Published in final edited form as:

Proc SPIE Int Soc Opt Eng. 2015 February 24; 9306: . doi:10.1117/12.2083652.

Image-guided removal of occlusal caries lesions with a $\lambda = 9.3\text{-}\mu\text{m}$ CO₂ laser using near-IR transillumination

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Abstract

Previous studies have shown that near-IR transillumination is well suited for imaging deep occlusal lesions. The purpose of this study was to determine if near-IR images can be used to guide a CO₂ laser for the selective removal of natural occlusal lesions on extracted teeth. Near-IR occlusal transillumination images of extracted human teeth with natural occlusal caries lesions were acquired using an InGaAs camera and near-IR light at wavelengths from 1290 to 1470-nm from a filtered tungsten halogen source. A CO₂ laser operating at 9.3- μm with a pulse duration of 10–15- μs and a pulse repetition rate of 100–300-Hz was used for caries removal. Optical Coherence tomography was used to confirm lesion presence and serial scans were used to assess selective removal. Teeth were also sectioned for histological examination using polarized light microscopy. This study suggests that near-infrared transillumination is a promising method for the image guided laser ablation of occlusal caries lesions but the use of serial near-IR transillumination imaging for monitoring lesion removal was limited.

Keywords

light scattering; enamel; near-IR imaging; selective laser ablation

1. INTRODUCTION

In order to minimize the loss of healthy tissues during the removal of carious tissue it is advantageous to combine methods for caries imaging and caries removal. Since lasers can be operated in a non-contact mode they are ideal systems to use in conjunction with caries imaging.

Compact sealed CO₂ laser marking/engraving systems incorporating complete 3D computer programmable scanning systems are already available from several manufacturers and it is feasible to modify the wavelength and pulse duration of such low cost systems for the efficient removal of dental hard tissues [1]. Moreover, unlike other lasers systems, the CO₂ laser can be used effectively for a large range of soft and hard tissue procedures including gingival surgery, caries removal and caries prevention treatments, therefore only a single

laser system would be needed by the clinician. For selective removal, a small laser spot size should be used to minimize the amount of tissue removed per laser pulse. Therefore the laser has to be operated at high pulse repetition rates for practical removal rates. Transverse excited atmospheric pressure (TEA) and radiofrequency excited carbon dioxide lasers can be operated efficiently at high repetition rates at wavelengths that are coincident with the strongest absorption of dental hard tissues near $\lambda=9.3$ and $9.6\text{-}\mu\text{m}$ due to the phosphate ion in hydroxyapatite. Therefore, the irradiation intensities required for ablation of dental hard tissue are markedly lower at those wavelengths than for other laser wavelengths and are likely to lead to less accumulation of heat in the tooth for typical irradiation conditions. We have demonstrated that enamel and dentin can be most efficiently ablated using laser pulses of $10\text{--}15\text{-}\mu\text{s}$ duration [2–4]. Another important advantage of CO_2 laser irradiation is that it also imparts improved morphological and chemical changes to dental enamel [5] that increases resistance to acid dissolution.

Fluorescence has been combined with laser ablation for caries removal. Eberhard et al. [6–8] have combined laser ablation with an Er:YAG laser with red fluorescence (655-nm excitation, 680+ detection) and they claim this combination can be used to selectively remove infected dentin. Lennon and Buchalla et al. [9–11] have developed red fluorescence aided caries excavation (FACE) for use with the conventional handpiece and claim it has superior performance than conventional excavation, caries detector dyes and chemomechanical removal.

Recent studies suggest that near-IR imaging methods are highly promising due to weak light scattering from sound enamel and the lack of interference from stain [12–17]. Stains greatly interfere with visible reflectance and quantitative light fluorescence (QLF) measurements, therefore such measurements are of limited effectiveness in the occlusal surfaces [18, 19] and are not suitable for guiding laser ablation. Stains do not interfere at near-IR wavelengths of $1300\text{--}1750\text{-nm}$. Last year LaMantia et al. [20] demonstrated that near-IR reflectance measurements were well suited for guiding the removal of caries lesions using a $9.3\text{-}\mu\text{m}$ CO_2 laser. Laser modification of the enamel surface did reduce the contrast between sound and demineralized enamel but the contrast was still sufficiently high to enable use for sequentially removal of lesion areas in subsequent scans.

