in the dentine matrix in the mixed layer between the two groups after application of the adhesive.

4 Discussion

The success of resin restoration mainly depend on the solid bonding interface. A good bonding system not only form a durable and effective bonding interface, but also effectively repair and seal the cavity and dentin pulp complex, prevent postoperative sensitivity and the occurrence of micro-leakage[16-19]. The condition of bonding surface influence the bonding effect. Dentin caries is divided into outer caries infected dentin and inner caries-affected dentin. Although the caries-affected dentin is partially demineralized, there is no bacterial invasion. Pediatric dentists have adopted more minimally invasive dental treatment concepts, like single tooth anesthesia, stainless steel preformed crowns technology and the most alternative way is to retain partially hardened and carious-affected dentin. The incomplete dentine caries removal technique is one of the relatively easy preservation dentistry methods. The reports on the influence of carious-affected dentin on bond effects were contradictory. Nicoloso et al. [10]reported that artificially-induced caries-affected deciduous dentin showed about 19.6 MPa bond strength. Lenzi et al. [11]found that the bond strengths of several adhesives on sound deciduous dentin were better than the one on artificially-induced caries-affected dentin. This study focused on the influence of caries-affected deciduous dentin on the bond effect.

Since the 1950s, Buonocore's acid-etched technique was proposed[20]. After that, dental adhesives have evolved from the first generation of adhesives that attempt to chemically bond teeth structure to the 8th-generation one-step self-etching system[21, 22]. The bonding strength has been greatly improved and technical sensitivity decreased. Previous researches have shown that the Single bond Universal adhesive SBU can achieve strong bonding strength with both the self-etching and full-etching bonding technique[23, 24] . Because of its simple operation steps and short operation time, universal adhesives are popular among pediatric dentists. Xuan et al.

[25] found that both for caries-affected dentin and normal dentin, the bonding strength of the full acid etching bonding system was acceptable. However, Paranhos et al. [26] believed that the micro-tensile adhesive strength of caries-affected dentin was significantly lower than the original adhesive strength. Tachibana et al. [27] showed that for caries-affected dentin with self-etching bonding system, different caries removal methods have no effect on the bonding strength. The contradictory results may come from the different adhesive systems.

In the present study, the 8th-generation one-step self-etching system was used. The results of micro-tensile bond strength showed that there was no statistical difference in bond strength between the caries-affected deciduous dentin and the normal deciduous dentin. The adhesion mechanism of dentin adhesive systems is thought to be related to the creation of a hybrid layer[28]. In our study, the structure of dentin under electron microscope observation showed that there was no significant difference in morphology between caries-affected dentin and normal dentin. The minerals and smear layer were removed after acid etching, and the collagen network of dentin tubules and intertubular dentin was exposed. The surface of caries-affected dentin was relatively rough and partially decomposed.These structural properties of two groups may have little influence on the hybrid layer characteristics and the bond strength values of deciduous dentin.

5.Conclusions

In vitro, caries-effected deciduous tooth dentin could achieve similar bond strength as normal deciduous tooth dentin when Single Bond Universal adhesive was applied. The results of this study may provide theoretical basis for partial caries removal method treatment of deciduous tooth caries. There were also some limitations in this study. Firstly, because only retained deciduous teeth were extracted clinically, the number of second deciduous molars in the present study was limited. Secondly, although the second deciduous molar is the largest deciduous tooth in the deciduous dentition, there are few slices that can obtain in the end due to the special anatomical structure factors such as root absorption and high pulp Angle in the pulp cavity. The immediate and long-term microleakage of in vitro deciduous teeth after filling and repairing remains to be further studied.

