Effect of various laser irradiations on the mineral content of dentin

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ABSTRACT

Objective: The aim of this study was to evaluate the mineral content of dentin irradiation with Erbium: yttrium-aliminum-garnet (Er:YAG), Neodmiyum:yttrium-aliminum garnet (Nd:YAG) and potassium titanium phosphate (KTP) laser used for in the treatment of dentin hypersensitivity.

Methods: Six extracted wisdom, unerupted molar teeth were used in this study. The enamel of the teeth was removed with a conventional bur under water cooling to expose the dentin surface. The teeth were mounted in a slow-speed, diamond-saw, sectioning machine. Two dentin slabs were obtained from each tooth and each slab was sectioned so that 4 slabs were made from each teeth. Then dentin slabs were randomly divided into four groups. Group A: Control Group, Group B: Er:YAG laser, Group C: Nd:YAG laser, Group D: KTP laser. The levels of Ca, K, Mg, Na,P and Ca/P mineral ratio in each dentin slab were measured by inductively coupled plasma-atomic emission spectrometry (ICP-AES). Data were analysed by one way analysis of variance (ANOVA) and Tukey HSD tests. One sample from each group was prepared for scanning electron microscopy (SEM).

Results: There were no significant differences between the groups for Ca, K, Mg, Na,P and Ca/P mineral ratio ($P>.05$). SEM photographs indicated that there were melted areas around the exposed dentin tubules in groups treated with Er:YAG and KTP lasers.

Conclusion: This study demonstrated that laser etching with the Er:YAG, Nd:YAG, KTP laser systems did not affect the compositional structure of the dentin surfaces. (Eur J Dent 2013;7:74-80)

Key words: Ca/P ratio; laser irradiation; dentin hypersensitivity; ICP-AES, mineral content

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INTRODUCTION

Laser irradiation of dental hard tissue causes morphological and chemical changes. The extent of these changes is affected by the absorption characteristics of the tissue, and hence, the changes are likely to be varied according to the type of laser and dental tissue.[1]

Dentin hypersensitivity (DH) is characterized by short, sharp pain arising from exposed dentine in response to stimuli, typically thermal, evaporative, tactile, osmotic, or chemical, which cannot be ascribed to any other form of dental defect or pathology.[2] This sensitivity is also characterized by an exaggerated response to a sensory stimulus that usually produces no response in a normal, healthy tooth. DH causes chronic irritation that affects eating, drinking and breathing.[3]

Most of the DH treatments aim to block exposed dentin tubules; however, none of these treatments have produced consistently effective or long-lasting results.[4] To date, most of the therapies have failed to satisfy the patients; however, some authors have reported that laser irradiation may now provide reliable and reproducible treatment.[5,6]

Laser technology has gained popularity over the recent years, and many applications of laser technology in dentistry and medicine have been proposed. The first use of laser for the treatment of DH was reported by Matsumoto et al[5] by using Nd:YAG laser. Laser therapy has also been recommended by Kimura et al[7] to treat DH and is reportedly effective in 5.2% and %100. Er:YAG laser was the first laser approved by the Food and Drug Administration (in 1997) for application on dental hard tissue.[8] This laser emits light at 2.94 μm, and is strongly absorbed by water and less absorbed by hydroxyapatite, thus enabling the enamel cutting by the ablation process, which involves absorption of the laser energy by water droplets contents in the enamel; this results in the water micro-expansion an ejection of the hard tissue.[9] Treatment of DH by Er:YAG laser is highly effective in reducing the diameters of dentin tubules under specific conditions and also partially obliterates the tubules below the ablation threshold.[10]

The other laser treatment for DH involves the use of Nd:YAG laser. It has been suggested that the effect of Nd:YAG laser on DH is related to the laser-induced occlusion or narrowing of the dentin tubules.[11] Direct nerve analgesia[12] and suppressive effect achieved by blocking the depolarization of Ad and C fibers[13] are also considered to be the possible mechanisms by which Nd:YAG laser irradiation reduces DH.

