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MicroCT Analysis of a Retrieved Root Restored with a Bonded Fiber-Reinforced Composite Dowel: A Pilot Study

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Abstract

Purpose: This evaluation aimed to (1) validate micro-computed tomography (microCT) findings using scanning electron microscopy (SEM) imaging, and (2) quantify the volume of voids and the bonded surface area resulting from fiber-reinforced composite (FRC) dowel cementation technique using microCT scanning technology/3D reconstructing software.
Materials and Methods: A fiberglass dowel was cemented in a condemned maxillary lateral incisor prior to its extraction. A microCT scan was performed of the extracted tooth creating a large volume of data in DICOM format. This set of images was imported to image-processing software to inspect the internal architecture of structures.
Results: The outer surface and the spatial relationship of dentin, FRC dowel, cement layer, and voids were reconstructed. Three-dimensional spatial architecture of structures and volumetric analysis revealed that 9.89% of the resin cement was composed of voids and that the bonded area between root dentin and cement was 60.63% larger than that between cement and FRC dowel.
Conclusions: SEM imaging demonstrated the presence of voids similarly observed using microCT technology (aim 1). MicroCT technology was able to nondestructively measure the volume of voids within the cement layer and the bonded surface area at the root/cement/FRC interfaces (aim 2). *Clinical significance:* The interfaces at the root dentin/cement/dowel represent a timely and relevant topic where several efforts have been conducted in the past few years to understand their inherent features. MicroCT technology combined with 3D reconstruction allows for not only inspecting the internal arrangement rendered by fiberglass adhesively bonded to root dentin, but also estimating the volume of voids and contacted bond area between the dentin and cement layer.

Traditional methods for evaluating root/cement/dowel structures and their interfaces after cementation are mainly based on 2D imaging obtained from microscopic techniques, which require specimen sectioning and careful polishing.[1-3] The shortcomings of such methods include the prevention of further material characterization,[4] the potential resulting artifacts during specimen preparation (i.e., loss of adhesion from grinding, scratches on the surface),[5] excessive steps for specimen preparation,[5,6] limitations analyzing dissimilar planes,[4] the acquisition of only a few sections per tooth,[4] and that the sample can be irreversibly damaged during its preparation. Specific to the evaluation of the dowel/cement/root dentin interface, specimens should be replicated with an epoxy resin material to avoid artifacts during specimen preparation and from the scanning electron microscope (SEM) vacuum chamber, which adds further labor and time to the methodology.[7,8]

A potential method to nondestructively explore the dowel/cement/root interface and overcome the issues regarding SEM imaging is the use of micro-computed tomography (microCT) associated with 3D reconstructing software. Detailed quantitative and qualitative evaluation of the bone-to-implant contact, for instance, has been shown to be possible,[9] along with other evaluations of the hard or soft tissues.[10]

MicroCT seems to allow for comprehensive imaging of the spatial organization of the specimen structures, and quantitative analysis can also be attained from a 3D model rendered by a software package.[11] MicroCT imaging has been successfully employed in the dental field to evaluate tooth morphology[12] and root preparations,[13] generate finite element models (FEM),[14] assess and quantify periimplant bone morphology,[15-17] and track bone formation surrounding dental implants as well as to characterize bone density changes after dental implant placement.[18] Although nondestructive investigation through microCT imaging has been performed to measure voids and gaps in endodontic material fillings,[19] its use to characterize the dowel/cement/root canal structures after adhesive bonding in vivo has not been described to date.

The interest in this particular clinical situation comes from the challenging scenario posed by adhesively bonding a fiber-resin reinforced (FRC) dowel to the root canal. This procedure is commonly selected in reconstructive dentistry when the coronal remainder structure needs compensation for further treatment. As a result of luting agent manipulation and root insertion, voids may be introduced within the cement layer. Such inconvenience may affect the longevity of adhesive bonding in two ways: first, voids can be located directly at the interface between dentin and cement, decreasing the contact bonded area; second, as mechanical properties are highly dependent on flaw distribution and void formation, it would be expected that presence of voids decreases the strength of cement and creates sites for crack initiation and propagation. Thus, it would be of great interest to understand whether microCT imaging combined with 3D reconstruction would provide information of spatial distribution and volume area of potential voids produced during fiber dowel cementation.

The objectives of this pilot study were to: (1) validate the microCT method using SEM imaging, and (2) to quantify the volume of voids and bonded surface area resulting from fiber dowel cementation technique using microCT scanning technology/3D reconstructing software.

