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Abstract. The aim of this study was to use microchannels drilled by an Er:YAG laser into a human tooth through the enamel into the dentin for direct injection of hydrogen peroxide (HP) to produce a minimally invasive, rapid, tooth bleaching effect. The experiments were conducted *in vitro*. Five microchannels with a diameter of $\sim$200 μm and a depth of $\sim$2 mm were drilled through the palatal side of a human tooth crown using the microbeam of an Er:YAG-laser with a wavelength of 2.94 μm. After injection of an aqueous solution of 31%-HP through the microchannels, the tooth color was evaluated using a VITA shade guide and International Commission on Illumination $L*ab$ color parameters. A tooth model used for the evaluation of the distribution of HP concentration was created and the amount of HP which can be injected into tooth dentin to bleach it safely was estimated. Injection of 1.5 ± 0.1 mm^3 of 31%-HP into the tooth led to noticeable bleaching within 3 h and significant improvement of tooth color within 24 h. © *2016 Society of Photo-Optical Instrumentation Engineers (SPIE)* [DOI: 10.1117/1.JBO.21.12.125001]

Keywords: Er:YAG-laser; microchannels; tooth bleaching; color parameters; lightness.

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1 Introduction

Tooth color is determined by the spectral signature of endogenous or exogenous chromophores and their location within tooth structures. Two types of discoloration, extrinsic and intrinsic, can affect tooth appearance. The extrinsic is due to staining of the tooth surface by food products, such as tea, coffee, wine, tobacco, and so on. This staining can be effectively minimized by regular and correct home tooth brushing, or removed by professional cleaning in the dental office.

Intrinsic staining occurs as a result of natural pigmentation or blood and medical or chemical material penetrating into tooth dentin.[1–3] Discoloration also occurs due to the release of disintegrated by-products into the surrounding dentin from the dental pulp by bacterial, mechanical, or chemical irritants.[1]

Nathoo[2] has shown that intrinsic staining can be divided into pre-eruptive and posteruptive types. Endemic fluorosis, tetracycline stains, dentinogenesis imperfecta, amelogenesis imperfecta, and hematologic disorders are examples of pre-eruptive intrinsic staining. Posteruptive intrinsic staining tends to be due to factors such as pulpal hemorrhage, secondary and tertiary dentin deposition, pulp stones, metal release from amalgam restorations, and incomplete obturation of the pulp chamber.

Intrinsic staining is more difficult to treat and peroxide-based tooth bleaching [hydrogen peroxide (HP) or one of its precursors such as sodium perborate or carbamide peroxide, which are strong oxidants] is typically used.[1,4]

The mechanisms of peroxide-based tooth bleaching are not well understood. They differ according to the type of discoloration involved and the chemical and physical conditions at the site of the chemical reaction.[4] In general, these mechanisms include the oxidation of chromophores located within enamel and dentin and the degradation of the extracellular matrix. As a result of oxidation, the π-conjugated electronic systems of the chromophore molecules are broken, and their absorption bands are shifted from the visible to the UV region, resulting in a whiter appearance of the teeth.

Unfortunately, peroxide-based bleaching technology has a number of drawbacks, imposing serious limitations and concerns at application.[5–10] Raman and fluorescence spectroscopy has shown that compounds with a concentration of HP of about 30% may cause adverse effects to the mineral and organic matter of human tooth enamel, reducing enamel microhardness.[5] The drawbacks also include local effects on the oral mucosa including gingivitis,[6,7] erosion, and abrasion of enamel and dentin,[7,8] modification of dentinal permeability with posttreatment transient tooth sensitivity,[7,9] possible effects on pulpal tissues and dental restorations,[5,10] and resorption in the cervical area of the teeth.[5] Repeated treatment, which is usually required to reach the desired bleaching condition, adds to these adverse effects.[5]

To improve the efficacy of the oxidizer, to make the technology safer and faster, different physical and chemical enhancers for HP activation and better diffusion have been suggested and studied.[11–21] For example, teeth with an applied bleaching agent

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were subjected to local heating[11] or intensive white light and light-emitting diode, or laser.[11–18] Suemori et al.[16] and Calatayud et al.[18] have shown that effective bleaching of blood stained dentin was achieved using a chemical composition containing only 3.5%-HP and titanium dioxide photocatalytic nanoparticles at 405 nm-diode laser excitation.

A bleaching agent can also be applied externally to vital teeth—known as vital bleaching, or internally within the pulp chamber of nonvital teeth—known as nonvital bleaching.[22] The penetration of HP molecules when applied externally to tooth structures has not been well controlled until now, since[23] the effect largely depends on the individual aspects of the tooth. Ubaldini et al.[23] have shown that permeation is not merely a physical passage through enamel interprismatic spaces into the dentinal tubules, but that HP diffusion dynamics present a concentration gradient determined by the chemical affinity of the HP molecules to each specific dental tissue.

