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Laser Radiation as an Adjunct to Nonsurgical Treatment of Periodontal Disease

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1. Introduction

It is well known that biofilm and calculus responsible for periodontal disease can be of different nature depending on their supra or subgingival location, and that the physical or chemical methods used for their elimination, achieve different results in both places (Davies et al., 1998). Since the use of laser confocal microscope and the study of biofilms in their natural state, it has been observed that the behaviour of bacteria is quite different to the observed in traditional cultures. In their natural state, the bacterial colonies are constituted by several microcolonies included in a matrix, which has canals through which flow fluids transporting nutrients, metabolic wastes, enzymes, oxygen and other products enabling the presence of different environments (Costerton et al. 1987).

The biofilms adhered on the internal and external walls of the periodontal pocket, the free biofilm and the possibility of a bacterial penetration through the epithelium to the underlying connective tissue can cause a gingival inflammatory reaction. This inflammation may progress with vasodilation, cellular migration and release of mediators, thus increasing the inflammatory response and perpetuating the disease. This situation makes microorganisms more resistant to drugs, which frequently are unable to reach the colonies protected by the matrix and by the presence of resistent bacteria (Donlan & Costerton, 2002). The inflammatory phenomena triggered by the bacteria and their waste products attracts macrophages that produce, among others, interleukin 1 (IL-1) and tumor necrosis factor alfa (TNF-a), which have the ability to activate osteoclasts and produce bone resorption. TNF-a activates the adhesion molecules of the endothelial cells of the vessels, favouring the adhesion of monocytes and diapedesis. It also stimulates the arrival of T lymphocytes, which contribute with receptor activator of nuclear factor kappa B ligand (RANKL) to the bone, consequently favouring the bone loss (Kong et al., 1999). But this process is more complex as it needs some proteins such as nuclear factor kappa B (NF-kB), receptor activator of nuclear factor kappa B (RANK), RANKL and osteoprotegerin (OPG), among others, which may change the answer of the osteoclast precursors and therefore modify the osseous destruction. The NF-kB plays a basic role as activator of immunoglobulins during the infectious process (Gilmore, 2006). So, as IL-1 and TNF-a favour the synthesis of RANK-L

and thus the activation of RANK, it allows the differentiation of the preosteoclast into osteoclast. But this process may be hindered when OPG (a soluble protein expressed in numerous tissues and in osteoblastic cells) appears (Aubin & Bonnelye, 2000), blocking the union between RANK and RANKL, stopping the process of bone destruction (Fig. 1). OPG is a tumor necrosis factor receptor –like molecule, which is produced by gingival fibroblasts, ligament and epithelial cells (Sakata et al., 1999), that can be modulated by several inflammatory cytokines.

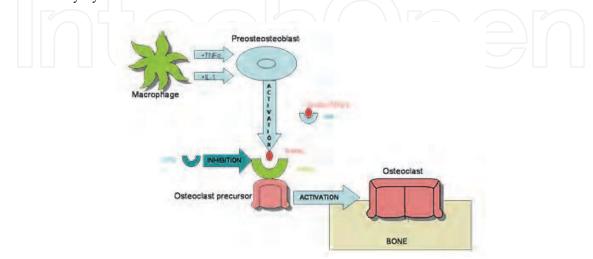


Fig. 1. Activation by RANKL and inhibition by OPG of bone resorption.

These proteins can be detected at the periodontal pocket and are related to the degree of evolution of the periodontal disease (Bostanci et al., 2007). Its monitoring allows us with much more precision than the clinic to detect the possible biological effects over the periodontal status, once applied the treatment.

Searching for effective techniques for the elimination of the biofilm and reduction of the inflammation, the mechanical treatment is still considered the gold standard. Using scaling and root planing (SRP), ultrasonic scalers and adequate hygiene techniques, acceptable results can be achieved, but these treatments alone are unable to eliminate completely all the bacteria due to radicular morphologic factors, deep pockets with a difficult access (Adriaens & Adriaens, 2004), bacterial invasion of adjacent gingival tissues and fast variation of the bacterial colonies (Costerton et al., 1999). Nowadays, antibiotics are used as complementary elements but its use should be restricted to the minimum due to the frequent development of resistances, and to the difficulty of maintaining stable and effective levels during a long period of time (Socransky & Haffajee, 2002). For this reason it becomes necessary the additional research of substances or techniques that can modify the pH, the oxygen concentration or the nutrient disposition of the dental plaque in order to modify the microflora of the biofilm. We also need to find systems able to interfere with the bacterial genetic signals and to modify the inflammatory response in the periodontal tissues.

An alternative to be considered is the use of laser technology. Several studies guarantee its beneficial effects such as sulcular and/or pocket debridement, reduction of subgingival bacterial load and decrease of inflammation (Ando et al., 1996; Folwaczny et al., 2002; Schwarz et al., 2008). Recently, in the periodontal field, it has also been introduced the photodynamic therapy (Chan & Lai, 2003; Maisch et al., 2007). Nowadays, the use of lasers within the periodontal pocket has become a promising field in the periodontal therapy.