Occlusal near-IR transillumination can be used to acquire high contrast images of deep occlusal lesions, and studies show that the area and contrast of the lesion in near-IR images at 1300-nm increases significantly with lesion depth and severity [13]. In our first study of image guided laser ablation, Tao et al. [21] created patterned lesions on blocks of bovine enamel and demonstrated that images of the patterned lesions acquired using near-IR transillumination measurements could be used to guide a $9.3\text{-}\mu\text{m}$ CO_2 laser for highly selective laser ablation. In the following year, Tao et al. [22] also demonstrated the approach can be used for natural lesions.

The purpose of this study was to follow up on the studies initiated by Tao et al. and employ near-IR transillumination near 1300-nm for the removal of deeply penetrating natural occlusal lesions on extracted teeth.

2. MATERIALS AND METHODS

2.1 Sample Preparation

Ten human teeth with suspected lesion in the occlusal surfaces were collected and sterilized with gamma radiation. Teeth were visually examined and those that appeared to have non-cavitated lesions were selected and mounted in black orthodontic acrylic blocks. Samples were stored in a moist environment of 0.1% thymol to maintain tissue hydration and prevent bacterial growth. The outlines of 5x5 mm windows approximately 50- μ m deep were cut on the occlusal surface of each tooth using a CO₂ laser (Impact 2500, GSI Lumonics Rugby, UK) around the suspected lesion area as shown in Fig. 1 (red box). The channels cut by the laser serve as reference points for serial sectioning and are sufficiently narrow that they do not interfere with calculations of the image contrast. The reference boxes were cut into the samples using a spot size of 200- μ m, a pulse spacing of 50- μ m and a pulse repetition rate of 50-Hz with 30-mJ per pulse (See Sec. 2.2 below).

Optical coherence tomography (OCT) was used to confirm lesion presence and severity before removal.

2.2 Laser Irradiation

Samples were irradiated using an industrial marking laser, Impact 2500 from GSI Lumonics (Rugby, United Kingdom) operating at a wavelength of 9.3 μ m. The laser was custom modified to produce a Gaussian output beam (single spatial mode) and a pulse duration between 10–15- μ s. The laser energy output was monitored using a power meter EPM 1000, Coherent-Molelectron (Santa Clara, CA), and the Joulemeter ED–200 from Gentec (Quebec, Canada). Computer-controlled XY galvanometers 6200HM series with MicroMax Series 671 from Cambridge Technology, Inc. (Cambridge) were used to scan the laser beam over the sample surfaces. A repetition rate of 100-Hz was used with point to point scanning with a pulse spacing of 100- μ m. The laser beam was focused to a spot size of ~350- μ m using a ZnSe scanning lens of 90-mm focal length for an incident fluence of 20 J/cm² (19 mJ per pulse). An air-actuated fluid spray delivery system consisting of a 780S spray valve, a Valvemate 7040 controller, and a fluid reservoir from EFD, Inc. (East Providence, RI) was used to provide a uniform spray of fine water mist onto the tooth surfaces at 2 mL/min.

2.3 Near-IR Transillumination

Near-Infrared transillumination images were acquired at intervals of 20 scans (iterations) to a maximum of 160 iterations. A NoblePeak Vision Triwave Imager, Model EC701 (Wakefield, MA) that employs a Germanium enhanced complementary metal oxide semiconductor (CMOS) focal plane array (640x480 - 10- μ m pixel pitch), sensitive in the visible and NIR from 400–1600-nm with a Infinimite™ video lens (Infinity, Boulder, CO) was used to capture the near-IR images. Light from a 150-W fiber-optic illuminator FOI-1 from E Licht Company (Denver, CO) coupled to an adjustable aperture and a BP1380-90 bandpass filter from Spectrogon (Parsippany, NJ) centered at 1380-nm with a bandwidth of 90-nm was used. Near-IR light was delivered by a low profile fiber optic with dual line lights, Model P39-987 from Edmund Scientific (Barrington, NJ), with each light line directed at the cementoenamel junction beneath the crown on the buccal and lingual sides of

each tooth. The angle and position were extremely important. If the light is directed too high on the tooth or at an upward angle the light does not enter the dentin of the crown and the contrast of the lesion is greatly reduced. A simple box filter was applied to the image over a range of 8 pixels to remove the small dots on the image which were caused by nonlinear gain in certain pixel groups of the Triwave Imager. Note that contrast measurements were carried out prior to the addition of filters.

