

## A Morphological *in Vitro* Study of the Effects of Nd:YAG Laser on Irradiated Cervical Dentin

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### ABSTRACT

**Objective:** The purpose of this *in vitro* study was to verify the efficacy of Nd:YAG pulsed laser at 1064 nm in sealing dentinal tubules *in vitro*, with its resulting morphological changes using clinical parameters applicable to the treatment of dentin hypersensitivity. **Background Data:** Although many papers have been written on the subject, no rapid, efficient, and long-lasting treatment for this problem has been developed. **Materials and Methods:** Twenty healthy extracted human teeth were kept in saline solution. Cervical cement was removed with a no. 57FG bur to expose approximately 4 mm<sup>2</sup> of dentin. Two groups were in the buccal aspect (A and B—irradiated) and one group in the lingual aspect (C—control not irradiated) of the same tooth. Samples with a remaining dentinal thickness between 1 and 1.5 mm were selected. Irradiation parameters were 30 mJ, 0.3 W (Group A) and 40 mJ, 0.4 W (Group B); 7 Hz was used in both groups in two 43-sec applications each, with a 10-sec interval. In 10 samples, the dentinal surface underwent SEM examination for study. The other 10 remaining irradiated samples were centrally cleaved to study laser penetration depth in dentin (SEM). **Results:** Obliteration of dentinal tubule openings and solidification of the dentin surface were observed in all irradiated samples. Laser penetration depth in dentin (SEM) varied from 1 to 7 μm, depending on irradiation parameters used in each group. **Conclusion:** We concluded that the use of the Nd:YAG laser was a very effective measure for obliterating dentinal tubule openings.

### INTRODUCTION

THE MECHANISM OF PAIN TRANSMISSION in dentin hypersensitivity is still not completely understood. With the exposure of dentin surface, a connection between the oral cavity and pulp appears through the openings of the dentinal tubules. According to Brännström,<sup>1</sup> fluid crosses dentinal tubules in a bidirectional movement, acting like a transport means for mechanical, thermal and chemical irritants.<sup>2</sup>

Both dentin permeability and hypersensitivity are reduced when dentinal tubules are obliterated.<sup>3–8</sup>

Microscopic examinations following extraction of teeth with hypersensitive dentin by Absi et al.<sup>9</sup> have revealed that tubular diameters are increased twofold (0.9 μm) when compared to those in non-sensitive dentin.<sup>10</sup> The average diameter of the tubules is of great importance, as according to Poiseuille's law,

doubling the diameter can per se result in a 16-fold increase in fluid flow. Similar evidence for surface characteristics of hypersensitive dentin was obtained *in vivo* using the replication technique.<sup>11</sup> This technique in essence offers the first objective method for monitoring the potential effects of etiological agents and therapeutic components.

Based on these theories and on clinical, microscopic, and laboratory observations, dentinal tubule sealing agents<sup>12</sup> that reduce their permeability have been developed, as suggested by Pashley.<sup>4</sup>

In this context, the laser is used as a coadjutant to hypersensitive dentin therapy,<sup>13,14</sup> or as an essential and unique technique in the treatment of this condition.<sup>15</sup>

The objectives of this study were to assess the efficacy of using Nd:YAG laser to seal dentinal tubule orifices *in vitro* and to verify dentinal morphological changes identified by clinical

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parameters suggested and used by several authors<sup>2,6,15,16</sup> for the treatment of dentin hypersensitivity.

## MATERIALS AND METHODS

Twenty healthy freshly extracted human third molars from patients ranging in age from 18 to 25 years, which had been stored in physiological saline in order to remain hydrated, were used.<sup>17</sup> Initially, teeth were subjected to washing, debris removal, scaling and polishing with a rubber cup, Robson's brush and pumice. Teeth with a wider cervical body in the mesio-distal and occluso-apical direction were selected for the preparation of three groups. Cross-sectional sections were made 1 mm from the cemento-enamel junction, at the cervical portion of the dental crown, and at the cervical third of the root.

