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Optical Coherence Tomography Combined with the Confocal Microscopy Method and Fluorescence for Class V Cavities Investigations

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Abstract—The purpose of this study is to present a non invasive method for the marginal adaptation evaluation in class V composite restorations. Standardized class V cavities, prepared in human extracted teeth, were filled with Premise (Kerr) composite. The specimens were thermo cycled. The interfaces were examined by Optical Coherence Tomography method (OCT) combined with the confocal microscopy and fluorescence. The optical configuration uses two single mode directional couplers with a superluminiscent diode as the source at 1300 nm. The scanning procedure is similar to that used in any confocal microscope, where the fast scanning is en-face (line rate) and the depth scanning is much slower (at the frame rate). Gaps at the interfaces as well as inside the composite resin materials were identified. OCT has numerous advantages which justify its use in vivo as well as in vitro in comparison with conventional techniques.

Keywords**—**Class V Cavities, Marginal Adaptation, Optical Coherence Tomography Fluorescence, Confocal Microscopy

I. INTRODUCTION

No matter their chemical composition, tooth colored materials used for restoration of class V cavities are subjected to marginal leakage. This phenomenon has been widely investigated by a great number of authors.

For the evaluation of marginal adaptation in class V composite restorations, several methods have been developed (bacterial penetration, fluid transport, clarification, penetration of radioisotopes, electrochemical methods and gas chromatography). Dye penetration tests (microleakage tests) however, are to be the most widely used due to their ease of application and low costs. Their main disadvantage is that the samples are sectioned. The purpose of this study is to present an alternative non invasive/non destuctive method instead of the dye penetration evaluation.

Along with the well known dye penetration technique, optical coherence tomography (OCT) was used for detection of voids and gaps at the interfaces which could have an impact on both the marginal seal and microleakage. To our knowledge this method was never employed before in the study of the marginal seal between composite and tooth structure in class V cavities.

II. MATERIALS AND METHODS

A. *Sample Preparation*

202 human extracted teeth crack-free were randomly selected for this study. The teeth were stored in physiological saline solution prior to use. [2–Haller, 1993, 3–Li, 2002] A standardized class V cavity (mesio-distal width 3 mm, occluso-gingival width of 2 mm and a depth of 1,6 mm) was prepared on the buccal surface of each tooth using a regular grit fissure diamond bur (no. 835, ISO 806 314 108 524 018, Hager&Meisinger, Neuss, Germany). According to Uno et al, the cavities were prepared with 90 degrees cavo-surface angle. [7–Uno, 1997] The preparations were parallel to the cementoenamel junction, with the occlusal half of them extending approximately 1 mm above the junction. The cavities were conditioned as follows: total acid etching, 15 seconds with 37,5% phosphoric acid Gel Etchant (Kerr), than aplication of OptiBond Solo Plus (Kerr) adhesive. All cavities were bulk filled with Premise (Kerr) composite. All materials were used according to manufacturer's instructions. The composite restorations were finished and polished [4–Lim, 1999], using a fine grit diamond bur (no. 858F, ISO 806 314 165 514 014, Hager&Meisinger, Neuss, Germany) and Kerr-Hawe polishing discs (Dentsply DeTrey, Konstanz, Germany). The specimens were then stored one week in distilled water (37°C) and thermocycled (1000 cycles) with a dwell time of 25 seconds in each bath (transit time 5 seconds) between 5-55 °C. The restored teeth were stored in distilled water (7 days). The occlusal and gingival interfaces were examined by the Optical Coherence Tomography method (OCT) combined with the confocal microscopy. [6–Podoleanu, 2004] The optical configuration (fig.1) uses two single mode directional couplers with a superluminiscent diode as the source at 1300 nm. The scanning procedure is similar to that used in any confocal microscope, where the fast scanning is *en-fa*ce (line rate) and the depth scanning is much slower (at the frame rate).