2. Lasers employed in periodontics

Currently, different equipments of laser radiation (Er:YAG, Er,Cr:YSGG, Nd:YAG, diode, CO₂), are available in Periodontics, each one with particular features and diverse effects, making necessary the selection of the most suitable for each type of application (Table 1). Some of these lasers are effective in eliminating the residual calculus and detoxifying the radicular cementum (Er:YAG) (Aoki et al., 2004; Ishikawa et al., 2004; Schwarz et al., 2008); on the contrary others are unable to eliminate the calculus but can act over the soft tissues reducing the inflammation, as they modify the tissue oxidation systems and the cytokines which mediate in inflammation (Nd:YAG, diode) (Gómez et al., 2011). Although these effects over the tissues are difficult to evaluate clinically, they are guaranteed by molecular biology techniques. The results seem to be variable, but the investigation should help us to select the wavelength of the radiation, pulse duration, energy/power applied, pulse shape, repetition rate, time of exposure, sequence, type of wave, continuous (cw) or pulsed, type of applicator (cutout or rigid fiberglass), and other factors which can provide the desired objectives.

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635 nm					
diode	**	**			
650 nm					
diode	**	**			
675 nm					
diode	**	**			
690 nm					
diode	** a		**		
810 nm					
diode	** a		**	*	
980 nm					
Nd:YAG	* a		**	*	
1,064					
nm/cw					
Nd:YAG					**
1,064 nm/pulsec					
Er,Cr:YS					**
GG					
2,790 nm					**
Er:YAG					ጥጥ
2,940 nm					
CO ₂				**	
10,600 nm					

Biostimulation Photodynamic Photocoagulation Photovaporization Photoablation

a= at low-level laser power

Table 1. Periodontal laser systems and tissue interaction.

The therapeutic application of laser radiation can be clinically useful only if the appropriate instrumentation is available. Since the laser has been introduced into medicine, and so into dental discipline, a number of different applicators have been developed for day-to-day clinical use. The types of applicators most used in Periodontics are shown next. For instance, those with rigid fiberglass are set over handpieces. They should be used sliding them almost parallel to the radicular surface (Fig. 2), with a 20° angle in a coronoapical sense, as the perpendicular application produces damage in the cementum. Due to their size, the applicators with cutout fiberglass (Fig. 3) allow intrasulcular insertion and can reach deep areas. The displacement is also done in a coronoapical sense, outlining the whole radicular surface following the depth of the periodontal pockets, in the same manner as in a periodontal probing. Other rigid and thin applicators should be used with spiral or circular movements instead of coronoapically, in order to optimize results.



Fig. 2. Application of rigid fiberglass.



Fig. 3. A) Application with cutout fiberglass. B) Cutout fiberglass.

In Periodontics we need treatments to eliminate the plaque and calculus, and to eliminate and/or reduce the gingival inflammatory phenomena. We have to operate therefore over the soft and hard dental tissues. For this reason, the basic effects of periodontal lasers over these soft and hard tissues are presented next, in order to show the possibilities of this technology applied alone, and mainly in combination with SRP.

3. Lasers on dental soft tissues

The earliest clinical studies mentioning the application of lasers in the non-surgical treatment of periodontitis began in the early 1990s using a Nd:YAG laser with the development of flexible optical fibres. Since then, many studies have been carried out to evaluate the possible advantages of the use of lasers (Nd:YAG, diode (GaAlAs, InGaAsP), Er:YAG, Er,Cr:YSGG and CO₂) with wavelengths ranging from 635 to 10,600 nm. Recently, systematic reviews have compiled different clinical and microbiological effects of different

types of laser radiation used as monotherapy or adjunctive therapy compared with SRP. However, less information to demonstrate the anti-inflammatory effect of the laser radiation is available from the literature.

3.1 Nd:YAG laser radiation

The Nd:YAG laser is nowadays the most important solid-state laser. The essential advantages of the Nd:YAG laser are its simple and compact construction and its high average power output. The neodymium (Nd+) ion, implanted into various host crystals, is the source of the laser radiation. Among the many crystals which have been investigated, one material stands out: Y₃Al₅O₁₂, yttrium-aluminium-garnet, a crystal with a garnet structure, which enables a laser emission at 1,064 nm, in the near-infrared spectral region. This allows its transmission through 200 to 400 micron of diameter optical fibre, making easier its use and reaching even the internal side of the periodontal pocket (Fig. 3). Unlike other infrared lasers with a strong absorption by water, such as Er:YAG or CO₂, the wavelength of Nd:YAG laser presents a poor absorption by water, thus increasing scattering and infiltration of its energy into the biological tissues. The photothermal effects of Nd:YAG laser are useful in soft tissue surgeries. Thanks to its great penetration depth and thermogenesis properties, this type of laser produces a thick coagulation layer in the irradiated area, presenting a great haemostatic capacity, being efficient for the ablation of potentially haemorrhagic soft tissues (Perry et al., 1997, Romanos, 1994).

