

INFLUENCE OF CO₂ LASER POWERS IN THE TEMPERATURE OF PRIMARY ENAMEL**Rodrigo Alexandre Valério¹, Caio Cesar Pereira Simon², Luciano Bachmann³, Silmara Aparecida Milori Corona⁴ and *Maria Cristina Borsatto⁵**¹DDS, MD, Postgraduate Student, Clinical Pediatric Dentistry Department, Ribeirão Preto School of Dentistry/ São Paulo University, Café Avenue, Monte Alegre, Ribeirão Preto - SP, Brazil, CEP: 14040-904.²DDS, Clinical Pediatric Dentistry Department, Ribeirão Preto School of Dentistry/ São Paulo University, Café Avenue, Monte Alegre, Ribeirão Preto - SP, Brazil, CEP: 14040-904.³DDS, MD, PhD Doctor Professor, Physics Department, School of Philosophy, Science and Literature/ São Paulo University, 3900 Bandeirantes Avenue, Ribeirão Preto - SP, Brazil. CEP: 14040-901.⁴DDS, MD, PhD, Associate Professor, Restorative Dentistry Department, Ribeirão Preto School of Dentistry/ São Paulo University, Café Avenue, Monte Alegre, Ribeirão Preto - SP, Brazil. CEP: 14040-904.⁵DDS, MD, PhD, Full Professor, Clinical Pediatric Dentistry Department, Ribeirão Preto School of Dentistry/ São Paulo University, Café Avenue, Monte Alegre, Ribeirão Preto - SP, Brazil, CEP: 14040-904.***Corresponding Author: Maria Cristina Borsatto**

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ABSTRACT

Objectives: Different types of CO₂ laser power are used to irradiate primary teeth, however, the study of these powers has fundamental importance, since, the use of correct parameters prevent the occurrence of thermal damage to the pulp tissue. The aim of the present *in vitro* study was to evaluate the temperature change on specimens of primary enamel irradiated with different powers of CO₂ laser. **Methods:** Fifteen sound primary molars were sectioned mesiodistally, resulting in 30 specimens (3.5x3.5x2.0 mm). Two small holes were made on the dentin surface using round bur in high speed turbine (1/4), in which K-type thermocouples were installed to evaluate thermal changes. Specimens were individually fixed with wax in Plexiglass cylinders and randomly assigned in 3 groups (n=10) according to the powers used: (A) 0.5 W, (B) 1.0 W and (C) 1.5 W. The CO₂ laser ($\lambda=10.6 \mu\text{m}$) was applied in an ultrapulse mode (non-contact), 4 mm focal length, 20 Hz frequency, and beam diameter of 0.4 mm. Temperature reading was performed every 0.3 seconds for 30 seconds. Analysis of variance (ANOVA) were performed for the statistical analysis ($p \leq 0,05$). **Results:** CO₂ laser powers provided no difference on the temperature changes on specimens of primary enamel, in which the following means were observed: (A) $49.32^\circ\text{C} \pm 11.87$, (B) $44.75^\circ\text{C} \pm 8.3$ and (C) $47.48^\circ\text{C} \pm 6.88$. **Conclusions:** It can be concluded that the CO₂ laser was not influenced by different powers (0.5W, 1.0W, 1.5W), used in the irradiation of primary enamel teeth fragments.

KEYWORDS: Lasers, temperature, dental enamel, primary teeth.**INTRODUCTION**

CO₂ laser emits infrared light ($\lambda = 9.3, 9.6, 10.3$ and $10.6 \mu\text{m}$), outside the visible spectrum to the human eye, in a non-contact mode, continuous or pulsed. This high power laser equipment has a delivery system made by articulated arm with mirror, facilitating its use in the oral cavity.