Cervical cement was removed with a no. 57 bur to expose approximately 4 mm<sup>2</sup> of dentin. This procedure was performed in three areas of the same tooth, two on the buccal aspect (groups A and B—irradiated) and one on the lingual aspect (group C—control, not irradiated).

All the samples were split in half in a mesio-distal direction, so that the thickness of the remaining dentin (TRD) could be measured with the use of a thickness gauge.<sup>18</sup> According to White et al.,<sup>19</sup> this is a variable parameter of great importance to determine pulp temperature, and consequently, pulp vitality. The sections were kept in vials containing saline, until laser application. In order to standardize TRD, samples with remaining dentin thickness between 1.0 and 1.5 mm were selected, as this variable changes according to pulp volume and anatomy. The samples were then numbered for identification and etched with EDTA (pH 7) for 2 min, to remove the smear layer; finally, they were washed in 10 mL of physiological saline.<sup>20</sup>

The Nd:YAG laser was chosen because, according to the literature,<sup>19,21–23</sup> it is the method that has demonstrated greater effectiveness and caused the least number of undesirable effects to teeth (cracks and pulp damage due to heating) when used with adequate parameters.

### Specifications of the laser equipment used

Laser delivery system = quartz fiber  
 Fiber diameter = 320 µm  
 Wavelength  $\lambda$  = 1064 nm  
 Waveguide He-Ne,  $\lambda$  = 632 nm  
 Temporal pulse width = 450 µsec  
 Fiber area = 0.08 mm<sup>2</sup>

### Irradiation characteristics

Laser beam diameter  $\cong$  320 µm  
 Application form: a light contact with a continuous mesio-distal and occluso apical scan  
 Direction: perpendicular to the surface  
 Emission type: focused pulsed mode

### Parameters and irradiation conditions

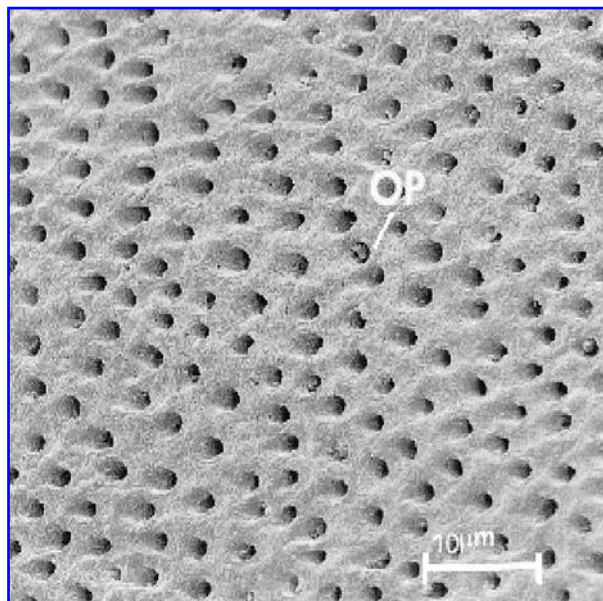
The 20 samples were irradiated according to the parameters previously mentioned. Ten samples were kept intact to

assess sealing of dentinal tubule openings, and 10 were cleaved in order to assess laser penetration depth into dentin. Cryofracture was used to cleave some samples which had previously been frozen in liquid nitrogen.<sup>22,24</sup> The remaining samples were cleaved using the oriented fracture technique, usually employed in surgery with dental sectioning. Dental sectioning was accomplished by placing two grooves next to the irradiated area, without contacting it, and using a double-faced diamond disk attached to a handpiece. Next, a chisel was inserted into one of the grooves and lateral lever movements caused fragment separation. All Group A, B, and C samples were then dehydrated in an ascending alcohol sequence of 70%, 80%, 90%, and 100%, 10 min in each concentration, and left to dry.<sup>25</sup> Finally samples were glued to stubs and metallized in gold 99.9% vacuum purity. Metallization was done for 100 sec, resulting in an approximately 25-nm gold film.

