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Effect of Irradiation with 980-nm Diode Laser on the Microhardness of Young and Old Root Canal Dentin after Treatment with Chemical Solutions

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ABSTRACT

Purpose: The purpose of this study was to evaluate the effect of diode laser-980nm on microhardness of young and old canal dentin after treatment with NaOCl, NaOCl/EDTA and distilled water solutions. **Material and Methods** Forty-eight single rooted premolars were divided into three groups according to the chemical solutions used (16 each) (8 for young teeth <30 years old and 8 for old teeth >50 years old). Group I: irrigated with 5ml of 2.6% NaOCl activated with diode laser, group II: irrigated with 5ml of 2.6% NaOCl without laser activation followed by 5ml of 17% EDTA activated with diode laser and group III: irrigated with 5ml of distilled water activated with diode laser. Then microhardness was evaluated at coronal, middle, and apical thirds of each root canal. **Results:** In all young and old groups, the coronal third had the highest mean value, followed by the middle, and the apical third had the lowest mean value, with a statistically significant difference between thirds of the groups (p0.001<). There was a statistically significant difference between young and old teeth at all levels (coronal, middle, and apical) for each group (I, II, and III), with old teeth having a greater mean microhardness value than young teeth. **Conclusion:** Irradiation of NaOCl/EDTA solutions with 980-nm diode caused reduction in microhardness of young dentin more than old dentin, and NaOCl only or distilled water in all thirds of groups.

KEYWORDS

Laser, Microhardness, Solutions.

INTRODUCTION

Dentin is the hard tissue that makes up the tooth's basic structure and protects the dental pulp. Dentin also has a microstructure that includes dentinal fluid-filled microtubules, an odontoblastic process, and mineralized intertubular and peritubular dentin. However, with

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aging, the thickness of the peritubular linings increases, reducing the tubule lumen diameter. These modifications take place until the majority of tubule lumens have calcified and the sclerotic tissue is apparent. Changes in mechanical parameters like fracture resistance, flexure strength, and microhardness are also recorded, in addition to microstructural changes(1).

Microhardness is a measure of local deformation resistance based on the degree of induced persistent deformation on the surface that continues after the load is removed. Any alteration in the microhardness of root dentin can impair the ability of dental materials, like root canal sealers, to adhere to the dentin(2). Instrumentation throughout the root canal is easier with a lower microhardness. However, it is possible that it will impair the root structure. Microhardness testing can be used to demonstrate the loss or gain of any mineral substance in the tooth hard tissues(3).

Endodontic irrigating solutions have the potential to influence the chemical and structural composition of radicular dentin, which could affect its permeability, solubility, and microhardness (4). Sodium hypochlorite solution is the most often utilized irrigant during root canal therapy because it can dissolve organic components, particularly collagen, reducing the mechanical characteristics of root dentin. Furthermore, inorganic dentin remaining in the root canal does not disintegrate(5).

The use of NaOCl in combination with 17% ethylenediaminetetraacetic acid (EDTA) was recommended as a way to dissolve the inorganic part of dentin. This is the most common irrigation method nowadays, especially in root canals that are narrow and calcified. Although EDTA is used to remove the smear layer, it can also remove calcium ions from the dentin. The chemical composition of dentin may differ from the main proportion of organic and inorganic components due to changes in ca/p ratio. As a result, dentin's solubility, permeability, and microhardness would alter(6).

Various irrigant techniques are used to provide superior irrigation solutions in root canals. Among the alternatives are needle irrigation, sonic and ultrasonic devices, and NaOCl and EDTA solutions(7). Endodontic laser devices have recently been used for various purposes and activation in treatment of root canal(8). The diode laser is a portable and compact device used in clinical applications. It has wavelengths that range from 655 to 980 nm, which are suitable for dental use. The diode laser (810-980nm) has the capacity to penetrate dentinal tubules and reduce bacteria counts up to 500 µm depth(9).

The laser parameters, such as application mode and output power frequency, determine the morphological changes in root dentin which irradiated with diode laser(10). In actuality, these variables are related to a rise in the temperature of the dental tissues. As a result, the chemical and physical properties of dentin may change and microhardness may alter as a result of laser activation(9). Accordingly, the study was intended to evaluate the effect of diode laser 980-nm on the microhardness of young and old root canal dentin after treatment with chemical solutions.