Potassium-titanyl-phosphate (KTP) laser, a type of Nd:YAG laser, is absorbed well by hemoglobin and melanin[14] but not by hydroxyapatite or water.[15] This laser does not appreciably increase temperature. Its photons have high energy that facilitate chemical and photodynamic reactions without damaging either the hard or pulp tissues.[16] Like the Nd:YAG laser, KTP laser has also been used in other dental applications such as root canal disinfection, DH treatment and soft tissue surgery.[17]

Dentin composition is determined based on its organic and inorganic components. Calcium (Ca) and phosphorus (P) present in hydroxyapatite crystals are the major inorganic components of the dental hard tissue. The calcium-to-phosphorus mineral ratio (Ca/P ratio) of hydroxyapatite in dentin, which implies the basic composition of the dental hard tissue surfaces, was found to be ~1.67, depending on the crystal type, availability of Ca, anatomical location, and the technique of determination.[18] It has been reported that some chemical agents cause alterations in the chemical structure of human dentin and change the Ca/P mineral ratio of the dentin surface.[18]

In contrast, the dentin surface produced by laser etching is acid resistant. Laser radiation of the dental hard tissues modifies the Ca/P mineral ratio, reduces the carbonate-to-phosphate ratio, and leads to the formation of more stable and less acid-soluble compounds, thus reducing susceptibility to acid attack and caries.[19] Alterations in the Ca/P mineral ratio can change the original ratio between organic and inorganic components which in turn changes the permeability and solubility characteristics of dentin and affects the adhesion of dental materials to hard tissues[18]; however limited studies have been performed on how lasers change the elemental composition. Moreover, small amounts of magnesium are always detectable in dentin in addition to calcium and phosphorus. Magnesium is considered to influence the mineralization process, especially crystal growth.[20]

Inductively coupled plasma technique (ICP-AES) uses a sample that is passed through argon in a ray fluorescence (RF) field. When the sample is introduced into the plasma, the atoms are excited and emit very stable light of varying wavelengths that permits the identification of the elements. This technique has become highly popular for element analysis.[21] The null hypothesis tested was that there would be differences in the compositional changes (Mg, P, Ca, K and Na) of dentin surfaces prepared using Er:YAG, Nd:YAG and KTP laser irradiation for the treatment of DH. The second aim of this study was to compare the Ca/P mineral ratios of the groups using the ICP-AES techique.

MATERIAL AND METHODS

Preparation of the dentin slabs

Six, unerupted molar (wisdom) teeth, free of dental caries or restoration were cleaned with gauze and a fine brush and stored in distilled water at room temperature immediately after extraction. The teeth were then mounted on quadrangular molds with an autopolymerizing acrylic resin (Meliodent, Bayer Dental Ltd., Newbury, UK). The enamel of the teeth was removed using a conventional bur under water cooling to expose the superficial dentin surface. Occlusal thirds of the crowns were cut with a slow-speed diamond-saw, sectioning machine (Isomet, Buehler Ltd., Lake Bluff, IL, USA) under water cooling. To prepare the dentin slabs of 0.60-mm thickness, cuts were made perpendicular to the long axis of the tooth. Four dentin slabs were obtained from the same tooth. Then dentin slabs were randomly divided into 4 groups (n=6 per group). The treatment groups were as follows:

Group A (Control): The specimens in this group were prepared using 19% ethylenediaminetetraacetic (EDTA) gel (MD-Chel Cream, Meta Biomed Co., Ltd., Mandaluyong, Korea) for removing the smear layer and similating the hypersensitive tooth. The specimens were then washed with distilled water and dried.

Group B (Er:YAG group): After the EDTA gel had been applied to dentin, the surfaces were manually irradiated to simulate clinical conditions. Surfaces were irradiated with an Er:YAG laser (Smart 2940 d plus, Deka, Calenzana, Italy) using scanning movements perpendicular to the surface, in the defocus mode 60 s/cm^2, 10 Hz, 100 mJ and 60 s with air and water cooling. Irradiation was conducted twice, in long pulse (LP) mode using an R14 handpiece.[22,23] The distance between the fiber and tooth was about 1 mm in the perpendicular position.

Group C (Nd:YAG group): The Nd:YAG laser device (Smarty A 10, Deka, Calenzano, Italy) had a 300-µm quartz fiber optic delivery system. After EDTA gel had been applied to the dentin, the surfaceswere manually irradiated using scanning movements at 60 s/cm^2, 25 Hz, 1 W and 60s. This was conducted twice without cooling and in a very short pulse (VSP) mode (100 µs). The distance between the fiber and tooth was about 1 mm in the perpendicular position.[23,24]

Group D (KTP group): After the EDTA gel had been applied to the dentin; we manually irradiated the surfaces to simulate the clinical conditions. A KTP laser (Smartlite D, Deka, Calenzano, Italy) was used and conducted scanning movements (twice) perpendicular to the surface in continuous mode at 60 s/cm^2, 1 W, 119 j/cm^2 and 60s. The distance between the fiber and tooth was about 1 mm in the perpendicular position.