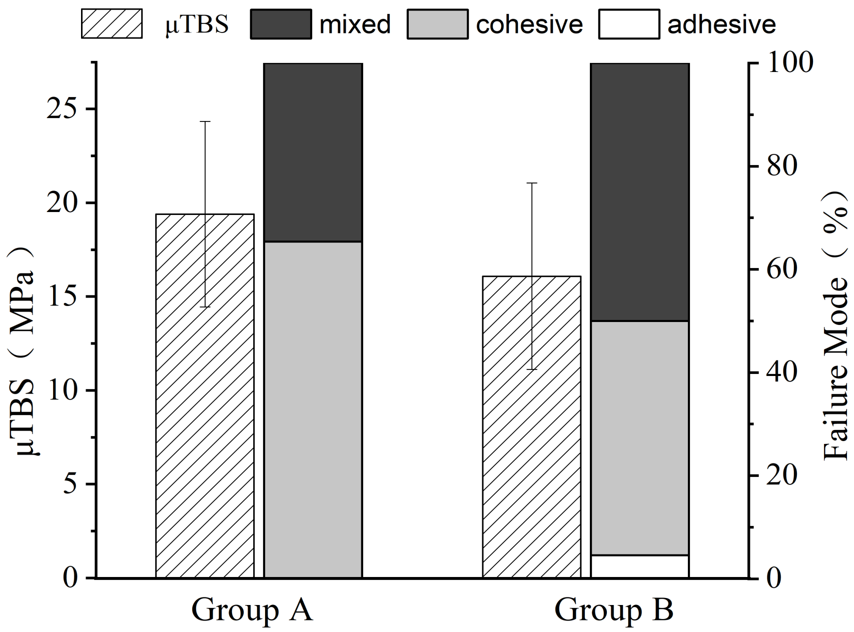

Figure 1. Micro tensile bond strength of different groups in MPa (means ± SD) and Graphic of percentage distribution of the different failure modes

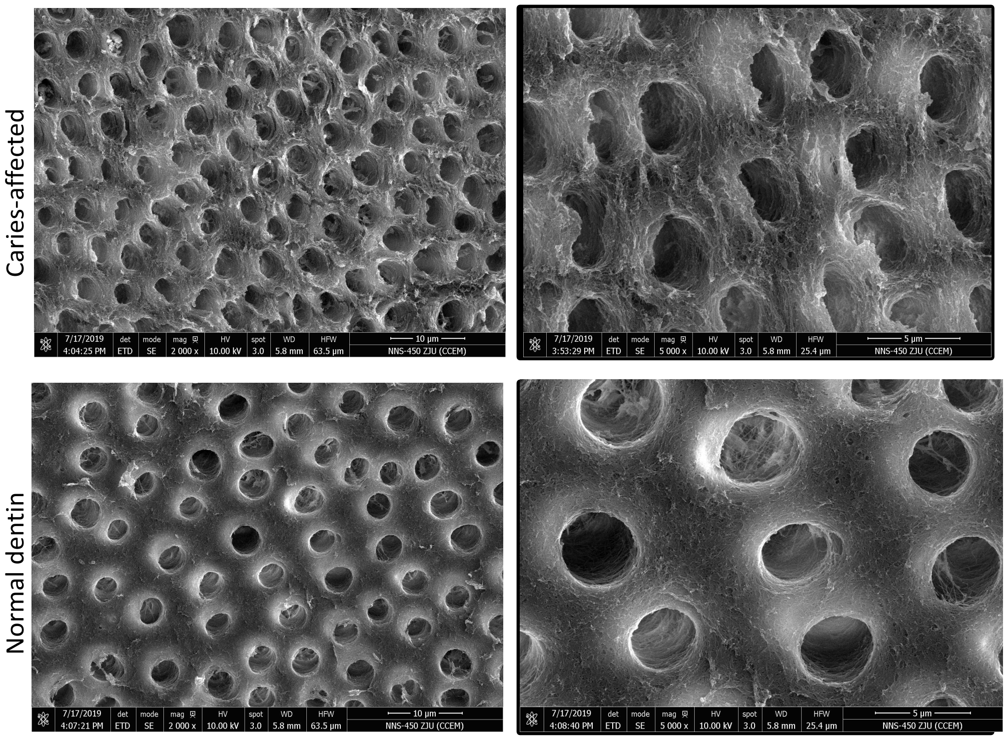

Figure 2. SEM of the caries-affected deciduous tooth dentin and normal deciduous tooth dentin surface remaining after treatment at 2000 and 5000 magnification.