MATERIAL AND METHODS

Sample size estimation and statistical power

The calculation was estimated using CDC Epi Info program version 7.2.0.1 (Atlanta, USA) assuming a power of 80% and alpha=0.05 to detect significant difference in microhardness of root dentin of young and old root canal in 3 groups according to the final irrigation solution (NaOCl, combined NaOCl+EDTA and control using distilled water solution)(11).

A total sample of 48 teeth (16 in each group) is needed based on an estimated mean microhardness of 48.3±2.32 in NaOCl compared to 43±3.25 and 54.56±3.13 in EDTA and control, respectively.

Sample selection

Forty-eight freshly extracted single rooted anonymous teeth from young patients (30< years old) due to orthodontic reasons and old patients (>50 years old) due to periodontally diseases were used in this study. Ethical approval in the use of extracted human teeth was obtained in accordance with guidelines from the Research Ethic Committee of Faculty of Dental Medicine for Girls Al-Azhar University (REC-EN-21-08). Teeth with root caries, abnormal anatomy, cracks, previous endodontic treatment, internal (using periapical radiograph), external resorption and calcification were excluded. The collected teeth were rinsed in tap water to remove blood, tissue and debris cleaned from hard attached tissues. They were kept in distilled water solution throughout the study.

Samples preparation

Access cavity preparation was done through the deroofing of pulp chamber of selected teeth. Inserting a #10 size K-file (MANI Inc, Japan) into the canal and measuring the point at which the file exited the apical foramen of the root established the working length. The glide path was then established after the file length was reduced by 0.5 mm. ProTaper Universal rotary Ni-Ti files (Dentsply Maillefer, Ballaigues, Switzerland) were used for cleaning and shaping of canal. Preparation of root canal was performed on all experimental samples, starting with the SX file as an orifice opener and ending with the F4 file as a master apical file (MAF), with a standardized irrigation protocol of (2 ml of 2.6 % NaOCl for 1 minute)[5]. Between each file using a universal 27-gauge needle to complete apical preparation, the needle was inserted about 2mm shorter than the working length without binding. Distilled water was used in the control group as an irrigating solution at a rate of 2ml/1minute between each file.

Samples grouping

Samples were randomly divided into 3 groups (group I-III) according to the final irrigating solution and the activation protocol with 980-nm diode laser (Photon Plus, Zolar Technology & Mfg Co. Inc, Mississauga, Canada), with a 200μ fiber optic tip with setting at average power 1.2-W in pulsed wave mode. Up to the apical area the fiber tip was inserted 2mm short of working length. Each group was subdivided into 2 subgroups young and old teeth.

- **Group I (16 samples):** irrigated with 2.6% sodium hypochlorite (NaOCl) as final rinse in a total volume of 5 ml, divided into 5 cycles each cycle include 1 ml of 2.6% NaOCl irradiated with 980-nm diode laser for 5 seconds followed by 20 seconds pause, with a total activation time 25 seconds and 100 seconds' pause.
- **Group II (16 samples):** Using 5 ml of 2.6% NaOCl for 2 minutes without laser activation followed by 5 ml distilled water to deactivation of NaOCl solution then 5 ml of 17% EDTA as final irrigating solution which irradiated with 980-nm diode laser, divided into 5 cycles as group I.

Finally, group I, II rinsed with distilled water after laser activation to stop action of solutions.

- **Group III (16 samples) control:** Using 5 ml of distilled water as final irrigating solution irradiated with 980-nm diode laser, divided into 5 cycles as group I.

All groups (I, II, III) were subdivided into two subgroups (A, B).

- Subgroup (**A**): (n = 8) for evaluation of Microhardness in young teeth.
- Subgroup (**B**): (n = 8) for evaluation of Microhardness in old teeth.

Microhardness evaluation

A water-cooled diamond disc was used to remove the crowns of the teeth at the cemento-enamel junction (Komet, Brasseler). In cold cured clear acrylic resin blocks, the roots were vertically positioned and centered (Acrostone Co., Industrial

Zone, 15 km northwest of Cairo, Egypt). Each root was horizontally sectioned with a circular diamond disc at low speed and fresh cooling water (Microdont LDA. Brazil). The sectioning was done in a horizontal plane perpendicular to the main canal's long axis (Fig.1a).

