Dentinal Microcrack Development after Canal Preparation: A Longitudinal *in Situ* Micro–computed Tomography Study Using a Cadaver Model

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Abstract

Introduction: The purpose of this study was to evaluate the development of dentinal microcracks after root canal preparation with Reciproc and ProTaper Universal systems using an *in situ* cadaver model by means of a micro–computed tomography (micro-CT) imaging system. **Methods:** At autopsy, 8 maxillary bone blocks having at least the first and second premolar teeth (n = 16) were excised, scanned at a resolution of 13.18 μm, and randomly distributed into 2 groups (n = 8) according to the preparation protocol: Reciproc and ProTaper Universal systems. Root canals were prepared up to R25 and F2 instruments in the Reciproc and ProTaper Universal groups, respectively. After the preparation procedures, the specimens were scanned again, and the registered preoperative and postoperative cross-section images of the roots (n = 19,060) were screened to identify the presence of dentinal defects. **Results:** In the Reciproc group, 9176 cross-section images were analyzed, and no crack was observed. In the ProTaper Universal group, 244 of 9884 cross-section slices (2.46%) had dentinal defects; however, all defects were already present in the corresponding preoperative images, indicating that no new microcrack was created after canal preparation. **Conclusions:** *In situ* root canal preparation of maxillary premolars with Reciproc and ProTaper Universal systems did not induce the formation of dentinal microcracks in a cadaver model as observed by micro–CT. *(J Endod 2017;■:1–6)*

Key Words

Cadaver model, dentinal defects, instrumentation, microcracks, micro-CT, root canal preparation

Significance

This study highlighted that the response of root dentin to root canal instrumentation with Reciproc and ProTaper Universal systems did not cause microcracks in an *in situ* cadaver model.

In recent years, the occurrence of root fracture in either sound or endodontically treated/restored teeth has become a major concern in endodontics (1–3). Root fracture has been defined as a devastating clinical event (4), and it is currently one of the leading causes of tooth loss (5). Throughout the years, several hypothetical etiologies for root fracture were suggested including hypotheses that the root fracture would start from dentinal microcracks caused by dentin dehydration, post placement and corrosion, spreader design, or excessive forces during filling procedures (6–8). Some years later, Bier et al (9) and Shemesh et al (10) also correlated dentinal microcrack formation to root canal preparation performed by motor-driven nickel-titanium (NiTi) instruments. Because the mechanical preparation of the root canal has become the mainstream for root canal shaping, it is not a surprise that this phenomenon has increasingly gained importance in the endodontic research field (9–16).

Overall, the methodology used in most of the *ex vivo* studies on dentinal microcrack formation includes the sectioning of the sample, followed by postoperative observation of the exposed dentinal surface by using optical microscopic devices (9–15). However, this experimental model has some critical limitations that reduce its overall reliability, such as the destructive nature of the method, the two-dimensional observation, the absence of a full-tooth range inspection, and the lack of longitudinal follow-up, because it does not allow for the screening of the non-prepared sample. In this way, it is unlikely that the results reported in most of these studies, in which cracks

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were observed in more than 40% of the samples (16), would reflect the clinical reality. Scientific logic behind this inconclusive scenario would dictate that limitations of the conventional methods are indeed prone to systematic analysis errors and, consequently, far from an ideal experimental model.

Recent technological advances in the field of imaginology, such as the introduction of micro–computed tomography (micro-CT) in dental research, have led to a more comprehensive understanding regarding dentinal microcrack formation. Micro-CT is a highly accurate and nondestructive technology that allows the longitudinal assessment of the specimens throughout the experimental procedures; consequently, each tooth serves as its own control, hundreds of slices can be evaluated per specimen, and all extension of the defects can be tracked (17–20). By using this method, De Deus et al (17) showed a clear lack of causal relationship between dentinal microcrack development and canal preparation with rotary and reciprocating systems. This conclusion was later confirmed by other studies using the same methodology (18, 19). However, authors reported a significant number of preexistent defects on the roots probably caused by excessive extraction forces and/or storage condition of the teeth. Consequently, these conditions also do not stand for a close-to-ideal experimental model. Thus, even with a considerable body of evidence accumulated during the last 30 years, several aspects regarding crack formation and endodontic procedures remain inconclusive, and critical questions are still open. Recently, a cadaveric model was suggested as an ideal methodological approach for a comprehensive evaluation of dentinal microcrack formation (14) because the viscoelastic properties of the attachment apparatus would absorb the forces applied to the dental tissues during root canal preparation procedures.