There is little evidence to support the efficacy of Nd:YAG laser treatment as an adjunct to nonsurgical periodontal treatment in adults with periodontal inflammation. In the last decade, there are barely clinical studies published analyzing the clinical evolution and the inflammatory mediator levels in the gingival crevicular fluid (GCF) after irradiation with Nd:YAG laser in the affected sites in patients with chronic periodontitis. The results obtained in four clinical studies, performed by three different research groups, should be emphasized (Gómez et al., 2011; Miyazaki et al., 2003; Qadri et al., 2010, 2011). The overheating of the irradiated tissues and the consequent damage of the oral hard and soft tissues, could explain the limited support to this kind of laser radiation from the scientific community (Miserendino et al., 1994). For this reason, to avoid thermal damage, the irradiation parameters employed in these clinical studies were selected according to the results obtained in previous *in vitro* investigations, where potential morphological alterations of root surface irradiation were assessed under standardized conditions (Bader, 2000; Gómez et al., 2009).

Concerning the evolution of the clinical parameters, the application of Nd:YAG laser both as monotherapy or as an adjuvant to scaling and root planing, did not offer significant advantages versus the treatment with ultrasonic devices, both at 8 (Gómez et al., 2011) (Fig. 4) and at 12 weeks (Miyazaki et al., 2003).

Nevertheless, Qadri et al., in their split mouth trial, found better clinical results in the test side (SRP + Nd:YAG) than in the control side (SRP) during the long term follow up (up to 20 months) (Qadri et al., 2011).

When analyzing the inflammatory mediators, Miyazaki, in a 12 weeks study, found a non statistically significant decrease of IL-1 β in Nd:YAG group used as monotherapy in comparison with ultrasonic devices for the non surgical treatment of chronic periodontitis (Miyazaki et al., 2003). On the contrary, Gómez did find in a short term study (4 and 8

weeks) significant decreases both in IL-1 β as in TNF- α when using Nd:YAG as an adjuvant to SRP versus SRP alone (Fig. 5). In this same study, the total antioxidant status (TAS) of the gingival fluid, gradually increased until the eighth week after the treatment with SRP + Nd:YAG, while it remained stable when the treatment consisted of SRP, being these differences statistically significant (Gómez et al., 2011). The total antioxidant capacity of the gingival fluid decreases in periodontitis as a consequence of the inflammatory lesion, and it recovers after non surgical therapy (Brock et al., 2004; Chapple et al., 2007; Tsai et al., 2005). Thus, Gómez observed that the total antioxidant capacity of GCF was influenced by the reduction of periodontal inflammation after successful non-surgical therapy complemented by Nd:YAG radiation (Gómez et al., 2011). Finally, in the study of Qadri in which the longterm effects of a single application of a water-cooled pulsed Nd:YAG in combination with SRP were investigated, the authors found less bone loss and a lower GCF volume than in the control group (SRP alone), and thus the severity of periodontal inflammation seemed to be reduced (Qadri et al., 2011).

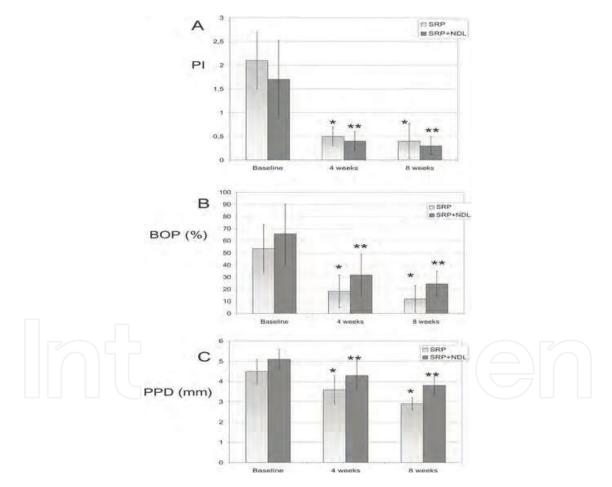


Fig. 4. *A*) Plaque Index (PI), B) Bleeding on Probing (BOP) and C) Probing Pocket Depth (PPD): Mean Scores (± SD), (n=30 patients) at baseline, 4 and 8 weeks post-therapy. (*) Significance of differences compared to baseline within SRP group at different points of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of differences compared to baseline within SRP+NDL group at different points of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametric Wilcoxon test (p<0.05). (**) Significance of time by a non-parametri

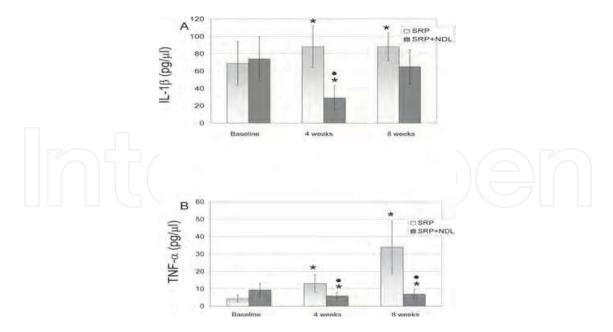


Fig. 5. A) Interleukin 1 β (IL-1 β) and B) Tumour Necrosis Factor α (TNF- α): Mean Scores (± SD), (n= 30 patients) at baseline, 4 and 8 weeks post-therapy. (*) Significance of differences compared to baseline within the groups at different points of time by a non-parametric Wilcoxon test (p<0.05). (•) Significance of differences compared to baseline between the groups at different points of time by a non-parametric Mann-Whitney test (p<0.05). (Gómez et al., 2011)