The apical, middle, and coronal thirds are represented by 3mm thick sections extending from apex to coronal (Fig.1b). Each section's exact thickness was measured with a digital caliper and the coronal surface of each segment was coded. On a circular grinding machine, the specimens were flattened with SiC abrasive papers of increasing grit between 400 and 1000 grit (BIGO, Dent Product. Germany) using a rotary felt disc with constant water irrigation. Then, with the same load and time, each test condition was repeated three times at distances of 100, 150, and 200 µm from the canal lumen of each sample with the average of the three readings collected for each distance (Fig.1c)[2]. Each measurement was performed with a 200g load applied perpendicular to the surface for 10 seconds. Each residual impression was examined with an optical microscope using a Vicker microhardness machine (Model HVS-50 BUEHLER, Germany) with a pyramid diamond indenter tip (Fig.1d).

The monitor used the calculation HVN = 1854 (F/d2) to convert the readings into a Vickers hardness number (VHN). It is F is the indentation load (g), and d is the indentation diagonal (mm). Each specimen nine readings were averaged to generate a single hardness value. The VHN value of a specimen is calculated using the average of the three measurements for each test condition.

RESULTS

The significance level was set at $P \leq 0.05$. Statistical analysis was summarized in Table1.

1. Comparison between groups at each level of subgroup A (young teeth):

Regarding all levels (coronal, middle, apical) in subgroup A (young teeth) the highest mean value in the microhardness was found in group III control (distilled water+laser), followed by group I (NaOCl+laser). While the least mean value was in group II (NaOCl+distilled water+EDTA+laser).

2. Comparison between levels at each group in subgroup A (young teeth):

Regarding all groups (I,II and III) the highest mean value of microhardness was found in coronal level followed by middle, while least mean value in apical level.

3. Comparison between groups at each level of subgroup B (old teeth):

Regarding all levels (coronal, middle, apical) in subgroup B (old teeth) the highest mean value in the microhardness was found in group III

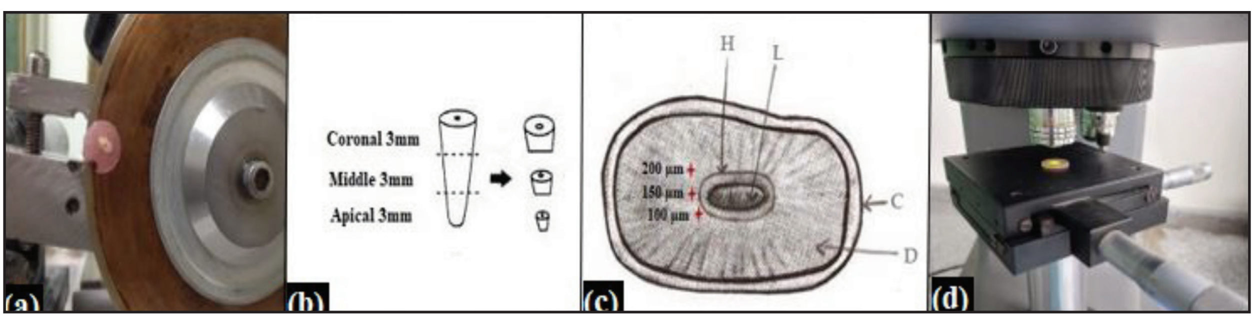

Figure (1) (a) The root sectioned in horizontal plane (b) Schematic representation of root sectioning (c) Root cross section showing the area where the microhardness of root canal dentin was measured D: dentin, C: cementum, L: lumen of the root canal, H: area where the microhardness was measured (100,150,200m from the lumen) (d) Vickers microhardness machined.

control (distilled water+laser), followed by group I (NaOCl+laser). While the least mean value was in group II (NaOCl+distilled water+EDTA+laser).

4. Comparison between levels at each group in subgroup B (old teeth):

Regarding all groups (I,II and III) the highest mean value of microhardness was found in coronal level followed by middle, while least mean value in apical level.

5. Comparison between subgroups A (young) and B (old) teeth at different levels for each group:

The results revealed that there was a statistically significant difference between young and old teeth at all levels (coronal, middle and apical) for each group (I, II and III).

Old teeth showed a higher mean value of microhardness than the young teeth in group III followed by group I, while least microhardness was in group II at all levels coronal, middle and apical thirds respectively (Fig 2).