2.4 Radiographs

Digital radiographs of each tooth were acquired prior to ablation using Kodak K-2200 X-ray unit (Carestream Dental LLC, Atlanta, Georgia) on the following setting: 7 kVp, 7 mA, and duration of 0.074 second. The dental radiographs are taken in a bitewing view on GX-S700 digital sensor (Gendex Dental System, Hatfield, Pennsylvania).

2.5 Optical Coherence Tomography (OCT)

Serial OCT scans were taken at ablation intervals of 0 (pre-ablation), 40, 80, and 160 laser scans (post-ablation) using a single-mode fiber, autocorrelator-based Optical Coherence Domain Reflectometry (OCDR) system. The OCDR system was coupled with an integrated fiber probe, high efficiency piezoelectric fiber-stretchers and two balanced InGaAs receivers that was designed and fabricated by Optiphase, Inc. (Van Nuys, CA). The OCDR system was integrated with a broadband high power superluminescent diode (SLD), DL-CS313159A Denselight (Jessup, MD) with an output power of 19-mW and a bandwidth of 83 nm and a high-speed XY-scanning system, ESP 300 controller & 850-HS stages, Newport (Irvine, CA) and used for *in vitro* optical coherence tomography. The fiber probe was configured to provide an axial resolution at 9- μ m in air and 6- μ m in enamel and a lateral resolution of approximately 50- μ m over the depth of focus of 50- μ m. The all-fiber OCDR system has been previously described in greater detail [23, 24]. The OCT system is completely controlled using Labview™ software (National Instruments, Austin, TX). Acquired scans are compiled into *b-scan* files. Image processing was carried out using Igor Pro™, data analysis software (Wavemetrics Inc, Lake Oswego, Oregon).

2.6 Polarized Light Microscopy

After sample imaging was completed, approximately 200 μ m thick serial sections were cut using an Isomet 5000 saw (Buehler, IL), for polarized light microscopy (PLM). PLM was carried out using a Meiji Techno RZT microscope (Meiji Techno Co., LTD, Saitama, Japan) with an integrated digital camera, Canon EOS Digital Rebel XT (Canon Inc., Tokyo, Japan). The sample sections were imbedded in water and examined in the brightfield mode with crossed polarizers and a red I plate with 500-nm retardation.

2.7 Image Analysis and Statistics

Line profiles were extracted at the same position from the most severe area of each image across each 5×5 mm window and lesion or image contrast was calculated using the equation $(I_S I_L)/I_S$; where I_S is the mean intensity of the sound enamel outside the window area, and I_L is the mean intensity of the lesion. The lesion is darker, i.e., lower intensity in transillumination.

All image analysis was carried out using Igor pro software (Wavemetrics, Lake Oswego, OR). A repeated measures one-way analysis of variance (ANOVA) followed by the Tukey-Kramer post-hoc multiple comparison test was used to compare groups employing Prism software (GraphPad, San Diego, CA).

3. RESULTS

Figure 2 shows a series of OCT images that were taken before removal and at different time-points during lesion ablation: 0 (pre-ablation), 40, 80, and 160 scans (post-ablation) for one tooth. The pre-ablation OCT image shows a diffuse area of higher reflectivity near the dentinal-enamel junction on the left side of the pit which suggests a lesion that has penetrated to the dentin and spread laterally. The OCT image after 40 scans of the same area, shown in Fig. 2B, continues to show the presence of subsurface demineralization and even shows higher reflectivity in areas not visible before. Further ablation in the area, Fig. 2D, indicates the removal of the subsurface area of higher reflectivity, suggesting that the demineralized area of the lesion was removed. Figure 3 shows a PLM image of a section at the same location of the OCT scan of Fig. 2. The tooth surface has been significantly modified by the series of ablation using the CO₂ laser. The subsurface demineralization was removed. The depth of removal reached the dentin-enamel junction (DEJ) as shown in Fig. 3 and the demineralized lesion identified in the serial OCT scans was completely removed.