ICP-AES technique

The dentin slabs were dehydrated in plates at 70°C in a cabinet desiccator (Ventisell, Italy) until they reached a fixed weight. Their weights were recorded using an electronic balance (Electronic Balance AX200, Shimadzu Corporation, Japan). Five milliliters of nitric acid (HNO3) and 2 mL of hydrogen peroxide (H_2O_2) were added to the specimens, and the specimens burned at 180°C in a microwave (CEM, MarsXpress, USA) until they were dissolved. After calibration of the ICP-AES instrument (Vista AX, Varian, Australia), we took 2 mL of the solution. In this technique, the solutions are carried in a nebulizer with the help of a peristaltic pump. The specimens were aerosolized and carried in the form of an argon spray. The aerosols were heated by conduction and radiation to achieve a temperature of approximately 10,000°C, at which temperature they are completely atomized, and energy was released. The light was transferred to a detector, and every element was described according to its wavelength.[25] In this study, we performed 3 measurements of every element of each solution. The means of the measurements were calculated in milligrams per liter (parts per million) by using a computer. The levels of the 5 elements (Mg, P, Ca, K, and Na) in each specimen were measured using ICP-AES. The mineral contents were calculated as percentage weights.

Scanning Electron Microscopy Evaluation

An additional specimen from each group was prepared for SEM (Jeol JSM-5600; Jeol Ltd., Tokyo, Japan). After surface treatment, the specimens were sputter-coated (Polaron SC500 Sputter Coater, VG Microtech, E. Sussex, England) with gold-palladium alloy under high vacuum, and photomicrographs were obtained. The data were analyzed using 1-way analysis of variance (ANOVA) and Tukey's HSD tests. Statistical differences were determined at a 95 % confidence level (P=.05) All the statistical analyses were performed using the Statistical Package for Social Sciences, ver. 13.0 (SPSS, Chicago, IL).

RESULTS

The mean percentage weights of the 5 elements (Ca, K, Mg, Na, P) in the dentin after laser treatment are shown in Table 1. One-way ANOVA showed that there were no significant differences between the groups with respect to Ca, K, Mg, Na, P concentrations or Ca/P mineral ratio (P>.05). Mean percentage weights of Ca, P, Mg, and Na were found to be increased in Groups B, C, and D, but these differences were not significant (P>.05).

SEM images of control, Er:YAG, Nd:YAG, and KTP laser-irradiated dentin surfaces are shown in Figure 1. The surface texture of the dentin irradiated by Er:YAG laser was clearer than that of the dentin from other irradiated groups, and melted areas around the exposed dentin tubules are apparent.

There were some differences between SEM images of Nd:YAG and KTP laser-irradiated dentin surfaces and those of the control group. Recrystallization of the melted areas was more distinct in the treated surfaces. The laser-induced occlusion and narrowing of the dentin tubules can be easily seen on the images.

DISCUSSION

In this study, the compositional change in the dentin surfaces irradiated by Er:YAG, Nd:YAG, and KTP lasers was evaluated, and the Ca/P mineral