Altshuler et al.[24] found that the chemical interaction of HP with enamel is minor, whereas the interaction with dentin is noticeable. Intrinsic bleaching can be performed when the pulp is removed and a large access cavity is cut, to place a medium with HP for a prolonged exposure.[22] It is much more effective, because the HP molecules release active oxygen and other radicals in the immediate vicinity of the target chromogenic molecules. This technique cannot, however, be used for vital teeth.

The importance of the use of a high-quality Er:YAG laser beam for well controlled intrinsic bleaching of vital teeth was discussed by Altshuler et al.[24] and Belikov et al.[25] The aim of this paper is to prove the feasibility of tooth bleaching using an aqueous solution of HP via microchannels drilled by an Er:YAG laser through the enamel into the dentin in a manner which will not damage the pulp.

The concentration of HP that will not damage the pulp is described using the results of Bowles et al.,[26] who showed that a direct application of up to 2.5% HP when injected into pulpal extract does not inhibit its enzymes at normal body temperature. On this basis, it can be concluded that if the concentration of HP in dentin at the pulpal surface does not exceed 1% during the entire bleaching procedure, no adverse effects should occur.

This study was conducted on freshly extracted human teeth *in vitro* using an aqueous solution of 31%-HP. The bleaching process was allowed to continue for a period of 48 h and the color changes were determined using the VITA shade guide and International Commission on Illumination (CIE) $L*ab$ color parameter determination: L (lightness), a (redness), and b (yellowness). A tooth model for numerical simulation of 31%-HP aqueous solution diffusion into the tooth dentin through laser drilled microchannels within the tooth crown is presented.

2 Materials and Methods

2.1 In Vitro Experiment

The teeth used for the experiments were intact human maxillary central incisors, extracted as these teeth were to be replaced with implant supported crowns. The extracted teeth were then stored in an aqueous solution of 0.1%-thymol for no >4 weeks. The age of the patients was 45 to 50 years (ITMO, LTBMO IRB approval #04 at 23/04/2015). Immediately prior to treatment, the teeth were taken from the thymol solution, the crowns were cleaned using a mixture of abrasive pumice powder (medium coarse #3, Kerr Dental) and water for 60 s with a brush (RA Junior Cup White Bristle Martins) in a slow-speed drill (8000 rpm).

After cleaning, 1 to 2 mm of the apex of tooth root was sectioned using a diamond disc to facilitate fixation of the tooth into a stainless steel container, black in color. The tooth was then cemented into this container using a dental cement (Meron, Combipack, VOCO GmbH, Germany) and both tooth and container were placed into a 0.1%-thymol solution for a period of 5 to 10 min. The container with the tooth was taken from the thymol solution and fixed into a holder for photographing. The holder was designed so that the tooth was always kept in the same position to provide comparable photographs before and after tooth processing. The appropriately matching tooth from the shade guide (VITAPAN® classical, VITA Zahnfabrik H.Rauter Gmb&Co.KG, Germany) was placed next to the tooth under observation, into a similar holder.

A digital camera (Nikon D80) with a flash lamp (Sigma EM-140DG) was placed at a distance of 50 cm from the holder. The camera had an external power supply, remote control, and USB cable for online reading via a computer. All camera and flash settings (aperture, focus, exposure, white balance, ISO, and so on) were set manually for each photograph. A standard 18% gray card was placed in the field of view for all photographs for digital image calibration and verification as described by Bengel.[27]

The camera, flash lamp, tooth in its container, VITA shade guide tooth, and 18% gray card in its container were covered with a black velvet cloth to eliminate ambient light. They were photographed then the container with the tooth was removed from its holder, and laser drilling was carried out. Five microchannels with a diameter of ~200 μm and a depth of ~2 mm (see Fig. 1) were drilled through the palatal surface of the tooth crown using an Er:YAG laser with a wavelength of 2.94 μm and a beam propagation ratio of 1.5 ± 0.1. The beam propagation ratio indicates how close the laser beam is to being in the fundamental Gaussian mode, which allows for the smallest spot size and minimal beam divergence.[28]
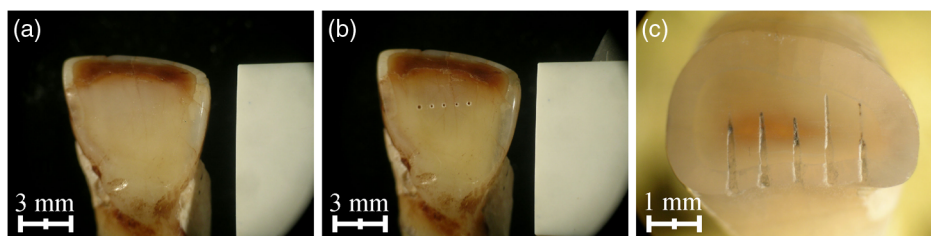
Fig. 1 Photographs of a tooth crown (a) before and (b) after laser drilling, and (c) a cross-sectional view of the drilled microchannels after tooth cut along channels.