Table (1) *The mean value and standard deviation (SD) of subgroups A and B (young and old) teeth at different levels for each group.*

	Subgroup A (young teeth)		Subgroup B (old teeth)		*p*-value
	Mean	SD	Mean	SD	
Group I (NaOCl+laser)					
Coronal	44.35	0.67	47.61	0.74	<0.001*
Middle	42.43	1.00	44.77	1.54	<0.001*
Apical	39.94	0.53	41.17	0.96	0.019*
Group II (NaOCl+ distilled water+EDTA+laser)					
Coronal	41.19	0.56	42.88	0.31	<0.001*
Middle	38.65	0.25	41.72	0.29	<0.001*
Apical	34.52	1.40	38.81	0.55	<0.001*
Group III (distilled water+laser)					
Coronal	48.81	1.16	50.82	1.21	0.014*
Middle	45.01	1.14	48.26	0.74	<0.001*
Apical	41.92	0.50	43.69	1.22	0.009*

**; significant (p<0.05)*

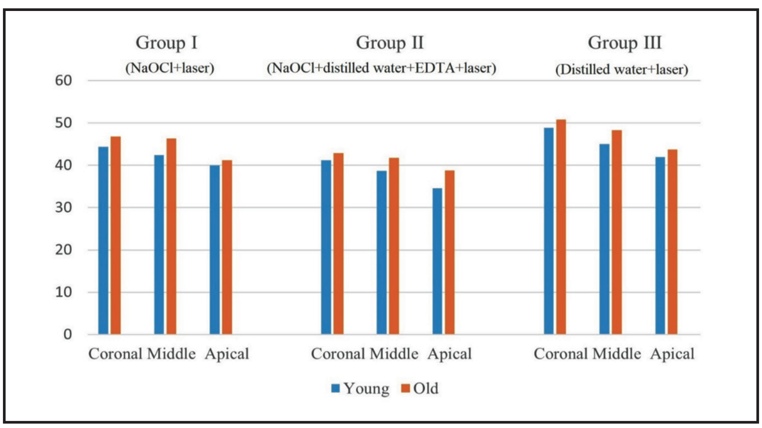

Figure (2): Bar chart representing microhardness of young and old groups at a different level for each group.

DISCUSSION

Cleaning and shaping of the root canal system is essential for root canal treatment while irrigation of the root canal system provides multiple advantages; including lubrication, elimination of microbes, dissolution of tissues and cleaning of areas inaccessible for mechanical preparation. During irrigation dentin of root canal is exposed to irrigating solution, which may cause alteration on properties of dentin and may affect interaction with material used for obturation(12).

The current study was performed on single rooted mandibular premolar with single root canal (Type I), to reduce variables as the anatomy of root canal has many geometrical possibilities in cross section; round, oval, long oval, flattened or irregular. These complexities in anatomy might represent physical constraints and a serious challenge to adequate root canal instrumentation and disinfection(13).

ProTaper Universal rotary files were used for canals instrumentation. They have a convex triangular cross-section, with an increasing taper design. This alleviates friction between the blade of file and the canal wall while increasing its cutting efficiency. Full cleaning and shaping of the root canal may be attained with apical preparation of the canals ending with F4 ProTaper Universal file(14).

Moreover, irrigation was used (2ml of 2.6% NaOCl for 1 min), with rotary mechanical preparation of root canals, between each file increasing permeability of the chelating agent to the radicular dentin. This has reduced bacterial count, dissolved collagen, and caused oxidation of the organic portion of radicular dentin(15). Hence, it was limited to 1 minute as prolonged usage of NaOCl can affect dentinal tissue. It may bring about undesirable microcrack formation and lower fracture resistance, flexural strength and microhardness(16). A universal 27-gauge needle was used in delivery of irrigation solution to complete apical preparation, as it corresponds to international standardization organization size 0.42mm(17).

As the purpose of this study was to evaluate how NaOCl/EDTA with diode laser 980-nm affected microhardness of young and old radicular dentin, diode laser activates 2.6% NaOCl were used as a final rinse in one of the experimental group and 2.6% NaOCl/17% EDTA in the second group. In the control group distilled water was used. Sodium hypochlorite was selected as it is more capable of removing organic material from root dentin and EDTA solution was used due to its capability in removing smear layer from dentin when used as a final rinse. The amount and time were selected to be 5ml for 2 min, as longer time exposures can affect mechanical properties of radicular dentin negatively(18).

