Root Surface Morphological Changes After Focused Versus Defocused CO_2 Laser Irradiation: A Scanning Electron Microscopy Analysis

A. Barone,* U. Covani,* R. Crespi,* and G.E. Romanos†

Background: Many studies have observed damages to root surfaces treated by CO_2 laser in continuous mode with a focused beam. The morphologic changes observed were always associated with temperature increase induced by high energy release.

Methods: The purpose of this study was to analyze by scanning electron microscopy (SEM) the effects of CO_2 laser in 2 different modes on root surfaces. Study samples consisted of 30 extracted single-rooted periodontally compromised human teeth. Root specimens were randomly assigned to 3 groups: group A (12) treated with CO_2 laser in continuous mode with a focused beam of 0.8 mm; group B (12) treated with CO_2 laser in pulsed mode with defocused beam of 4 mm; and group C (6), untreated controls.

Results: Group A (continuous mode) showed severe damages to dentin surfaces such as craters and fissures. Group B (defocused mode) did not result in any damages to the root surfaces, showing flat and smooth surfaces with apparent fusion of the smear layer and dentinal tubules almost completely sealed. The untreated control group was characterized by irregular and amorphous surfaces with several shallow depressions.

Conclusions: Although both laser modes resulted in changes to the treated root surface specimens, the changes resulting in a smooth surface from use of defocused pulsed beam may present an advantage in periodontal treatment. *J Periodontol 2002; 73:370-373.*

KEY WORDS

Comparison studies; lasers/adverse effects; tooth root.

* School of Dental Medicine, University of Genoa, Italy.
† Department of Oral Surgery, Dental School Frankfurt, Frankfurt, Germany.

Bacterial colonization of root surfaces is recognized as the primary cause of periodontal disease. Cementum from root surfaces exposed to bacterial colonization could act as a reservoir for bacterial endotoxins, causing further loss of attachment apparatus.[1-3]

Traditional treatment to detoxify periodontally diseased root surfaces is performed with scaling and root planing using hand instruments and/or ultrasonics.

Some authors reported that the CO_2 laser has a bactericidal activity[4] and could be useful in detoxification of dentin and alveolar bone.[5,6]

Several studies have shown dramatic changes on the lased root surface includings cracks, charring,[7-9] and crater formation with melting of the root surfaces.[10,11]

Anic et al.[12] observed craters with a fungoid-like and lace-like formation on the cementum on root surfaces following use of a CO_2 laser. Other authors evaluated the effects of Nd:YAG laser on root surfaces in vitro.[13] The results from this study showed a significantly lower number of fibroblast attachment in the endotoxin-treated samples exposed to laser compared to control and non-lased endotoxin-treated groups. The authors concluded that laser treatment could modify the biocompatibility of cementum to fibroblastic attachment. The same group[14] examined chemical changes at the root surfaces after Nd:YAG laser treatment using photoacoustic spectroscopy. They concluded that laser treatment could induce a reduction of the protein/mineral ratio and a contamination of root surfaces with protein by-products which may inhibit fibroblast adhesion. The changes in the root surface after Nd:YAG laser irradiation may be extensive and dependent on the power setting.[15]

Laser treatment might cause thermal side effects with morphologic and chemical modifications of dental hard tissues. Shariati et al.[16] have studied the effects of CO_2 laser on root surfaces in continuous mode with a beam size of 1 mm. This laser treatment, using different power energies and exposure times, induced charred craters and an apparent melting of dentin. The increase in power setting and exposure time resulted in an increase in the width and depth of damages to dental hard tissues.

Most of the studies reporting extensive changes to tooth structure used the CO_2 laser in continuous focused mode. The use of pulsed laser may induce less thermal damages to root surfaces, because the tissue cools down between the pulses. The spot size seems to influence the power density. The larger the spot, the greater the power required to maintain the same power density.

The purpose of the present study was to observe and compare in vitro 2 types of laser treatment, continuous focused mode and pulsed defocused mode, on dental hard tissues using scanning electron microscopy (SEM).

