Titanium Deposition After Peri-implant Care with the Carbon Dioxide Laser

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Purpose: *Titanium endosseous implants are becoming increasingly important in dentistry because of their excellent long-term results. However, it has been reported that these implants may lead to higher concentrations of titanium, especially in the lungs and kidneys. The purpose of this study, therefore, was to determine whether CO_2 laser–assisted decontamination of exposed implant surfaces is associated with an increase in titanium release.* ***Materials and Methods:*** *In 6 beagle dogs, a total of 60 implants were placed. After osseointegration and second-stage surgery, peri-implantitis was induced by cotton floss ligatures for 12 weeks. Surgical treatment consisted of granulation tissue removal, including decontamination of the implant surface with 3 different methods. Twenty implants were decontaminated conventionally by an air-powder abrasive for 60 seconds. Another 20 implants were decontaminated by laser treatment alone. The last 20 implants were treated conventionally by air-powder abrasive and then lased. Four months later, fresh tissue samples of various tissues were evaluated by histologic and chemical analysis.* ***Results:*** *Quantitative analysis indicated that titanium accumulation could be detected, especially in the spleen, liver, oral mucosa, regional lymph nodes, lung, and kidney in the beagle dog model.* ***Discussion:*** *The concentrations found did not exceed those previously reported in the literature.* ***Conclusion:*** *These results support the hypothesis that CO_2 laser–assisted therapy of ailing implants will not result in excessive titanium concentrations in tissues. Accordingly, CO_2 lasers appear suitable and safe for peri-implant gingival surgery.* (Int J Oral Maxillofac Implants 2002;17:707–714)

Key words: *dental implants, lasers, peri-implantitis*

Endosseous implants are becoming increasingly important in dentistry. Their excellent long-term results are attributable to a superficial titanium dioxide layer (TiO_2), which prevents corrosion of the metal in the body.[1–3] However, Schliephake and coworkers[4,5] reported that machined dental implants caused increased titanium loads in the organs of mini-pigs. Moreover, Weingart and coworkers[6] noted that titanium was found in the regional lymph nodes of beagle dogs after the placement of plasma spray-coated implants. Fischer-Brandies and coworkers reported that titanium osteosynthesis in the maxillofacial region resulted in increased peri-implant titanium accumulation.[7] It was proposed that microabrasion could be a potential cause of titanium release.[4–7] It is also well known that thermal laser energy used for decontaminating ailing implants may alter the superficial structure and chemistry of implant surfaces.[3,8–10] Accumulated laser heat on the implant surface can cause chemical reactions, converting TiO_2 to compounds that are

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Table 1 Therapies Performed in Dogs

Dog no.	Right hemimandible	Left hemimandible
4118	Group 3	Group 1
4333	Group 3	Group 2
4332	Group 1	Group 2
4132	Group 2	Group 3
4322	Group 2	Group 1
4320	Group 1	Group 3

Group 1 = Conventional decontamination (Prophy-Jet); group 2 = laser-assisted decontamination; group 3 = laser-assisted decontamination after using the Prophy-Jet.

less resistant to microabrasion and chemical aggression in the body.[10,11] Because of that, laser parameters chosen for the decontamination process should not destroy the superficial TiO_2 layer, which plays a significant role in the healing and generation of host tissue cells adjacent to the metallic implant.[1]

Nevertheless, comparatively little is known about the titanium release of laser-irradiated titanium implants in the body. Therefore, the aim of the present study in 6 beagle dogs was to determine whether laser irradiation of ailing implants will result in an increased release of titanium, which could cause higher titanium presence in the organs of the body.

MATERIALS AND METHODS

Dental Laser

The CO_2 laser (λ = 10.6 µm) employed was model 20 C manufactured by the Sharplan Company (Freising, Germany). This laser has a power output range from 1 to 20 W and can be operated in continuous, pulsed, or so-called superpulsed modes of laser beam delivery. In the superpulse mode, mean power is generated by increasing the frequency of pulses; the energy of each single superpulse is 20 mJ.

In addition, an accessory system, the Swiftlase scanner (Sharplan), was used. The scanner allows the focused CO_2 laser beam to sweep in 0.1 second over an area with a diameter of 3.0 mm, thereby reducing the heat accumulation on the lased tissue.