B. *Optical Coherence Tomography*

To obtain 3D information about the object, any imaging system is equipped with three scanning means, one to

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scan the object in depth and two others to scan the object transversally. Depending on the order these scanners are operated and on the scanning direction associated with the line displayed in the raster of the final image delivered, different possibilities exist. B-scan images, analogous to ultrasound B-scan are generated by collecting many reflectivity profiles in depth (A-scans) for different and adjacent transverse positions. The transverse scanner (operating along X or Y) advances at a slower pace to build a B-scan image. The majority of reports in literature refer to this way of operation. In longitudinal OCT, the axial scanner is the fastest and its movement is synchronous with displaying the pixels along the line in the raster, while the lateral scanning determines the frame rate. In this case, the transverse scanner (or scanner) determine(s) the fast lines in the image. We call each such image line as a T-scan. This can be produced by controlling either the transverse scanner along the X-coordinate, or along the Y-coordinate with the other two scanners fixed. This procedure has a net advantage in comparison with the A-scan based B-scan procedure as it allows production of OCT transverse (or 2D *en-face*) images for a fixed reference path, images called C-scans. In this way, the system can be easily switched from B to C-scan, procedure incompatible with A-scan based OCT imaging. C-scans are made from many T-scans along either of X, Y, repeated for different values of the other transverse coordinate, Y, X respectively in the transverse plane. The repetition of T-scans along the other transverse coordinate is performed at a slower rate than that of the T-scans, which determines the frame rate. In this way, a complete raster is generated. Different transversal slices are collected for different depths Z, either by advancing the optical path difference in the OCT in steps after each complete transverse (XY) scan, or continuously at a much slower speed than the frame rate. The depth scanning is the slowest in this case. It is more difficult to generate *en-face* OCT images than longitudinal OCT images as the reference mirror is fixed and no carrier is produced. Therefore, in order to generate T-scans and T-scan based OCT images, a phase modulator is needed in order to create a carrier for the image bandwidth. This complicates the design and introduces dispersion. Research has shown that the X or Y-scanning device itself introduces a path modulation which plays a similar role to the path modulation created by the longitudinal scanner employed to produce A-scans or A-scan based B-scans. The optical configuration uses two single mode directional couplers with a superluminiscent diode as the source. The scanning procedure is similar to that used in any confocal microscope, where the fast scanning is *en-fa*ce (line rate) and the depth scanning is much slower (at the frame rate)[4]. The *en-face* scans provide an instant comparison to the familiar sight provided by direct view or via a conventional microscope [5]. Features seen with the naked eye could easily be compared with features hidden in depth. Sequential and rapid switching between the *en-face* regime and the cross-section regime, specific for the *en-face* OCT systems developed by us, represents a significant advantage in the non-invasive imaging methods.

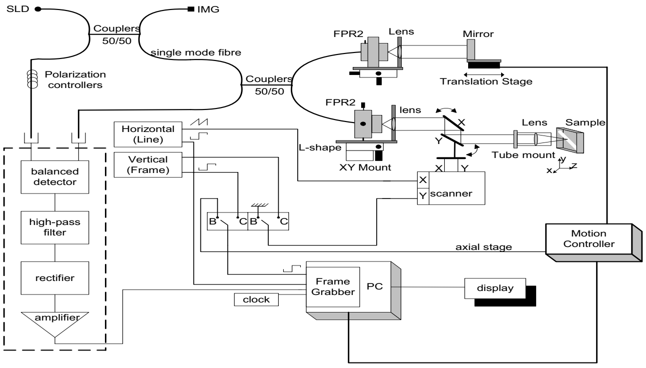

Fig. 1. Optical coherence tomography system architecture. SLD = superluminescent diode, IMG = index matching gel.

C. Flourescence and Confocal Microscopy

Two *en-face* OCT systems have been used. Both use similar pigtailed super-luminescent diodes (SLD) emitting at 1300 nm and having spectral bandwidths of 65 nm which determine an OCT longitudinal resolution of around 17.3 microm in tissue. The first OCT system performs OCT only, in both C-scan and B-scan regimes, with low NA, allowing 1 cm lateral image size. The second system, equipped with a confocal channel at 970 nm, uses a high NA interface optics allowing 1 mm image size. The configuration of the second system, as shown in Fig. 1, uses two single mode directional couplers. For each sample there was an OCT investigation, a combine OCT / Confocal investigation and OCT / Confocal / Fluorescence investigation after the fluorescence agent was added to the sample.