Materials and methods

A maxillary lateral incisor with combined endodontic and periodontal problems (due to previously failed endodontics and localized bone resorption at palatal side greater than 10 mm) was referred for extraction (Fig 1). After receiving consent from the patient and prior to extraction, dowel canal space was prepared with #1-2 intra-canal drills (RelyX™ Fiber Post Drill, 3M ESPE, St. Paul, MN) attached to a slow-speed handpiece. After preparation, the dowel space was etched with a 37% phosphoric acid (Total Etch, Ivoclar Vivadent, AG, Schaan, Liechtenstein) for 15 seconds and rinsed with distilled water for 30 seconds with an endodontic irrigating syringe. Excess water was removed with paper points (Tanari, Tanariman Industrial Ltda, Manaus, Brazil). Subsequently, primer-adhesive material (Excite DSC, Ivoclar Vivadent, AG) was applied into the root canal with a micro-brush provided by the manufacturer, excess was removed with paper points, and light curing was performed for 20 seconds (750 mW/cm^2, Ultraled, Dabi Atlante, Ribeirão Preto, Brazil). The dual-cured resin cement (Variolink II, Ivoclar Vivadent, AG) was manually mixed, injected into the root canal with a syringe (Centrix Sistema, DFL Ind. E Com. SA, Rio de Janeiro, Brazil) to ensure minimal void incorporation, and applied onto the FRC (RelyX™ Fiber Post #2, 3M ESPE) dowel surface. The FRC dowel with cement was placed into the canal, and the resin cement light activated for 40 seconds. The dowel core was reconstructed with resin composite (Filtek Z100, 3M ESPE), and a full-crown preparation was performed. All materials used for the present study (fiber dowel, luting agent adhesive system, and composite resin) were bought and used in 2009. A temporary crown was fabricated and cemented (Provy, Dentsply, Petropolis, Brazil). A periapical X-ray of the tooth was taken. Thirty days after the FRC dowel cementation, the tooth was extracted, decontaminated, and sterilized using gamma radiation.[20] The specimen was stored in water with 0.1% Thymol until microCT and SEM imaging.

Prior to microCT scanning, the extracted tooth was embedded in a methacrylate-based resin[21] (Technovit 9100, Heraeus Kulzer GmbH, Wehrheim, Germany). A microCT scan (μCT 40 Scanco Medical, Brüttisellen, Switzerland) was used to image the embedded tooth with the voltage set at 70 kV and anode current of 114 μA. The microCT resolution was set to 20 μm increment/slice thickness with a small-angle cone beam operating at room temperature. The microCT produced a large volume of data in DICOM (Digital Imaging and Communications in Medicine) format ($\sim$ 850 DICOM images). This set of images was imported to the image-processing software Amira (Amira, Version 5.3.3, TGS, San Diego, CA), and an external overview of the tooth was rendered.

To estimate the volume of voids and bonded surface areas, the 3D model of the tooth was digitally sectioned by the apical aspect of the FRC dowel and above the cementoenamel junction, acquiring a total of 361 DICOM images. The Amira software processed the image segmentation. This process uses pixel contrast rates to assign what pixel belongs to each object of interest. The resolution limit employed was from 0 to 255 (gray scale contrast). After all structures were segmented, a 3D model was rendered to visualize the internal architecture arrangement. Five DICOM project files were obtained by the Amira software using the threshold tool, corresponding to: (1) FRC dowel; (2) cement layer associated with FRC dowel; (3) top portion of the FRC dowel; (4) top portion of the cement layer; and (5) voids. The DICOM files were imported into ScanIP (ScanIP, Version 4.2 Build 135, Simpleware Ltd, Exeter, UK) to estimate the bonded area between root dentin and cement as well as the volume of voids.

For each file, a mask was created in which was applied the Cavity fill filter to fill the "virtual voids" within the mask. To fill the small gaps and holes around the edges, a morphological close filter was also applied. For the cement layer + fiber dowel files, two masks were generated (one for cement layer and one for fiber dowel) and joined before applying the abovementioned filters. After that, the surface area and void volume were acquired through 3D statistics. The surface areas from both tops of the cement layer and fiber dowel were summed, and the results were subtracted from the total cement layer (cement layer and FRC dowel combined), to estimate only the bonded area between root dentin and cement.

