A MICROSCOPIC STUDY ANALYSING THE EXTENSION OF A BURN LESION PRODUCED „IN VITRO" ON RADICULAR DENTIN USING THE DENTAL LASER

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Abstract

This "in vitro" study is intended to demonstrate the possible negative effects on dentin structure during decontamination of the endodontic system, assisted by the dental laser.
An upper central incisor (21), that was extracted due to severe endo-periodontal pathology, was treated endodontically, using rotary files, Hyflex EDM, Coltene, up to 25 ISO with taper 04, and then was sectioned longitudinally with the help of a fine diamond disc. On each half of the tooth, three burn lesions were artificially created, by activating the laser tip from 3 W to 6.5 W, for 10 seconds. The dental laser used was Epic X, Biolase, and the two pieces of tooth were investigated by stereomicroscopy and optical microscopy in reflected light. For the extension of the burn lesion, we have studied the depth of the dental structures damaged and the width compared to the surface of the root canal.
The outcomes confirms the safe use of the EpicX diode laser, Biolase, in endodontic treatments, as long as the manufacturer's instructions are followed (< 1.5 W). Thus, the thermal risk of damaging the radicular dentin is avoided.

Keywords : in vitro, burn lesions, diode laser, root canal decontamination, radicular dentin.

Introduction

An advanced and modern technique used for decontamination the endodontic system is the laser-actived irrigation.
This device generates a coherent beam of light, with the following properties [1]:

- monochromaticity, a narrow spectrum of wavelengths,
- high intensity of the light beam,
- collimation, reducing the divergence of the light beam,
- it can propagate long distances with a small divergence, which means it can be focused on a small surface.

Under normal conditions, at the tissue level, the light beam of the dental laser is reflected, transmitted, dispersed and absorbed in different proportions. The most important aspect of the laser wavelength is the amount of energy absorbed, as it turns into thermal energy and thus increases the temperature of the tissue [2].
By using this technique, studies have shown an improvement of the disinfection of the root canal due to the fact that the penetration of the irrigant into the dentinal tubules is greater than in case of the conventional technique (100 μm) [3-5]. Also, the activation of Sodium hypochlorite by the diode laser (after the mechanical preparation of the root canal) intensify the bactericidal effect [6,7].
Demidova et al demonstrates, in vitro, the bactericidal effect of the laser light wave at the level of the root canal previously inoculated with Escherichia coli and Enterococcus faecalis.

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The power of the dental laser being set to 3 W, having activation periods of the irrigation solution of 5 sec. It was also found that by increasing the power to 4 W, the bactericidal effect is stronger, but the temperature of the dental structures increases by 6 degrees [8].
Besides the cavitation effect of the dental laser, there is also a photothermal effect causing the temperature of the sodium hypochlorite solution to increase. High temperature levels can produce irreversible damage to the nearby dentine structure. A negative aspect, that it is highly recommeded to be avoided by following the manufacturer's instructions for the 3 parameters used: frequency, power and energy [3].
Burn lesions in endodontic spaces can be accidentally done with diode laser, during root canal decontamination, through the direct contact of the active tip with the tooth structure
Da Fonseca et al., studied the effects of the laser-activated irrigation technique on the nearby dental structures, by apllying increased intensities (1.5 – 3.5 W). Thus, it was found that the risk of raising the temperature at the radicular dentin level is much lower if the power of the device is below 3 W [9].
Burn lesions can occur in two situations:
- in the situation where the intensity of the laser radiation is incorrectly adjusted to a too high level;
- in the situation where the tip of the device is kept activated for a long time in direct contact and in a constant position at the level of the root canal wall.
In the present study, the burn lesions were artificially generated using increasing intensities of the dental laser. These lesions can affect the endocanal dentin.

The investigation of these lesions is important for two reasons:

- the "burnt" dentin tissue is mostly carbonized, these portions being friable and easy to detach from the root canal wall, leading to the appearance of small excavations. These can have consequences on the endodontic space obturation procedures and possibly in the realization of the composite resin coronal restoration;
- the detached carbonized dentine segment can be drawn into the apical area during root canal instrumentation.