With the evolution of this technology, there was a greater interest from the scientific community in the development of research involving the CO₂ laser in all areas of dentistry. In pediatric dentistry has been used to increase the acid resistance of tooth enamel.^[1,2]

CO₂ laser irradiation's more appropriate to dental enamel because produces radiation that coincides closely with

some of apatite absorption bands, mainly phosphate and carbonate group absorption.^[3]

Higher effectiveness in caries prevention could be achieved with lower occurrence of harmful effects to dental tissues.^[3] The CO₂ laser energy is absorbed in few micrometers of the external enamel surface and converted into heat, causing loss of carbonate from mineral and fusion of hydroxyapatite crystals, reducing the interprismatic spaces.^[4] Furthermore, it increases its acid resistance, decreasing the mineral reactivity and promoting caries-preventive effect.^[1,5]

The thermal effect promotes melting of the irradiated dental surface^[6], resulting in chemical and structural alterations regarding to calcium and phosphorus loss^[7-9],

calcium and phosphorus concentration on the surfaces^[10] and alterations in organic matrix.^[11]

The higher thermal conductivity of primary dental enamel compared to dentin^[12,13], can produce thermal damage to the irradiated structures by the spread of temperature towards the pulp organ, since, the anatomical characteristics of the teeth^[14] provide different timed thermal declines. It has been reported in the literature that thermal variations higher than 5.5°C could cause permanent damage to the pulp tissue.^[15]

To investigate the short and long-term pulpal effects to cavity preparations in healthy human third molars using carbon dioxide (CO₂) laser, the laser parameters included a pulse energy of 40 mJ and a repetition rate of 100 Hz with a water-cooling system.^[16]

In the literature there are no studies that evaluate the thermal variations in the specimens of primary enamel teeth after irradiation with CO₂ laser using different powers that can be used to increase the acid resistance.

The thermal variation in the pulp chamber and tooth enamel of primary canines were verified by Arrastia *et al.*^[17], concluding that parameters of 176 J/cm² in the continuous wave mode and 264 J/cm² in single pulses are appropriate for pulpotomy and promotes minimum temperature of the irradiated dental hard tissues with the CO₂ laser.

Due to the increased research, related to the use of laser technology in pediatric dentistry and regarding the heat generation produced on dental substrates during irradiation, more studies evaluating the thermal changes in specimens of primary teeth irradiated with CO₂ laser using different powers (0.5 W, 1.0 W, 1.5 W), become necessary.

MATERIAL AND METHOD

Experimental Design

The study factor method was the powers of CO₂ laser employed during the irradiation of enamel specimens of primary molars at 3 levels: (A) 0.5 W (19.89 J/cm²), (B) 1.0 W (39.78 J/cm²) and (C) 1.5 W (59.68 J/cm²). The experimental sample was composed by 30 specimens of primary human enamel, which were randomly assigned (n=10), according to the design in randomized complete blocks. The quantitative response variable was the temperature change, in Celsius degrees, of the primary tooth substrate subjected to the CO₂ laser irradiation.

Teeth selection and preparation of specimens

First and second upper primary human molars newly exfoliated were examined with an explorer probe #5 (Duflex, SSWhite, Rio de Janeiro, RJ, Brazil), using a stereomicroscope (Leica S6 D Stereozoom, Mycosystems Leica AG, Switzerland), with increase of 20X. Those which presented cracks or hypoplasia were discarded. Fifteen teeth were selected and cleaned with

periodontal cures, being polished with Robinson brushes mounted in low speed turbine (Dabi Atlante, Ribeirão Preto, SP, Brazil) embedded in pumice and water, washed and kept in 0.9% saline solution containing 0.4% sodium azide at 4°C.^[18]

The teeth were individually fixed by the coronary portion with thermoplastic wax (Wax Sculpture Fixed Prosthodontics, Aspheric Chemical Industry Ltda., São Caetano do Sul, SP, Brazil) in acrylic plates and taken to the section machine (Minitom, Struers A/S, Copenhagen, Denmark) in which the root portion, if it was present, there was sectioned 1mm below the cement-enamel junction. Then, the coronary portions were sectioned mesiodistally and from the buccal and lingual surfaces of each tooth. It was obtained specimens of 3.5x3.5x2.0 mm of thickness of enamel and dentin. The dimensions of the specimens were determined using a digital caliper (Myamoto, Tokio, Japan) and the thickness were established with a specimeter (BioArt, São Carlos, SP, Brazil).