3.2 Fluorescence-controlled Er: YAG laser radiation

In erbium lasers, erbium ions (Er³⁺) are implanted in the solid-state materials yttrium aluminium garnet (YAG, Y₃Al₅O₁₂). Its pulsed infrared radiation, at 2,940 nm, is characterized by being highly absorbed by water, therefore it is particularly indicated for a precise and located ablation of the biological tissues with a high water content. Theoretically, the absorption coefficient of water for the Er:YAG laser is 10,000 cm⁻¹, and thus 15 and 2,000 times higher than for CO₂ and Nd:YAG lasers, respectively. This high absorption coefficient results in extremely small optical penetration depths and therefore in tissue ablation with minimal thermal damage. Additionally, the OH groups, as components of the hydroxyapatite, show their higher absorption around 2,800 nm, explaining thereby its ablation capacity over the enamel, dentin and bone, so this type of laser is indicated both for soft and hard tissues. The energy transport from the laser system to the patient is done by an articulated arm or by a flexible waveguide made of zirconium fluoride or crystalline sapphire. A variety of new applicators are continuously extending the potential dental uses. Based on the results of in vitro studies, Watanabe performed the first clinical application of an Er:YAG laser for debridement in 1996 (Watanabe et al., 1996). Nowadays, after the investigation carried out in the clinical field with this type of laser radiation, it seems that Er:YAG laser emerges as the most adequate laser system as an alternative or adjuvant tool of SRP. This is due to the lower thermal damage generated on the hard tissues (Schwarz et al., 2008). Although the in vitro capability of calculus and plaque removal of Er:YAG laser has been displayed, clinical studies have shown divergent clinical outcomes in the initial treatment of chronic periodontitis. Crespi (Crespi et al., 2007) reported a significant reduction in clinical parameters at 6 months in the Er:YAG group compared to the group

treated by SRP with ultrasonic scalers, while a recent study has found that adjunctive use of Er:YAG laser to conventional SRP did not reveal a more effective result than SRP alone in the short term of 6 months (Rotundo et al., 2010).

A new Er:YAG laser equipment introduced to improve the results is a device that allows the control of Er:YAG laser radiation by incorporating a feedback system that selectively detects subgingival calculus (Fig. 2). Few clinical studies have evaluated the treatment outcomes after laser debridement by using fluorescence controlled Er:YAG radiation. In the split-mouth study of Sculean it was observed that fluorescence-controlled Er:YAG radiation led to clinical improvement at 3 and 6 months, similar to ultrasonic debridement (Sculean et al., 2004). Tomasi evaluated the clinical and microbiological outcomes after a feedback-controlled Er:YAG laser and ultrasonic device debridement during periodontal supportive therapy (Tomasi et al., 2006). They observed that mean PPD reduction and CAL (clinical attachment level) gain were significantly higher in the laser group after 1 month of healing. However, both treatments resulted in a significant reduction of subgingival microflora, although no significant differences were observed between groups at each time point investigated. Derdilopoulou compared the microbiological effects of SRP, Er:YAG laser with feedback, sonic and ultrasonic scalers in patients with chronic periodontitis over a period of 6 months. The treatment methods employed resulted in a comparable reduction of the evaluated periodontopathogens, where Er:YAG laser did not demonstrate to be superior (Derdilopoulou et al., 2007). Finally, more encouraging were the results obtained by Domínguez, when Er:YAG laser was employed as an adjuvant to SRP. Though no statistically significant differences were observed between both groups in any of the investigated clinical parameters (Domínguez et al., 2010) (Fig. 6), the cytokine levels in the GCF were reduced with the feedback-controlled Er:YAG laser radiation (Fig. 7). Since the outcome in the SRP+ Er:YAG group was only slightly better than in the SRP group, previous mechanical subgingival debridement is necessary. Despite the above described advantage of using this laser prototype, no additional effect of the Er:YAG laser therapy was found on the local (GCF) total antioxidant capacity.

Finally, in a study reported by Schwarz, immunohistochemical characterization of wound healing following non-surgical periodontal treatment revealed that fluorescence-controlled Er:YAG laser radiation was effective in controlling disease progression, and may support the formation of a new connective tissue attachment (Schwarz et al., 2007).

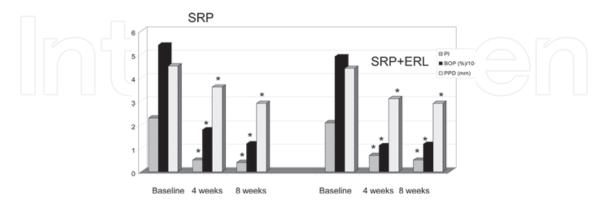


Fig. 6. Plaque Index (PI), Bleeding on probing (BOP) and Probing Pocket Depth (PPD): Mean Scores (± SD), (n=30 patients) at baseline, 4 and 8 weeks. (*) Significance of differences compared to baseline within the groups at different time points by Wilcoxon test (p<0.05). (**) Significance of differences compared to baseline between the groups at different time points by Mann-Whitney test (p<0.05). (Domínguez et al., 2010)