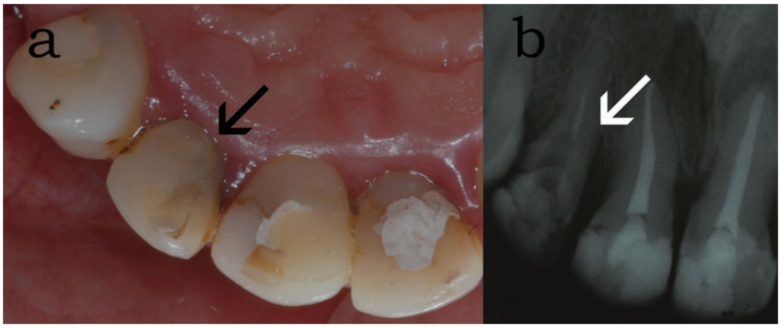

Figure 1 Images showing the initial clinical (a) and radiographic (b) aspect of the tooth referred for extraction (white and black arrows) before the FRC dowel cementation.

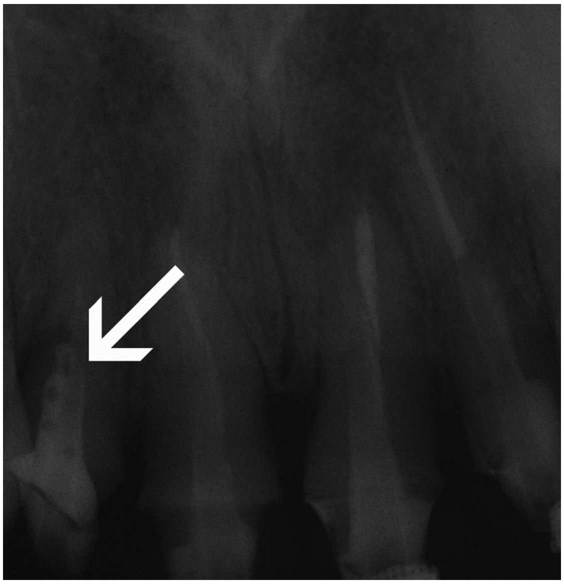

Figure 2 Radiographic aspect of cemented FRC dowel. Large voids incorporated during cementation are noticeable within the radiopaque resin cement (white arrow).

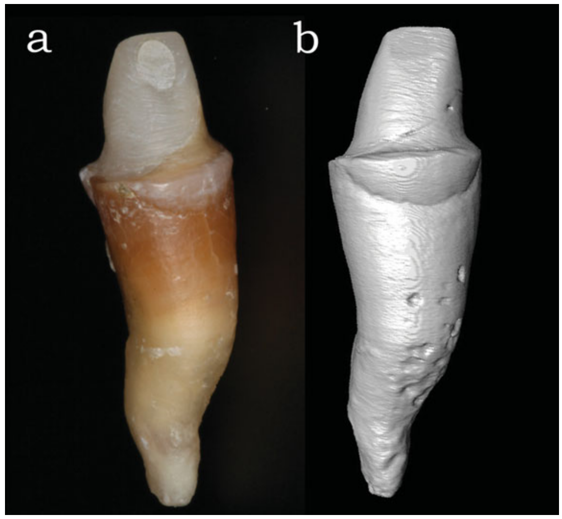

Figure 3 (a) Macro picture of buccal side of the extracted tooth and (b) 3D model reconstructed after microCT scanning, where surface topography is visualized.

After software calculations, the specimen was longitudinally sectioned in the center of the tooth/cement/FRC dowel with a precision diamond saw (Isomet 2000, Buehler Ltd., Lake Bluff, IL). The cut surfaces were polished under water irrigation by means of a series of silicon carbide (SiC) papers (400, 600, 800, 1200, and 2400 grit) (Buehler Ltd.).[22] Upon completion of the polishing step, the specimen was imaged in both polarized light (PLM) (MZ-APO stereomicroscope, Carl Zeiss MicroImaging, Thornwood, NY) and environmental SEM (Carl Zeiss AG – EVO® 50 Series, Carl Zeiss). For PLM, the specimens were ultrasonically cleaned with alcohol for 10 minutes, dried, and then attached to a specimen holder. The specimen surface was oriented 90° to the objective. An external two-arm spotlight illumination was adjusted to achieve the desired light incidence (best contrast found) over the specimen surface. The magnification was set to 8X, and the focus was manually adjusted. Before image acquisition, the software Leica QWin Standard (Leica QWin Standard V 2.5, Leica Microsystems, Buffalo Grove, IL) was employed to find the best sharpness, contrast, and brightness. For SEM, specimens were once again