Figure 4 shows a series of OCT images for another sample for which the removal was incomplete. The areas of higher subsurface reflectivity on the OCT images show the area of the demineralized lesion. The removal of the demineralized lesion at each ablation interval is shown in the serial OCT scans, however in contrast to the 1st sample there are areas of high subsurface reflectivity remaining even after 160 scans as shown in Fig. 4D, particularly in areas peripheral to the deepest area of removal.

Fig. 5 shows a PLM image of the same lesion indicated in Fig. 4. The image shows that the demineralized areas were not completely removed which is consistent with the corresponding OCT image, Fig. 4D. After 160 scans the depth of the cut did not reach the dentin-enamel junction. However, sizable portions of the demineralized lesion were removed. The image contrast between the demineralized areas and the sound enamel for each sample were measured at ablation intervals of 20 scans from 0 to 160. Dark areas on the near-infrared images indicate regions of demineralization while light areas indicate regions of sound enamel. The mean lesion contrast was determined for each ablation interval. The change in mean lesion contrast for each ablation interval showed no significant difference ($p > 0.05$).

A total of forty-seven lesions were identified in pre-ablation OCT images. Upon analysis of the post-ablation images, twenty-one of the forty-seven lesions were identified to be completely removed from the enamel. Removal was incomplete after ablation ceased after 160 scans for 26 out of the 40 lesions.

4. DISCUSSION

The results of this pilot study suggest that it may be feasible to utilize near-infrared transillumination to guide a carbon dioxide laser for selective removal of carious lesions. Our studies show that 45% of the total number of demineralized lesions identified were completely removed down to the dentin-enamel junction (DEJ) with 160 scans. However, 55% of the lesions identified were only partially removed from the enamel after 160 scans. The small spot size and limited depth of focus combined with the highly convoluted occlusal surface profoundly influences the rate of removal and this study suggests that a dynamic focusing system is required.

The lesion contrast was monitored at intervals during removal and there was no significant change after 160 scans. This is surprising and we anticipated a decrease in contrast as the lesions were removed. The chemical modification of enamel and dentin by the laser causes an increase in reflectivity and a decrease in contrast. However, as the depth increases and the dentin is approached the contrast increases due to the higher reflectivity of the dentin. Therefore, even though the near-IR transillumination images are useful for identifying areas of deep lesion penetration this study suggests that serial imaging scans during lesion removal are of more limited value.

In contrast, the serial OCT were extremely useful, and more of the lesion was exposed in OCT images as removal progressed. An example of this can be seen in Fig. 2. Part of the underlying subsurface lesion was hidden in the pre-ablation OCT image, shown in Fig. 2A. After 40 iterations of ablation, the hidden area of the subsurface lesion became visible as shown in Fig. 2B. Removal of the surface enamel shortened the total penetration depth the OCT system needed to reach the DEJ. Doing so, improved the ability of the OCT imaging system to detect demineralized lesions.

In summary, this pilot study suggests that near-infrared transillumination is a promising method for the image guided laser ablation of occlusal caries lesions but the use of serial near-IR transillumination imaging for monitoring lesion removal was limited.

ACKNOWLEDGMENTS

This work was supported By NIH/NIDCR Grants R01-DE14698 and R01-DE19631 and the UCSF School of Dentistry.

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Fig. 1. Images taken before ablation; (A) visible light, (B) NIR-transillumination, (C) standard clinical dental x-ray in bitewing orientation, and (D) OCT. Red box demarcates the region of interest. The yellow arrow shows the lesion position in each image. Lesion is not visible on radiograph (C).

Fig. 2. Serial OCT images taken (A) 0 (pre-ablation), (B) 40, (C) 80, and (D) 160 repetitions (scans) (post-ablation). The arrows mark the positions of the lesions on the OCT scans.

Fig. 3.
PLM image of tooth cross-section from same tooth/position shown in Fig. 2 after the removal of the lesion.

Fig. 4. Serial OCT images taken (A) before ablation, (B) after 40, (C) 80, and (D) 160 repetitions (scans) after ablation. The arrows mark the positions of the lesions on the OCT scans.

Fig. 5.
PLM image for the sample shown in Fig. 4.