The laser beam was focused onto the palatal side of the crown using a lens with a focal length of ~38 mm to drill the microchannel. The target area of the crown was irrigated with a drop of water of ~1 mm^3 immediately prior to laser drilling. Each microchannel was formed using M^2 laser technology,[29] with 80 laser pulses at a repetition rate of 1 Hz ($F = 1$ Hz). The free-running laser pulse duration of $\tau_p = 150 \pm 15$ μs, pulse energy of $E_p = 28.0 \pm 0.5$ mJ, and the beam diameter at e^{-2} intensity level of 120 ± 10 μm were used. The distance between the microchannel centers was ~500 μm.

After laser drilling, the container with the tooth was placed under a microscope at 16× magnification and 96%-ethyl alcohol was injected into the microchannels for 20 to 30 s using a syringe with a needle, with an external diameter of 110 μm (World Precision Instruments, Inc.), to clean the microchannels from any debris. They were then dried using air and each one was filled with 31%-HP heated to 40°C. A needle with an external diameter of 110 μm (World Precision Instruments, Inc.) was used to fill the microchannels with the HP. The needle was pushed to the bottom of microchannel so that the HP could displace the air trapped inside the channel and minimize the probability of air bubbles. A microscope was used to visually check that each microchannel was filled correctly, with injection ceasing once it was assessed that the microchannel was full. The total amount of HP at 31% concentration that was used to fill the five microchannels was 1.5 ± 0.1 mm^3.

The container with the tooth was then placed in a 0.1%-thymol solution for 1 min and fixed into the appropriate holder to take the photograph of the tooth, VITA shade guide, and gray card. Photographs were taken at 3, 24, and 48 h after 31%-HP injection. The containers with the teeth were stored in a 0.1%-thymol solution at a temperature of 37°C between photographs.

Tooth shade was evaluated using the VITA shade guide (VITAPAN® classical, VITA Zahnfabrik H. Rauter Gmb&Co. KG, Germany) prior to and at 3, 24, and 48 h after HP-injection. All images were processed using Adobe Photoshop CS6

following the method described by Bengel[27] and color CIE $L*ab$ color parameters: L (lightness), a (redness), and b (yellowness) were calculated for the VITA shade guide and teeth. The differential color parameters $\Delta(\text{CIE}\,L*ab)$ could then be calculated for the 3, 24, and 48 h time periods after HP-injection, to compare the difference in appearance between the commencement and the various time periods.

2.2 Statistical Analysis

Fifteen human maxillary central incisor teeth were used for the study. Five teeth which were not treated by laser and peroxide were used as the control group (C). Five teeth which were subjected to laser drilling only were used as the test1 group (Tx1). Five teeth processed with laser drilling and exposed to 31%-HP were used as a test2 group (Tx2). The software package StatGraphics Plus 2.1 (Statistical Graphics Corp.) was used for statistical data processing. The means and confidence interval for Δ of CIE $L*ab$ color parameters in the control (C) and test (Tx1 and Tx2) groups were calculated. The Kolmogorov–Smirnov test to estimate the statistical difference for Δ of CIE $L*ab$ color parameters between the control (C) and test (Tx1 and Tx2) groups, with p-value <0.05 was used.

3 Results

The typical digital images of the VITA shade guide tooth, 18% gray card, and treated (Tx2) tooth (sample #1Tx2) before and after injection of 31%-HP through the microchannels drilled by Er:YAG laser radiation at varying time intervals are shown in Fig. 2.

The results of the shade evaluation using the standard VITA shade guide are shown in Fig. 3.