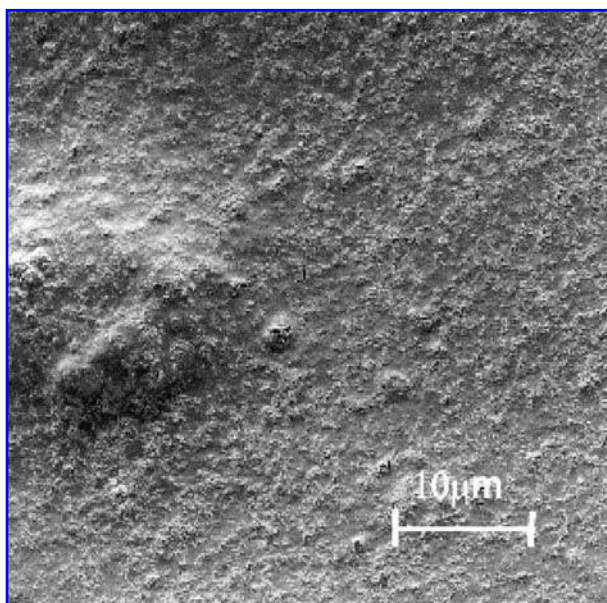
## RESULTS AND DISCUSSION

Evaluation was carried out using Scanning Electron Microscopy to assess the obliterating effect of pulsed Nd:YAG laser on human extracted teeth cervical dentinal tubule openings following sample preparation, comparing different energy densities in groups A and B (Table 2). Initially Group C, which was not irradiated, was assessed with open dentinal tubules being observed (Fig. 1).

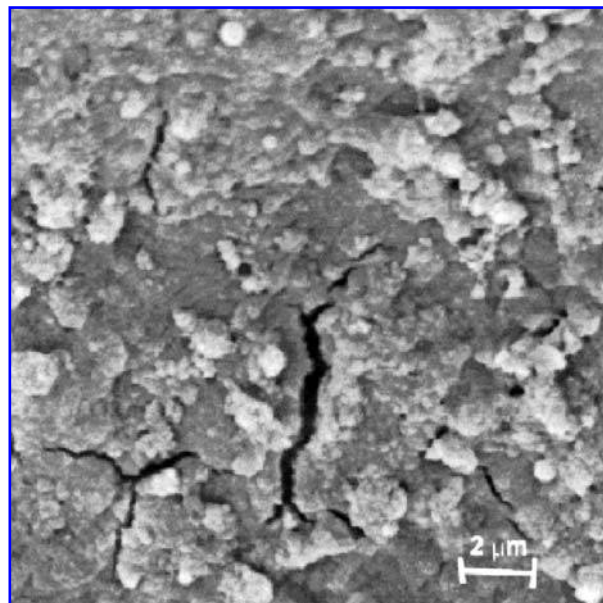
In groups A and B, dentinal tubule sealing was demonstrated by means of Nd:YAG laser irradiation with melting of dentin, a result also reported by Lan and Liu<sup>16</sup> (*in vitro* study) and Liu et al.<sup>22</sup> (*in vitro*) (Figs. 1–3).



**FIG. 1.** Group C, control, showing open dentinal tubules before irradiation of the Nd:YAG and odontoplastic process (OP).



**FIG. 2.** Group A showing dentinal tubules sealing with an energy of 30 mJ, 7 Hz, and 0.21 W.



**FIG. 4.** Group B with a higher magnification for crack and recrystallization granule visualization (10 kV, 11 mm, 2 μm).

There were morphological changes in irradiated dentin both in groups A and B, with dentin fusion, recrystallization, and dentinal tubule obliteration.