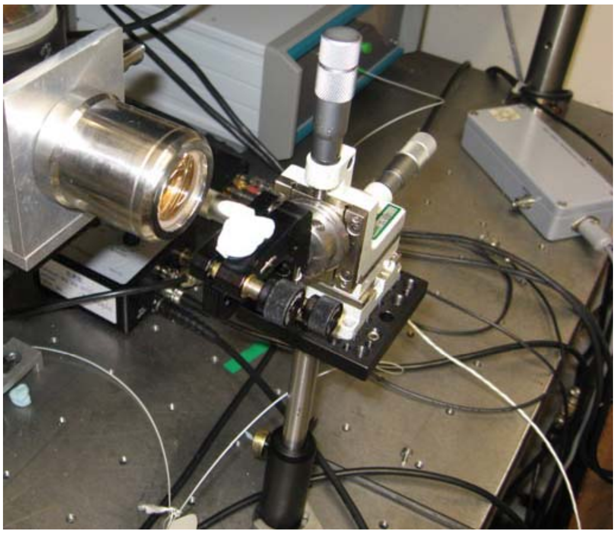

Fig.2. Aspects from the OCT/Confocal investigation system used in our study.

The OCT scanning at 18 or 8 degree (zoom) in air lead to stacks of 98 slices, with 33 microns between the slices. The

OCT/Confocal scanning lead to 3D reconstructions that allow localizing and evaluating the magnitude of the defects. After that the fluorescence agent (Fluoresceina) was applied on the teeth / resin interfaces and than another OCT scan was performed.

III. OCT RESULTS

Marginal adaptation at the interfaces and gaps inside the composite resine materials were identified by means of optical coherence tomography (fig. 2, 3). Exploration of the recent advances in OCT in terms of different excitation wavelengths and wider bandwidths can lead to state-of-the-art imaging systems in odontology.

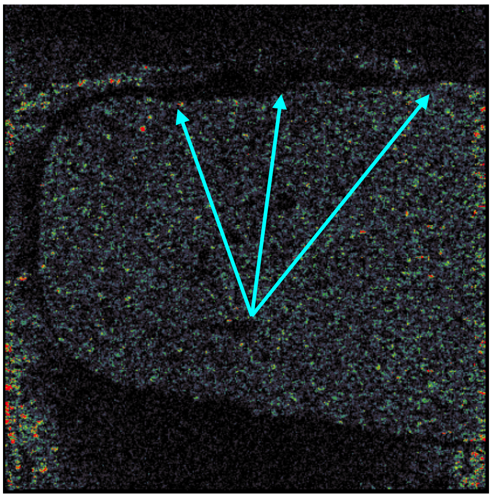

Fig.3. OCT investigation of sample 22, class V cavity filled with composite resin. In the upper part of the tooth resin interface a big gap can be spotted. Slice 43 from 98, 33 microns between slices, 8 degree scanning in air.

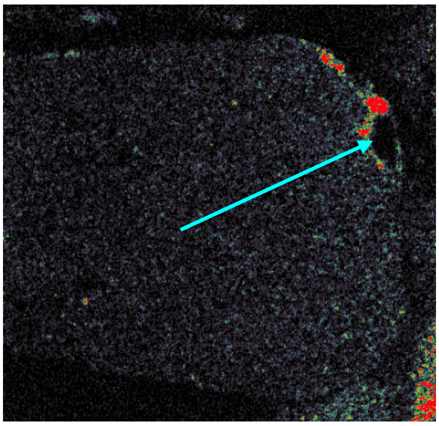

Fig. 4. OCT investigation of sample 46, class V cavity filled with composite resin. In the upper part of the tooth resin interface a big gap can be spotted. Slice 35 from 98, 33 microns between slices, 8 degree scanning in air.

