Laser Use in Endodontic for Increase the Adhesion of Root Canal Filling

A synchrotron radiation micro tomography study

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The aim of the present paper is to explore, through a high-resolution synchrotron radiation-based micro computed tomography experiment, the efficiency of the Er:YAG laser involvement in the debridement of the root canal of pathologic teeth, applied after the chemo-mechanical treatment of the canal. The adhesion quality of root canal filling is the consequence of the root canal cleaning and shaping, thus preventing the leakage. Twelve extracted single-rooted teeth were considered. The first 5 mm of the roots, corresponding to the apical region of the teeth, were initially investigated through high-resolution synchrotron radiography phase contrast micro computed tomography. Afterwards, a chemo-mechanical endodontic treatment was performed and the same region of the teeth was imaged again. Finally, an Er:YAG laser with PIPS tip treatment was applied on the same teeth and a last tomographic scan was done. The chemo-mechanical treatment properly removed the necrotic tissue only in the upper part of the apical area of root canal, but tissue debris were found in the lower and partially also in the medium and upper part of the apical region, especially when the apical area exhibited a larger curvature. The laser treatment applied afterwards gave good results in terms of removing the debris from the apical area of root canal, mainly when they remained in the main canal. The present study proves the capability of Er:YAG with PIPS tips laser treatment to enhance the removing of tissue debris from the apical region of root canals after a chemo-mechanical treatment. The high-resolution synchrotron radiation-based phase contrast micro computed tomography is the most powerful tool for a non-destructive 3D investigation method of the root canal morphology analysis and a proper characterization technique for the assessment of the endodontic treatment.

Keywords: Phase contrast micro-CT, Er:YAG Laser root canal debridement, Dental morphology, Photon Induced Photoacoustic Streaming

The aim of endodontic treatment is to preserve functional teeth without to prejudice the patient health. Endodontics includes diagnosis and treatment of oral pain of pulpal or periradicular origin, vital pulp therapy, and nonsurgical treatment of root canal systems, with or without periradicular pathosis of pulpal origin and the obturation of these root canal systems [1]. Currently, a successful root canal treatment is based on a few basic principles: diagnosis and treatment planning, knowledge of anatomy and morphology, traditional concepts of debridement, well-compacted and tightly adapted root filling. The quality of root canal filling is the consequence of root canal cleaning and shaping, thus preventing the leakage. The process of cleaning and shaping determines both the degree of disinfection and the ability to obturate the radicular space [2]. Endodontic treatment is necessary when the pulp becomes inflamed or infected. Preparing the root canal system requires that all inorganic debris and organic substrates and microorganisms are eliminated, together with shaping the root, which facilitates the placement of a long-term tri-dimensional filling [3]. After biomechanical preparation of the radicular spaces, the next important step in the endodontic treatment is represented by the root canal filling. Three-dimensional obturation of the radicular space is essential to long-term success. The canal system should be sealed apically, laterally, and coronal. Subsequently, the tooth is restored with a crown or filling for protection. After restoration, the tooth continues to function like any other tooth, often for a lifetime. The endodontic space is composed by the main canal, an easily accessible area to hand root instruments and not easily accessible or inaccessible spaces represented by lateral and accessories canals, dentinal tubules, isthmus, apical delta [4]. However, because of the complexity of root canal system, it has been shown that the complete elimination of debris and achievement of a sterile root canal system is still an ongoing challenge [5].

A recent study reported that chemo-mechanical preparation only managed to remove necrotic and/or vital tissue from the entrances of lateral canal and dentinal walls in 75% of the teeth investigated [3]. Despite the development of the instrumentation technique and the use of intracanal irrigants, failures of the endodontic treatment are still reported in the literature [6]. Therefore, in case of microbial resistance to routine treatment procedures, laser can assist the endodontic treatment for elimination and reduction of microorganisms existing in the main canal, lateral canals and dentinal tubules. A successful endodontic treatment requires both mechanical and chemical cleaning [7]. During the mechanical preparation, the surface of the root canal wall is not totally exposed to instrument blades in an efficient way. The preparation of canal with specific instruments implies accumulation of debris, blocking isthmus, fins, accessory canals and lateral canals. Studies investigating the cleaning ability of endodontic instruments, typically have been performed by light- or scanning electron microscopy. Instrumentation involving larger file sizes using rotary NiTi