Energy Dispersive X-ray and Electron Spectroscopy for Chemical Analysis

Forty new standard plasma spray-coated Frialit-2 titanium implants (Friadent AG, Mannheim, Germany), with a length of 11 mm and a diameter of 3.8 mm, were provided for tests on the superficial chemical compound of lased titanium surfaces using energy dispersive x-ray (EDX) and electron spectroscopy for chemical analysis. In these implants, a maximum surface roughness of 61 µm was identified in a previous study by metallographic cuts.[10] With all other factors held constant, rough implants would be likely to render an increase in the ionic leakage as compared to machined implants, especially after laser irradiation.

The implants were irradiated with different powers ranging from 1 to 5 W in 0.5-W steps in both continuous wave mode and superpulse mode. All irradiations were applied for 5 and 10 seconds.

EDX allows detection, in particular, of nitrogen and oxygen in the superficial implant layer. Moreover, electron spectroscopy for chemical analysis enables quantitative detection of atomic concentrations related to the sputter depth. Therefore, an x-ray beam stimulates the emission of photo-electrons; their energy is characteristic for each element detected. Relative intensities of the energies detected allow a quantitative analysis of the elements.[12,13]

In Vivo Study

Six 2-year-old female beagle dogs from the same pedigree were used in this study. Five 11-mm-long titanium plasma spray-coated Frialit-2 implants with a diameter of 3.8 mm were placed bilaterally in each animal in the premolar and molar region of the mandibles (n= 60 implants overall). The implants were uncovered 3 months after placement. After 4 weeks of oral hygiene, cotton floss ligatures were positioned around the implants.[14] Gross plaque accumulation around the implants was undisturbed for 3 months, which resulted in circumferential peri-implant bone defects.

Surgical treatment consisted of granulation tissue removal, including decontamination of the implant surface with 3 different methods. Twenty implants (group 1) were decontaminated conventionally by an air-powder abrasive,[14] the Prophy-Jet, for 60 seconds. Another 20 implants (group 2) were decontaminated by laser treatment alone (continuous wave, 2.5 W, duration of 6 times 10 seconds). The last 20 implants (group 3) were treated conventionally by the Prophy-Jet for 60 seconds and then lased with the specified parameters for another 60 seconds.

In each hemimandible, only 1 mode of treatment was performed (Table 1). Consequently, each treatment method was carried out in 4 hemimandibles, resulting in a total of 20 implants per group.

Histology and Determination of Titanium

The animals were sacrificed 4 months after treatment. Fresh specimens of oral mucosa, regional lymph nodes, spleen, liver, lung, and kidney were

Table 2 Titanium Concentration (µg/g Fresh Weight)

	Group 1 (n = 4)				Group 2 (n = 4)				Group 3 (n = 4)			
Location	Mean	Min	Max	SD	Mean	Min	Max	SD	Mean	Min	Max	SD
Local oral mucosa	0.74	0.12	1.48	0.68	1.69	0.16	4.88	2.2	2.79	0.11	8.78	4.04
Regional lymph nodes	1.85	0.17	4.77	2.53	1.51	0.54	3.33	1.28	2.02	0.27	6.06	2.71

Group 1 = Conventional therapy; group 2 = laser-assisted therapy; group 3 = combined therapy.

obtained for both histologic and chemical analysis. For the histologic evaluation, standard procedures for soft tissue examination were used.

The titanium concentration in the fresh specimens was determined chemically by the ICP-AES technique (inductively coupled plasma atomic emission spectroscopy).[13] Tissue samples were freeze-dried, mixed with 1 mL of a suprapure solution of nitric acid, and then incinerated to ash in a quartz tube at a temperature of 170°C for 8 hours. Finally, the contents of titanium determined were converted to concentrations of the fresh tissue (µg titanium/g fresh weight).

Statistical Analysis

Statistical analysis was performed using a commercial computer program (Microsoft Excel, version 97, Munich, Germany). Data are presented as maxima, minima, and means ± standard deviation. A P value less than .01 in Student 2-sample t test was considered to indicate statistical significance.

To understand the correlations that were performed, it should be kept in mind that in each beagle dog, the 2 hemimandibles were treated with a different method (Table 1). Accordingly, correlations between the 3 different methods were possible only with regard to the oral mucosa and the regional lymph nodes, ie, tissues that could be definitely assigned to 1 treatment modality (Table 2).