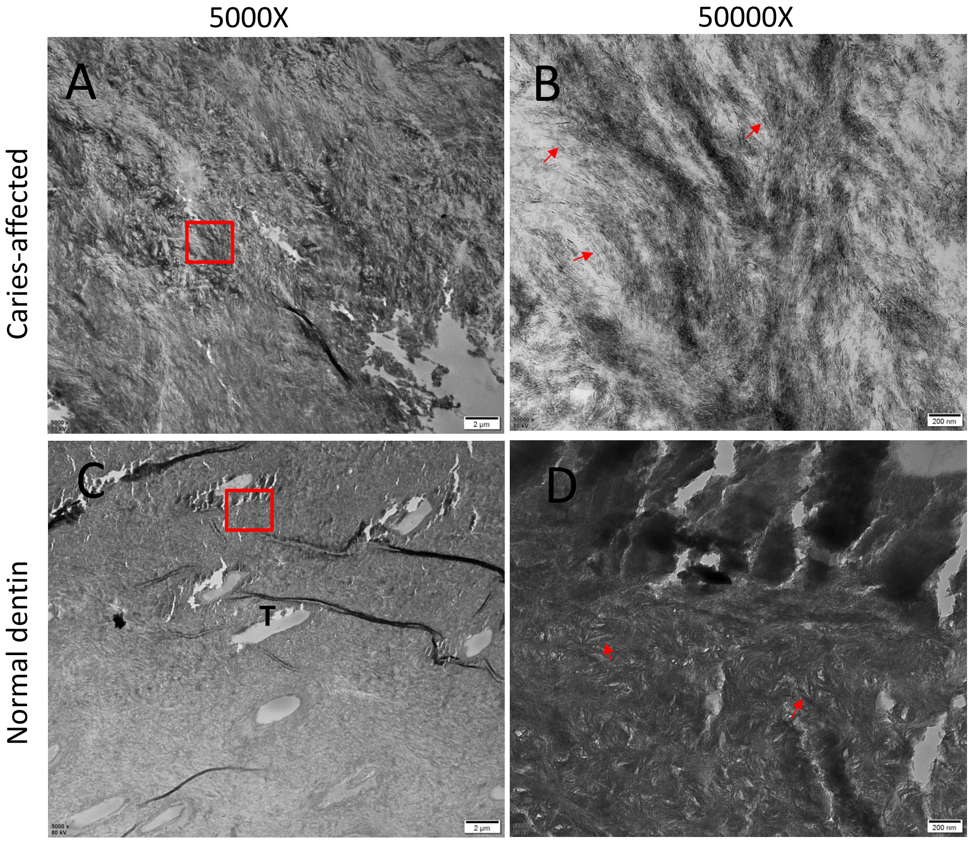

Figure 3. Unstained TEM images of cross-section of the caries-affected deciduous tooth dentin(panels A-B), normal deciduous tooth dentin(panels C-D) .Panels B and D are the higher magnifications of the marked areas (red boxes) in panels A and C respectively.