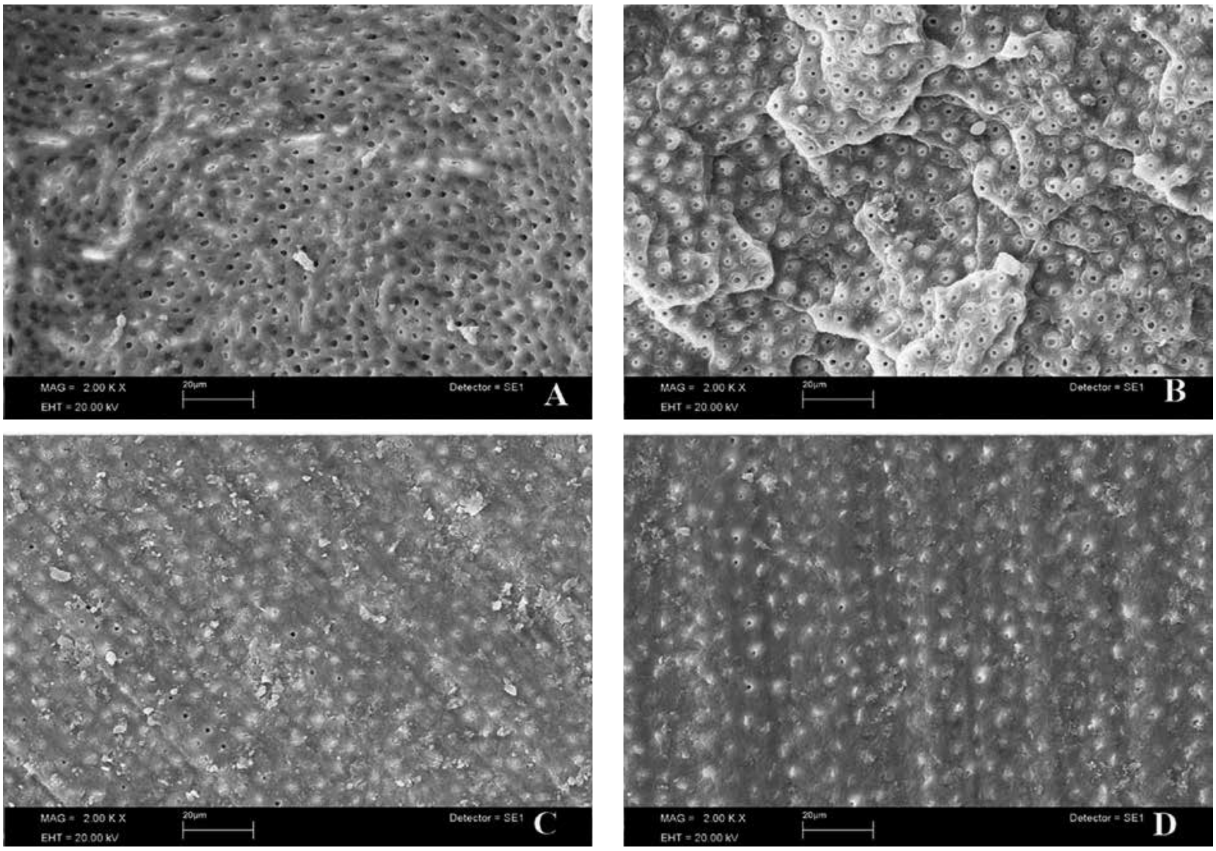

Figure 1. Scanning electron microscope (SEM) images of laser-irradiated dentin (x200). A.Control (19% ethylenediaminetetraacetic [EDTA] gel); B. Er-YAG; C. Nd-YAG; D. KTP

Table 1. Mean percentage weights of the five elements (Mean±SD) and Ca/P ratio for each group (n=6).

Groups	Ca	K	Mg	Na	P	Ca/P
Control	20,14±0,34	0,04±0,03	0,49±0,35	0,79±0,55	9,66±0,64	2,08±0,05
Er:YAG	26,51±0,03	0,005±0,00	0,70±0,09	1,09±0,19	12,79±0,37	2,07±0,04
Nd:YAG	26,08±0,13	0,04±0,01	0,63±0,04	0,95±0,03	12,25±0,13	2,12±0,07
KTP	25,36±0,13	0,05±0,01	0,67±0,08	0,91±0,09	11,88±0,79	2,13±0,05

ratio of the groups were compared using the ICP-AES technique. Mean percentage weights of Ca, K, Mg, Na, P and the Ca/P mineral ratio of the groups showed no significant differences among the 4 groups. Therefore, the null hypothesis was rejected.

Treatment of DH by using the Er:YAG laser is highly effective in reducing the diameters of dentin tubules under specific conditions, with partial obliteration of the tubules below the ablation threshold.[10] According to the hydrodynamic theory, the decrease of dentin fluid movements effect directly in the reduction of DH.[22]

In this study, the Er:YAG laser parameters-100mJ/pulse,[10] Hz, and defocus mode were lower than the ablation threshold of dentin, and we expected that this treatment would partially seal the tubule orifices and decrease the fluid movements by evaporating the superficial layers of the dentin fluid. This mechanism involves modification of the dentin tubular structure by melting and fusing the hard tissue or smear layer and subsequently sealing the dentin tubules. However, the mechanism of action of Er:YAG laser for the treatment of DH remains ambiguous and needs further study.