MATERIALS AND METHODS

Sample Collection and Preparation

Thirty freshly-extracted single-rooted human teeth without crown or root fractures were used in this study. The teeth were periodontally compromised and selected based on the following criteria: 1) sites with probing depth (PD) >6 mm and attachment level (AL) >5 mm; 2) no visible root fractures or anatomical abnormalities of the root; 3) root surfaces carious free and not endodontically involved; and 4) absence of restorative materials.

Immediately after extraction, teeth were scaled with ultrasonic scaler[‡] to remove soft tissues and plaque.

Thirty root specimens (5 × 5.0 mm and 3.0 mm thick) were cut longitudinally with a water cooled-low speed handpiece using a diamond bur. Each specimen was prepared from each root surface of one extracted tooth.

Treatment Groups

Specimens were randomly assigned to 1 of the 3 groups: group A (12) treated with a CO_2 laser[§] in a focused (0.8 mm spot size) continuous mode with 8.0 W power setting; group B (12) treated with the same laser in non-focused (4.0 mm spot size) pulsed mode with a frequency of 4.0 Hz and 2.0 W of power setting; and group C (6) untreated controls.

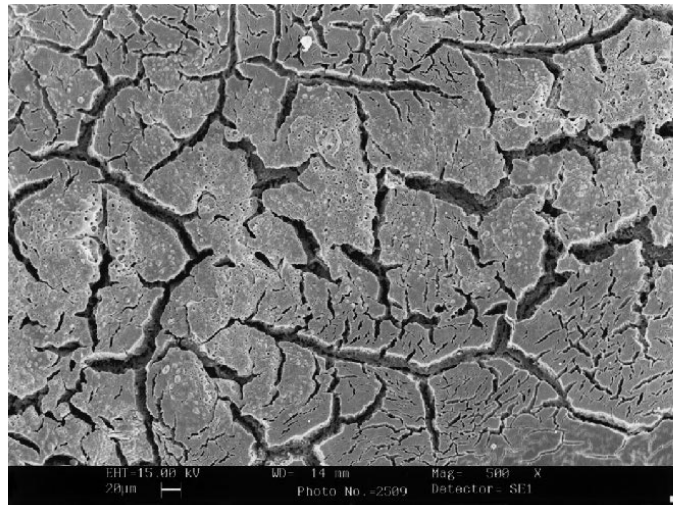

Figure 1.
Severe damages of root surface after CO_2 laser exposure in continuous focused mode (original magnification, 500×).

Scanning Electron Microscopy Analysis (SEM)

Specimens were fixed with a 2% glutaraldehyde solution in phosphate buffered saline (PBS). Fixed specimens were then dehydrated in an ascending order of ethanol for 30 minutes. After dehydration in hexametyldisiloxane for 30 minutes, the fixed and dehydrated specimens were sputter coated before SEM examination. All specimens were then examined and photographed with SEM[‖] by 2 examiners blinded to treatment.

RESULTS

All root specimens treated in the continuous focused mode (group A) exhibited zones of heat cracking, fissuring, and pronounced roughness under SEM examination. The central area of the lased surfaces was characterized by melted mineral surface (Fig. 1) alternating with fissures and craters. This mode of treatment induced severe changes of the dentin layer and no remnants of bacterial cells were observed by SEM.

The surfaces of specimens treated by laser in pulsed non-focused mode (group B) showed areas without cementum with completely sealed dentinal tubules (Fig. 2). The dentin appeared as a melted layer (Fig. 3), showing a flat and smooth surface with an apparent fusion of the surface of the smear layer. This characteristic feature of the dentin layer gave the appearance of a "glazed dentin," the dentinal tubule orifices appeared almost completely plugged (Fig. 4). No remnants of bacterial cells were observed after the laser irradiation. In contrast, the untreated specimens (group C) were substantially different compared with lased specimens, exhibiting a relatively irregular and amor-

‡ Piezon Master 400, EMS, Milan, Italy.
§ Elan Co., Florence, Italy.
‖ Leo 420, Electron Microscopy, Frankfurt, Germany.

