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In-vitro Comparison of Occluding Effect of Fluoride Varnish and Diode Laser Irradiation with Fluoride Varnish and Er,Cr:YSGG Laser Irradiation on Dentinal Tubules of the Cervical Root Area of the Tooth

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ABSTRACT

Background and purpose: *Dentin hypersensitivity (DH) is a sharp, short dental pain which originates from exposed dentin surfaces in response to thermal, evaporative, tactile, osmotic, chemical or electrical stimuli. Research showed that dentin tubule occlusion can lead to pain remission. Our goal was to evaluate the dentinal tubules in the cervical area of root teeth that are occluded by fluoride varnish, diode laser irradiation, and erbium laser irradiation.*

Materials and methods: *This is an in vitro single-blind study. Twenty-four samples of extracted third molars were divided into four groups: control (A), fluoride varnish (B), fluoride varnish and diode laser (C) and fluoride varnish and Er,Cr:YSGG laser (D). After applying varnish and different lasers, the tubule diameter and number of open tubules were examined by SEM. Data were analyzed by SPSS 23 software.*

Results: *In this research, there was no significant difference between groups C and D (ρ=0.999), although there were substantially more open tubules in the control group than groups C (ρ=0.004) and D (ρ=0.003). The mean diameter of tubules in the four groups was statistically different (ρ <0.001), and the descending order of tubule diameter was A > B > C > D.*

Conclusion: *Using diode and erbium laser in combination with sodium fluoride varnish had a significant effect on the reduction of dentinal tubule diameter and their occlusion; thus, these therapies can be used to treat dentin hypersensitivity.*

Keywords: diode laser, Er:Cr;YSGG laser, dentin hypersensitivity, scanning electron microscopy.

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INTRODUCTION

"Tooth hypersensitivity" is a sharp, short dental pain which originates from exposed dentin surfaces in response to evaporative, tactile, osmotic, chemical or electrical stimuli (1). Based on the hydrodynamic theory, fluid movement in dentin tubules stimulates nerve receptors at the level of odontoblasts and causes discomfort (1). It has been shown that hypersensitive teeth contained nearly eight times more dentinal tubules than normal teeth as well as larger tubular widths. When compared to teeth without symptoms, they are roughly twice as wide (2). Gingival recession, tooth fracture, erosion and abrasion expose dentinal tubules to oral environment (3). Therefore, neurotransmission reduction and physical closure of dentin tubules can relief pain (4). Dentin hypersensitivity is difficult to treat, although there are multiple available treatment options, including bonding materials, varnish, dental restoration (5), and oxalate containing dentifrices and mouthwashes (6). Use of gels and varnishes containing sodium fluoride is the most often used therapy. Tubules are shielded by a calcium fluoride coating created by fluoride varnish (7). Laser was first used in 1985 to treat tooth hypersensitivities (8). Low power lasers can depress nerve transmission by blocking afferent depolarization of C fibers (9), while high-level lasers occlude or narrow dentinal tubules by melting superficial layer of dentin at high temperatures (10), and after recrystallization they seal dentin tubules. We aimed to compare the effect of fluoride varnish in combination with diode laser and fluoride varnish with Er,Cr:YSGG laser on dentinal tubule occlusion in the cervical root region. ❑

MATERIALS AND METHODS

We conducted a single-blind in vitro study, which was approved by the ethics committee of Mazandaran University of Medical Sciences, Iran, under the certificate number of IR.MAZUMS.REC.1398.527. Twenty-four samples from 12 extracted third molars were collected. The sample size was obtained by the following formula, where alpha was 0.05 and beta 0.2, and it was calculated based on the study of Gholami *et al* (11):

$$n = 2\frac{\left(z_{1-\frac{\alpha}{2}}+z_{1-\beta}\right)^2\left(\sigma_1^2+\sigma_2^2\right)}{\left(\mu_1-\mu_2\right)^2}$$

Teeth with caries, fractures, cracks, abrasions, or restorations were excluded (11). Soft and firm tissues were removed with a scaler then teeth were brushed with fluoride-free pumice powder (Kimia Medium, Iran). Then, using a high-speed diamond disk and cooling water, the 12 teeth were first sectioned laterally with a 3 mm thickness, and then mounted in plaster.

In Figure 1, 24 samples were created by transversally dividing each specimen into two buccal and lingual halves.