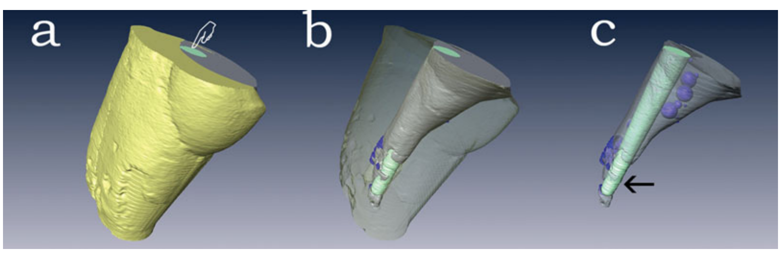

Figure 4 3D model images. (a) Shows the root dentin substrate (yellow), the cement layer (gray), and the white pointer depicts the FRC dowel (green). (b) By applying a semi-transparent layer on the root dentin surface, the spatial relationship between dentin, cement layer, FRC dowel, and voids could be illustrated. (c) Note the widespread voids (blue) within the resin cement and the absence of luting agent at the most part of apical region of the FRC dowel (black arrow).

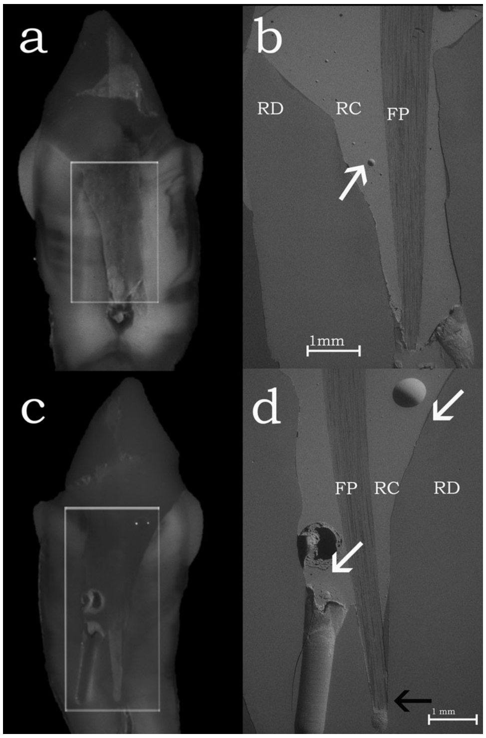

Figure 5 Images of tooth surface after being longitudinally sectioned and polished. (a) PLM view shows one surface, (b) ESEM view of the white rectangle from (a) depicts small voids indicated by arrow in the resin cement, (c) PLM image of the opposite longitudinal section and, (d) ESEM micrograph of the white rectangle shown in (c) depicts voids (white arrows) in the resin cement. The black arrow shows a gap at the resin/cement interface in the apical region. (RC) root dentin, (FP) FRC dowel, (RC) resin cement.

ultrasonically cleaned in alcohol for 10 minutes, dried, and placed in the SEM stage holder. Using environmental SEM gold coating was not necessary. The working distance was set to 25 mm, vacuum pressure was set to 50 Pa, and the signal employed was SE1.

Results

The periapical radiographic image showed the presence of several voids (unfilled space within the cement regardless of their location) within the resin cement layer (Fig 2). A total of ~850 DICOM files comprised the 3D-reconstructed tooth structure, each slice having a 20 μm increment. The 3D model produced was able to render the outer surface morphology (Fig 3) and display the spatial relationship among dentin, resin cement, FRC dowel, and voids within the cement layer (Fig 4). Microscopic images of tooth/cement/FRC dowel and voids present in the cement layer showed correspondence with the microCT images (Fig 5). The lack of bond integrity at the resin cement/root dentin interface at the apical region was observable in a 2D spatial configuration, whereas more detailed 3D imaging was obtained with the microCT. Apparently, most voids within the cement layer were formed in the most cervical aspect of the root, where the cement layer was thicker. Table 1 presents the quantitative volumetric and surface areas analyses.

Table 1 Volumetric and surface area estimates

Material	Outermost surface area (mm^2)	Volume (mm^3)
Cement layer	58.84	18.08
FRC dowel	20.23	4.81
Voids	–	1.78

Discussion

The main objective of this study was to present the potential advantages to using microCT imaging combined with quantitative and descriptive analyses using 3D reconstructing software for evaluating the root dentin/resin cement/dowel scenario of a retrieved tooth after short-term clinical function. To compare the microCT-scanned images with conventional 2D imaging, polarized-light and SEM images of the same longitudinally sectioned root were used.[1-3] In addition to being a nondestructive method, microCT imaging presents several advantages including reproducibility, agility, and impartiality when acquiring and reading specimens.[23] MicroCT morphometric results have been shown to be comparable with those obtained from the conventional 2D histomorphometry method.[4] Additionally, the software package used to reconstruct the file from microCT offers the potential to easily and quickly inspect any internal part of the sample via digital manipulation.[6]

Although the microCT method presents several advantages, one potential limit is related to discovering and quantifying smaller voids (e.g., 1 to 100 μm), which may not be discoverable from the present approach. Such potential limitation may be related to the segmentation process or associated with the resolution of the microCT equipment; however, the large amount of macro porosity may be the more important problem to deal with first.