The CIE $L*ab$ color parameters describing L (lightness), a (redness), and b (yellowness) of the tooth that were measured for the samples from the control (C) and test (Tx1 and Tx2) groups and presented in Table 1.
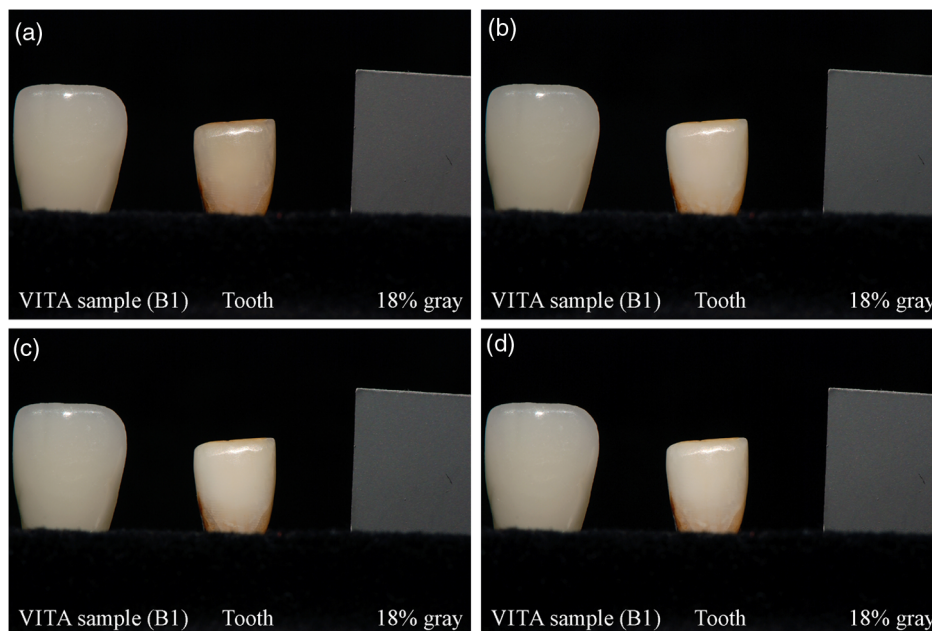
Fig. 2 Photographs of the VITA shade guide tooth, Tx tooth (sample #1Tx), and 18% gray card: (a) before, (b) at 3 h, (c) at 24 h, and (d) at 48 h after injection of 1.5 ± 0.1 mm^3 volume of 31%-HP through microchannels drilled by a 2.94-μm-Er:YAG laser.
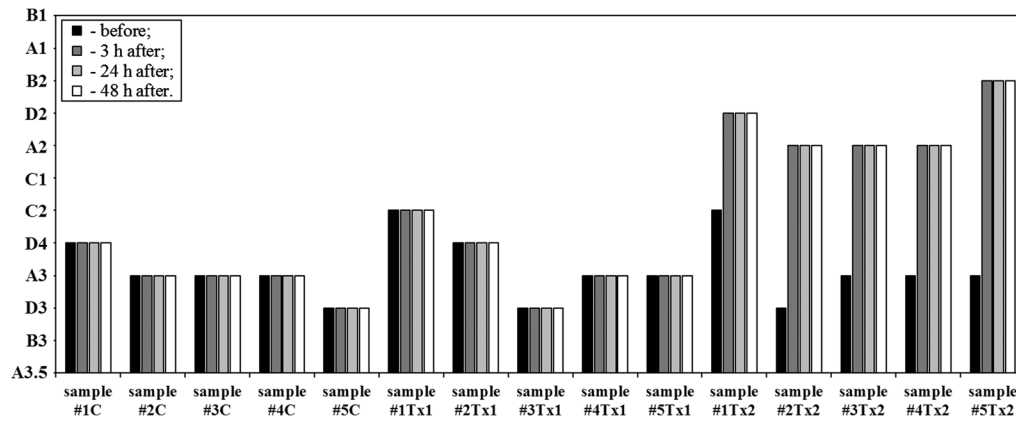
Fig. 3 Results of shade evaluation using the VITA shade guide for the control (C) and test teeth (Tx) samples before bleaching, at 3, 24, and 48 h after injection of 1.5 ± 0.1 mm^3 volume of 31%-HP through the microchannels drilled by Er:YAG laser radiation.

The differential color parameters $\Delta(\text{CIE } L * ab)$ for the samples from the control group (C) and test (Tx1 and Tx2) groups quantifying changes in the CIE $L * ab$ color parameters of the tooth before treatment and for the varying times after treatment are presented in Table 2.

4 Discussion

The VITA shade guide analysis presented in Fig. 3 shows that the VITA shade of the control group specimens (C) did not change. The VITA shade of the test1 group specimens (Tx1) did not change, i.e., drilling of microchannels into the tooth did not cause any color change. The VITA shade guide analysis shows that the appearance of the test2 tooth (Tx2) is significantly improved after HP-injection during the first 3 h and does not change for further observations during 24 and 48 h. There is an increase in the VITA shade in the range of 3 to 6 grades depending on the original shade of the tooth.

The CIE $L * ab$ color parameter analysis (see Tables 1 and 2) shows some minor variations for the controls (C) and for the test1 (Tx1) that are, however, within the limits of the

Table 1 CIE $L * ab$ color parameters for the samples from the control (C) and test (Tx1 and Tx2) groups before and 3, 24, and 48 h after HP-injection through the microchannels drilled by Er:YAG laser radiation.

Sample #	Before			3 h			24 h			48 h		
	L	a	b	L	a	b	L	a	b	L	a	b
1C	61.0	5.8	17.9	61.0	5.8	17.7	60.8	5.9	17.8	61.1	5.6	17.8
2C	64.5	3.9	19.1	63.9	3.9	19.5	63.9	3.7	19.6	63.2	3.8	19.7
3C	67.8	4.0	19.2	67.5	3.9	19.9	68.1	4.0	20.1	67.7	4.2	19.6
4C	68.8	4.4	20.6	68.5	4.9	20.5	68.9	4.1	20.8	68.3	4.5	20.3
5C	71.2	8.9	21.6	71.0	8.8	21.7	70.9	8.8	21.5	71.0	8.7	21.6
1Tx1	60.0	8.3	19.0	59.2	8.1	18.8	59.6	8.2	18.9	59.6	8.2	18.7
2Tx1	61.1	5.7	17.8	61.0	5.7	17.6	60.5	5.5	17.8	60.9	5.6	17.7
3Tx1	71.4	9.0	21.8	70.9	8.8	21.5	70.6	8.8	21.3	71.0	8.7	21.4
4Tx1	63.9	3.7	19.9	63.0	3.6	19.8	63.2	3.7	19.7	63.1	3.7	19.7
5Tx1	68.5	4.0	20.3	67.9	3.7	20.0	68.0	3.8	19.9	68.0	3.9	20.1
1Tx2	60.5	8.2	18.8	68.5	5.4	13.9	72.3	4.2	12.7	72.7	4.4	12.4
2Tx2	61.2	6.3	18.9	67.5	4.4	15.9	69.8	4.0	14.7	69.8	3.9	15.4
3Tx2	62.0	4.2	23.4	70.2	3.8	19.9	67.1	3.3	20.8	66.5	3.6	20.4
4Tx2	61.7	3.2	16.6	71.3	2.7	15.1	69.3	2.4	15.9	63.7	1.9	15.8
5Tx2	68.0	3.5	19.8	70.9	3.0	15.9	76.1	3.1	15.1	79.8	2.5	15.1