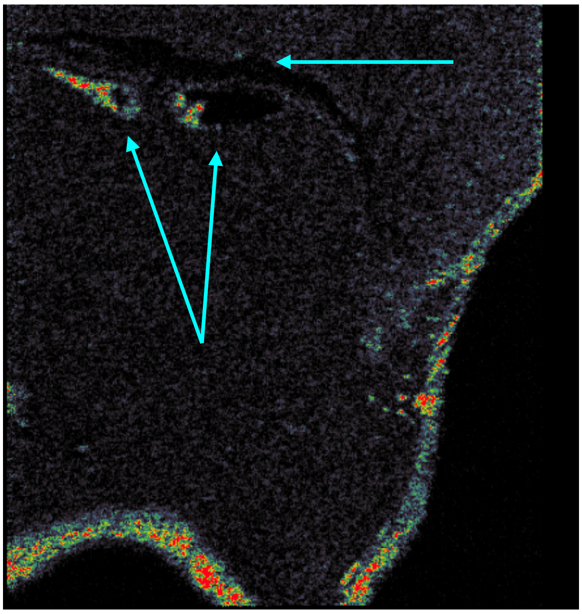

Fig.5. OCT investigation of sample 57, class V cavity filled with composite resin. In the upper part of the tooth resin interface a big gap can be spotted. Also two resin defects are located near the problematic interface. Slice 28 from 98, 33 microns between slices, 8 degree scanning in air.

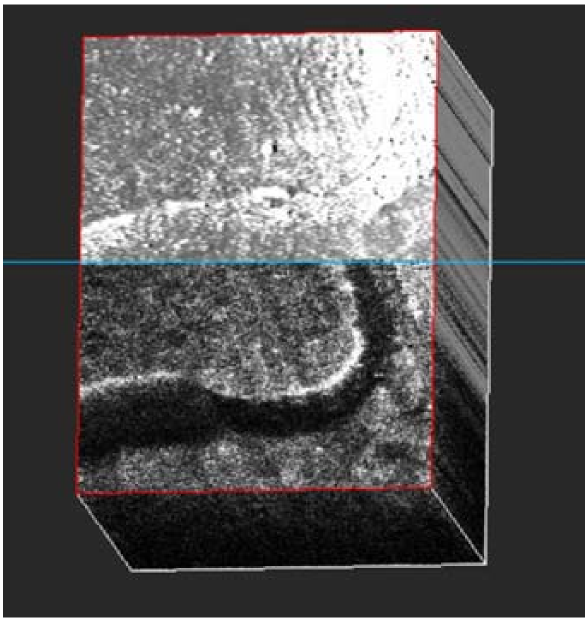

Fig. 6. Marginal gap of a class V cavity filled with the composite resin. 3D reconstruction of the OCT/ Confocal Investigation of sample 63. En-face scanning reveals the gap in depth. 1300 nm, slide 57 from 98, 18 degree in air. The upper image is the confocal projection on the investigated interface

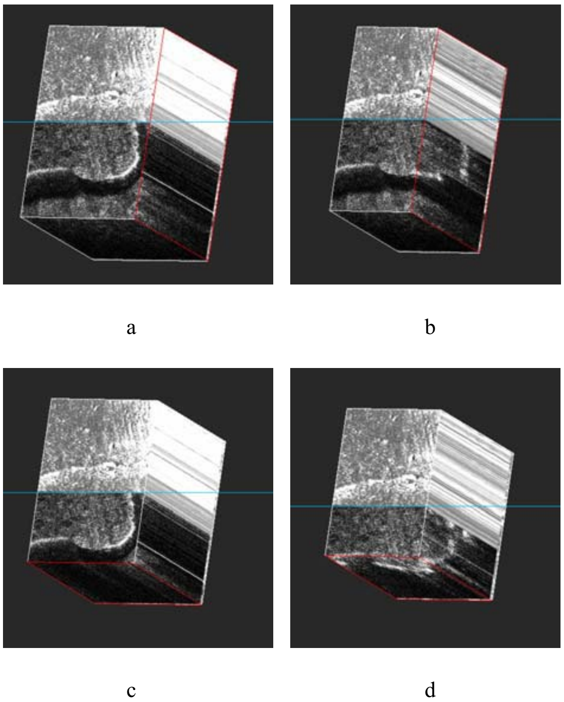

Fig.7. OCT/ Confocal Investigation of sample 63. Three dimensional reconstruction of the investigated interface allows to analize the sections in depth using the B scanned slices obtained by C scan images recorded in en face imaging. Proximal introspection (**a** and **b**) and cervical introspection (**c** and **d**) towards the composite filling. The confocal image remains the same in all slices

Fig. 8. Applying the fluorescence agent on the tooth resin interface for sample 79.

Fig. 9. OCT Fluorescence investigation in order to detect the micro leakage of the resin filling for sample 79.