To the best of the authors' knowledge, the current scientific literature lacks a nondestructive *in situ* longitudinal experimental report on this issue. Therefore, this study was designed to investigate the potential cause-effect relationship between root canal preparation performed by 2 motor-driven NiTi systems (Reciproc; VDW, Munich, Germany and ProTaper Universal; Dentsply Maillefer, Ballaigues, Switzerland) and dentinal microcrack formation in a cadaver model by using micro-CT technology.

Materials and Methods

Sample Size Calculation

The ideal sample size for this cadaver model on microcrack formation was calculated on the basis of the study of Arias et al (14). The estimated 3.125 effect size was input together with an alpha-type error of 0.05 and a power-beta of 0.95 into a *t* test for independent means statistical family (G*Power 3.1 for MacIntosh). The results pointed to a minimum total sample size of 8 teeth to observe differences in microcracks between the groups.

Sample Selection

Eight dentoalveolar maxillary bone blocks containing 3–5 adjacent teeth were collected from autopsy of different adult donors after family member's informed consent obtained under a research protocol approved by the local Forensic Department and the National Committee on Health Research Ethics (protocol #931.732). The age of the donors ranged from 19 to 30 years (mean age, 23 years). Inclusion criteria were the presence of non-carious maxillary first and second premolars surrounded by alveolar bone and periodontal ligament. Bone blocks were kept stored in −20°C and submitted to the experimental procedures within 1 month from their collection.

Before the scanning procedures, frozen bone blocks were removed from the freezer and put into a refrigerator at a constant temperature of 8°C for a slow defrost. After 3–4 hours, each bone block was scanned in a micro-CT device (SkyScan 1173; Bruker-microCT, Kontich, Belgium) by using an isotropic resolution of 13.18 μm at 90 kV and 88 mA through 360° rotation around the vertical axis, with a rotation step of 0.5°, camera exposure time of 1000 milliseconds, and frame averaging of 5. The x-rays were filtered with a 1-mm-thick aluminum filter. The acquired images were reconstructed into cross-sectional slices with NRecon v.1.6.10 software (Bruker-microCT) by using standardized parameters for beam hardening (15%), ring artifact correction of 5, and contrast limits (0.0095–0.03), resulting in the acquisition of 1100–1300 transverse cross sections per bone block.

Root Canal Preparation

After the scanning and reconstruction procedures, the first and second maxillary premolars from each bone block were selected for the experimental procedures (n = 16). The first premolars had 2 canals, whereas the second premolars had only 1 root canal. After conventional access cavity preparation, the working length (WL) was established 1 mm from the apical foramen by using a size 10 K-file (Dentsply Maillefer) with the aid of an apex locator (Root ZX; J Morita USA Inc, Irvine, CA) and confirmed by digital radiograph. After that, glide path was established by scouting a stainless steel size 15 K-file (Dentsply Maillefer) up to the WL. Then, teeth were randomly assigned to 2 experimental groups (n = 8). In the Reciproc group, R25 instrument (25/0.08) was activated in reciprocating motion (VDW Silver; VDW) and moved in the apical direction with light apical pressure by using a slow in-and-out pecking motion of about 3 mm in amplitude. After 3 pecking motions, the instrument was removed from the canal and cleaned. The WL was reached in the third wave of instrumentation for all teeth. In the ProTaper Universal group, SX instrument was used to one half of the WL, followed by S1, S2, F1, and F2 instruments to the full WL, with a gentle in-and-out motion (VDW Silver), according to the manufacturer's instructions (SX, S1, and S2, 300 rpm and 3 Ncm; F1 and F2, 300 rpm and 2 Ncm).

Each set of instruments was used to enlarge 2 teeth, and an experienced operator performed all experimental procedures after substantial training with the systems. During preparation, a total of 30 mL 2.5% sodium hypochlorite was delivered in each root canal by using a 31-gauge NaviTip double sideport needle (Ultradent Products Inc, South Jordan, UT). A final irrigation with 5 mL 17% EDTA and 5 mL bidistilled water, followed by drying with absorbent paper points (Dentsply Maillefer), was performed. Then, the bone blocks were submitted to a new scan and reconstruction applying the initial parameter settings.

Image Analysis

The reconstructed image stacks of the bone blocks before and after canal preparation were co-registered by using the affine algorithm of the 3D Slicer v.4.6.2 software (available from http://www.slicer.org) (21). CTVol v.2.3 (Bruker-microCT) was used for the three-dimensional visualization and qualitative analysis of the bone blocks (Fig. 1). Then, all cross-section images of the premolar teeth (n = 19,060) were screened from the cementoenamel junction to the apex by 3 previously calibrated examiners who were blinded to the experimental groups, aiming to identify the presence of dentinal defects. To validate the screening process, image analyses were repeated twice at 2-week intervals; in case of divergence, images were examined together until an agreement was reached (17–19).