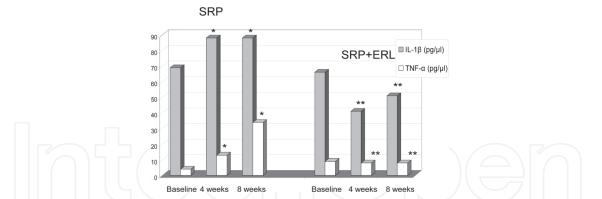


Fig. 7. Interleukin1 β (IL-1 β) and Tumour Necrosis Factor α (TNF- α): Mean Scores (± SD), (n=30 patients) at baseline, 4 and 8 weeks. (*) Significance of differences compared to baseline within the groups at different time points by Wilcoxon test (p<0.05). (**) Significance of differences compared to baseline between the groups at different time points by Mann Whitney test (p<0.05). (Domínguez et al, 2010)

3.3 Photodynamic therapy (PDT)

Photodynamic therapy basically involves three nontoxic ingredients: visible harmless light, a nontoxic photosensitizer (i.e. a photoactivable substance) and oxygen (Fig 8). It is based on the principle that the photosensitizer binds to the target cells and can be activated by light of a suitable wavelength. Following activation of the photosensitizer, singlet oxygen and other reactive agents that are extremely toxic to certain cells and bacteria are produced. This singlet oxygen might cause toxic effects on the microorganisms: damage of the membrane lipids, destruction of protein and ion channels, elimination of critical metabolic enzymes, cell agglutination and inhibition of exogenous virulence factors such as lipopolysaccharide, collagenase and protease. Photosensitizers for PDT are selected for their ability to rapidly penetrate bacterial biofilms and to selectively stain and kill the prokaryotic cells under illumination while avoiding damage to human tissues (Konopla & Goslinski, 2007).



Fig. 8. A) Periodontal pocket irrigation with methylene blue, B) Subsequent photosensitizer activation with diode laser at 670 nm.

Several studies have reported the use of PDT therapy as an adjunct to nonsurgical treatment for initial and supportive therapy of chronic periodontitis. In view of the published results, the adjunctive use of photodynamic therapy to SRP may result, on a short-term basis (up to 3 or 6 months), in higher reductions in bleeding on probing with PPD reductions and CAL gains comparable to those obtained after SRP alone (Chondros et al., 2009; Christodoulides et al., 2008; Ge et al., 2011; Polansky et al., 2009). The higher improvement in mean fullmouth bleeding scores (FMBS) might be attributed in part to the additional photobiomodulation effect mediated by the low-level laser irradiation during photodynamic therapy (Qadri et al., 2005).

Photodynamic therapy has been introduced as an important novel disinfection therapy in the field of dentistry. Taking into account the microbiological improvement of PDT, some research groups did not find additional benefit of SRP+PDT above SRP at any follow-up evaluation (Chistodoulides et al. 2008; Polansky et al., 2009; Yilmaz et al., 2002). Chondros (Chondros et al., 2009) found a significant reduction in *F. nucleatum* and *E. nodatum* in the group receiving the combined treatment (SRP+PDT) at the 3-month evaluation.

A recent study of 10 patients in supportive periodontal therapy with 70 residual pockets (PPD≥5mm) and a parallel group design, has confirmed positive effects of repeated adjunctive PDT to SRP treatment on those residual pockets treated, such as greater PPD reductions, CAL gains and decreased BOP percentages after 6 months post-therapy (Lulic et al., 2009). These positive results could be related with the study protocol: SRP of the residual pockets followed by immediate PDT application, repeating this sequence of treatment five times in 2 weeks.

The presence of BOP is considered an objective indicator of gingival inflammation (Chaves et al., 1993; Lang et al. 1990). The published results of SRP+PDT treatment generally show a tendency to reduce BOP or FMBS, and thus a tendency to reduce inflammation. It should be pointed out that there are hardly any studies in the literature relating PDT and proinflammatory cytokines, with the exception of de Oliveira's. These authors investigated cytokine levels in GCF of patients with aggressive periodontitis after PDT or SRP. The results showed that both treatment modalities significantly reduced TNF- α and RANKL levels following treatment (de Oliveira et al., 2009). Recently, histological examinations of periodontal tissues of rats treated 1 month before with toluidine blue-mediated PDT, revealed a remarkable reduction of inflammatory reactions (reduced infiltration of inflammatory cells, mainly lymphocytes), greater than in periodontal tissues conventionally treated (Qin et al., 2008). In addition, after the application of a single *in vitro* PDT treatment, an inactivation of host destructive cytokines (which impair periodontal restoration) was seen by means of a cytokine inactivation assay that measured E-selectin expression in response to IL-1 β and TNF- α (Braham et al., 2009).

Although the results published in the literature seem to indicate that the adjunctive use of PDT may improve some outcomes, randomized controlled clinical studies, evaluating the clinical, microbiological and immunological potential benefits of photodynamic therapy in the treatment of periodontitis, are still limited. The main drawbacks may be related to the rather limited number of patients, the short-term duration of studies (i.e. 3 or 6 months) and the fact that the most effective protocol of PDT has not yet been established.

Finally, we should point out that in few years PDT has progressed greatly in the biomedical field. Nowadays, numerous research groups are working in this field investigating its application. New photosensitizers are being developed searching its faster removal from healthy tissues, acting at lower doses and being able to absorb longer wavelengths. This would allow higher light penetration, requiring lower doses of photosensitizer, while new sources of irradiation are also being developed.