T: dentin tubuler   Arrow: hydroxyapatite

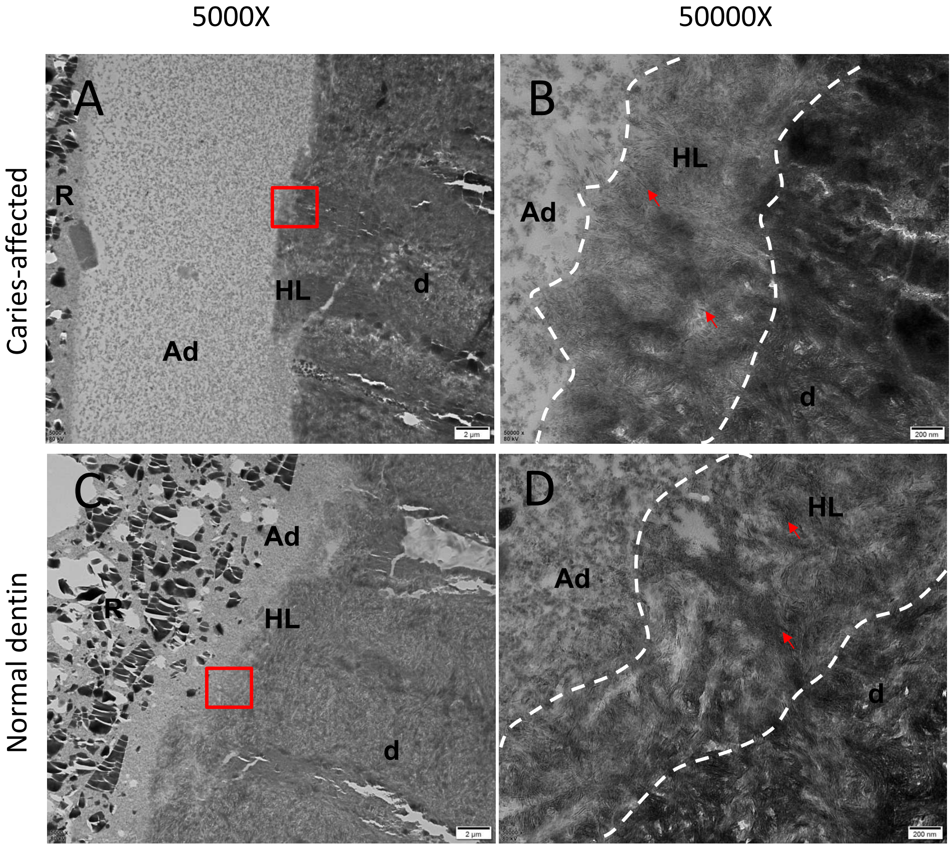

Figure 4. Unstained TEM images of cross-section of the interfaces between dentin (caries-affected deciduous tooth dentin (panels A-B), normal deciduous tooth dentin (panels C-D) and resin (5000×, 50000×). Panels B and D are the higher magnifications of the marked areas (red boxes) in panels A and C respectively.
R: resin; Ad: adhesive; HL: hybrid layer; d: dentin

Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

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Qiang Sun and Chengze Wang contributed equally to this work and should be regarded as co-first authors.

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Contributions

QS and CW executed the work and wrote the manuscript.MS and YC participated in experiment and data analysis,MW and QL revised the manuscript,JX and CS made the sample preparation for transmission electron microscopy and the preparation of figures.XL provided the conception and designed the work,supervised the study and revised the manuscript. All authors read and approved the final manuscript.

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Correspondence to Xiaodong Li.

Ethics declarationsEthics approval and consent to participate

Ethical approval for the study was obtained from the Ethics Committee of the Affiliated Stomatology Hospital, Zhejiang University, School of Medicine (2018-33). Written informed consent was obtained from each participant.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