For further analysis of titanium concentrations in the oral mucosa and the regional lymph nodes after conventional or laser treatment, all laser-treated specimens in group 2 and 3 together were compared to the conventionally treated specimens in group 1 (Table 3).

Visceral organs such as the lung, liver, spleen, and kidney could not be assigned to a specific treatment modality since both hemimandibles of each dog were treated in 2 different ways. Therefore, titanium concentrations of visceral organs were only compared descriptively.

Table 3 Titanium Concentration (µg/g Fresh Weight)

	Conventional treatment (group 1, n = 4)				Laser treatment (groups 2 and 3, n = 8)			
Location	Mean	Min	Max	SD	Mean	Min	Max	SD
Local oral mucosa	0.74	0.12	1.48	0.68	2.24	0.11	4.88	3.07
Regional lymph nodes	1.85	0.17	4.77	2.53	1.76	0.27	6.06	1.98

Group 1 = Conventional therapy; group 2 = laser-assisted therapy; group 3 = combined therapy.

RESULTS

Energy Dispersive X-ray and Electron Spectroscopy for Chemical Analysis

The analysis of implant surfaces irradiated in superpulse mode revealed in all cases titanium, oxygen, carbon (C1s.Is1), and nitrogen. Thickness of the superficial TiO_2 layer was increased considerably, to a maximum of 100 nm (Fig 1).

The surface sections of all continuous wave–irradiated plasma spray-coated titanium implants showed a chemical compound similar to that of superpulse-irradiated references. In contrast, the thickness of the superficial TiO_2 layer was increased only to a maximum of 50 nm (Fig 2).

Titanium Concentration of Tissues

To determine whether there was a correlation between therapy and titanium concentration of the local tissues, in every treatment group the mean values of titanium concentrations in the oral mucosa and regional lymph nodes were evaluated (Table 2). Even though the mean values of the titanium concentration in the oral mucosa were clearly higher in both laser groups than in the conventionally treated group, there was no statistically significant difference because of the wide range of values. Furthermore, overall there was no statistically significant

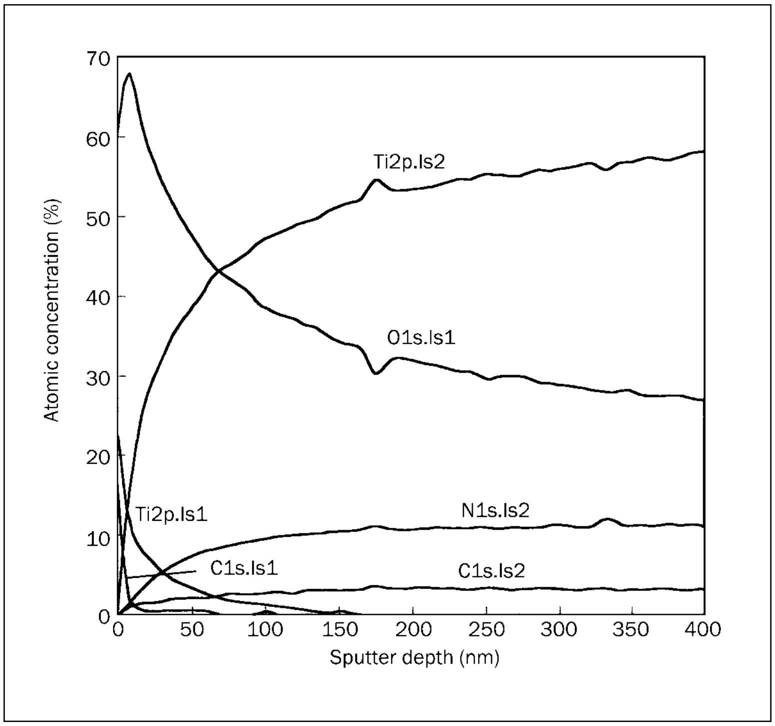

Fig 1 Chemical analysis of the surface of a CO_2 laser–irradiated dental implant (superpulse mode, P = 2.5 W, T = 10 s).