3.4 New research lines: Phototherapy/biostimulation

The variety of biological effects which laser radiation may produce in the oral tissues are not yet completely understood. Among the many physiological ones, it is important to recognize benefitial biostimulant effects of laser radiation in cells of the oral tissues during laser therapy, such as the contribution to a faster healing during the reparation process of the periodontium, which may not take place during the conventional mechanical therapy. These biostimulant effects have been associated with the use of low level laser radiation (Peplow et al., 2010). According to the first law of photochemistry, the biological effect observed after the application of laser radiation of low energetic level, can only be a result of the presence of a photoacceptor molecule, able to absorb the photonic energy emitted (Karu, 2007). Additionally, there are no photothermal nor photoacoustic mechanisms associated to this effect, so no heating is observed macroscopically. One target identified in laser phototherapy is a highly specialized enzyme, cytocrome C oxidase, which plays a crucial role in cellular bioenergy (Karu, 2007). Some studies indicate that after laser irradiation at 633 nm, an increment of both the mitochondrial membrane potential and the proton gradient occurs, causing changes in the optical properties of mitochondria, thus increasing the rate of exchange of adenosine diphosphate/adenosin triphosphate (ADP/ATP) (Alexandratou et al., 2002). Moreover, it has been proposed that laser irradiation reduces the catalytic center of cytocrome C oxidase, originating more available electrons for the reduction of dioxygen (Byrnes et al., 2005). The upregulation of ATP following low level laser therapy is also coupled with transient increases of the reactive oxygen species (ROS), participating afterwards in transduction of intercellular signals (Tafur & Mills, 2008). It has been observed that the modulation of cellular metabolism and transduction of signals alter the gene expression (Snyder et al., 2002), the cellular proliferation (Moore et al., 2005), the mitochondrial membrane potential (Alenxandratou et al., 2002), the generation of transient reactive oxygen species (Lubart et al., 2005), the level of calcium ion (Tong et al., 2000) the gradient of protons and the oxygen consumption.

The efficacy of low-level laser therapy in periodontal disease is still controversial. Ribeiro (Ribeiro et al., 2008) corroborated that the use of diode laser as an adjunct to SRP 4 times in the first two days of treatment did not provide any apparent clinical benefit in teeth with shallow to moderate pockets. Lai (Lai et al., 2009) also observed that phototherapy used as a complement of non surgical periodontal therapy eight times in the first three months of treatment did not improve healing response as assessed by both clinical and radiographic parameters. In the study of Yilmaz, laser used as a monotherapy did not affect the inflammatory response more than oral hygiene instructions (Yilmaz et al, 2002). The group treated with subgingival debridement plus low level laser therapy obtained the same results as that treated only with subgingival debridement, thus demonstrating that mechanical subgingival debridement is always necessary. However, other studies verify the efficacy of phototherapy. For example, Qadri (Qadri et al, 2005) observed significantly higher reductions in PPD, PI, GI, GCF and MMP-8 in the laser treated sites. Therefore, the use of a low level laser radiation as an adjunct to the periodontal treatment showed a positive influence on inflammation and healing. Pejčic (Pejčic et al., 2010) confirmed that low level laser radiation at 670 nm may be successfully used as an adjunctive treatment method, which, together with conventional periodontal therapy, achieves better long term results (up to 6 months after treatment). Thus the PI, GI and BOP decreased, demonstrating that the number of laser applications is of great importance in order to obtain the best results in the irradiated tissue, achieving a considerable antiinflammatory effect after the fifth application. In addition, Kreisler (Kreisler et al., 2005) when using a 809 nm GaAlAs semiconductor laser operated at 1 W power output as an adjunct to SRP, observed a significant improvement concerning the tooth mobility, PPD and CAL, not finding significant differences in PI, GI, BOP and GCF volume.

A higher number of well-designed long term randomized controlled clinical trials over a longer period of time may be required in order to evaluate the adjunctive use of low-level diode laser radiation during non-surgical periodontal therapy and to clarify controversy.

4. Lasers on dental hard tissues

Until the beginning of the 1990s, the use of laser systems in periodontal therapy was limited to soft tissue procedures, such as gingivectomy and frenectomy, as application to periodontal hard tissues had previously been shown to be clinically unpromising (Cohen & Ammons, 2002; Pick & Colvard, 1993). In the last decades, laser therapy has been proposed as an alternative or an adjunct to conventional non-surgical therapy, due to its capability to obtain tissue ablation and haemostasia, bactericidal effect against periodontal pathogens and detoxification of root surface (Aoki et al., 2004; Folwaczny et al., 2002; Ishikawa et al., 2004; Schwarz et al., 2008).

CO₂ and Nd:YAG lasers can produce carbonization and major thermal side effects when used on hard tissues such as root surface and bone (Israel et al., 1997; Wilder-Smith et al., 1995) (Fig. 10). The CO₂ irradiation is well absorbed by water, and by the main mineral components of hard tissues, especially phosphate ions (-PO4) of the hydroxyapatite (Koort & Frentzen, 1995). Since ablation is basically produced by heat generation, unlike other laser systems such as Er:YAG laser, carbonization occurs easily on the irradiated surface (Sasaki et al., 2002). When used with relatively low energy output in a pulsed and/or defocused mode, CO₂ lasers have been used to achieve root conditioning, detoxification, and bactericidal effects on contaminated root surfaces (Coffelt et al., 1997). However, at low energy outputs in a continuous mode, it is unable to remove subgingival calculus and when used with high energy outputs, especially in a cw mode, it is inappropriate for calculus removal due to major thermal side effects, such as carbonization, melting and cracking on the cementum and dentin (Misra et al., 1999).