Figures

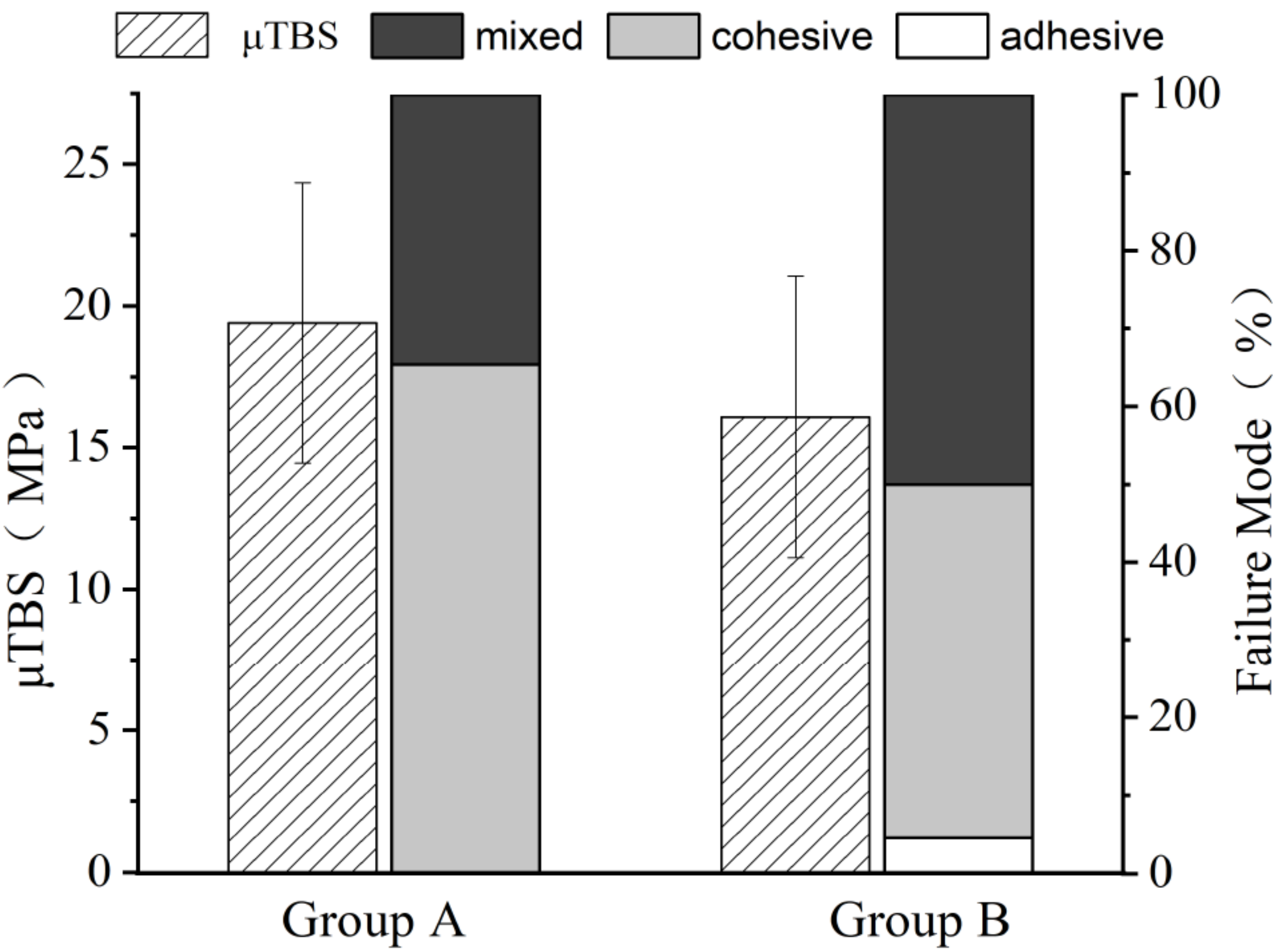


Figure 1

Micro tensile bond strength of different groups in MPa (means ± SD) and Graphic of percentage distribution of the different failure modes