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introduced in the canal, resulting in increased debris removal. Shock waves within the cleaning and debriding solutions increase the power of the Er: YAG laser to create photoacoustic waves. Laser-activated irrigation (LAI) is a revolutionary method for cleaning root canals. Photon Induced Photoacoustic Streaming (PIPS) is an innovative method for improving the effectiveness of irrigation, including sonic or ultrasonic devices and different types of lasers. The use of lasers in the field of endodontics represents an innovative approach to match the needed requirements for a properly cleaned root canal. In endodontics, laser is used in three main directions: reduction of the microbial colonies by means decontamination of the infected root canal, cleaning of the root canal surface and removing the smear layer. Recently, the interaction between laser and the irrigant in the root canal outlines a new area of interest in the field of endodontic disinfection. Laser-activated irrigation (LAI) has been introduced as an activation method of irrigation solutions by the transfer of pulsed energy. In 2010, Divito introduced another design of the erbium laser tip, the so-called PIPS-tip, which stands for Photon-Initiated Photocoustic Streaming. Pulsed laser operation induces photocoustic shock waves in the irrigant which travel throughout the RC system and allow their 3-D movement. Using low, sub-ablative pulse energies (only 10 or 20 mJ) the PIPS technique minimizes undesirable thermal effects on the dentinal walls. When activated in a limited volume of liquid, the high absorption of the laser in hypochlorite combined with the high peak power derived from the short pulse duration (50 μs) determines a photoacoustic phenomenon. Nowadays, the use of LAI has been found to be more effective in removing dentin debris and smear layer in comparison to passive ultrasonic irrigation (PUI) or hand irrigation. Recently, the use of laser energy has been also proposed for increasing the decontaminating action of the sodium hypochlorite. Er:YAG laser proved its ability and effectiveness in removing the smear layer and debriding the root canal. The Er: YAG laser of 2940 nm wavelength has a high absorption in water and a high affinity for hydroxyapatite. It works on the principle of transferring the pulsed energy to activate the irrigation solutions, which makes it suitable for use in root canal disinfection and cleaning. Photon Induced Photocoustic Streaming (PIPS) is a revolutionary method for cleaning and debriding the root canal system using Er:YAG laser energy at sub-ablative power levels. The PIPS tips harness the power of the Er: YAG laser to create photocoustic shock waves within the cleaning and debriding solutions introduced in the canal, resulting in increased debris removal with minimal or no thermal damage to the organic dentinal structure.

As the apical part of root canal is generally not straight, its proper cleaning is not an insignificant process. The removal of the necrotic tissue and debris from this part of canal is crucial for the future of restored teeth, so a proper evaluation of the endodontic treatments is fundamental. The use of a non-destructive analysis method for the investigation of the effectiveness of the various applied treatments is important because it does not affect the debris quantity and distribution. However, canal preparation may be adversely influenced by the highly variable root canal anatomy and the relative inability of the operator to visualize this anatomy from radiographs. Therefore, the need of having a clear 3D picture of the entire system of the root canal and its additional channels is fundamental. Same studies have evaluated the adhesive properties of root canal walls by measuring the microleakage between the dentinal wall and the filling material. These studies have investigated the effect of smear layer removal on sealer adhesion with controversial results. X-ray microcomputed tomography (micro-CT) involving synchrotron radiation X-rays is a powerful technique that can deliver non-destructive spatial information in the field of dentistry and implantology as it allows an easy reconstruction of the imaged specimens, together with a complete morphological analysis. Several groups studied the root canal morphology by micro-CT, as slices could be recreated in any plane, and data could be represented as rendered 3D images. Micro-CT was used to generate both qualitative and quantitative outcome measures for investigations of pulp cavity and root canal morphology. Another group quantified the inorganic dentine debris following internal tooth preparation by micro-CT. Anyhow, we have no knowledge of any work, involving synchrotron radiation micro-CT, done in the field of analysing the presence of necrotic tissue and dentine debris, as a result of endodontic treatment, involving laser cleaning of the root canal. This is probably due to the fact that it is difficult to visualize and quantify the organic tissue in the root canal by absorption micro-CT, produced by laboratory sources (desktop micro-CTs), which is the case of all the studies mentioned above. The use of X-rays delivered by synchrotron facilities has several advantages compared to X-rays produced by laboratory sources. It includes the possibility to take advantage of the high photon flux, which guarantees the achievement of high spatial resolution with good signal-to-noise ratio and reduced artefacts. Furthermore, third-generation synchrotron light sources produce brilliant photon beams which are suitable for routine application of phase sensitive X-ray imaging methods. In soft biological tissues, as the necrotic pulp tissue, the attenuation of highly energetic X-rays is weak, thus yielding poor absorption contrast. However, such phases introduce sizable phase shifts to X-ray wave fronts during a synchrotron radiation-based micro-CT experiment, creating intensity contrast downstream of the object due to a free-space propagated transmission function and the phase contrast can be 1000 times larger than absorption contrast. It has been opted for a high-resolution phase-contrast micro-CT experiment, because it provides high resolution and good signal to noise ratio images, allowing a good visualization of the roots morphology and a good evaluation of the effectiveness of cleaning level in terms of removing the necrotic pulp tissue and other debris, impossible to be properly observed by other non-destructive investigations methods, including desktop micro-CT, which does not allow a similar level of contemporary visualization.
of the tooth structure and organic phases present in the root canals.