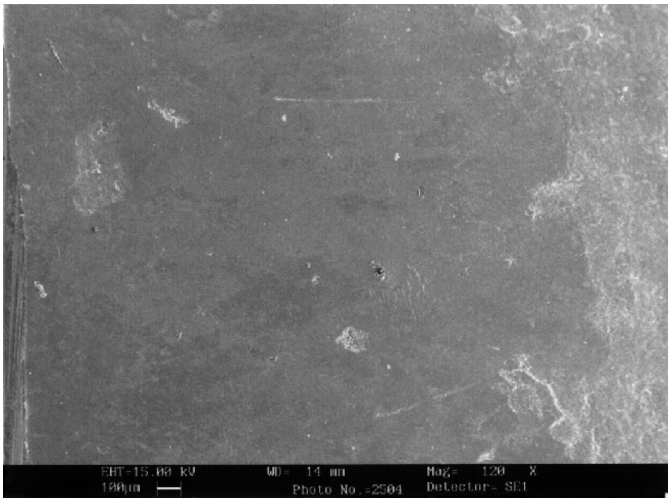

Figure 2.
Root surface exposed to CO_2 laser in pulsed defocused mode showing melted root surface (original magnification, 120×).

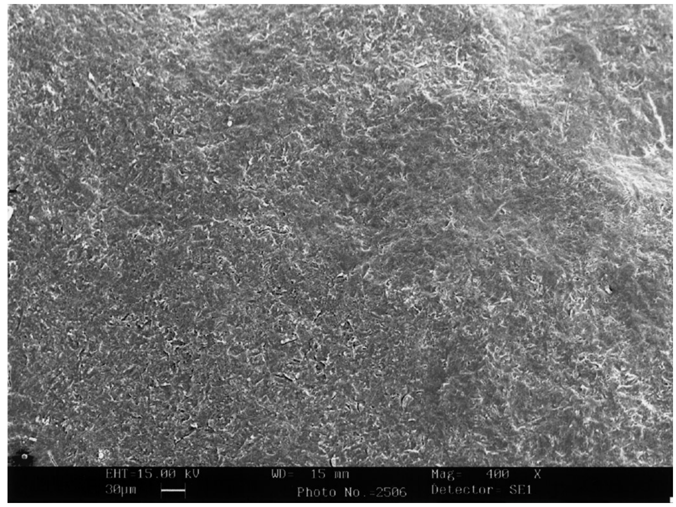

Figure 3.
Root surface treated by laser in pulsed defocused mode. Melt-down smooth surface is observed (original magnification, 400×).

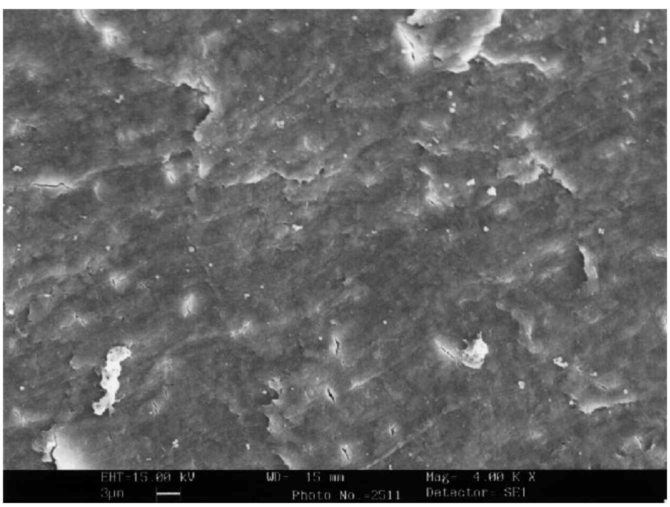

Figure 4.
Root surface treated by laser in pulsed defocused mode showing sealed dentinal tubules (original magnification, 4000×).

phous surfaces characterized by several shallow depressions. Such areas showed isolated islands of microbial plaque deposits composed of cocci and rods as well as a small number of collagen fibrils.