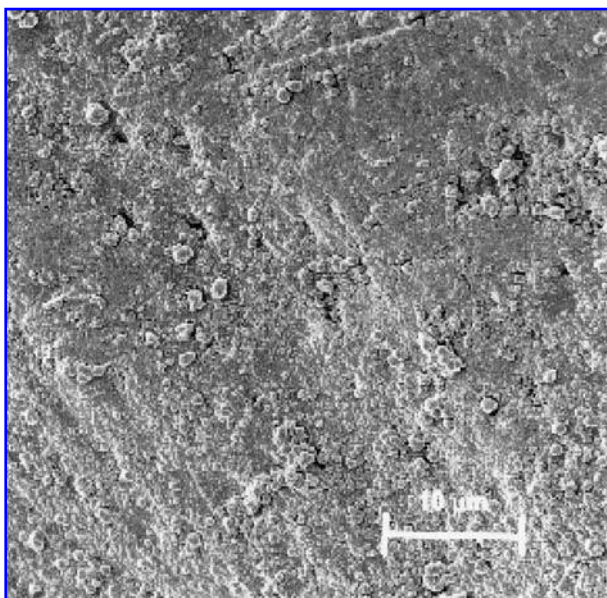
Cracks occurred more frequently in group B and a large amount of recrystallization granules and the presence of fused dentin debris was demonstrated. Higher energy (40 mJ) could add to dentinal hypersensitivity relapse because dentin would again become permeable (Fig. 4). In group A the irradiated sur-

faces seemed to be smoother and homogeneous, showing less dentin recrystallization granules than in group B.

Zach and Cohen<sup>26</sup> performed a study on pulp response to external application of heat; results have demonstrated that a few healthy pulps had not recovered from a temperature rise above 5.5°C. The lowest parameters found in the literature were selected to avoid teeth overheating and to remain within safety limits for *in vivo* use (Table 1), as shown below.

White et al.<sup>19</sup> discussed the importance of considering the remaining dentin thickness because of the possibility of heat caused by irradiation leading to irreversible pulp injuries.

Monitoring of temperature rises was not done in this study, as it was based on the studies by White et al.,<sup>19</sup> who studied temperature rises as a function of time and the thickness of remaining dentin. These authors emphasized that the operator should select the parameters within safety limits, as it would not be possible to measure the *in vivo* remaining dentin thick-



**FIG. 3.** Group B showing dentinal tubules sealing with dentin recrystallization granules, energy of 40 mJ, 7 Hz, and 0.28 W (10 kV, 18 mm, 10 μm).

TABLE 1. PARAMETERS USED FOR IRRADIATION

Groups	Parameters			
	Energy <sup>a</sup>	Repetition rate	Average power	Exposure time <sup>b</sup>
A	30	7	0.21	2 × 43 <sup>c</sup>
B	40	7	0.28	2 × 43 <sup>c</sup>

<sup>a</sup>The energy was measured at the fiber output, after the losses, using a commercial energy meter. (Coherente-Fieldmaster, Model FM).

<sup>b</sup>The time of 43 s was used to obtain the same total energy clinically used (GUTKNECHT et al., 1997), because the laser used has the maximum frequency of 7 Hz.

<sup>c</sup>An interval of 10 s between the applications was provided (time for thermal relaxation of the lased tissue).

TABLE 2. IRRADIATION CONDITIONS

<i>Irradiation conditions</i>	<i>Formula</i>	<i>Group A</i>	<i>Group B</i>
Average power density by application (intensity)	$\frac{\text{Average } P}{\text{total irradiated area}}$	5.25 W/cm <sup>2</sup>	7.0 W/cm <sup>2</sup>
Average power density by application (dose)	$\frac{\text{Average } P \times \text{exposure } t}{\text{total irradiation area}}$	226 J/cm <sup>2</sup>	301 J/cm <sup>2</sup>
Average individual energy density of each pulse	$\frac{E_{\text{pulse}}}{\text{Fiber area}}$	37.5 J/cm <sup>2</sup>	50 J/cm <sup>2</sup>
Individual peak power of each pulse	$\frac{E_{\text{pulse}}}{\text{temporal pulse width}}$	66.7 W	88.9 W

t = time, E = energy, P = power.