Experimental part
Materials and methods

Investigated teeth and experimental procedure

Twenty single-rooted mature teeth were extracted for periodontal reasons and were selected for this study. Each patient gave a written consent in order to use the tooth for the study. The study design (teeth collection and consequent investigation methodology) was approved by the Ethical Committee of University of Medicine and Pharmacy Victor Babes, Timisoara, Romania. The teeth were cleaned of debris and tissue remnants after that were stored in a 0.1% thymol solution until the experiment. The teeth were radiographed to confirm the presence of single root canal and absence of root canal filling, calcifications, or other foreign body. The inclusion criteria were endodontic sound teeth, no previous root canal treatment, root filling or restoration, single canal and no sign of resorption. All teeth were subjected to the same protocol. In the first phase, each tooth was microstructurally analysed by synchrotron radiation X-ray micro-CT before any mechanical preparation. After this examination, eight teeth with abnormality in the apical area were excluded from the next phase (fig. 1A, B).

In the second phase, twelve teeth were submitted to the chemo-mechanical preparation of the roots to apical size of 0.40 mm using ProTaper Rotary System (Dentsply Maillefer Ballaigues, Switzerland) and alternative irrigation with sodium hypochlorite (3 mL 2.5% between each file and after preparation). At the end of the procedure alternative distilled water and 3 mL 17% EDTA solution were used. Subsequently, a second investigation by synchrotron radiation X-ray micro-CT was performed. In the third phase, a LAI protocol was applied using an Er:YAG laser with a wavelength of 2940 nm (Fidelis Plus III, Ljubljana, Slovenia), using photon-induced photoacoustic streaming (PIPS®) fiber tips 300/14, with 400 microns in diameter, with cylindrical-conical shape at its apical end. The following parameters were considered: 20 mJ, 15 Hz, SSP 10-30 s, under 2.5% NaOCl irrigation; 30 s resting time between one cycle and another (2 cycles); at the end, 20 seconds irrigation with distilled water for neutralization was applied. The laser tip was introduced no further than 4 mm in the canal. Both phase 2 and phase 3 were performed at the Semez Srl dental practice in Trieste (I). Finally, the specimens were imaged again by synchrotron radiation micro-CT.

Synchrotron radiation X-ray phase contrast micro-CT

All samples underwent advanced microstructural characterization by phase contrast synchrotron radiation micro-CT. The experiment was performed on the SYRMEP beamline of the ELETTRA Synchrotron Radiation Facility (Trieste, Italy). A contrast phase setup was considered for a better visualization of both teeth morphology and organic tissue inside the root canals. The radiographs were acquired at an X-ray energy of 27 keV and a detector pixel size of 4.2 mm. In order to have a better flux at this energy and higher signal to noise ratio, the experiment was performed during a 2.4 GeV operation period of the ELETTRA synchrotron, which provides an increased higher energy photons flux. A total of 1200 radiographic images were recorded for each specimen. The sample-detector distance of 200 mm enabled to work in contrast phase mode. The tomographic reconstruction was performed by means of the common filtered back-projection method. The field of view for the experiment was about 5 mm in height, with no restrictions in the horizontal direction. The obtained data were analysed using the VGStudio MAX 1.2 software (Volume Graphics, Heidelberg, Germany). Quantitative parameters were calculated directly from 3-D images to characterize the full set of specimens, after segmentation of the different phases to separate them from the background. The grey scale inversion was obtained using the ImageJ software.