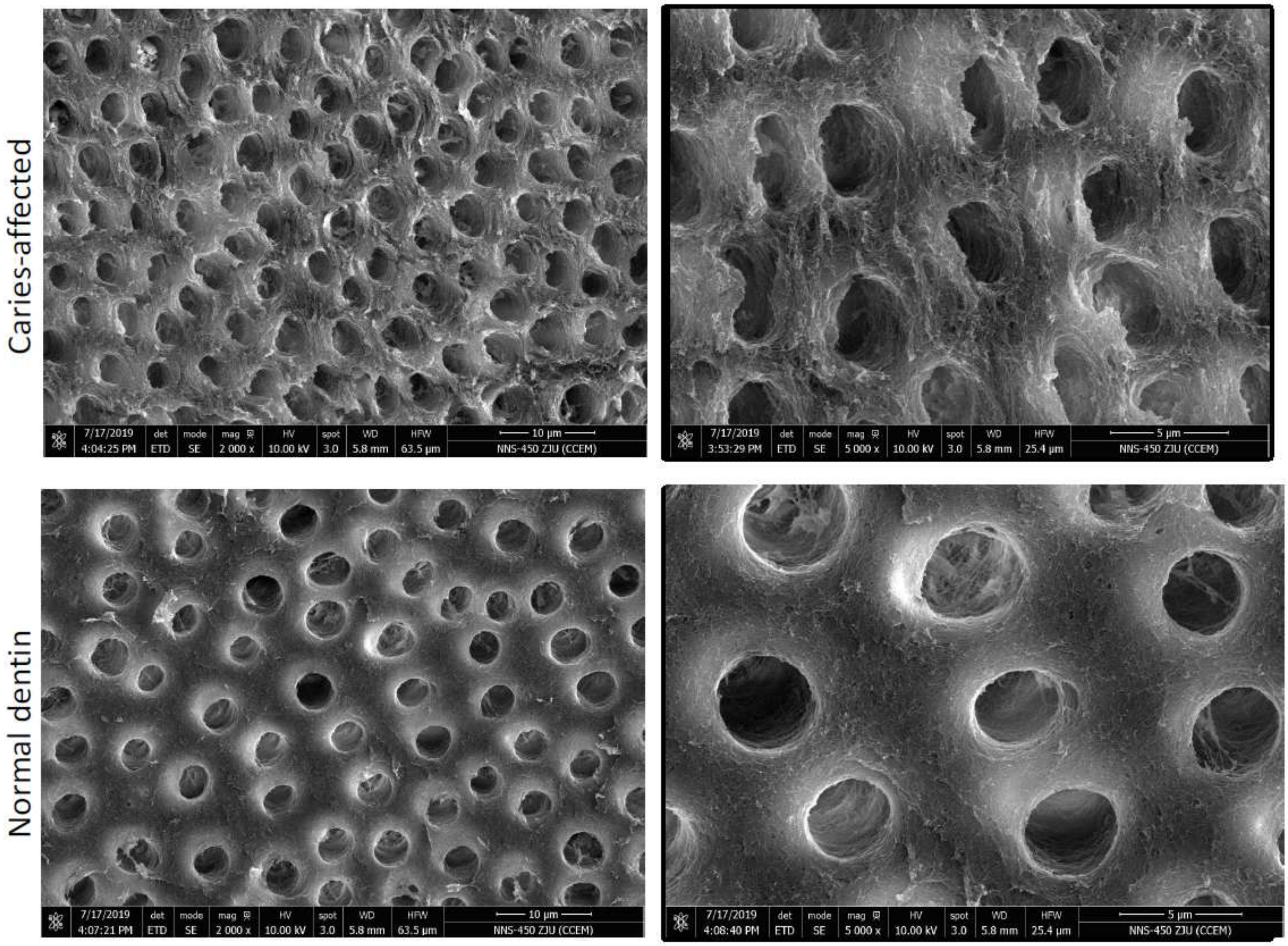


Figure 2

SEM of the caries-affected deciduous tooth dentin and normal deciduous tooth dentin surface remaining after treatment at 2000 and 5000 magnification.