ness. Energy level should also be set according to the total size of the tooth area subjected to laser irradiation and the area to be treated. They also stated that the effect of *in vivo* intrapulp temperature rises could be reduced by the presence of the periodontal ligament, bone and pulp blood flow. Similar results were also reported by Zezell et al.<sup>27</sup>

Lan and Liu<sup>16</sup> carried out an *in vitro* study using the Nd:YAG laser with different power settings, irradiating exposed dentin and assessing the effects of occlusion on dentinal tubules. Their aim was to find the most adequate parameters for the treatment of dentinal hypersensitivity. Our findings were similar to those of Lan and Liu,<sup>16</sup> according to the already mentioned parameters selected for sample irradiation (Table 1).

Yu et al.<sup>28</sup> studied the effects of pulp temperature rises following the use of Nd:YAG laser in the treatment of dental hard tissues and concluded that these thermal effects may be reduced by cooling the teeth with water and by moving the laser over the tooth.

An important point is that dentin should be homogeneously irradiated by laser to avoid flaws in dentin sealing, as hypersensitivity may persist if dentin is not irradiated over all its surface, which leaves areas with open tubules (Fig. 5).

A device could be designed to perform an automatic scan of the area to be irradiated to avoid the problem of irregular dentin irradiation. This may be the focus of future studies.

Nd:YAG laser dentin penetration depth was also studied in groups A and B. Figure 6 (Group A sample) shows a cross-

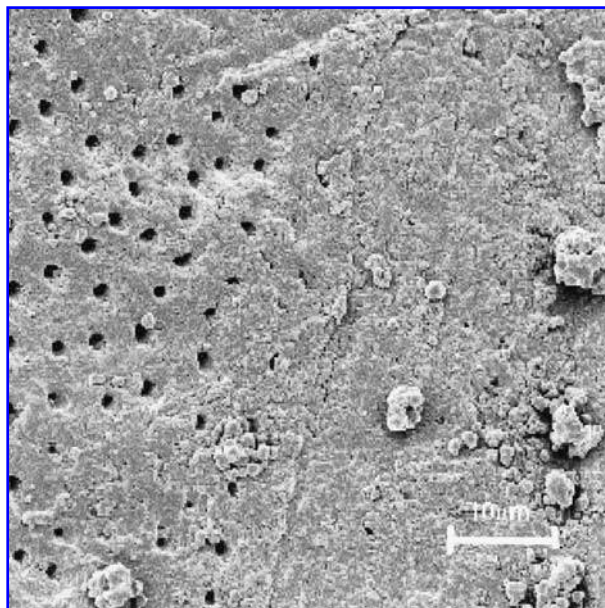


FIG. 5. Area of dentin partially irradiated.

