Chemical analysis and surface morphology of enamel and dentin following 9.6μ CO2 laser irradiation versus high speed drilling

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Summary
Objectives. The purpose of the present in vitro study was to determine whether there is a change in the chemical composition and surface morphology of enamel and dentin following 9.6μ CO2 laser irradiation and high-speed drilling.

Materials and methods. Ten permanent, non-caries, young premolars, extracted for orthodontic reasons, were selected. The crowns were separated longitudinally into two equal parts at their mesiodistal axis. Two areas on the inner enamel surface of each specimen and two on the dentinal surface were selected. A high-speed drill and 9.6μ CO2 laser irradiation were applied to the selected enamel and dentinal areas. A random area on the unlased enamel and on the unlased dentin of each specimen served as controls. The morphology of the specimens was evaluated using scanning electron microscopy. Calcium, phosphorus and oxygen levels were measured using an energy dispersive spectrometer.

Results. Mineral analysis revealed no significant difference in the mineral content of the enamel and dentin after laser irradiation or high speed drilling versus the control.

Use of the high-speed drill on enamel and dentin resulted in very clear cavity margins, with characteristic grooves, whereas laser irradiation of enamel and dentin did not produce clear margins and the floor of the cavity displayed an irregular surface.

Conclusions. The 9.6μ CO2 laser appears to be a promising tool in the clinical setting. However, further investigation is indicated to ensure maximum effectiveness.

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Introduction

Laser is the acronym for light amplification by stimulated emission of radiation. It differs from conventional light, as it is a single wavelength, collimated, coherent and intense. Laser photons interact with tissue in one of four ways: they are transmitted through tissue, reflected from tissue, scattered within tissue, or absorbed by tissue. When absorbed, light energy is converted into thermal energy.

The particular properties of each type of laser and the specific target tissue make them suitable for different procedures. The CO2 laser is highly absorbed by all biological soft and hard tissues and is most effective in tissues with a high water content, like the soft tissues of the oral cavity.

Enamel, dentin and cementum contain hydroxyapatite, which has absorption bands in the infrared region (9.0 through 11.0 μm) due to the presence of phosphate, carbonate and hydroxyl groups in the crystal structure. The absorption coincides closely with the radiation produced by the CO2 laser. This implies that dental hard tissues should efficiently absorb the radiation from the CO2 laser. The mechanism of action of CO2 lasers is the delivery of a great deal of energy to a small target area. If the energy is absorbed rather than reflected, the energy of the laser is converted into heat. When sufficient heat is produced on the dentin surface, it may burn, melt or vaporize the matrix. The effects of laser irradiation on dental hard tissues may cause chemical, thermal and/or mechanical changes. The latter effect is useful in removing carious dentin, while melting of dentin can seal the dentinal tubules and alleviate pain in cases of dentinal hypersensitivity.

CO2 laser energy (wavelengths 9.3, 9.6, 10.3 and 10.6 μm) is best absorbed by water and since 80–85% of body soft tissue volume consists of water, this beam does not penetrate the depth of the tissue but is immediately absorbed. Previous studies have shown that the energy absorbed in enamel by 9.6 μm CO2 laser irradiation is higher than the energy absorbed by other CO2 laser wavelengths.

We hypothesize that in spite of the reported morphological changes incurred by the use of the CO2 laser, there is no influence, as in the use of the high speed drill, on the chemical composition of enamel and dentin.

The purpose of the present in vitro study was to determine whether there is a change in the chemical composition and surface morphology of enamel and dentin following 9.6 μm CO2 laser irradiation and high-speed drilling.

Materials and methods

Ten permanent, non-caries, young premolars, which were extracted for orthodontic reasons, were selected. The teeth were stored in sterile saline until use. The roots of all the teeth were removed, using an E-1 high-speed diamond bur at the cementoenamel junction. The crowns were separated longitudinally into two equal parts at their mesiodistal axis, each half serving as an independent specimen. Thus, a total of 20 specimens were examined.