Whereas the conventional cross-sectional method has a limited capacity to acquire slices (i.e., 2 longitudinal slices were obtained here), the microCT produced ~850 slices.[4] This large amount of thin slices allowed detailed visualization using the rendered image of the internal architecture of the scanned tooth and the quantitative analysis of void volume within the resin cement as well as the contacted bond area.

In this investigation voids represented 9.89% of all cement layer volume. Previous studies also have pointed out the presence of voids in the dowel cementation layer.[1,24] However, they did not provide quantitative analysis. Nevertheless, a possible explanation for such a percentage of voids might be attributed to the cement mixing method (manually in this pilot study), which may have introduced air bubbles into the material.[25] Moreover, the large amount of voids might also be related to the large dowel space at the cervical region, since from a quantitative point of view the majority of them were present at this area. Results from a previous study using 2D imaging to evaluate dowel cementation accuracy showed that when the cement was applied directly into the root by means of an application aid, the presence of voids was statistically lower in comparison to conventional techniques.[26] However, inferring 3D structures such as voids from 2D imaging[19] may neglect the possible overlap of voids in the nonobservable areas of the cement layer. Finally, understanding the effects of voids located within the cement layer concerning strength and modulus of elasticity of cement is the ultimate issue that warrants further investigation.

The contact bonded area between root dentin and cement (58.84 mm^2) was 60.63% larger than that between FRC dowel and cement (20.23 mm^2). From this result, the commonly used approach of calculating the push-out bond strength of dowel systems based on the radius of the FRC dowel[27] may be inaccurate considering that the primary failure mode is not adhesive between the FRC dowel and cement. Conversely, adhesive failure between root dentin and resin cement has been extensively reported,[28] suggesting that the calculated area should be based on dentin/cement bonded area, instead of the FRC dowel radius. In addition, the lack of bond integrity also should be taken into account before addressing stress calculation. Thus, both failure mode (either between dentin and cement or cement and dowel) and the presence of voids in the adhesive interfaces should be considered when calculating the total surface area employed to accurately calculate the bond strength.

Several challenges are imposed when bonding fiber-reinforced dowels to the root canal, especially at the apical area where moisture control and adhesive system application are still critical,[2] and polymerization light access is limited and variable between translucent dowels.[29] Other aspects, including high resin cement contraction stresses (C-factor), large areas covered by smear layer, debris, and sealer/gutta-percha remnants even after root dentin cleaning with phosphoric acid,[30] have to be overcome to ensure suitable bonding at the referred area. Due to these issues, it is general consensus that clinical retention of FRC dowels highly relies on sliding friction.[28,31] In this report, the most apical part of the FRC dowel remained uncovered by the cement. Although it was not possible to quantify the uncovered surface area, it was apparently the largest area unbonded to the root dentin. This finding may be related to the narrow space available for cement filling combined with the limiting bonding factors cited above. This find may imply that the overall adhesive interface bond strength is negatively influenced, suggesting that it is an important role in the adhesive interface integrity maintenance.

The tapered configuration of the cervical cuff resulted in the thickest layer of cement, where most of the voids were concentrated. Despite the better polymerization light access and moisture control, it can be speculated that the presence of voids might be a contributing factor of root/cement/FRC interface instability, since it may provide a free surface area for resin cement contraction stress release during polymerization. Whether this would result in improved bond strength merits further investigation.

Relative to the microCT imaging method demonstrated here, it should be replicated several times to prove which cementation technique or luting agent produces fewer voids within the cement layer and try to find whether the void distributions are randomly dispersed along the cement layer. Another relevant possibility is the repeated scanning of the same intact specimen to monitor, for instance, the material degradation after mechanical fatigue and aging of cemented dowels in a laboratory scenario. Moreover, the method described here has the potential to provide 3D-rendered images from retrieved dental implants of varied designs, several restoration systems, root canals cemented with a multitude of cement, and dowel systems for the construction of FEM, which reproduce clinically relevant scenarios.

Conclusions

SEM imaging demonstrated the presence of voids similarly observed using microCT technology (aim 1). MicroCT technology was able to nondestructively quantify the volume of voids within the cement layer and the bonded surface area at the root/cement/FRC interfaces (aim 2). The qualitative and quantitative analyses obtained by the combination of microCT scanning and visualization software packages have the potential to represent a baseline to assess the outcome of bonded resin-reinforced fiber dowels in root canals.

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