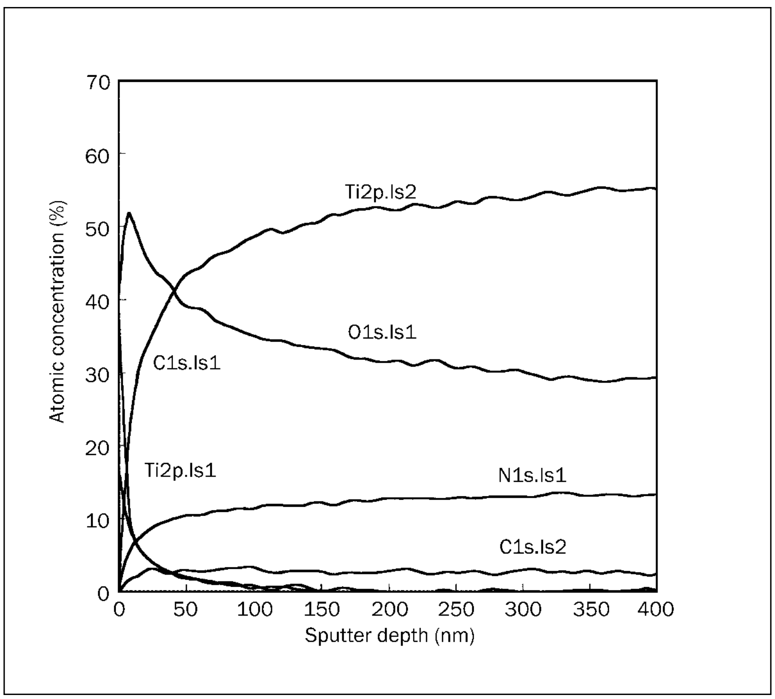

Fig 2 Chemical analysis of the surface of a CO_2 laser–irradiated dental implant (continuous wave, P = 2.5 W, T = 10 s).

difference between laser-treated specimens as compared to non–laser-treated specimens (Tables 3 and 4). Thus, there was no correlation between the titanium concentration of local tissues and the method of treatment used.

Whether or not there was a significant difference in the titanium concentration of visceral organs in dogs that had undergone laser irradiation on one or both hemimandibles could not be evaluated because of the small sample size. However, laser treatment on both sides seemed to yield the highest titanium concentrations in the lungs, whereas the highest values after irradiation on one side were found in the liver (Table 5). Accordingly, there were no correlations between titanium concentrations in organs and the quantity of laser-treated implants.

Table 4 Statistical Analysis According to Student 2-sample *t* Test

Tissues and groups compared	Mean ± SD	n	*t* value	Difference significant?
Oral mucosa				
Group 2/group 1	1.69 ± 2.20/0.74 ± 0.68	n = 4/n = 4	0.74	No
Group 3/group 1	2.79 ± 4.04/0.74 ± 0.68	n = 4/n = 4	0.87	No
Groups 2 and 3/group 1	2.24 ± 3.07/0.74 ± 0.68	n = 8/n = 4	1.29	No
Regional lymph nodes				
Group 2/group 1	1.51 ± 1.28/1.85 ± 2.53	n = 4/n = 4	–0.46	No
Group 3/group 1	2.02 ± 2.71/1.85 ± 2.53	n = 4/n = 4	0.11	No
Groups 2 and 3/group1	1.76 ± 1.98/1.85 ± 2.53	n = 8/n = 4	–0.12	No

Although the titanium concentrations in the oral mucosa seemed to be substantially greater in the laser-treated specimens compared to non-laser treated specimens, no statistical differences could be detected. Group 1 = Conventional therapy; group 2 = laser-assisted therapy; group 3 = combined therapy.

Table 5 Titanium Concentration (µg/g Fresh Weight)

	Laser on one side (n = 4)				Laser on both sides (n = 2)			
Location	Mean	Min	Max	SD	Mean	Min	Max	SD
Lung	0.71	0.64	0.80	0.07	3.03	0.98	5.08	2.89
Liver	2.84	0.33	7.37	3.10	0.37	0.30	0.44	0.09
Spleen	0.29	0.16	0.49	0.17	EF	EF	EF	EF
Kidney	1.86	0.14	7.41	2.97	0.18	0.10	0.30	0.09

EF = evaluation failed.

Table 6 Titanium Concentration (µg/g Fresh Weight)

	Spleen	Liver	Oral mucosa	Regional lymph nodes	Lung	Kidney
Mean (n = 6)	4.14	2.01	1.83	1.78	1.48	1.30
Minima	0.16	0.32	0.11	0.17	0.64	0.1
Maxima	15.7	7.37	8.78	4.77	5.08	7.41
SD	7.7	2.72	2.68	2.01	1.76	2.51

Mean values of titanium concentrations in all organs of all 6 dogs were evaluated. As can be seen in Table 6, the maximum concentration was found in the spleen, followed by the liver, oral mucosa, regional lymph nodes, lung, and kidney. This order was the result of an extremely wide range of the titanium concentrations in the spleen. It should also be noted that wide ranges were also found in all other tissues.