To standardize the thickness of enamel/dentin, specimens were taken to the polisher (Politriz DP-9U2, Struers A/S, Copenhagen, Denmark) and subjected to wear with sanding discs of silicon carbide #600 (Norton/Saint-Gobain Abrasivos Ltda., Guarulhos, São Paulo, SP, Brazil), aiming to plan and regularize the surfaces. Using drill #1/4 (KG Sorensen, Barueri, SP, Brazil), mounted in high speed turbine (Roll Air 3, Kavov do Brasil S.A, Joinville, SC, Brazil), two holes were made manually by the same operator, at dentin surface (medium depth 0.1 mm) corresponding to the roof of the pulp chamber, to accommodate the thermal-sensors during the specimens irradiation. After these procedures, specimens were individually fixed with thermoplastic wax at cylindrical Plexiglass® abutment (5.0 mm diameter) using a parallelometer to ensure that the enamel surface was kept parallel to the horizontal plane, aiming to sealing any space between the specimen and the plaque. The specimens were randomly assigned (n=10) and kept in distilled water at 4°C until 2 hours before the experiment start.^[2,19]

The irradiation distance was standardized by a device which couples the laser pen maintaining a fixed predetermined height of 4 mm^[20], while the specimens were placed in an adjustable base that allows its synchronized movement on the axes XYZ (right and left, forward and backward, up and down).

The irradiated area was delimited by insulating type (3M do Brasil Ltda, Campinas, SP, Brazil) with a 4.0 mm² central window. Each group was irradiated with CO₂ laser ($\lambda = 10.6 \mu\text{m}$) (PC 015-D CO₂ laser system, Shanghai Jue Hua Laser Tech. Development Co., Ltd., Shanghai, China) in the ultrapulse mode, focal length of 4 mm, 20 Hz frequency, and beam diameter of 0.4 mm, at non-contact mode with different powers and energy density: (A) 0.5 W (19.89 J/cm²), (B) 1.0 W (39.78

J/cm²) and (C) 1.5 W (59.68 J/cm²), were applied to the specimens of primary enamel, for 30 seconds.

The laser parameters of CO₂ laser irradiation used in this study were based on the favorable results obtained^[1,2], to increase the acidic resistance to demineralization. To evaluate the temperature change, a device consisting of a data acquisition card HI-Speed USB Carrier NI USB-9162 (National Instruments Corporation, Austin, Texas, USA), with thermal filaments sensors (K-type thermocouples, Omega Engineering Inc. USA), were used to check the temperature. The board has 4 input channels with full resolution of 24 bits and 12 Hz maximum total rate of data acquisition.

The thermocouples were made with the aid of a spot welding of carbon and presented 120 µm of diameter and 40 cm in length. The system was connected to a computer. The Measurement and Automation Software and VI Logger Lites (National Instruments Corporation, Austin, Texas, USA), supplied by the manufacturer of data acquisition card, was employed for data collection. Two thermo-sensors strands were placed in the niches at the dentin surface of each specimen, providing better thermal contact between the thermo-sensors and the specimens. A thermal paste based on water was employed (Implastec, Votorantim, São Paulo, SP, Brazil).

For each specimen, the temperature (°C) was registered from the first laser pulse emitted by the CO₂ laser and repeated every 0.3 seconds, for 30 seconds using the K-type thermocouples adapted to the dentin surface 1 mm under irradiated surface. All measurements were performed in a temperature/humidity-controlled room. Figure 1 represents the schematic design of the employed methodology.

SEM Evaluations

For the scanning electron microscopy (SEM) evaluation, two fragments of each group: (A) 0.5 W (19.89 J/cm²), (B) 1.0 W (39.78 J/cm²) and (C) 1.5 W (59.68 J/cm²) irradiated with CO₂ laser were cleaned by ultrasound (BioFree 06L, Gnatus Equipamentos Médico-Odontológicos, Ribeirão Preto, SP, Brazil) for 10 minutes. The fragments were then immersed in 2.5% glutaraldehyde solution buffered with 0.1 M sodium cacodylate, which was followed by rinsing in distilled water. Next, the specimens were dehydrated with ethanol in increasing percentage solutions, namely 25% (20 minutes), 50% (20 minutes), 75% (20 minutes), 90% (30

minutes), and 100% (60 minutes) and dried with absorbing paper.