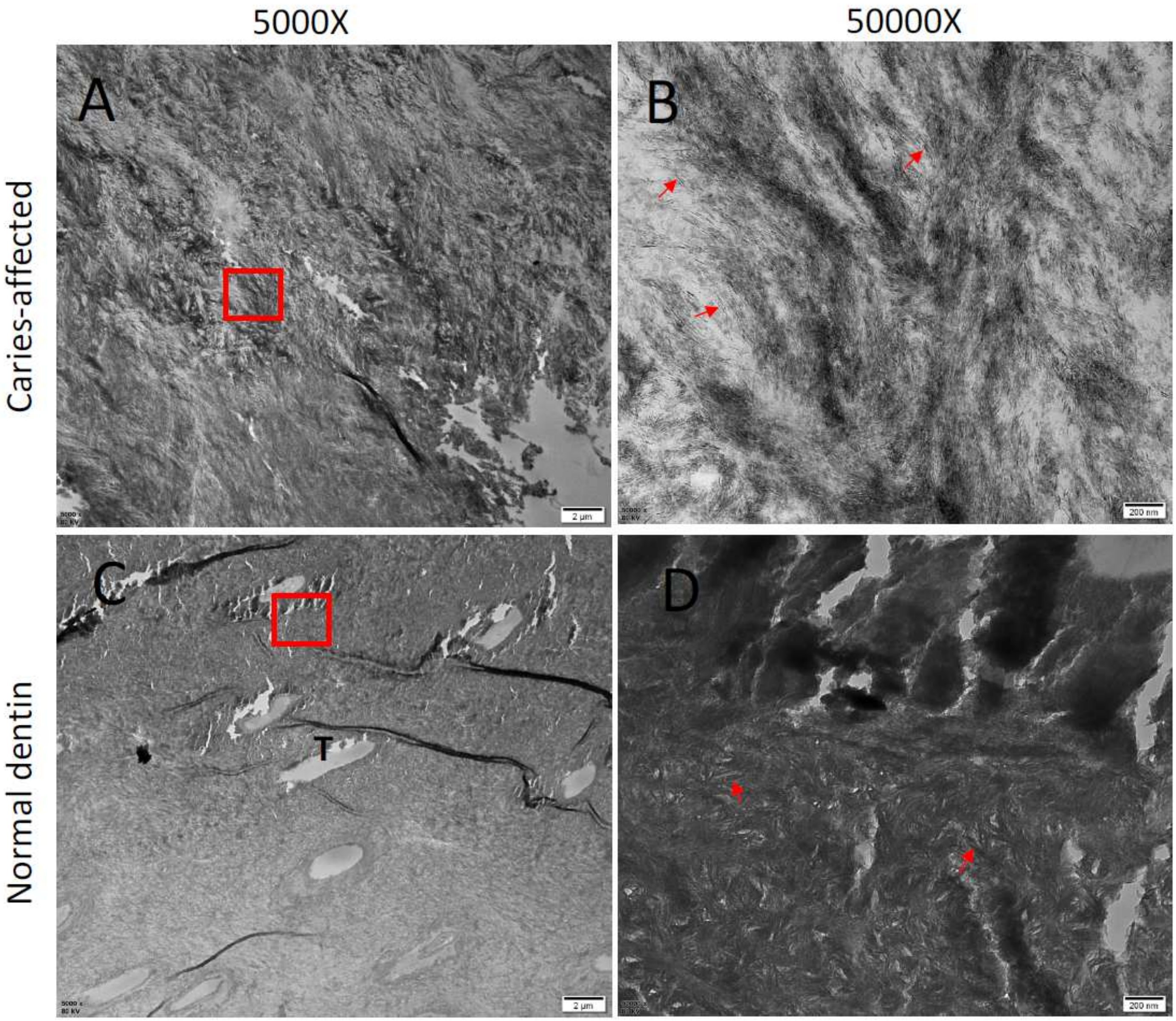
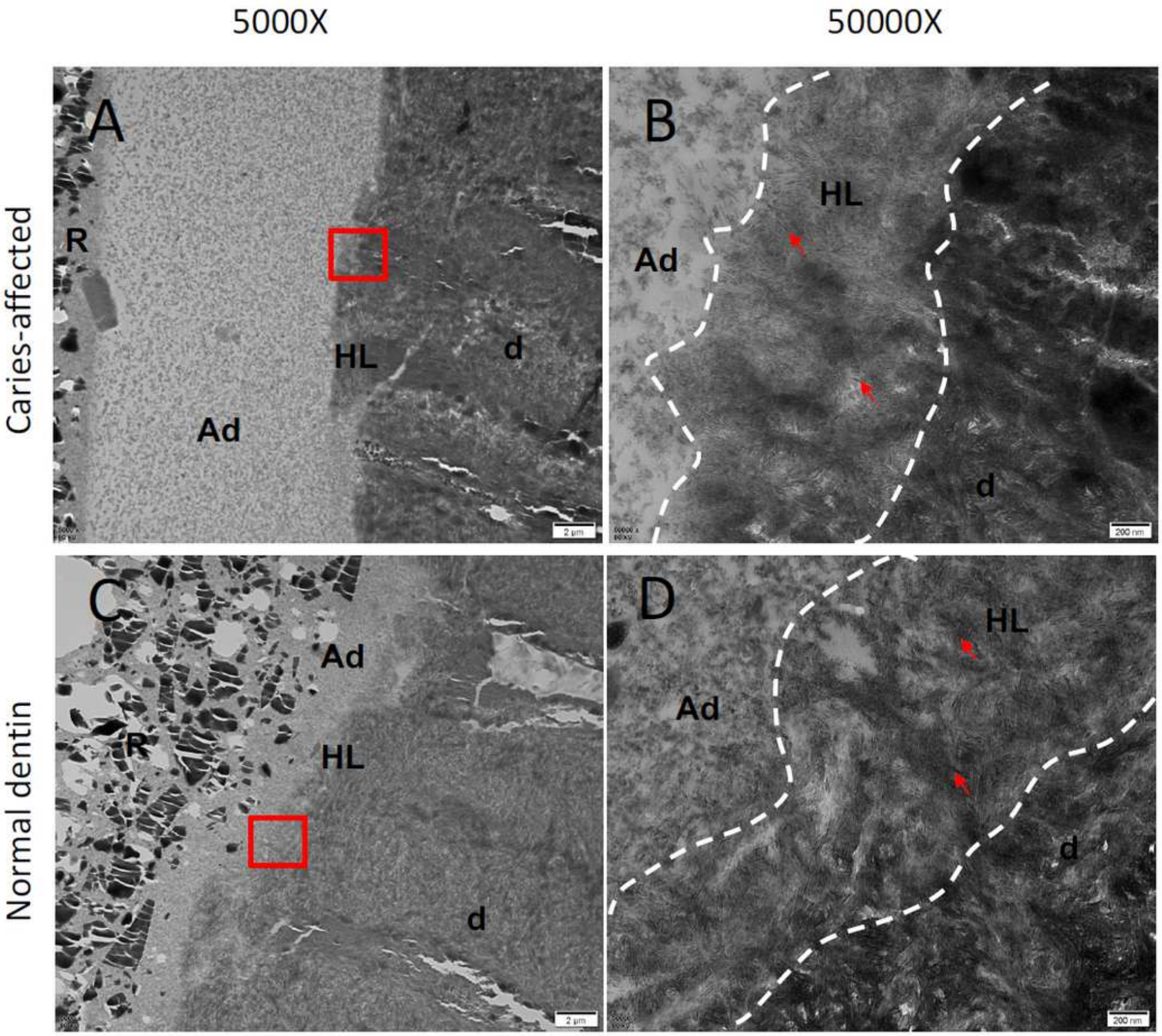


Figure 3

Unstained TEM images of cross-section of the caries-affected deciduous tooth dentin(panels A-B), normal deciduous tooth dentin(panels C-D). Panels B and D are the higher magnifications of the marked areas (red boxes) in panels A and C respectively.

T: dentin tubuler   Arrow: hydroxyapatite



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Unstained TEM images of cross-section of the interfaces between dentin (caries-affected deciduous tooth dentin (panels A-B), normal deciduous tooth dentin (panels C-D) and resin (5000×, 50000×). Panels B and D are the higher magnifications of the marked areas (red boxes) in panels A and C respectively. R: resin; Ad: adhesive; HL: hybrid layer; d: dentin