The other laser employed for occluding the dentin tubules is Nd:YAG. The mechanism of action of the Nd:YAG laser on dentin involves thermal energy absorption. Hydroxyapatite crystals melt in the presence of sufficient amount of energy leading to the closure of dentin tubules.[26] Because the interaction of laser and tooth surface liberates heat, investigations on the diffusion of heat across the dentin and its effect on pulp tissue are of particular importance. Given its ability to close or partially close the dentin tubules and decrease hydraulic conductance, the Nd:YAG laser would appear to have a potential to reduce root surface hypersensitivity without harming the dentin surface.[27]

Renton-Harper and Midda[28] conducted a clinical trial on 30 patients to evaluate the efficacy of Nd:YAG laser for reducing DH. The results indicated that application of Nd:YAG laser irradiation to sensitive teeth could significantly reduce the degree of sensitivity and alleviate this condition. The treatment was reportedly 90% effective, and the investigators concluded that the procedure could be performed easily and painlessly with a predictable response and considerable patient satisfaction. In the present study, the Nd:YAG laser parameters (25 Hz, 1 W, 60 s/cm^2, approximately 1 mm away from the dentin surface, 2 times, in VSP mode [100 µs]) were calibrated for this hypersensitivity.

ICP-AES is one for of the most attractive detection systems for determining the levels of trace elements in the dentin. With this technique, polishing is not necessary and dentin chips are sufficient for element detection.[18] The mineral content of dentin can be measured using SEM/EDS (SEM with X-ray microanalysis), with amounts detectable at parts per million (milligram per liter) level. However, with ICP-AES mineral content can be detected in parts per billion (micrograms per liter) level. In addition, multiple elements can be measured at the same time by ICP-AES. The measurements should be repeated for a second element in SEM and energy dispersive spectrometer.[18] We preferred the ICP-AES technique for this study because of the above mentioned advantages.

Rohanizadeh et al[1] evaluated the ultrastructural and compositional changes in dentin after irradiation with a short pulse laser (Q-switched Nd:YAG). The irradiated and non- irradiated areas of the lased dentin samples were investigated by SEM and transmission electron microscopy (TEM), micro electron diffraction and electron microscope analysis of dispersive energy (EDX). In the irradiated areas the Ca/P mineral ratio was lower than that in the non-irradiated areas. In the present study, the Ca/P mineral ratio was decreased in the tissue irradiated by Er:YAG laser, whereas it was increased in the tissue irradiated by Nd:YAG and KTP lasers.

Hossain et al[29] evaluated the compositional changes and knoop hardness of the cavity floor prepared using Er, Cr:YSGG laser irradiation and compared these with the compositional changes and knoop hardness of the cavity floor prepared using conventional bur and subjected them to atomic analysis by using SEM/EDX. Their results showed that the quantities of Ca (Ca weight %) and P (P weight %) were significantly increased in the laser cavity floor; no significant differences were found in the Ca /P mineral ratios between laser and bur cavities. Sazak et al[30] evaluated the effect of laser-etching by using Nd:YAG laser, air abrasion, and acid etching on the mineral content of dentin and enamel using SEM /EDS. They found that Nd:YAG laser-irradiated dentin had an apparently melted surface with partial obstruction of the dentin tubules, and showed cracks along the lased surfaces. Furthermore, laser irradiation caused changes in the mineral content of dentin in this study.

Lee et al[31] demonstrated that Er:YAG laser-irradiation with water spray does not significantly change the structure and composition of the dentin. Secilmis et al[25] observed that laser treatment at 2 and 3W did not significantly affect the mean percentage weight of any of the elements. In this study there were no significant differences in the Ca/P mineral ratios between the assessed groups.

CONCLUSION

In this study we demonstrated that laser irradiation of dentin surfaces using Er:YAG, Nd:YAG and KTP lasers did not affect the compositional structure of the dentin surface. The mean percentage weights of Ca, K, Mg, Na, P and Ca/P mineral ratios of the dentin in different groups were not affected by the various laser treatments.

CONFLICTS OF INTEREST

No conflicting financial interests exist.

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