The fragments were then fixed with aluminium stubs and their treated surfaces were positioned with upwards faces. Specimen coating with a gold-palladium layer was conducted by means of a sputtering device (SDC 050). The surfaces were examined under the scanning electron microscope EVO (Carl Zeiss, Oberkochen, Baden-Wuttemberg, Germany – Chemical Sciences Department, Ribeirão Preto School of Science and Letters, Ribeirão Preto, SP, Brazil), operating at 20 kV. A standardized series of photomicrographs was taken on representative areas, with different magnifications. A consensus was reached for selection of representative illustrations for each group.

Statistical Analysis

The temperature mean value of each specimen was analyzed using the Kolmogorov-Smirnov test and presenting normal distribution and homogeneity of variance. Thus, Analysis of Variance (ANOVA) was used. Statistical analysis was performed with SPSS software for Windows, version 12.0 (SPSS Inc., Chicago, IL, USA).

RESULTS

Results showed that temperature changes during CO₂ laser irradiation, using different powers in specimens of primary enamel was statistically similar among themselves, ($p \leq 0.05$), as shown in Table 1.

The initial temperature and thermal changes during CO₂ laser irradiation at specimens of primary enamel are shown in Figure 2, since the initial temperature in the environment was 25°C at all periods of the experiment. From this temperature was measured the thermal changes with different energy densities, in which it wasn't verified statistical difference between them.

SEM of primary enamel fragments irradiated with a CO₂ laser has demonstrated that the power of 0.5 W did not promote morphological changes in the irradiated surface (Figure 3A). Using power of 1.0 W were observed minor morphological changes related to the increase of rugosity on the surface of the irradiated primary enamel with CO₂ laser (Figure 3B). Morphological changes related to the formation of cracks on the surface of the irradiated primary enamel with CO₂ laser were verified with power 1.5 W (Figure 3C, yellow arrows).

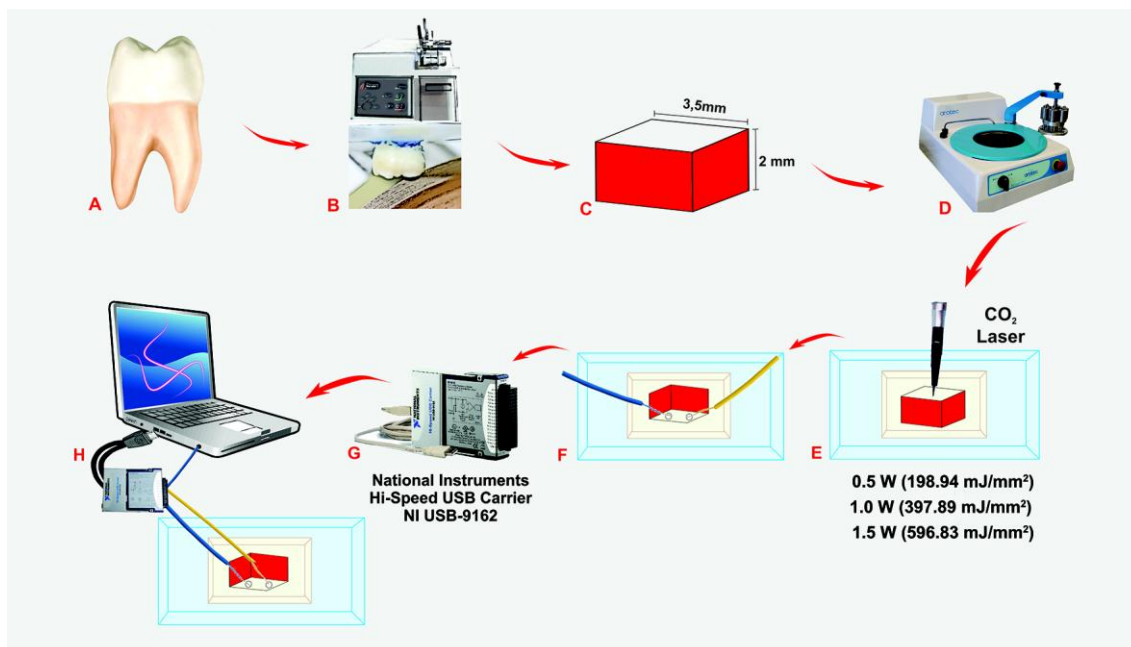


Figure 1 - Schematic design of the employed methodology.