DISCUSSION

Several authors have reported observing severe damages to dentin surfaces after Nd:YAG and CO_2 laser exposure. Tseng and Liew[17] in an in vitro study noted heat penetration and damage on cementum and dentin surfaces after laser treatment. Morlock et al.[10] used a Nd:YAG laser on root surfaces followed by root planing and observed characteristic morphologic changes such as charring, carbonization, fissures, and craters. These morphologic changes to the root surface after the Nd:YAG laser irradiation may be dependent on the power setting used and not on the laser application.[15] Furthermore, Nd:YAG laser irradiation may have a negative effect on fibroblast attachment on the root surface.[10,13,18]

Shariati et al.[16] studied the effects of a CO_2 laser at 2 power levels and 6 exposure times on root dentin. The observations showed surface cratering and fissures followed, at a deeper layer, by melting of the dentin and partial sealing of the tubules. The authors noted that with the increase of the exposure time the craters quickly expand causing severe changes of the tooth structures.

As with the results of the above studies, SEM analysis from our study revealed that the continuous laser mode with a focused beam caused severe changes to the dentin surfaces such as cracks, fissures, and pronounced roughness. In contrast, the pulsed laser mode with a defocused beam did not induce any extensive modifications to root surfaces. The surface of experimental specimens which received CO_2 laser irradiation in defocused pulsed mode showed an almost completely melted dentin layer. The scattering of the root surface in the pulsed mode is more homogeneous without deep craters or melting due to thermal damage, which has been observed after use of the continuous mode with the focused beam. SEM observations revealed an absence of residual bacterial cells on all laser irradiated specimens; specimens that did not receive laser treatment showed some residual bacterial cells probably because of insufficient initial mechanical instrumentation with the ultrasonic scaler. These findings were in agreement with results from other studies.[19,20] Misra et al.[21] observed that CO_2 laser irradiation at low power setting (3 watts) for 1 second was able to expose dentinal tubule orifices removing the smear layer without extensive damage to the root surfaces. The smear layer, normally obtained after root planing, always contains residual dental calculus as well as microbial plaque bacterial endotoxins[19,22] which negatively affect new connective tissue attachment. Conditioning of the root surfaces by different methods has been recommended as an adjunctive tool in periodontal therapy to enhance regeneration of periodontal tissues.[23,24]

Adriansen et al.[3] showed bacterial invasion in the root cementum and dentinal tubules of periodontally diseased caries-free human teeth. Since this bacterial invasion cannot be eradicated by mechanical debridment alone and may be, in part, responsible for a rapid recolonization of periodontal treated sites, it would appear necessary to combine mechanical therapy with the use of adjunctive techniques.

The use of CO_2 laser has been suggested for detoxification of titanium surface in vivo.[25] Mouhyi et al.[26] examined different power settings of the CO_2 laser for optimal detoxification: the laser was used in pulsed mode at 8.0 W with pulse duration of 10 ms and repetition rate of 20 Hz to detoxify the titanium surface keeping the temperature below the critical levels of bone damage.

In conclusion, the findings presented from this study show that laser irradiation in the pulsed mode with a defocused beam induced some morphologic changes without causing extensive changes in the root surface. The role of CO_2 laser as an adjunctive tool in periodontal therapy should be investigated with more extensive and well-controlled studies. Further research must be conducted to test attachment of fibroblasts to the root surfaces after CO_2 laser irradiation.