Table 2 Differential color parameters $\Delta(\text{CIE } L * ab)$ for the samples from the control (C) and test (Tx1 and Tx2) groups before and 3, 24, and 48 h after HP-injection through the microchannels drilled by Er:YAG laser radiation.

Sample #	Before			3 h			24 h			48 h		
	ΔL	Δa	Δb	ΔL	Δa	Δb	ΔL	Δa	Δb	ΔL	Δa	Δb
1C	0.0	0.0	0.0	0	0	−0.2	−0.2	0.1	−0.1	0.1	−0.2	−0.1
2C	0.0	0.0	0.0	−0.6	0	0.4	−0.6	−0.2	0.5	−1.3	−0.1	0.6
3C	0.0	0.0	0.0	−0.3	−0.1	0.7	0.3	0	0.9	−0.1	0.2	0.4
4C	0.0	0.0	0.0	−0.3	0.5	−0.1	0.1	−0.3	0.2	−0.5	0.1	−0.3
5C	0.0	0.0	0.0	−0.2	−0.1	0.1	−0.3	−0.1	−0.1	−0.2	−0.2	0
Mean (C)	0.0	0.0	0.0	−0.3	0.1	0.2	−0.1	−0.1	0.3	0.7	0.0	0.1
±Δ (C)	0.0	0.0	0.0	0.3	0.3	0.5	0.4	0.2	0.5	0.3	0.2	0.5
1Tx1	0.0	0.0	0.0	−0.8	−0.2	−0.2	−0.4	−0.1	−0.1	−0.4	−0.1	−0.3
2Tx1	0.0	0.0	0.0	−0.1	0.0	−0.2	−0.6	−0.2	0.0	−0.2	−0.1	−0.1
3Tx1	0.0	0.0	0.0	−0.5	−0.2	−0.3	−0.8	−0.2	−0.5	−0.4	−0.4	−0.4
4Tx1	0.0	0.0	0.0	−0.9	−0.1	−0.1	−0.7	0.0	−0.2	−0.8	0.0	−0.2
5Tx1	0.0	0.0	0.0	−0.6	−0.3	−0.3	−0.5	−0.2	−0.4	−0.5	−0.1	−0.2
Mean (Tx1)	0.0	0.0	0.0	−0.6	−0.2	−0.2	−0.6	−0.1	−0.2	−0.5	−0.1	−0.2
±Δ (Tx1)	0.0	0.0	0.0	0.4	0.1	0.1	0.2	0.1	0.3	0.3	0.1	0.1
1Tx2	0.0	0.0	0.0	8.0	−2.8	−4.9	11.8	−4.0	−6.1	12.2	−3.8	−6.4
2Tx2	0.0	0.0	0.0	6.3	−1.9	−3.0	8.6	−2.3	−4.2	8.6	−2.4	−3.5
3Tx2	0.0	0.0	0.0	8.2	−0.4	−3.5	5.1	−0.9	−2.6	4.5	−0.6	−3.0
4Tx2	0.0	0.0	0.0	9.6	−0.5	−1.5	7.6	−0.8	−0.7	2.0	−1.3	−0.8
5Tx2	0.0	0.0	0.0	2.9	−0.5	−3.9	8.1	−0.4	−4.7	11.8	−1.0	−4.7
Mean (Tx2)	0.0	0.0	0.0	7.0	−1.2	−3.4	8.2	−1.7	−3.7	7.8	−1.8	−3.7
±Δ (Tx2)	0.0	0.0	0.0	3.2	1.3	1.6	3.0	1.8	2.6	5.6	1.6	2.6

fluctuations associated with flash power and measuring instrumentation signal instabilities.

A statistical analysis of the differential color parameters $\Delta(\text{CIE } L * ab)$ between the samples from the control (C) group and from the test1 (Tx1) group showed no statistically significant difference between $\Delta(\text{CIE } L * ab)$ determined after 3, 24, and 48 h (p-value = 0.08 to 082 > 0.05). Thus, it can be concluded that the drilling of microchannels into the tooth had no statistically significant influence on the CIE $L * ab$ parameters.