Results and discussions

The entire field of view (5 mm in height) was divided into 4 regions: one from the anatomical apex to the apical foramen (about 0.5 mm), the lower apical region (1.5 mm height), the medium apical region (1.5 mm) and the higher apical region (1.5 mm) (fig. 2A, B, C). Different analysis of the first region was not considered as no necrotic tissue was present in that region. This partition was determined by the different chemo-mechanical treatment range of effectiveness in function of height, connected to the natural curvature of the roots and access of the rotary system files. All investigated teeth had initially a certain amount of necrotic tissue inside the root canal (fig. 3A, B, C). While for the upper part of the apical region, the chemo-mechanical intervention produced better results in terms of necrotic tissue and debris removal, in the lower part, the mechanical instruments had limited access and a higher amount of tissue and debris was observed (fig. 4 A, B, C).

As presented in figure 5 and the results from table 1, the endodontic treatment was very efficient in properly removing the necrotic tissue in the higher apical region of the root canal. In fact, only a smallest quantity of tissue and debris were found in this area after the chemo-mechanical treatment, most of the root canal being free
of tissue and debris (fig. 5B). Furthermore, the laser treatment almost completely removed the necrotic tissue and debris thus the canal remains clean (fig. 5C).

The chemo-mechanical endodontic treatment was averagely less efficient in the middle and lower apical regions as tissue and dentine debris, pushed by the rotary system files during the treatment, were found in all the investigated roots in the middle-lower apical regions. As it results from figure 6, the rotary system files took out part of the necrotic tissue meanwhile, pushed part of it, together with some dentine debris in parts of the canal that were not reached by the rotary system files.

Figure 7 presents an evident difference between curved roots and straight roots in terms of tissue and debris cleaning in the lower apical region. If for the straight roots a good cleaning efficiency was observed, for the curved roots the mechanical cleaning instrument was not able to follow the root canal up to its end, leaving or pushing debris, especially in the lower apical region.

The Er:YAG laser treatment applied afterwards gives good results in terms of cleaning the tissue and dentine debris from the root canal, especially when the debris remain in the main canal, although some residual debris were still visible in our micro-CT images. The laser treatment proved its efficiency in the lower apical region, also in the case of a curved root.

The laser treatment for one tooth had a lower efficiency when the tissue, together with dentine debris were pushed by the mechanical treatment and formed a bottleneck (fig. 8). As the tissue and debris were stuck inside, only a small part was removed. Furthermore, the chemo-mechanical cleaning of the root could not reach the secondary canals, from where the tissue could not be removed by the cleaning actions. In an inverted image (fig. 7), is visualized a lateral canal, containing tissue that remains there both after the chemo-mechanical and laser treatments.

The ability of root-canal sealers to adhere to dentine root canal wall is expected to be a result in superior debriding, which in turn should reduce leakage in clinical situations. Adhesion depends on a multitude of interacting factors including the surface energy of the adherent (dentine or gutta-percha), the surface tension of the

<table>
<thead>
<tr>
<th>Tissue &amp; debris</th>
<th>Tissue &amp; debris after chemo-mechanical treatment</th>
<th>Tissue &amp; debris after laser treatment</th>
<th>Endodontic treatment efficiency</th>
<th>Laser treatment efficiency</th>
</tr>
</thead>
<tbody>
<tr>
<td>Higher apical region</td>
<td>5.4%</td>
<td>1.4%</td>
<td>0.4%</td>
<td>74%</td>
</tr>
<tr>
<td>Medium apical region</td>
<td>5.3%</td>
<td>2.1%</td>
<td>0.7%</td>
<td>60%</td>
</tr>
<tr>
<td>Lower apical region</td>
<td>5.9%</td>
<td>5.1%</td>
<td>2.1%</td>
<td>14%</td>
</tr>
</tbody>
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adhesive (sealer), the adhesive’s ability to wet the surfaces and the cleanliness of the adherent surface [35]. Two mechanisms of adhesion may be distinguished: chemical and mechanical. In case of chemical bonding a smooth surface generally results in better adhesion. Micromechanical bonding, on the other hand, requires the presence of irregularities on the surface of the adherent into which the adhesive can penetrate.