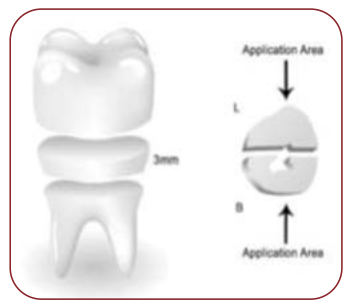

FIGURE 1. Cutting for tooth preparation

The enamel and cement were then removed from the tooth surface using a high-speed carbide bur with cooling water. The teeth were kept in distilled water until the day of examination, which lasted between one day and two days. Samples were then cleaned with EDTA (Trissodico Biodimanica, Portugal) solution for two minutes to remove smear layer on the dentin tubules (12). Samples were divided into four groups: (A) control group, (B) varnish group, (C) varnish and diode laser group, and (D) varnish and Er,Cr:YSGG laser group. In group B, after drying the samples, 5% sodium fluoride varnish (FlouroDose, Centrix, USA) was applied on the surface using a micro-brush for 15 seconds. Similar to the prior approach, varnish was applied to both group C, before being exposed to diode laser radiation, and group D, before being exposed to Erbium laser radiation. Laser irradiation was performed as follows:

- Diode laser [Doctor Smile dental laser (Simpler), Italy], wavelength 980 nm, power 1.5 W, irradiation mode: continuous, power density of 21.23 W/cm², time: 30 seconds, fiber tip: 300 µm, non-contact mode, distance: 1 mm, fiber reciprocating movement (11);

- Er,Cr:YSGG laser (Waterlase, Biolase Technology, Irvine, CA): wavelength: 2780 nm, irradiation mode: free-running pulse, power 0.25 W, F 20 Hz, pulse width 140 µs, time: 30 seconds, fiber tip: 600 µm, non-contact mode, distance: 1 mm, power density of 88.46 W/cm² with 0% water and 0% air, fiber reciprocating and scanning movement, perpendicular to the dentin surface (13).

Until SEM analysis, samples were kept in distilled water. The dried samples were then mounted on a SEM (Electrolux, Iran). Afterwards, samples were coated with gold using a coater sputter (Polaris, South Korea) device. The gold-plated specimens were examined by scanning electronic microscope (SEM) (SNE 4500M, South Korea) at the central laboratory of Noshirvani University of Technology in Babol, Mazandaran, Iran. The photomicrograph was taken from the central region of each sample with a 2000x magnification. Images revealed dentin tubules that were completely clogged, partially obstructed, and open. An examiner who was unaware of the study groups was able to determine the total number of open tubules and the average diameter of open tubules in the photomicrograph by utilizing scale-bar software. Software called SPSS version 23 was used to examine the data. Central indices (average), and dispersion indices (standard deviation) were used to describe data. Kruskal-Wallis and Mann-Whitney tests were used to compare the average diameter of the tubules. The post-hoc Tukey test was used to compare the number of open tubules. ❑

RESULTS

Twenty-four samples were evaluated by SEM in all four groups. The normal data distribution was evaluated using Shapiro-Wilk test and homogeneity was evaluated with Levene's test. With a 2000x magnification, Table 1 shows the abundant distribution of open tubules, and Table 2 displays the relationship between them using a post-hoc Tukey test. Table 3 summarizes the average diameters of open tubules in µm. Open tubules in different groups can be seen in Figure 2 to 5. Table 4 shows a comparison among mean ratios of open tubular diameters by Mann-Whitney test. ❑

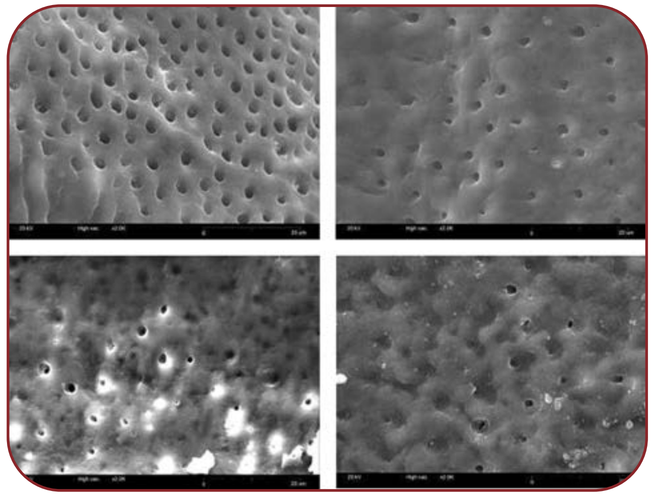

FIGURE 2. View of open tubules in groups A (control) (2), B (3), C (4), and D (5)

TABLE 1. Average number of open tubules based on group type with 2000x magnification

Group	Number of open tubules	Mean number of open tubules
A	354	59
B	235	39.1
C	178	29.6
D	171	28.5