Material and method

In this study, we aimed to study the extension of burn lesions produced in vitro, correlated with the radiation intensity of the laser wave used.

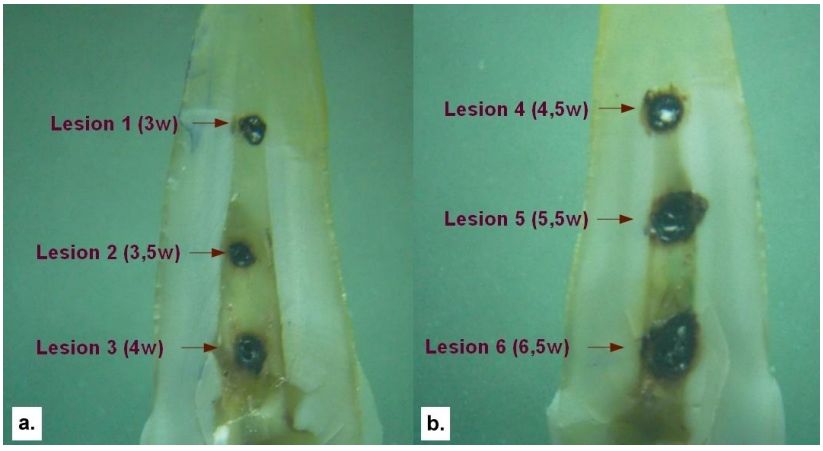

Fig. 1 The two root fragments resulting from the longitudinal sectioning of the front central incisor (tooth 21) :
a. the mesial fragment

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b. the distal fragment

Three burn lesions were done in vitro on each root canal half; the intensities, at which they were generated, are marked accordingly along with their numbering on the microscopic images. (stereomicroscopy in reflected light, 8x).

In order to have access to the endodontic space so that the tip of the dental laser can be maintain in a constant position with certainty, two dental section surfaces were used. A maxillary central incisor (21), that was extracted due to severe endo-periodontal syndrome, without endodontic treatment done :

- was treated endodontically, using rotary files, Hyflex EDM, Coltene, up to 25 ISO with taper 04 and, then,
- was longitudinally sectioned, in the vestibulo-oral direction, into two halves with a microtome, Leica Biosystem Microtome RM2125RTS, in specialized histopathology service.

The sectioning was done after making special markings and an additional stereomicroscopic investigation under strong transillumination conditions of the tooth. As shown in fig. 1 a mesial half and a distal half were separated. On each such half, in the space corresponding to the root canal, three burn lesions were produced, at a distance from each other, using increasing intensities of laser radiation. Each lesion was generated at an intensity specified below by holding the tip of the dental laser in place for 10 sec.

To measure the depth of the burn lesions were made 12 new cross-section surfaces, transversely on the tooth halves already obtained by longitudinal cutting with the microtome. The two new sections detached a small slice of tooth relatively perpendicular to the root canal, with an approximate thickness of 750μm.

Before making the 12 transversal sections, we have filled the root canal halves with a special epoxy resin, used for specimens consolidation in technical microscopy. The resin is inert and has no properties that would modify the carbonized dentin. The way in which the 12 additional section surfaces are placed compared to the longitudinal one, is shown in fig. 2.

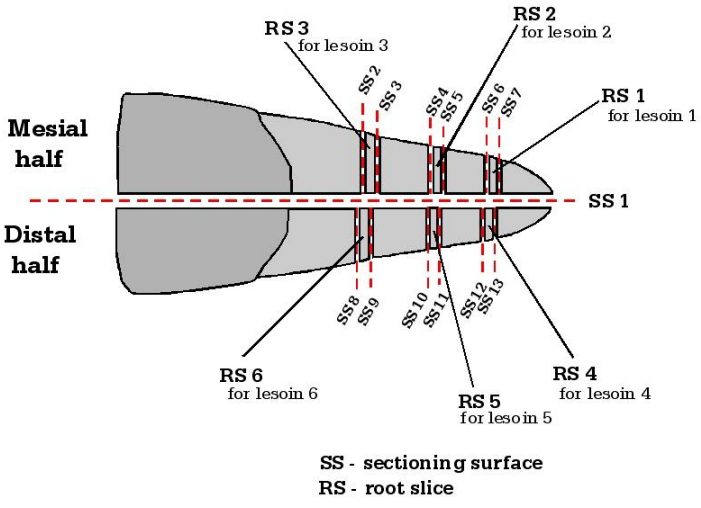

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Fig. 2. The sketch of how the sections of the investigated tooth were made for the burn lesions:

a) sectioning surface 1 (SS1) was performed before making the lesions, with the aim of opening the endodontic space and exposing the walls of the root canal;
b) sectioning surfaces 2 to 13 were made after making the lesions, with the aim of detaching six slices (root slice 1 to root slice 6 - RS1 to RS 6), to be able to investigate microscopically the depths of the affected dentin.

For each lesion, two digital microscopic photos were taken:

- one for the surface extension on the root canal wall at 40x magnification,
- one for the deep extension in the root dentin at 100x magnification.

Calibrations in micrometers (μm) were placed on both categories of pictures. The total number of pixels corresponding to each lesion was related to the total number of pixels of the image.

For interpretation we've selected four exemples, in which the burning pattern is different.

Data obtained following microscopy investigations :

1. the extension of the lesion compared to the surface of the root canal :

The measurements were made before the sectioning surfaces SS2 to SS13.
Two values were established:

- lesion's height : measured along the root canal and
- lesion's width : measured in a direction perpendicular to the walls of the root canal.

Microscopic images are shown in fig. 3 to 6 and the data obtained are presented in table 1.

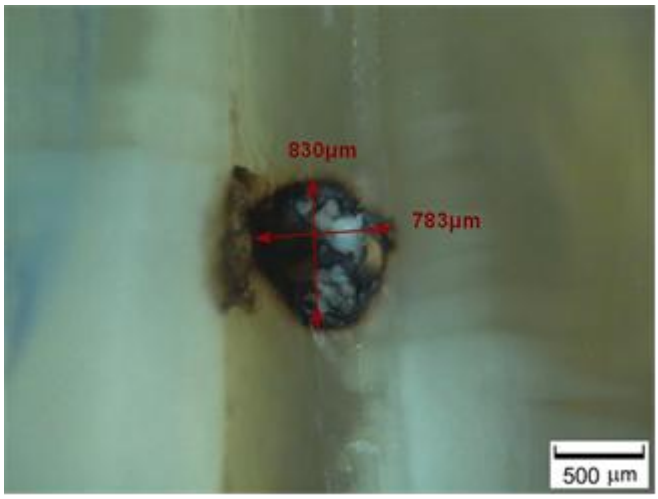

Fig. 3. Surface image /The height and width of lesion no. 1 on the wall of the root canal of the central incisor selected for the study (the lesion is produced using a laser radiation intensity of 3W) (Reflected light microscopy image, 40x)

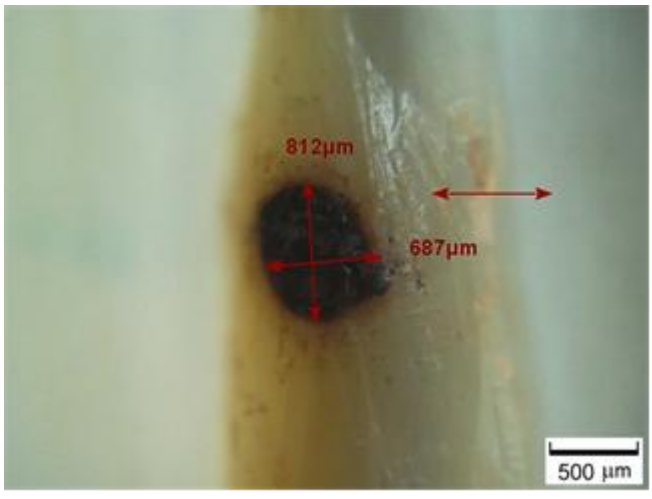

Fig. 4. Surface image/ The height and width of lesion no. 2 on the wall of the root canal of the central incisor selected for study (the lesion is produced using a laser radiation intensity of 3.5W) (Reflected light microscopy image, 40x)