The present study used a diode laser 980-nm for endodontic therapy. The diode laser device has both continuous and pulsed modes due to its practical application and prospective applicability. The continuous wave delivers laser energy to the tissue in a continuous stream. There are pauses of heat dissipation in the pulsed phase. The quantity of energy settled in tooth tissue in the pulsed mode is approximately half that of the continuous mode, hence it was chosen(19).

In case of having disinfection of the root canals with laser irradiation, appropriate parameters and protocols are needed to prevent thermal damage to the surrounding tissues. However, higher temperature in the periodontal ligament has not exceeded the safe limit (7 to 10°C) when 20-second rest periods were allowed after each cycle of laser therapy. This comes in accordance with an in vivo study showing disinfection efficacy of 1.05W diode laser after a 15-second cycle. Results were not substantially different from the control group(20). Dentinal melting, partial to complete obliteration of dentinal tubules, have been brought about with higher output powers 2W, urging us to use output powers of 1.2w to avoid melting areas and total occlusion of tubules(21).

It was proposed that 200 μm fiber optic tip be kept 2-3 mm distant from the anatomic apex to minimize irrigant extrusion and to prevent apical constriction eradication. The laser fiber tip was used 2 mm distant from the anatomic apex in the current study[22].

The microhardness of dentin is determined by the density of tubular changing from one area to another on the surface of root dentin as evaluated by the Vickers tester. Dentin microhardness decreases as tubular density increases. The Vickers microhardness tester was chosen over the Knoop hardness tester because of its applicability and feasibility for evaluating surface changes in deeper dental hard tissues. The Knoop hardness tester is used to measure the hardness of superficial dentin rather than deep dentin. Each surface treatment was done with three indenters. Microhardness is measured at the 100,150,200 m level from the root canal lumen in three parts (cervical, middle, and apical) of radicular dentin. The average of the data for the three indentations was used to calculate microhardness values[23].

The results of the present study showed that, among all levels there was the highest microhardness was found in Group III (distilled water+laser) followed by Group I (NaOCl+laser). While the least microhardness was in Group II (NaOCl+distilled water+EDTA+laser). This is probably attributed to the effect of NaOCl dissolving action on the organic collagen components of dentin. Furthermore, EDTA solution is efficient in eliminating the smear layer, which causes calcium/phosphate loss and lead to softening of the calcified dentin components [24]. The absorption of laser energy helps increase the penetration and efficacy of the irrigating solution by activating it with laser in the root canal resulting in the creation of vapor bubbles which are then collapsed resulting in sonic streaming, eventually leading to cavitation. This process enables the irrigant to exert shear force on the root canal walls and enhance the irrigant effectiveness[25]. However, we achieved similar results to those reported in a previous study on dentin microhardness decrease following irrigating with NaOCl and then EDTA[26].

Regarding the results of the present study showed that, the coronal one third has higher microhardness when compared to middle and apical thirds. This could be attributed to the number and diameter of dentinal tubules decreasing from cervical to apical dentin[27]. Moreover, variations in the structure at the apical region of the human teeth such as, varying quantity of irregular secondary dentin, accessory canals, cementum-like tissue, dentin sclerosis, and reduced amounts of noncollagenous proteins, decrease the microhardness in the apical region than that of the coronal one third[28].

The comparison between young and old teeth at different levels for each group revealed that there was a statistically significant difference between young and old teeth in all groups. Old teeth showed higher microhardness than the young teeth. This may be attributed to the physiochemical changes related to the aging process of dentin represented by increase in inorganic part of mineral content and decrease in organic part of collagen. Deposition of peritubular dentin increases leading to the narrowing of the dentinal tubules and complete obliteration of others which has a direct effect on the efficacy of the irrigating solutions[1]. Laser irradiation can cause morphological and chemical alterations in dentin represented by the evaporation of the dentin organic matrix leaving pores and spaces[29]. Thus, wider dentinal tubules of young teeth with higher irrigation efficacy have more pores and spaces than those of older teeth. This may explain the higher levels of microhardness of old teeth compared to young teeth[30]. Moreover, lower microhardness may weaken the root structure and limit the sealer ability to adhere to the root dentin walls and seal them [31].

CONCLUSION

Within the limitations of this study; it is concluded that using a diode laser with a wavelength of 980-nm on young and old dentin influences microhardness. This might affect the quality of obturation.

RECOMMENDATION

More research is recommended to determine the effect of 980-nm diode laser irradiation with chemical solutions for irrigation on the adaptability of obturating material.

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Conflict of Interest

The authors declare that there is no conflict of interest.

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