Two areas on the inner enamel surface of each specimen and two on the dentinal surface were selected, (Fig. 1). A D-1 high-speed drill was applied for two seconds to the first selected dentinal area and 9.6 μm CO2 (Opus 96, OpusDent,

![9.6 μm CO2 Laser application](image)

Figure 1 Scheme showing the areas on the inner enamel surface of each specimen and on its dentinal surface.
Israel) laser irradiation, emitting 22 mJ of radiation in 60 µs-long pulses and with an energy level of 3.5 W, was applied for five seconds to the second area. The beam was positioned via an optical scanner that moved the 250 µm diameter focused beam over a 2.5 mm diameter circular region. The same procedure was applied to the enamel surface, using the same high-speed drill as above for the first selected area and 9.6 CO2 laser irradiation with an energy level of 7 W to the second area. A random area on the unlased enamel and on the unlased dentin of each specimen served as controls. The specimens were then rinsed, dried and prepared for scanning electron microscopy and energy dispersive spectrometric analysis. The treated specimens were mounted on aluminum stubs and coated with a 20 nm layer of carbon for surface morphology evaluation, using a JSM-840A scanning electron microscope (JEOL, Tokyo, Japan). The calcium, phosphorus and oxygen levels of each specimen were measured to a minimum detectable level of 300 ppm, using an AN 10,000 energy dispersive spectrometer (Link-Oxford, High Wycombe, UK).

Changes in mineral level were recorded and the differences between the groups were analyzed statistically using the Kolmogorov-Smirnov test (to verify the normality of the data), and ANOVA. The level of significance was set at \( P < 0.05 \).

### Results

#### Chemical analysis

The normality of the data was verified by the Kolmogorov-Smirnov test. The mean concentrations of calcium (Ca), phosphorus (P), oxygen (O) and the Ca/P ratio in enamel and dentin following laser irradiation and high-speed drilling, are shown in **Table 1** and **Table 2**. Mineral analysis revealed no significant difference in the mineral content of the enamel and dentin after laser irradiation or after high speed drilling versus the control \( (P > 0.05, \text{ANOVA}) \).

#### Morphological changes

The specimens were examined at low \( (\times 25-50) \) and high \( (\times 900) \) magnifications. Round craters

| Table 1 Mean concentration (percent) of Ca, P and O, and Ca/P ratio in enamel following the application of laser and a high-speed drill. |
|---|---|---|---|
| Reference (N=20) | Laser (N=20) | Drill (N=20) |
| Calcium (Ca) | 33.8±2.3 | 35.0±4.3 | 32.9±4.3 |
| Phosphorous (P) | 19.2±0.8 | 19.1±1.0 | 18.6±1.4 |
| Oxygen (O) | 43.3±2.7 | 43.2±5.1 | 45.1±5.2 |
| Ca/P | 1.8±0.1 | 1.8±0.1 | 1.8±0.1 |

| Table 2 Mean concentration (percent) of Ca, P and O, and Ca/P ratio in dentin following the application of laser and a high-speed drill. |
|---|---|---|---|
| Reference (N=20) | Laser (N=20) | Drill (N=20) |
| Calcium (Ca) | 31.2±2.5 | 31.9±2.8 | 29.7±2.8 |
| Phosphorous (P) | 17.4±1.0 | 19.9±1.4 | 16.9±1.1 |
| Oxygen (O) | 46.1±3.1 | 47.0±4.4 | 48.8±3.4 |
| Ca/P | 1.8±0.05 | 1.8±0.1 | 1.7±0.06 |

**Figure 2** Enamel surface after high-speed drilling showing round craters measuring approximately 2 mm in diameter. Note clear cavity margins (magnification ×50).