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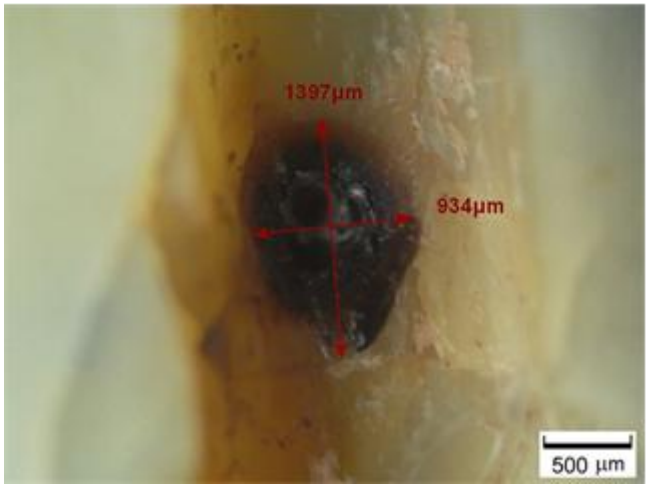

Fig. 5. Surface image / The height and width of lesion no. 3 on the wall of the root canal of the central incisor selected for the study (the lesion is produced using a laser radiation intensity of 4W) (Reflected light microscopy image, 40x)

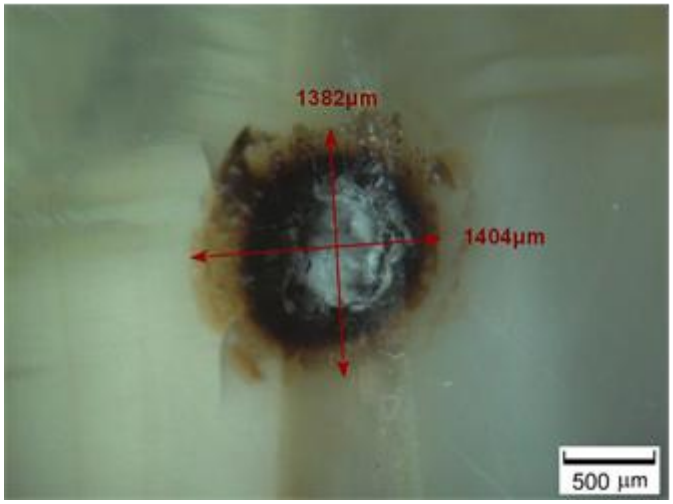

Fig. 6. Surface image / The height and width of lesion no. 4 on the wall of the root canal of the central incisor selected for the study (the lesion is produced using a laser radiation intensity of 4.5W) (Reflected light microscopy image, 40x)

Table 1 : Measurements related to the extension of the lesion on the surface of the root canal of the central incisor selected for the study

Lesion no.	Intensity of laser radiation (W)	Lesion height (µm)	Lesion width (µm)
1	3	830	783
2	3.5	812	687
3	4	1397	934
4	4.5	1382	1404
5	5.5	1944	1762
6	6.5	1994	1870

2. the extension of the lesion in the depth of the radicular dentin :

After making the transverse root slices (root slice 1 to root slice 6 – RS1 to RS 6), microscopic investigations and micrometric determinations could be made on each. It was measured the extension of each lesion in the depth of the radicular dentin. The microscopic images are presented from fig. 7 to fig. 10. As it can be seen in these cross-sectional images, the lesion is composed of two domains:

- one of carbonization, with a blackish appearance, becoming opaque, centrally located and being the largest part of the lesion;
- one with a brown appearance with reduced transparency (compared to unaffected dentin), which appears as a peripheral border of the lesion, being a small part of the lesion.

We considered the burn lesions to represent not only the charred part, but the entire dentin area in which the changes were found from an optical point of view (both color and transparency changes). Bidirectional transillumination was used to assess the

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change in transparency as visually as possible. The measurements are presented in table 2. The two values shown in the table for each lesion are noted as : carbonized zone depth, measuring only the expansion of the carbonization area, and lesion total depth, for the entire affected zone.