A statistical analysis of the differential color parameters $\Delta(\text{CIE } L * ab)$ between the samples from the control (C) group and from the test2 (Tx2) group showed a statistically significant difference between $\Delta(\text{CIE } L * ab)$ determined after 3, 24, and 48 h (p-value = 0.01 < 0.05). A statistical analysis of the differential color parameters $\Delta(\text{CIE } L * ab)$ between the samples from test1 (Tx1) group and from the test2 (Tx2) group also showed a statistically significant difference between $\Delta(\text{CIE } L * ab)$ determined after 3, 24, and 48 h (p-value = 0.01 < 0.05). Thus, it can be concluded that the HP-injection

had a statistically significantly influence on the CIE $L * ab$ parameters.

The CIE $L * ab$ color parameter analysis shows that for the test2 samples (Tx2) the L parameter (lightness) is significantly increased due to HP-injection through microchannels. This parameter increases within 3 h after injection of HP, continues to increase during 24 h, and reaches saturation after 48 h.

It should be noted that the increase in the L parameter (lightness) occurs with a decrease in parameters a (redness) and b (yellowness) as a result of chemical oxidation of pigments which give the red and yellow shades to dentin and tooth. According to Geenwall,[30] the molecules of these pigments have double bond groups and are organic in origin.[31] Irreversible discoloration of organic pigments is achieved by reduction of the molecular double bonds.[31] The main reaction in this case is the impact on the conjugated bonds ($-CH=CH-$) by HP or reactive oxygen species with epoque-compound formation.[32] Under more severe oxidation conditions, the formation of diols or complete C-C bond reduction is possible.[33]

The bleaching process causes a breakdown of molecular conjugation with the discolored product molecules. A single molecule of HP is consumed for each double bond of organic molecule. The oxidation products do not absorb light in the visible region, therefore, they are colorless.[31]

The above-mentioned alterations in the VITA shade guide and the CIE $L * ab$ color parameters are accompanied by visible variations in the whiteness of the test2 tooth (Tx2) crown which can be seen in the photographs of the tooth before and after HP-injection (Fig. 2).

The results described above show that a volume of 1.5 ± 0.1 mm^3 of 31%-HP injected into the dentin of a human tooth through microchannels with a diameter of ~200 μm and a depth of ~2 mm, which was drilled within the tooth crown using a high-quality beam of Er:YAG laser, is enough for bleaching of the teeth. Other types of lasers, but with a wide-beam diameter at the treatment area of 300 to 1000 μm, such as Er:YAG, Er:YSGG, or CO$_2$ multimode lasers were used for treatment of dental hard tissue.[34–39] The cavities created using such lasers with a wide-beam spot are characterized by a low aspect ratio, which is the ratio of cavity depth to cavity diameter, and are intended for traditional fillings. A multimode laser does not allow for formation of laser beams of a sufficiently small diameter for the cutting of the microchannels. The smaller the beam size, the greater the safety level.

Small size beams (microbeams) can form highly accurate cavities whose diameter is significantly smaller than the diameter produced by mechanical instruments (burs). Cavities with a high aspect ratio (microchannels) can also be formed using microbeams.

Another important feature of the laser radiation is the ability to form single spikes (micropulses) with a pulsewidth shorter than the thermal relaxation time of the treated tissue layer. The process of laser hard tooth tissue removal can be significantly improved by controlling the duration of the spikes, their duty cycle, and repetition rate.

Optimization is possible by supplying the laser energy at a particular time corresponding to minimal losses associated with laser beam attenuation by a water irrigation system or tooth debris. The simultaneous use of micropulses and microbeams is known as M^2 laser technology.[29,40] Single-mode TEM$_{00}$ lasers with a beam propagation ratio close to unity should be used to create these beams.[41,42] Figure 4 shows a tooth model which was developed to calculate the distribution of HP concentration at the dento–pulpal interface. The model was developed on the basis of the anatomical structure of a tooth as described by Nanci.[43] The tooth is surrounded by air and its root is composed of dentin and has a cylindrical shape with a diameter of 6.2 mm. The enamel has a thickness of 0.44 mm and uniformly covers the upper part of the tooth dentin to a height of 10 mm (similar to a natural tooth). The pulp chamber is cylindrical in shape and has a diameter of 2 mm.

A single channel was drilled at a distance of 5 mm from the tooth apex, perpendicular to the tooth axis, and its diameter and depth can be varied according to need. The modeling used a depth of 4.4 mm and a diameter of 0.66 mm, providing a volume of 1.5 mm^3 which was equivalent to 5 microchannels, as used in the experimental part of this study. In fact, the surface area of the laser drilled inlet microchannels [see Fig. 1(c)] was larger than that in the model. This difference may have a significant influence on the HP concentration distribution in the vicinity of the channel only within a rather short period of time after injection.
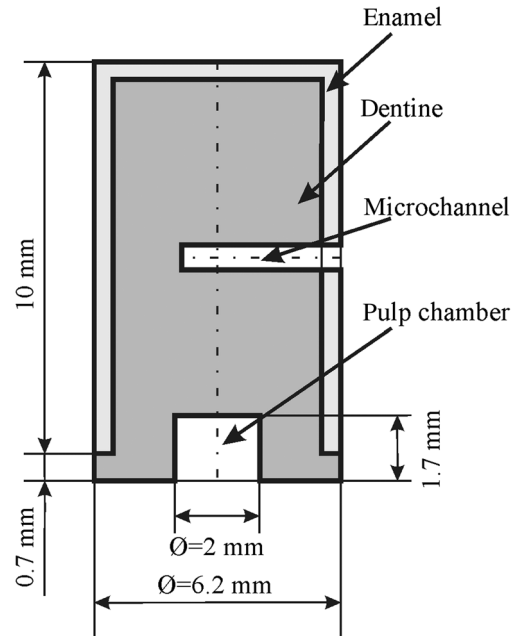