Histologic Findings

All histologic sections of all 6 dogs showed regular microanatomic structures. Signs of neither inflammation nor dysplasia could be detected in the oral mucosa, regional lymph nodes, spleen, liver, lung, and kidney. Even though titanium was found in all organs by the ICP-AES technique, no titanium-related pigment was found in tissues, either in the regional lymph nodes or in the liver (Fig 3), or in the lungs after laser treatment on one or on both hemimandibles (Fig 4).

DISCUSSION

The purpose of this study was to determine whether CO_2 laser irradiation might enhance the amount of titanium release from dental implants, because recent literature has implied that the thermal influence on titanium can have adverse effects on its biocompatibility.[2,3] Keller and coworkers[3] demonstrated that autoclaving commercially pure titanium

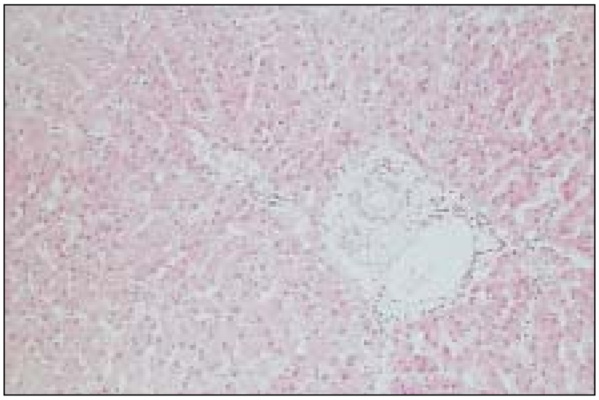

Fig 3 Section of a liver of a beagle dog after laser-assisted peri-implant care on both hemimandibles. No signs of alterations were evident, although a high concentration of titanium was found (hematoxylin-eosin; magnification ×50).

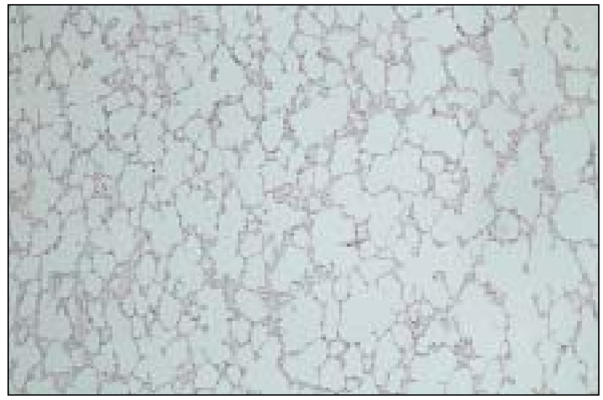

Fig 4 Section of a lung of a beagle dog after laser-assisted peri-implant care on both hemimandibles. No metallic particulates were seen, although a high concentration of titanium was found (hematoxylin-eosin; magnification ×20).

specimens caused an increase in the thickness of the superficial titanium oxide layer from 3 nm to 25 nm. On such surfaces, human fibroblasts showed significantly less adhesion than on titanium surfaces that were sterilized by ethylene oxide (sterilization with ethylene oxide caused an increase in the superficial titanium oxide layer from 3 nm to 5 nm). Although the authors could not explain why the fibroblasts behaved in a different manner, it was concluded that the thermal influence on titanium can have biologic consequences. On the other hand, with passing years, the superficial oxide layer grows in the body. McQueen and coworkers have shown that the oxide layer grew from 5 nm to 200 nm over a period of 6 years.[12]

Since titanium is known to be a very reactive metal under atmospheric conditions, laser irradiation could generate a thickened layer of titanium oxides on the implant. These oxides, again, are less resistant to mechanical stress,[11] thereby possibly imposing a higher titanium load on the tissues. According to chemical analysis of the irradiated implant surfaces, continuous wave irradiation does not appear to exert adverse effects on the surface chemistry. Superpulse irradiation, however, resulted in sufficient heat accumulation to melt the surface of the rough plasma-sprayed titanium. Thus, because of the chemical reaction with an atmospheric environment, the titanium oxide layer can be enlarged by about 50 times. Consequently, it seems that superpulse irradiation has a significant influence on the surface chemistry, which is not desirable for the purpose of decontaminating ailing implants.