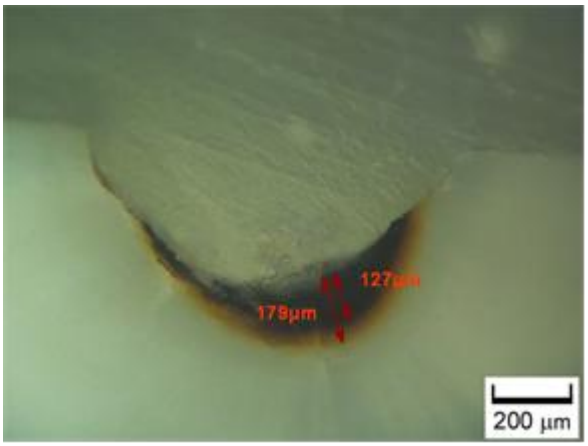

Fig. 7 Image of lesion no. 1 in the depth of the radicular dentin of the central incisor selected for the study (the lesion is produced using a laser radiation intensity of 3W)
(Related reflected light microscopy image with bidirectional transillumination, 100x)

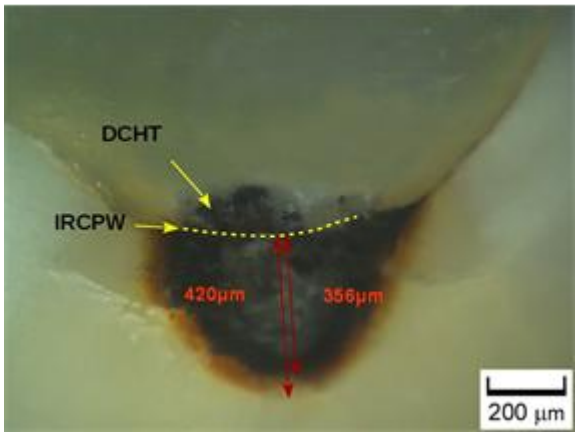

Fig. 8 Image of lesion no. 2 in the depth of the radicular dentin of the central incisor selected for the study (the lesion is produced using a laser radiation intensity of 3,5W)
DCHT – detached carbonized hard tissue
IRCPW – initial prepared root canal wall
(Related reflected light microscopy image with bidirectional transillumination, 100x)

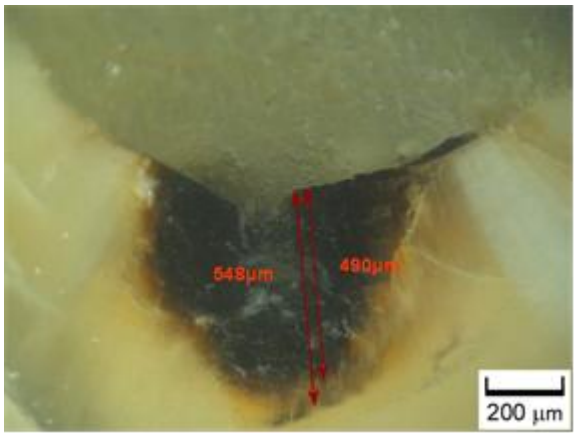

Fig. 9 Image of lesion no. 3 in the depth of the radicular dentin of the central incisor selected for the study (the lesion is produced using a laser radiation intensity of 4W)
(Related reflected light microscopy image with bidirectional transillumination, 100x)

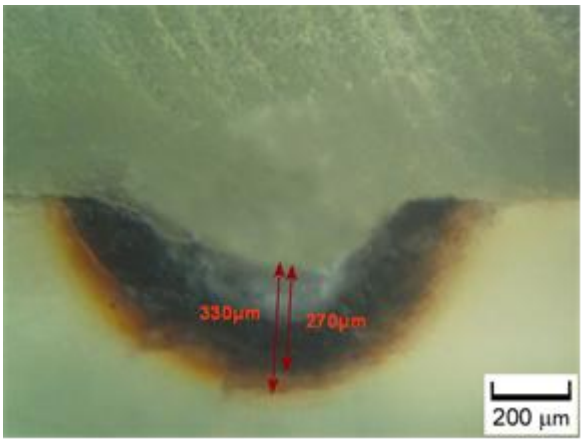

Fig. 10 Image of lesion no. 4 in the depth of the radicular dentin of the central incisor selected for the study (the lesion is produced using a laser radiation intensity of 4,5W)
(Related reflected light microscopy image with bidirectional transillumination, 100x)