Fig. 4 Tooth model.

As we are really looking for the effect of HP on the entire tooth over a several hours, it can be hypothesized that the above-mentioned difference would not alter the results significantly.

The channel could then be filled with an aqueous solution of HP of different concentrations and the injection volume could be varied. The modeling used a concentration of 31%-HP and the injected volume was equivalent to the total volume of the channel, i.e., 1.5 mm^3.

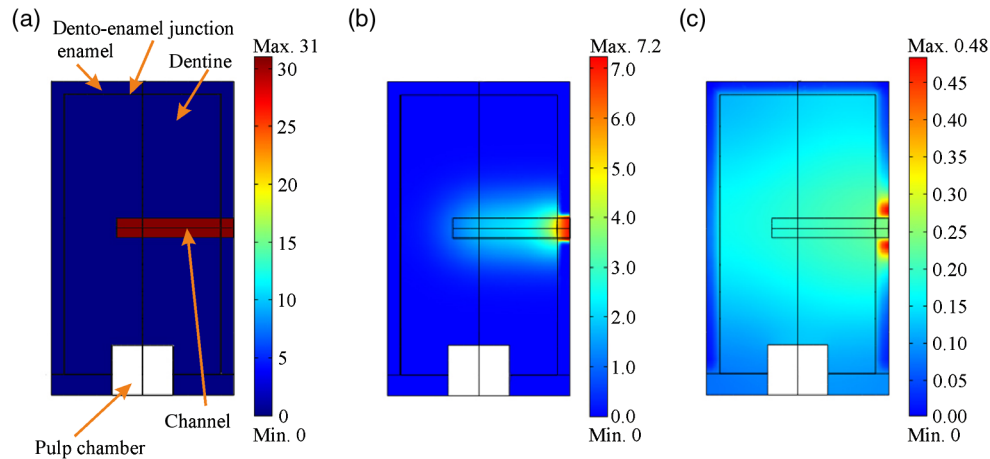The concentration of HP was calculated using the following matter diffusion equation:[44]

$$\frac{dC(\vec{r}, t)}{dt} = D\nabla^2 C(\vec{r}, t) - \gamma C(\vec{r}, t), \tag{1}$$

where $\vec{r} = (x, y, z)$ is the molecule coordinate, C is the HP concentration, D is the HP diffusion coefficient (m^2/s), and γ is the absorption factor (due to HP interaction with tissue organic molecules) (s^{-1}).

It was assumed that at the beginning of the process, there was no HP in the dentin and enamel, therefore, its concentration in these tissues could be expressed as $C_{e,d}(\vec{r}, t)_{t=0} = 0$, and the concentration of HP in the channel as $C_{ch}(\vec{r}, t)_{t=0} = C_0(\vec{r})$, where $C_0(\vec{r})$ is the initial concentration of HP in the solution (31%). The chemical interaction of the HP with the organic components of the tooth tissues was also not accounted for because of its insignificant effect on HP distribution within tooth tissues, i.e., it was assumed that $\gamma = 0$.

The diffusion coefficient of HP in dentin was taken as $D_d = 3.6 \cdot 10^{-7}$ cm^2/s (Ref. 45) and the diffusion coefficient of HP in enamel as $D_e = 3.6 \cdot 10^{-9}$ cm^2/s.[23] The results of numerical modeling of the HP-concentration distribution in a human tooth model are shown in Fig. 5.

Figure 5 shows that the concentration of HP decreases toward the bottom of the channel and does not exceed 2% at a depth of 2 mm from the channel inlet. The maximum concentration of HP in the channel is seen immediately after injection [dark red color in Fig. 5(a)]. At 3 h after injection, the maximum concentration of HP is seen at the entrance to the channel [red

Fig. 5 Numerical modeling of the HP-concentration distribution within tooth tissues calculated using the proposed tooth model: (a) immediately after, (b) 3 h after, and (c) 48 h after injection of a 31%-HP solution with a volume of 1.5 mm³ into the channel (volume of the channel is equivalent the volume of 5 micro-channels drilled by Er:YAG laser radiation in experiment). The figures on the right of each diagram show the calculated distribution of HP as expressed in a percentage of its concentration. In Fig. 5(a), because the picture shows the concentration of HP at commencement, no difference is seen between the dentin and enamel. However, in the channel cut by laser, it is at its maximum (31%) and a dark red color.

color in Fig. 5(b)]. At 48 h, the maximum concentration of HP is seen at the entrance to the channel in the enamel [red color in Fig. 5(c)]. It is distributed more uniformly in dentin and its concentration at the dento–pulpal interface is less than that at the dento–enamel junction.