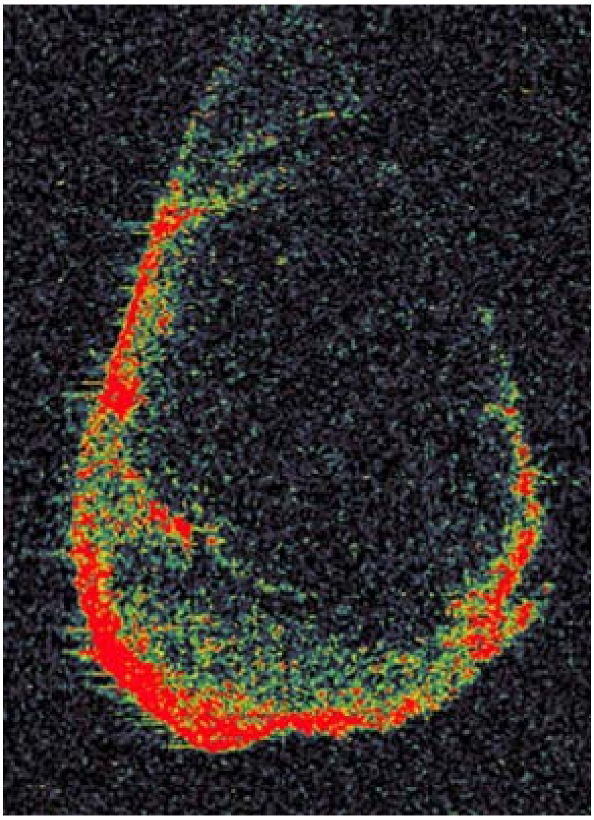

Fig. 10. OCT Fluorescence investigation with the micro leakage of the resin filling for sample 79.

IV. DISCUSSION

The test of microleakage at the tooth–restorative interface using various penetrating dyes has been widely used due to its ease of application . The results however, are subjected, partly or totally, to different influences of the methods that are applied. In addition, the comparison of results obtained by different authors is difficult and may even lead to doubful interpretation [14].

The usage of organic dyes is the most used method for the investigation of microleakage . The most commonly used dyes are methylene blue and basic fuchsine, with various concentrations and immersion times. Radioisotopes can also be used with different techniques, particle sizes and solution pH.

The interpretation of the results is based on previously established scores, probably due to their ease of examination. Another way of interpretation is to quantify the dye penetration measurements and to transform them into percentages.

The measurement of dye penetration in volume with a spectrophotometer method is another method of interpretation of micro leakage, but this technique requires a specific devices and professional training [14].

Other methods such as scanning electron microscopy (SEM) or dye staining gap tests have been used for evaluation of the marginal adaptation of the tooth colored restoratives. This quantitative method allows the assessment of the entire. The artifacts formed as a result of sectioning or dehydration can be minimized by utilizing replicas.

Another test using 1% red propylene glycol acid solution for 5 s to has been used to detect marginal gaps. The short time period of dye penetration allows only the capillary action thus preventing the diffusion into the adhesive layer.

The methods which evaluate microleakage by using sections may underestimate the extent of dye penetration. In order to overcome this major disadvantage, the clearing method was introduced, initially for the evaluation of root canal treatments. For the evaluation of leakage patterns around class V restorations, this method was used in combination with the silver staining method [15]. This method is unsuitable for detecting microleakage in enamel, but it can be used for cavities with margins below the cementoenamel junction. Other authors compared the silver staining/clearing technique with a fluid filtration method, which was found to be more sensitive than the clearing method. The clearing protocol with India ink as a tracer was found to be a supplementary aid regarding the visualization of microleakage [15].

The OCT investigations point out many tooth resin filling interfaces with problems along with material defects inside the resin filling. Both those can lead to micro leakage and failure of the treatment of class V cavities. The OCT/Confocal Microscopy scanning allows to develop 3D images that are very useful in evaluating the location and magnitude of the defect. Finally the Fluorescence OCT point out the micro leakage for the samples investigated. Finally, as demonstrated in the literature, in vivo and real-time OCT images can be obtained, and therefore this method of assessment is potentially useful for clinical diagnostics.

V. CONCLUSION

OCT has numerous advantages which justify its use in the oral cavity in comparison with conventional dental imaging. OCT can achieve the best depth resolution of all known methods (in principle 1 micron if the source exhibits a sufficiently wide spectrum) and is safe.

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