A- Primary molar; B- Section machine and sectioned tooth; C- Specimens (3.5x3.5x2.0mm); D- Polisher; E- Fixation of specimens at acrylic plates, delimitation of irradiated area with insulating tape (4.0mm²), irradiation

with CO₂ laser; F- Holes to accommodate thermal-sensors (0.1mm depth); G- Acquisition plate; H- System connected to the computer.

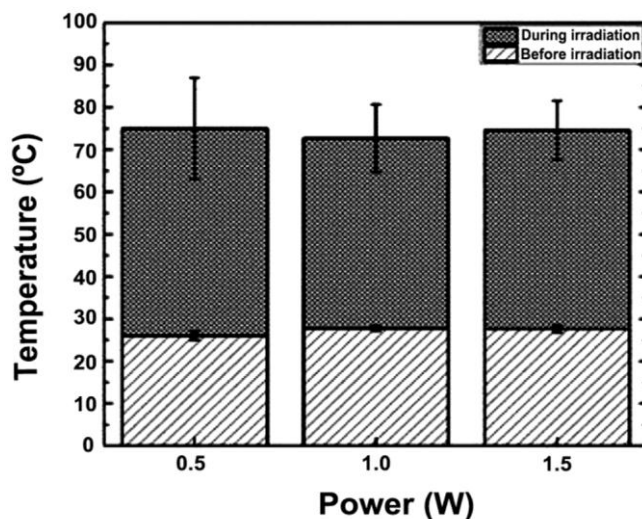


Figure 2 – Initial temperature analyzed before irradiation and thermal changes (°C) measured at specimens of primary enamel during the CO₂ laser irradiation.

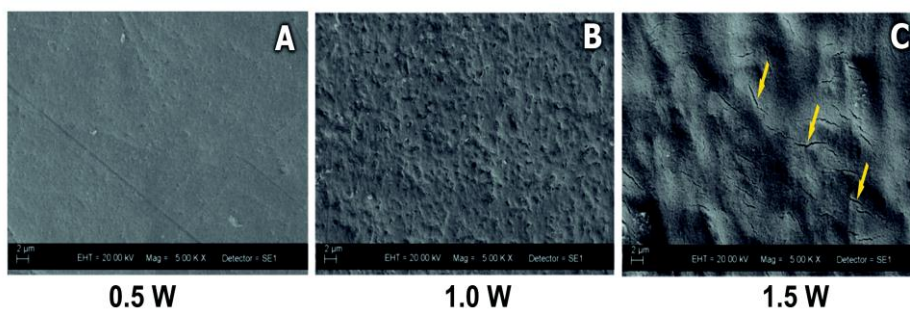


Figure 3- Scanning electron microscopy (SEM) of fragments irradiated with CO₂ laser using different powers.

Table 1 – Temperature changes (°C) in specimens of primary enamel, during CO₂ laser irradiation.

Different powers of CO ₂ laser	Mean ± SD
0.5 W (19.89 J/cm ²)	49.32 ± 11.9 a
1.0 W (39.78 J/cm ²)	44.75 ± 8.3 a
1.5 W (59.68 J/cm ²)	47.48 ± 6.9 a

Same letters indicate statistical similarity (p=0.05).

DISCUSSION

Inadequate knowledge of the physical properties of high power lasers by dentists and the generation of heat produced by the irradiation of the tooth surfaces can cause severe heat stress pulp^[15], thus, the pulp response to this thermal damage is directly related to the intensity of the damage.^[21] The increase in temperature should not exceed 5.5°C^[15,22], 11.2°C^[23] which would result in permanent / irreversible damage to dental pulp. In dentin of teeth mice irradiated with CO₂ laser^[24] the temperature exceeded 5.5°C pathways inducing pathological inflammatory cytokines and to repair damaged tissue pulp. In Macaca mulatta primate and beagle dog teeth^[25] irradiated with CO₂ laser, produces a fast and constant reactionary dentinogenesis without alteration of the necrotic pulp in animal tests.

This study compared changes in temperature during irradiation with different powers: 0.5 W (19.89 J/cm²), 1.0 W (39.78 J/cm²) and 1.5 W (59.68 J/cm²) in primary enamel teeth fragments using the CO₂ laser, where there was no found statistically significant difference.