The calculated concentration of HP in the channel and dento–pulpal interface for varying amounts of HP injected into the channel and absorbed by tooth dentin is shown in Fig. 6. Figure 6(a) shows that HP concentration in the channel decreases over time, but at the dento–pulpal interface it increases, as shown in Fig. 6(b). The results show that an amount of 1.5 mm³ of 31%-HP solution is sufficient to bleach a human incisor in *in vitro* conditions. At a distance of 2 mm from the inlet, HP concentration decreases to 10% in 0.5 h, to 5% in 1.2 h, and to 1% in 7 h. It reaches a maximum saturation level at the dento–pulpal interface in ~30 h and is almost an order of magnitude lower than the safety level as described by Bowles et al.[26] These results correspond to published *in vivo* pilot studies.[24,40]

Altshuler et al.[40] presented results of tooth vitality studies before and after injection of 35%-HP through microchannels formed by Er; Cr:YSGG laser radiation. The vitality of the dental pulp was assessed using an electric pulp tester. According to Prosvetov et al.,[46] the readings of the pulp tester reach 15 to

20 μA in intact teeth. With inflammation, the readings usually increase to 40 to 80 μA depending on the type and intensity of the inflammatory process. In the case of pulpal necrosis, readings are always >100 μA. In the study presented by Altshuler et al.,[40] the readings were 0.6 to 6.4 μA before laser treatment and immediately after laser treatment there was a slight increase of pulp tester readings but it did not exceed 20 μA. In a week, the readings returned to the baseline. The H&E histological analysis[40] of the intact tooth pulp after internal bleaching using laser microchannels for HP delivery did not show any histological abnormalities in the pulp after bleaching. Tissue necrosis or acute inflammatory infiltration with the accumulation of segmented leukocytes was not found in any of the specimens,[40] i.e., significant pulpal damage was not observed. From Altshuler et al.'s study,[40] it can be concluded that the effect on a vital tooth through microchannels formed by Er:YAG laser radiation leads to tooth bleaching which does not produce pulpal damage.

The open microchannels produced by this procedure will need to be restored in an appropriate manner. In principle, it would be possible to use a flowable composite material of which there are many which are currently available on the market. An alternative consideration could be given to a remineralizing technique, but this would be the subject of a further study.
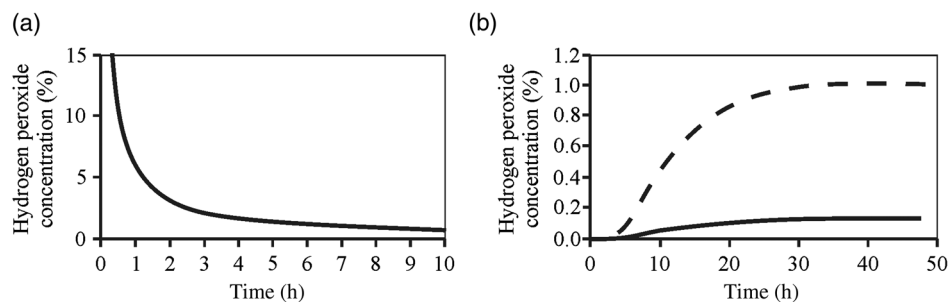
Fig. 6 The calculated concentration of HP on delivery time: (a) in the channel at its central axis, 2 mm apart from the inlet and (b) in the dentin at the central axis on the dento–pulpal interface. The calculations were made for two different volumes of 31%-HP: 1.5 mm³ (solid line) and 11.5 mm³ (dashed line).

5 Conclusions

A minimally invasive method of controlled intrinsic bleaching of teeth with HP delivered through laser microchannels from the palatal surface of the tooth which can potentially be used for vital tooth bleaching was presented. Laser microchannels were drilled into the tooth crown using multipulse tissue ablation by the radiation of an Er:YAG laser. These microchannels were then used for the delivery of HP directly into the tooth dentin.

The bleaching process is facilitated by the relatively uniform distribution of the bleaching compound into the dentin via free diffusion. Computer simulation showed that the amount of HP necessary for adequate tooth bleaching is well below the safety limit which would cause pulpal damage. The bleaching occurred predominantly during the first 3 h after HP injection, with a VITA shade increase in the range of 3 to 6 grades depending on the original shade of the tooth.

This result is consistent with the more objective color evaluation using the CIE $L*ab$ color system, where lightness (L) significantly increases after 3 h of treatment with some saturation at 24 and 48 h, and the two other parameters a (describing behavior in the red-green wavelength range) and b (describing behavior in the yellow-blue wavelength range) decreasing with time.

Disclosures

No conflicts of interest, financial or otherwise, are declared by the authors.

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