REFERENCES

1. Hatfield CG, Baumhammers A. Cytotoxic effects of periodontally involved surfaces of human teeth. *Arch Oral Biol* 1971;16:464-468.
2. Aleo JJ, DeRenzis FA, Farber PA, Varboncoeur AP. The presence and biologic activity of cementum bound endotoxin. *J Periodontol* 1974;45:672-675.
3. Adriansen PA, Edwards CE, DeBoever JA, Loesche WJ. Ultrastructural observations on bacterial invasion in cementum and radicular dentin of periodontally diseased human teeth. *J Periodontol* 1988;59:493-503.
4. Zakariasen KL, Dederich DN, Tulip J, DeCoste S, Jensen SE, Pickard MA. Bactericidal action of carbon dioxide laser radiation in experimental dental root canals. *Can J Microbiol* 1986;32:942-946.
5. Miserendino LJ. The laser apicoectomy: endodontic application of the CO_2 laser for periapical surgery. *Oral Surg Oral Med Oral Pathol* 1988;66:615-619.
6. Miserendino LJ, Pick R, Kos W. In vitro sterilizations of dental caries by CO_2 laser irradiation. *Laser Surg Med* 1988;8:148(Abstr. 26).
7. Wigdor H, Abt E, Ashrafi S, Walsh Jr JT. The effect of lasers on dental hard tissues. *J Am Dent Assoc* 1993; 124:65-70.
8. de Road M, Paschoud Y, Holz J. Action of CO_2 lasers on dental tissues. *J Biol Buccale* 1988;16:137-150.
9. Watanabe I, Lopes RA, Brugnera A, Katayama AY, Gardini AE. Effect of CO_2 laser on class V cavities of human molar teeth under a scanning electron microscope. *Braz Dent J* 1996;7:27-31.
10. Morlock BJ, Pippin DJ, Cobb CM, Killoy WJ, Rapley JW. The effect of Nd:YAG laser exposure on root surfaces when used as an adjunct to root planning: An in vitro study. *J Periodontol* 1992;63:637-641.
11. Read PP, Baumgartner JC, Clark SM. Effects of a carbon dioxide laser on human root dentine. *J Endod* 1995; 21:4-8.
12. Anic I, Dzubur A, Vidovic D, Tudja M. Temperature and surface changes of dentine and cementum induced by CO_2 laser exposure. *Int Endodont J* 1993;23:284-293.
13. Trylovich DJ, Cobb CM, Pippin DJ, Spencer P, Killoy WJ. The effects of Nd:YAG laser on in vitro fibroblast attachment to endotoxin-treated root surfaces. *J Periodontol* 1992;63:626-632.
14. Spencer P, Trylovich DJ, Cobb CM. Chemical characterization of lased root surfaces using fourier transform Infrared photoacoustic spectroscopy. *J Periodontol* 1992;63:633-636.
15. Romanos GE, Renner PJ, Everts H, Nentwing GH. Veranderungen an der Wurzeloberflache nach Anwendung eines Nd:YAG-Laser. Eine in vitro Untersuchung. *Quintessenz* 1998;49:497-500.
16. Shariati S, Pogrel MA, Marshall GW, White JM. Structural changes in dentin induced by high energy, continuous wave carbon dioxide laser. *Lasers Surg Med* 1993;13: 543-547.
17. Tseng P, Liew V. The potential applications of a Nd:YAG dental laser in periodontal treatment. *Periodontol* (Australia) 1990;11:20-22.
18. Thomas D, Rapley J, Cobb C, Spencer P, Killoy W. Effects of the Nd:YAG laser and combined treatments on in vitro fibroblast attachment to root surfaces. *J Clin Periodontol* 1994;21:38-44.
19. Blomlof JPS, Blomlof LB, Lindskog SF. Smear removal and collagen exposure after non-surgical root planning followed by etching with an EDTA gel preparation. *J Periodontol* 1996;67:841-845.
20. Tucker D, Cobb CM, Rapley JW, Killoy WJ. Morphologic changes following in vitro CO_2 laser treatment of calculus-ladened root surfaces. *Lasers Surg Med* 1996; 18:150-156.
21. Misra V, Mehrota KK, Dixit J, Maitra SC. Effects of carbon dioxide laser on periodontally involved root surfaces. *J Periodontol* 1999;70:1046-1052.
22. Blomlof J, Jansson L, Blomlof L, Lindskog S. Etching at neutral pH promotes periodontal healing. *J Clin Periodontol* 1996;23:50-55.
23. Terranova V, Wikesjö UM. Extracellular matrices and polypeptide growth factors as mediators of functions. *J Periodontol* 1987;58:371-380.
24. Lynch SE. Methods for evaluation of regenerative procedures. *J Periodontol* 1992;63:1085-1092.
25. Coffelt DW, Cobb CM, MacNeill S, Rapley JW, Killoy WJ. Determination of energy density threshold for laser ablation of bacteria. *J Clin Periodontol* 1997;24:1-7.
26. Mouhyi J, Sennerby L, Nammour S, Guillaume P, Van Reck J. Temperature increase during surface decontamination of titanium implants using CO_2 laser. *Clin Oral Implants Res* 1999;10:54-61.

Accepted for publication October 22, 2001.