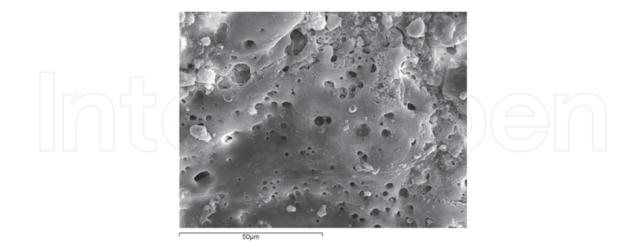


Fig. 9. Scanning electron microscope obtained image of a radicular surface treated with Nd:YAG (1,5 W, 10 Hz).

Nd:YAG laser presents low absorption by water, which produces scattering of its energy and thus a great penetration into the tissues. *In vitro* and *in vivo* studies have shown that this laser is inefficient to successfully achieve root surface debridement, due to its poor ability to

remove calculus. Nd:YAG should be employed as an adjunct to conventional mechanical treatments, rather than as a primary instrument in the treatment of periodontal pockets (Liu et al., 1999). The same as CO₂ laser, Nd:YAG produces different root surface alterations, induced by heat generation during irradiation (Fig. 9) (Cobb, 1996; Gómez et al., 2006; 2009). Nevertheless, several *in vitro* studies have demonstrated the ability of these lasers to create a compatible surface for soft tissue attachment (Israel et al. 1997; Wilder-Smith et al., 1995).

In 1989 Hibst and Keller (Hibst & Keller, 1989; Keller & Hibst, 1989) reported the possibility of dental hard tissue ablation by Er:YAG laser irradiation, which is highly absorbed by water. Since then, numerous studies on hard tissue treatment using the Er:YAG laser have indicated the ability of this laser to ablate dental hard tisses and carious lesions without producing major thermal side effects.

The absorption of Er:YAG laser in water is the highest because its 2,940 nm emission wavelength matches with the large absorption band for water. Additionally, as part of the apatite component, OH- groups show a relatively high absorption at 2,940 nm. As the Er:YAG laser is well absorbed by all biological tissues that contain water molecules, it is indicated not only for the treatment of soft tissues but also for ablation of hard tissues.

In vitro studies have shown that the Er:YAG laser application is effective in eliminating subgingival calculus, with similar results when compared with ultrasonic instrumentation (Aoki et al., 2000; Folwaczny et al., 2000; Herrero et al. 2010). However, some authors showed a greater amount of residual calculus in the areas treated with the Er:YAG laser (Eberhard et al. 2003). Factors such as the quantity and quality of the initial calculus (texture, thickness and water content) and root anatomy, together with the individualized instrumentation technique, may influence the results independently of the mode of implementation (Herrero et al., 2010). Due to their similar composition, calculus and cementum have similar ablation thresholds; therefore, it would be impossible to carry out selective and effective removal of calculus with the use of Er:YAG laser application without causing root damage (Aoki et al., 1994).

Different energy settings have been reported to be more efficient for calculus removal without damaging the root cementum. Most of the studies suggest the use of energies between 100-160 mJ (Crespi et al., 2006; Folwaczny et al., 2000; Frentzen et al., 2002). Higher energies may damage the radicular surface and lower energies are unable to eliminate the calculus effectively (García et al., 2001). Folwaczny et al. reported that the angulation tip to the root surface has a strong influence on the amount of root substance removed during Er:YAG laser irradiation (Folwaczny et al, 2000). Moreover, the use of water coolant minimizes heat generation by cooling the irradiated area and absorbing excessive laser energy. In addition, a water spray facilitates hard tissue ablation by eliminating the target moist (Burkes et al., 1992). A secondary effect of calculus removal is the elimination of cementum and subsequent dentinal tubular exposure (Fig. 10). Even though the amount of cementum on the root surface is highly variable and depends on factors such as patient age and previous periodontal treatment, in vitro studies have described that the number and diameter of exposed dentinal tubules was significantly higher in areas treated with laser than with ultrasonic scalers (Gómez et al., 2009; Herrero et al., 2010; Theodoro et al., 2003). The last generation of Er:YAG laser includes a calculus detection device based on the signal of the fluorescence emitted from the mineralized tissues (feedback system). Preliminar in vitro and clinical studies show that the Er:YAG laser debridement, when performed with automatic calculus detection, allows an efficient calculus removal similar to the obtained with ultrasonic scaling, without the partial ablation of subjacent cementum observed with

the Er:YAG laser prototypes without feedback system, thus resulting in almost no dentinal tubule exposure (Herrero et al., 2010; Krause et al. 2003; Schwarz et al., 2006).

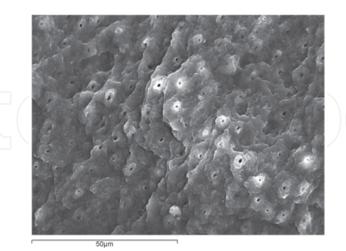


Fig. 10. Scanning electron microscope obtained image of a radicular surface treated with Er:YAG (120 mJ/pulse, 10 Hz).