Quantitative data (Figs 1 and 2) showed that laser-irradiated implant surfaces can also become enriched in nitrogen. Nevertheless, there is no danger in creating a laser-based reaction resulting in new compounds such as nitrosamines, which are known to be mutagenic and possibly carcinogenic. In this study, there was no evidence of nitrosamines after laser irradiation. Laser-irradiated zones were still covered by a titanium dioxide layer (TiO_2), which confers biocompatibility. The source of the nitrogen could be from the cooling gas (compressed air). On the other hand, grade 2 titanium normally has a nitrogen content of up to a maximum of 0.03 mass%. Moreover, it is well known from the literature that nitrogen is always found in electron spectroscopy for chemical analysis (ESCA), originating from organic molecules that are absorbed during the production of the implants.[15]

Special consideration must be given to laser-char deposits, which might have a mutagenic effect on contacting intraoral tissues over the long term. Even though the wavelength of the CO_2 laser (λ = 10.6 µm) is absorbed in water to a significant extent, char-free ablation is still not possible. On the other hand, ablation of intraoral premalignant lesions with this type of laser has been an established procedure for more than 20 years[16] and is still state of the art.[17] Consequently, CO_2 (λ = 10.6 µm) laser-char deposits do not appear to have mutagenic effects on contacting intraoral tissues over the long term.

Concerning titanium levels in the tissues, it has been stated that in mini-pigs the lungs seem to play an important role in the accumulation of titanium.[4,5] In the present study, titanium concentrations in local tissues such as oral mucosa and regional lymph nodes were not statistically significantly increased even after laser irradiation on both sides (Table 4). In contrast, very high titanium concentrations were found in the lungs of dogs with laser irradiation on both sides (Table 5). Since surgery was performed with endotracheal intubation, aspiration of small particulates or condensation of vapor is unlikely. Therefore, these concentrations must be attributed to the wide interindividual range of titanium tissue concentrations mentioned above. This assumption is supported by the unremarkable histology of the lungs, which showed no pigmented granulates (Fig 4).

In another study in beagle dogs, titanium concentrations of almost all organs were under the detection limit, ranging from 0.01 to 0.21 µg/g.[6] However, titanium was found in 12 of 19 regional lymph nodes, with concentrations between 0.16 and 9.0 µg/g.[6] In the present study, titanium concentrations in the visceral organs ranged from 0.14 to 7.41 µg/g; the concentrations in the regional lymph nodes ranged from 0.11 to 8.78 µg/g. Thus, these results are very similar to those of Weingart and coworkers concerning titanium levels in the lymph nodes, but they exceed those of the visceral organs.[6] Nevertheless, these levels are still lower than those determined by Schliephake and associates in the mini-pig, which had resulted only from placing dental implants alone without any decontamination procedure.[4,5] Therefore, the titanium levels after CO_2 laser decontamination cannot be regarded as increased, even though rough plasma-sprayed implants were used in this study. In addition, these outcomes support the results of Schliephake and associates, which were criticized by Steinemann[18] for presenting unrealistically high values of titanium leakage. To enhance clarity, Schliephake and associates stressed in their response that the criticism was not justified since the discrepancy could be accounted for by disparate titanium concentrations in fresh or freeze-dried specimens.[19]

With regard to the methods of treatment used, it is important to note that none of the histologic sections showed titanium-related pigments or any alterations caused by continuous wave CO_2 laser irradiation of the implants. Since none of the implants were loaded, microabrasion was almost impossible. Therefore, this finding is not surprising. Nevertheless, it can be concluded from the present chemical and histologic results that laser irradiation in continuous wave mode generates a titanium oxide layer very similar to that in the physiologic situation.

CONCLUSION

Laser decontamination did not raise the titanium concentration in the oral mucosa and the regional lymph nodes compared to the conventional decontamination technique. Titanium concentrations in visceral organs were also not increased in dogs that were irradiated on both sides, compared to dogs with irradiation on one side only. From these results, it can be concluded that continuous wave CO_2 laser decontamination does not enhance the amount of titanium release in vivo and seems to have no adverse effects on the reaction of local tissues or visceral organs.

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