Inter-group comparison	p-value
A/B	0.69
A/C	0.04*
A/D	0.03*
B/C	0.594
B/D	0.502
C/D	0.999

TABLE 2. Relationship between the average number of open tubules in each group

Group	Mean	Standard deviation	p-value
A	1.97	0.6	<0.001
B	1.4	0.5	
C	1.13	0.6	
D	0.73	0.4	

TABLE 3. Average open tubule diameter in µm based on group type

Inter-group comparison	p-value
A/B	>0.001*
A/C	>0.001*
A/D	>0.001*
B/C	>0.001*
B/D	>0.001*
C/D	>0.001*

TABLE 4. Comparison of mean ratios of open tubular diameters by Mann-Whitney test

DISCUSSION

The samples were evaluated by SEM in groups A (control), B (varnish), C (varnish + diode laser), and D (varnish + Er,Cr:YSGG laser). As the use of erbium laser on hard tissues had been approved by the FDA, it was chosen by us too. Furthermore, studies show that the ratio of calcium, potassium, magnesium, sodium, or phosphorus in the dentin structure is unaffected by erbium laser irradiation. Dentine tubule blockage may be achieved by exposing tissue to varying energy of the 1480 nm diode laser; however, 810 nm diode laser has a minimal impact. Moreover, 980 nm diode laser was selected in terms of its availability (15). Calcium-phosphorus deposition, such as calcium fluoride and fluorapatite, shows reduction in the movement of the tubular fluid (7). Therefore, fluoride varnish was chosen for the present study. Based on our findings, the average diameter of open tubules varied between 0.1–4.5 µm and it was significantly reduced in the four groups as follows: A > B > C > D. The average diameter of the tubules in the control group was 1.97 µm, which was in accordance with findings from other studies (16). Differences in average number of open tubules were only statistically significant among groups A, C, and A, D. Our results showed that a large number of dentin tubules became entirely or partially closed, and combination of fluoride varnish with both lasers was successfully used in tubular obstruction.

The rate of reduction in the diameter of tubules in our study was higher than that reported by Gholami *et al* (11), who evaluated the effect of lasers alone; they compared the occluding effect of 810 nm diode laser (P: 2 W, Pd: 30 ms) with that of Er;Cr:YSGG laser (P 0.25 W, F 20 Hz, Pd 140 mS). This difference may be due to the beneficial impact of varnish on tubular diameter reduction. Similar to our finding, Gholami *et al* found that the Er;Cr:YSGG laser significantly reduced the mean tubule diameter, with 810 nm diode laser sealing tubules to a much smaller extent. It is somewhat similar to and somewhat different from our result, because the exact diode laser wavelength used in our study was different from Gholami's study, 980 nm *versus* 810 nm. Furthermore, adding fluoride to the 980 nm laser improved the lase occluding effect.

Cakar *et al* compared the occluding effect of topical sodium fluoride gel 2% alone, the irradiation of Er:YAG (30 Hz, 60 mJ, for 10 seconds) alone, CO_2 (1 W, continuous-wave mode, for 10 sec) alone and gel, and laser combination. Dentinal tubule counts and diameters did not significantly vary between the laser-alone and combination groups, but they did differ when the control and gel groups were compared to the laser-alone or combination groups and each other (17). Regarding the superiority of combination of lasers and gel, results are similar to our study. We have also applied fluoride varnish, but Er;Cr:YSGG was evaluated, and despite this slight difference, the result was similar.

Jayaram *et al* compared the effect of dentinal tubule occlusion provided by 810 nm diode laser irradiation (0.25 W, CW) with 1.23% acidulated phosphate fluoride gel application and their combination (18). They used a low-power laser (0.25 W) and a different diode (810 nm *vs.* 890 nm), but their results were comparable to ours; also, in their study, phosphate fluoride gel was applied together with a low-level, diode 810 nm laser that was operated at a reduced power. On the other hand, Gholami *et al* (11) used diode 810 nm with a higher power, but not associated with a fluoride agent. It means that there is a synergism effect between any kind of fluoride agent (phosphate fluoride, sodium fluoride, etc) and laser irradiation (regardless of the parameters) on dentin tubule occlusion.

The aim of our in vitro study was only to investigate the occluding effect of varnish and laser on dentinal tubules, while the effect of laser on nerves was not considered. ❑

CONCLUSION

Based on the results of our study, combination of fluoride varnish with Er:Cr:YSGG laser irradiation or diode laser irradiation can partially or entirely occlude dentine tubules. Moreover, these methods are recommended for dentin hypersensitivity treatment. ❑

Conflicts of interest: none declared.
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