**Figure 3** Dentin surface after high-speed drilling showing round craters measuring approximately 2 mm in diameter. Clear cavity margins are evident (magnification ×50).
measuring approximately 2 mm in diameter were observed when the specimens were examined under magnifications of ×25-50. Application of the high-speed drill to enamel (Fig. 2) and to dentin (Fig. 3) resulted in very clear cavity margins, with characteristic grooves. In contrast, laser irradiation of enamel (Fig. 4) and dentin (Fig. 5) did not produce clear margins and the floor of the cavity displayed an irregular surface, without evidence of carbonization. In most of the specimens, artifactual cracks of varying sizes were located at the center of the crater or alongside the walls.

High magnification (×900) of the lased enamel (Fig. 6) and dentin areas (Fig. 7) revealed melted material, which resembled glazed interconnected droplets. Resolidification of the melted droplets appeared be discontinuous rather than uniform and glazed. A distinct border between the area of the melted material and the adjacent normal enamel (Fig. 8) and dentin (Fig. 9) was clearly observed.

Discussion

The use of scanning electron microscopy together with energy dispersive spectrometry enabled us to examine longitudinally section crowns of ten non-carious premolars and to determine the mineral content of their enamel and dentin following 9.6 μ CO2 laser irradiation and high-speed drilling. This analysis is based on bombarding the specimen with a beam of high voltage electrons that are refracted at different energy levels from the individual minerals. The change in the energy returned from the specimen reflects the change in its mineral content.9 This method allowed us to analyze the specimens accurately and non-invasively.
In our study, there was no statistically significant difference in mineral content between the laser group and the high-speed drill group versus the control. These findings are in accordance with our previous results, showing that Nd:YAP laser irradiation did not have any effect on the mineral content of enamel and dentin.\(^{10}\)

Our present data, however, are not in agreement with those of Dankner et al.,\(^9\) who found a significant decrease in P following irradiation with the XeCl-308 nm excimer laser. They also demonstrated a decrease in Ca and increased K and S levels, albeit these changes were not statistically significant. The authors speculated that the changes in mineral content were attributable to the effect of the laser on the inorganic components of the hydroxyapatite crystals following their melting and recrystalization on the dentin surface. The discrepancy between our findings and those of Dankner et al. could be anticipated due to the different properties of the lasers used (i.e. wavelength and energy level). Also, it has been suggested that the excimer laser is capable of breaking chemical bonds.\(^{11}\)

It is not surprising that there was no difference in mineral content between the high-speed drill group and the control group since the high-speed bur exerts only a mechanical force.\(^{12}\) However, irradiation with the 9.6\(\mu\)m CO\(_2\) laser, which affects the dentinal surface by releasing heat, also had no influence on the mineral content, causing melting and resolidification within a very short period of time.\(^{13}\) As previously shown by Wigdor and Walsh,\(^{14}\) irradiation with this laser does not cause pulpal damage since an elevation in pulp temperature is not generated during the process. In our study we used laser energy levels, 7 W on enamel and 3.5 W on dentin, for cavity preparation since, in a pilot study, we found that the ideal energy for drilling enamel should be twice as high as for dentin and also higher than those published by Wigdor and Walsh.

With regard to the observed changes in surface morphology, we found clear margins when using the high-speed bur, whereas the 9.6\(\mu\)m CO\(_2\) laser produced an irregular surface. Again, this was due to the rapid heating and cooling of the treated surface, which is characteristic of CO\(_2\) lasers.

The melting and resolidification may even lead to sealing of the dentinal tubules, thus reducing the sensitivity resulting from cavity preparation. It also bears mention that when the effect of the 9.6\(\mu\)m CO\(_2\) laser was compared with that of the Er:YAG, we noticed that Er:YAG is more efficient in abating the hard dental tissues.

Our study faces some limitations. Energy dispersive spectrometer to examine chemical changes, may be a method which is much less informative than XPS or Raman spectroscopy, since re-arrangements could occur which would not be evident with the energy dispersive spectrometer. However, this system is the only available system in our institution.

In summary, the 9.6\(\mu\)m CO\(_2\) laser appears to be a promising tool in the clinical setting. However, further investigation is indicated to ensure maximum effectiveness.

References