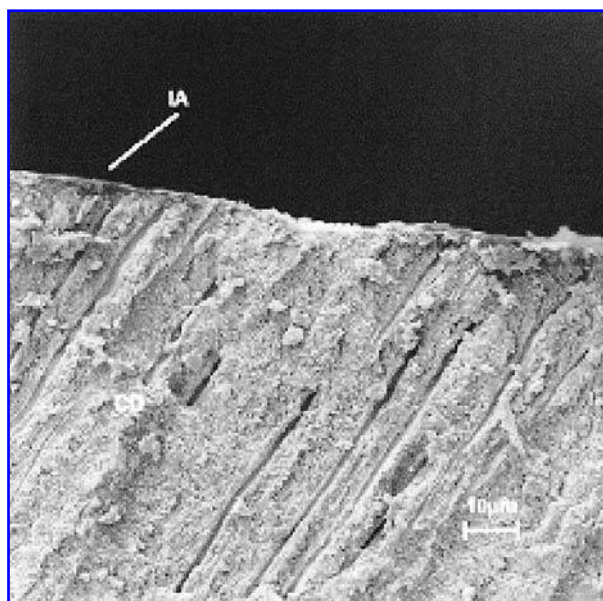
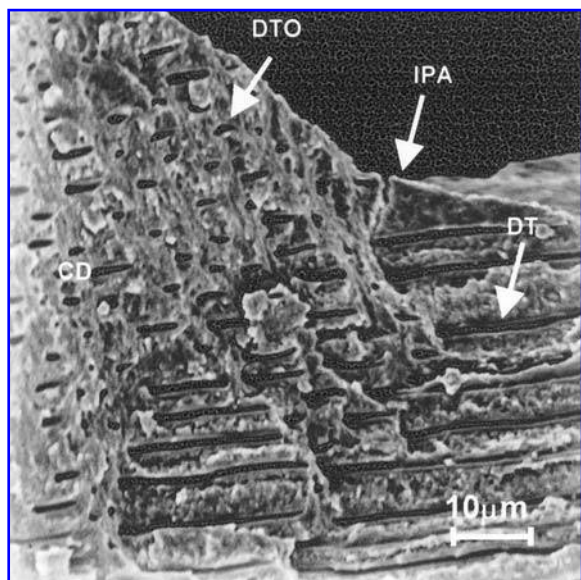
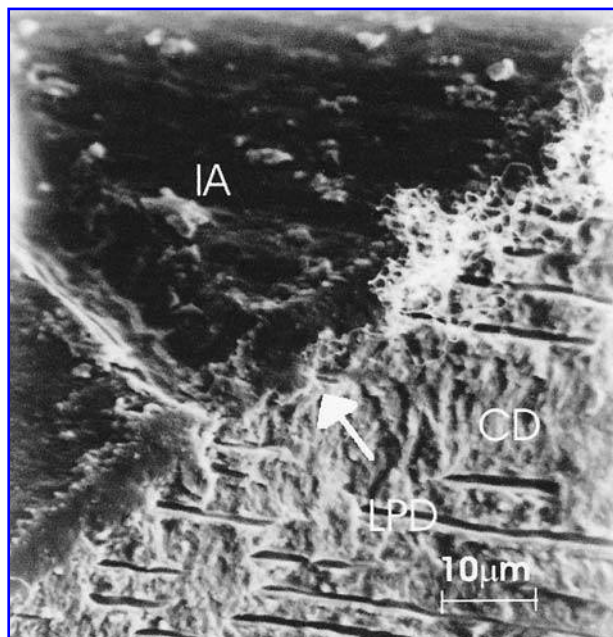


FIG. 6. Penetration depth of the Nd:YAG in the center of a sample in group A (10 kV, 11 mm, 10  $\mu$ m). IA = irradiated area, CD = cleaved dentin.



**FIG. 7.** Penetration depth of the laser on the periphery of the irradiated area (10 kV, 11 mm, 10  $\mu$ m). DTO = dental tubule opening, DT = dental tubule, IPA = irradiated peripheral area, CD = cleaved dentin.

sectional view of the floor of the cavity after application of the laser, showing a vitrified aspect. A penetration depth of approximately 1–3  $\mu$ m was demonstrated in this group which underwent lower irradiation energy (30 mJ; Table 1). Similar results were observed by Liu et al.<sup>22</sup>



**FIG. 8.** Penetration depth of the Nd:YAG laser in the irradiated surface central area in a sample of group B (10 kV, 11 mm, 10  $\mu$ m). IA = irradiated area, LPD = laser penetration depth, CD = cleaved dentin.

Laser penetration depth in the periphery of a sample in group B area, following laser application, is shown in Figure 7.

Figure 8 shows the penetration depth of Nd:YAG laser in the center of the irradiated area in different group B samples.

In group B, in which higher laser energy was used (40 mJ, Table 1), increased laser penetration depth up to 7  $\mu$ m was observed. This could be explained by the higher energy used and consequently a higher power level, with the other parameters in group A being kept constant.

## CONCLUSION

The Nd:YAG laser was effective in sealing dental tubules. The whole exposed dentin surface must be homogeneously irradiated to avoid areas left with open tubules. Laser penetration depth may vary from approximately 1 to 7  $\mu$ m, using, respectively, 30 and 40 mJ energy levels, with the other parameters being kept constant.

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