The results of this study demonstrated that the temperature rise in the primary enamel teeth fragments using the CO₂ laser exceeded 5.5°C. One factor that may have contributed to thermal variation founded is the fact that irradiation with the CO₂ laser is carried out without the presence of air and water. SR-FTIR spectra of CO₂ laser irradiated enamel manifested asperities after laser ablation if a water-spray was not used and also indicated that such structures did not have an apatitic CaP phase composition.^[26] Such asperities are more susceptible to acid dissolution.^[26] The cooling method (air and water) significantly reduces the temperature during irradiation^[27-29], avoiding thermal damage to pulp^[28] and pulp necrosis.^[30] Although the results of this study shows that there was no statistically significant difference in the temperature of the tested groups, the SEM showed morphological changes in the primary enamel surface irradiated with an CO₂ laser. Using power of 1.0 W, rugosity was observed on the surface of the irradiated enamel cracks and crevices using power of 1.5 W.

On the other hand, the increase in temperature found in this study may be related to mineralization of primary teeth^[31] and the smallest thickness of the primary enamel.^[32] This same relationship was observed for^[33-36], which minor structures temperature variation was verified with a largest enamel and dentin thickness, furthermore, based on the mineral content alone, one would expect that the temperature rise on the surface of enamel would be a factor of two higher than for dentin.

This apparent contradiction can be explained partly on the basis of the higher reflectance of enamel 37% vs. 9% for dentin at 9.3 μm, therefore, 30% less of the incident laser energy is actually absorbed by enamel than for dentin. In addition, the thermal diffusivity is almost a factor of three higher for enamel than for dentin, 0.0047 vs. 0.0018 cm²/second.^[37]

The average internal temperature increases in the pulp chamber of permanent molars irradiated with the CO₂ laser was 37.46°C^[38], revealed extensive carbonization, isolated balls of recrystallized material, and the presence of smear layer at some dentinal tubule orifices for the CO₂ and Nd:YAG lased teeth observed by scanning electron microscopy. The pulp chamber temperature rise ranged from 0.5°C to 19°C and the SEM revealed crystal fusion in both enamel and dentin permanent molars irradiated with the CO₂ laser.^[39]

Irradiation with a CO₂ laser in third molars produce chemical and morphological changes and reduce the acid reactivity of enamel without compromising the pulp vitality^[40] and can ablate enamel safely without harming the pulp^[41] and remove micrometer layers while underlying tissues, especially the pulp, are safe from thermal effects.^[42]

The comparison of these results with those found in the literature it is difficult, since the only work published in the literature monitored thermal events occurring in the pulp chamber and on the tooth surface during direct exposure of the pulp chamber to CO₂ laser irradiation and hence to determine safe and effective parameters for its application in pulpotomy^[17] with maximum temperature increase of 16.93°C and of 15.70°C was observed on the enamel surface and within the pulp chamber respectively.

In the pulp chamber the temperature found after irradiation with the CO₂ laser may be different, due to the supporting structures present around the teeth and the blood flow of the pulp tissue dissipate this heat.^[43,44] The amount of applied energy and the time of high power lasers exposure in the irradiated tissue, also contribute to the increase of pulp temperature. High energy densities over short periods of time promotes less pulp damage^[33] since the thermal relaxation is inversely proportional to the square of the irradiated volume.^[43]

In young and deciduous teeth with open apices and large pulp chambers, the loose connective tissue and large blood supply of the pulp can better serve as a heat sink

than in mature teeth with smaller pulp chambers and closed apices.^[17]

The low thermal conductivity of the enamel and the rapid temperature drop in the lower layers of the irradiated enamel can also contribute to the lack of pulp damage due to the high absorption of this substrate by the appropriate wavelength of 10.6 μm CO₂ laser, making the dental enamel immune to thermal effects.^[42]

Thermal variations produced by using a CO₂ laser in dental enamel promote reduction of the water content and carbonate which is converted into phosphate followed by decomposition of proteins with temperatures of 100 - 650°C, thermal recrystallisation (650 and 1100°C) and phenomena destructive as fusion hydroxyapatite (> 1100°C).^[44,45]

CONCLUSION

Based on results obtained, it was concluded that:

- The CO₂ laser was not influenced by different powers (0.5W, 1.0W, 1.5W), used in the irradiation of primary enamel teeth fragments.

Conflict of interest: The authors declare that they have no conflicts of interest.

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