The Er:YAG laser does not cause carbonization or melting of the irradiated root surface, but it has been demonstrated that the surface presents micro-irregularities, is slightly rougher and chalky, probably due to the mechanical ablation effect (Aoki et al., 1994; Aoki et al., 2000; Crespi et al., 2006; Folwaczny et al., 2000; Herrero et al., 2010). Some craters may be formed as a result of directing the laser beam too perpendicularly to the root surface or by using a non beveled tip, which hampers uniform radiation (Folwaczny et al. 2000; 2002). There is no clear indication in the literature of which is the ideal root surface to improve healing. It is well accepted that a rough surface does not negatively affect periodontal healing (Kathiblou & Ghoddsi, 1983). According to several authors (Eberhard et al., 2003; Folwaczny et al., 2000), the surface treated with Er.YAG laser is similar to the one observed after EDTA (Lasho et al., 1983) or citric acid conditioning (Wen et al., 1992), which for many years have been used as root conditioners improving the results of periodontal treatment (Schwarz et al., 2003a). On the contrary, the presence of a rough root surface may favour plaque retention and, therefore, limit the results of cause-related periodontal therapy (Adriaens & Adriaens, 2004).

5. Lasers for the treatment of periimplantitis

Bacterial colonization in the implant surface is considered the main aethiological factor of implant failure (Becker et al., 1990). The bacterial presence in the implant surface may cause inflammation of the peri-implant mucous membrane, and, if not treated, it may progress apically, resulting in periimplantitis and bone resorption. Therefore, removal of bacterial plaque is the main objective in the therapy of periimplant infections (Mombelli & Lang, 1994). However, the debridement of implant surfaces is difficult to achieve, particularly in the rough ones. Different mechanical and chemical methods have been proposed (Augthum et al., 1998; Ericsson et al., 1996) in order to reach this objective. Recent in vitro studies suggest the use of specific devices, made of materials with lower hardness than titanium (plastic scalers, rubber cups) for the mechanical debridement (Matarasso et al., 1996). As mechanical methods are

inefficient when used alone, chemical agents, such as local or systemic antimicrobials, have also been used (Ericsson et al., 1996; Norowski & Bumgardner, 2009).

Different laser systems have also been proposed for the debridement of implant surfaces (Kreisler et al., 2002a; Kreisler et al., 2002b). Recent in vitro studies show that, due the radiation features, only CO2, diode and Er:YAG lasers are adequate for implants surface debridement. This is because their wavelengths are scarcely absorbed by titanium, increasing slightly the implant temperature during irradiation (Kreisler et al., 2002a; Oyster et al., 1995; Romanos et al., 2000). Nevertheless, Nd:YAG laser produces an important thermal damage in the titanium surface (Kreisler et al., 2002a; Romanos et al., 2000). Additionally, only CO2 and Er:YAG have demonstrated bactericidal effects in in vitro studies, so both systems could be useful in the decontamination and detoxification of the implant surface (Kreisler et al., 2002c). As CO₂ as well as diode lasers are not effective for calculus removal in radicular surfaces nor in titanium implants, they should only be employed as adjuvants of the mechanical procedures (Moritz et al., 1998; Schwarz et al., 2003b). However, recent studies show that non surgical instrumentation of implants with Er:YAG laser, when using a specific applicator, is effective in calculus and subgingival plaque removal, without producing thermal damage to the implant surface (Schwarz et al., 2003b). The results of this study indicate that Er:YAG laser radiation does not damage titanium surfaces and subsequently does not influence the attachment rate of cultured human osteoblast-like cells (SAOS-2). Results of a preliminar clinical study by the same author, showed that non surgical treatment of periimplantitis with Er:YAG at 100 mJ/pulse and 10 Hz produces a significant reduction of PPD and CAL gain (Schwarz et al., 2005). Recently, 940 and 980 nm diode lasers have created a great expectation, due to their excellent incision, excision and coagulation properties over the soft tissues, allowing

subsequently to proceed in low energy conditions (low-level laser therapy, described previously in the above section), decreasing simultaneously the inflammatory process and achieving a faster healing of tissues (Romanos et al., 2009). The use of lasers in the treatment of periimplantitis is promising, but more studies evaluating its real efficacy are necessary.

6. Conclusions

We have to take into account that laser treatments are in continuous evolution; possibly in the next years our scope will be to have equipments combining different photonic properties allowing us to choose the most adequate system for each necessity. Although there has been a great progress in the last years, most of the studies are difficult to be evaluated clinically, due to their short duration (2 to 3 months). More long term systematic studies are necessary to evaluate the clinical and biological effects of each type of laser, the time and application mode, unique/multiple doses and application frequency. It will also be essential to know the appropriate energies of each kind of laser, deepening in its knowledge, as its application comfort, the silence, the anesthesia reduction and other advantages make them attractive for society and professionals.

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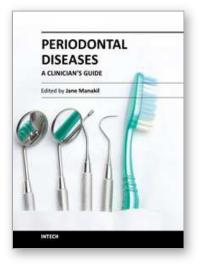
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