Bacteria are the major cause of pulpal and periapical diseases. Complexity of the root canal system, invasion of the dentinal tubules by microorganisms, formation of smear layer during instrumentation and presence of dentin as a tissue are the major obstacles for complete elimination of bacteria during cleaning and shaping of root canal systems. Microleakage is defined as the passage of bacteria, fluids, and chemical substances between the root structure and fillings of any type. This occurs because of microscopic gaps at the interface of the filling material and the dentinal wall of the root canal. The major inconvenient will be represented by the decrease in root canal sealer adhesion.

The mechanical preparation of the root canal with high taper instruments in the coronal part and small diameters in the apical region often leads to instrumented recesses, with accumulation of debris in the unprepared root canal area. Furthermore, using a tooth vacuum cleaner, Endo Vac for example, involving apical negative pressure, will optimize the removal of necrotic tissue and the debris produced during the chemo-mechanical treatment. This fact is motivated by the need to eliminate from the beginning as much debris as possible, in order to avoid their accumulation in the narrow parts of the apical region of the root canal or to avoid them to be stuck at the entrance of secondary canals. The diameter, length, shape, and undulation of the apical region vary among different tooth types, which may complicate or hinder thorough chemo mechanical debridement of those apical branches (fig. 9) [36].

Especially for this area, the use of the Er: YAG laser treatment with PIPS tips during the endodontic treatment proved to be very beneficial, increasing the cleaning efficiency of the endodontic treatment. In endodontics, in order to achieve a better adhesion of the different filling materials it is necessary to obtain a complete chemo mechanical cleansing of the complex root canal system. Intimate bond between the filling material and the root canal dentine will give the proper adhesion. In a previous research, it was proved by SEM examination the appropriate appearance of the dentin for this purpose [7]. Presence of the biofilm will determine the increase of microleakage between the root canal filling material and the dentinal wall which will conduced to the failure of the endodontic treatment. Another common clinical problem affecting root endodontically treated teeth is the possibility of root fracture. Factors that may predispose to fracture have been identified as changes in the mechanical properties and composition of dentine because of the action of irrigants.

The classic chemo-mechanical treatment should be associated with a laser-activated irrigation treatment for combined better results, as the shaking effect induced by the photoacoustic shock waves produced by the PIPS tips, will enhance the cleaning effects of the rotary system files. Recent studies demonstrate the effectiveness of laser assisted endodontic treatment compared to other irrigation techniques. Smear-layer removal was most effective when the root canals were irrigated using Er:YAG laser at low energy with 17% EDTA solution. Interestingly, removal of the smear layer along the entire canal was similar when the laser was inserted in the upper coronal third and at 1 mm short of the working length of the root canal. This
effect was not observed with the ultrasonic and positive-pressure techniques [37]. Another study showed that laser-activated irrigation (LAI) removed two times more antibiotic paste (DAP) from the root canal surface than other techniques [38]. Laser-activated irrigation removed more biofilm than ultrasonically activated irrigation when using sodium as the irrigant, indicating greater physical biofilm removal [39]. The use of NaOCl after or in combination with a chelator caused the greatest reduction of E. Faecalis. Diode laser and Er:YAG laser activation were superior to ultrasonics in dentinal tubule disinfection [40]. Eliminating debris from complex canal spaces found in mandibular molars was achieved at a significantly greater level using laser-activated PIPS irrigation [41].

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Conclusions

Therefore, further investigations are foresene, for developing a new study design, which will combine these two methods in preventing debris accumulation. This study proved the unique possibility of the phase contrast synchrotron radiation micro-CT to highlight, in a 3D non-destructive way, the quantitative kinetics of the organic debris removal from the most difficult area of the root canal to be instrumented - the apical region - after a laser assisted endodontic treatment, involving both chemo-mechanical and laser treatments. The limits of this study consisted in the absence of specimens with very similar morphology to allow a statistically significant investigation. It is difficult to identify a significant number of teeth with similar apical root morphology (curvature). A possible solution (a future study is foresene) would imply the use of a desktop micro-CT instrument for a preliminary investigation of the root canal geometry.

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