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Lesion no.	Intensity of laser radiation (W)	Carbonized zone depth (μm)	Lesion total depth (μm)
1	3	127	179
2	3.5	356	420
3	4	490	548
4	4.5	270	330
5	5.5	450	495
6	6.5	378	479

Discussions

Depending on the power of the light beam, the temperature of the nearby tissues can increase causing irreversible damage such as [10]:
• hyperthermia of the dental structure, reaching a temperature of 37-43° C,
• interruption of the enzymatic activity of the surrounding tissues. The measured temperature being 43-50° C,
• the collapse of collagen fibers from the dental structure, at temperatures of 70-80° C,
• carbonization of the dentin structure, at temperatures of 100-140° C,
• the destruction of the dental structure is achieved at temperatures of 500-800° C.

Therefore, it can be said that the burn lesions from the present study are influenced by the diode laser's radiation lightwave intensity.
In a study by da Fonseca et al., were investigated the temperature variations on the root surface at the level of the middle and apical third of mandibular incisors. Different laser intensities were used, and at 3.5W, the middle third showed a rise of more than 7°C compared to the apical area. It might suggest that a temperature increase of no more than 7° Celsius represents a safety limit for the dental structure and surrounding tissues [9].

In our study we found that :

- the burn lesions were clearly formed at a intensity over 3 W,
- the depth of the dentin substance degradation increases in figures 7 and 8, which represent lesions no. 1 and 2 on the cross-section. In comparison to the first lesion at 3W (179 μm), burn lesion no. 2 (420 μm) is nearly two times greater. It can be deduced that once the value of 3 W is exceeded, burn damage will unavoidably occur, and if the laser's intensity keeps increasing, leads to immediate depth expansion.

The depth of lesions no. 3 (total depth of the affected area: 548 μm, at 4W) and no. 4 (total depth of the affected area: 330 μm, at 4.5 W) differ significantly in table no. 2. This is supported by the values measured at the surface of the root canal corresponding to the two lesions, which are 812 μm and 1397 μm (figs. 5 and 6). Although the lesion in fig. 14 shows less damage in depth than the one in fig. 13, we may conclude that the affected surface's width is significantly greater, making it a more extended lesion in the longitudinal direction.

Since no such occurrence was seen in any of the other sections, the appearance in fig. 8, described as DCHT (detached carbonized hard tissue), cannot be understood as a tendency to expand in volume/depth along with the effect of dentin burning. I added a dotted line to the fig. 8, which, in our opinion, represents the limit of the root canal instrumentation.
Cho H et al., have evaluated, in their study, the permeability of radicular dentin after irradiation with CO_2 , 13J/cm^2 . They have proved, using a 3D digital microscopy1000x magnification, that the laser treated radicular

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dentin surface shows translucent globules with darker carbonized collagen, causing micro-cracks [11].

Conclusions

It would have been expected that the diode laser's increasing intensities, which were used in this investigation, would lead to a proportional gradual increase of the lesions in both ways : transverse and longitudinal. It can be observed that the lesions are not always proportionate.

These differences can be explained by the fact that while the depth's lesion unexpectedly decrease, the extension on the surface of the radicular dentin increases significantly. We consider that other factors, such as the laser tip appereance, the degree of deterioration after multiple applications, or the angle at which the laser tip is kept against the root canal wall, can also influence this relationship of expansion in depth and surface.

The extension of the burn lesion in the coronar direction, towards the access cavity, can present a challenge in applying the composite resin restoration because of the absence of adhesion at that level.

The use of the laser-activated irrigation technique is recommended during root canal decontamination, as long as the manufacturer's instructions are followed.

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