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Adjunctive effect of a water-cooled Nd:YAG laser in the treatment of chronic periodontitis

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Abstract

Objectives: To test whether use of a water-cooled Nd:YAG laser adjunctive to supra- and subgingival debridement (SRP) with hand and ultrasonic instruments results in greater clinical improvement than SRP alone. Another objective was to investigate the reduction in the number of microorganisms.

Methods: This study was an examiner-blind, randomized and controlled clinical trial using a split-mouth design. Nineteen subjects with moderate-to-severe generalized periodontitis were selected. Immediately following SRP in two randomly chosen contra-lateral quadrants, all pockets $\geqslant 4$ mm were additionally treated with the Nd:YAG laser (1064 nm, 6 W, 400 mJ). Clinical assessments (Plaque index, bleeding on pocket probing, probing pocket depth) were performed pre-treatment and at 3 months post-treatment. In each quadrant, one site was sampled for microbiological evaluation at pre-treatment, immediately post-instrumentation and 3 months post-treatment.

Results: At the 3-month visit, the clinical parameters had significantly improved for both regimens. No significant differences between treatment modalities were observed for any of the clinical parameters at any time. Immediately following instrumentation, the total colony forming units for both groups were significantly reduced as compared with pre-instrumentation. No significant differences between treatment modalities were observed.

Conclusions: Three months after SRP, no additional advantage was achieved with the additional use of the Nd:YAG laser. Microbiological findings reflect these clinical results.

Key words: laser; Nd:YAG; periodontitis; RCT; scaling and root planning; treatment; ultrasonics

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The goals of treatment of chronic periodontitis generally include reductions in pocket probing depth and supra- and subgingival microbial loads, gains in clinical attachment level and arresting of disease progression. Most treatment modalities used in periodontal therapy attempt to achieve these goals by reducing the amount of bacterial plaque on the root surface to levels compatible with periodontal health. The traditional periodontal treatment of supra- and subgingival debridement (SRP), which may be followed by periodontal surgery (Pihlstrom et al. 1983, Badersten et al. 1984a, b, Ramfjord et al. 1987, Kaldahl et al. 1996), is not always successful in eliminating all deep periodontal pockets around the teeth (Kaldahl et al. 1996). The residual pocket depth is positively related to the risk of future periodontal breakdown (Badersten et al. 1990, Claffey et al. 1990).

For many intraoral soft-tissue surgical procedures, the laser has become a desirable and dependable alternative to traditional scalpel surgery (Cobb et al. 2010). Gold and Vilardi (1994) evaluated the efficacy of a low-power pulsed Nd:YAG laser for removing pocket lining epithelium in humans with moderate periodontitis. The laser proved capable of removing pocket lining epithelium in moderately deep pockets. In addition, the Nd:YAG laser has shown a bactericidal effect (Kranendonk et al. 2010),

Conflict of interest and source of funding statement

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suppressing and eradicating putative periodontal pathogens from periodontal pockets (Cobb et al. 1992, Ben Hatit et al. 1996).

Debridement of the diseased root surface is usually performed by mechanical scaling and root planing using manual or power-driven instruments. Power-driven instruments such as ultrasonic scalers are frequently used for root surface treatment, as they are effective in removing plaque, calculus and endotoxin, and cause less root surface damage than hand scalers (Torfason et al. 1979, Loos et al. 1987, Folwaczny et al. 2004). Although the data are rather limited, the clinical outcome with the Nd:YAG laser appears to be comparable to the effect of SRP with regard to periodontal inflammation parameters (Slot et al. 2009). Investigators have also proposed using the Nd:YAG laser as an adjunct to SRP (Radvar et al. 1996, Neill & Mellonig 1997). Current evidence suggests that using the Nd:YAG laser for treatment of chronic periodontitis may be equivalent to SRP with respect to the reduction in subgingival bacterial populations (Cobb 2006, Schwarz et al. 2008). However, the Nd:YAG laser is not suitable for root planing or removal of mineralized accretions such as dental calculus (Cobb et al. 2010). Accordingly, this type of laser is indicated as an adjunct to SRP. Furthermore, improper use of the fibre tip may result in unfavourable thermal changes (Aoki et al. 2004, Schwarz et al. 2008).

Among dentists and dental hygienists in the Netherlands, the Genius Nd:YAG-pulsed laser with water and air coolant (Genius, Mølsgaard Dental, Copenhagen, Denmark) is used as an adjunct to ''non-surgical'' treatment of periodontitis, as suggested by Lioubavina-Hack (2002). This is a water-cooled laser that releases energy in short interrupted time intervals (pulsed). It has an optical fibre tip that approximates the diameter of a periodontal probe. The flexible fibre optic cable provides good operability, making it suitable for reaching the bottom of the periodontal pocket.

Use of an air–water spray for irrigation during laser irradiation provides a thermal gradient for removal of heat from tissue surfaces. The process of surface cooling is a direct result of the extensive heat capacity of water, which absorbs a significant amount of the surface heat generated by the laser, and thus, effectively limits collateral tissue damage. In addition, due to continual renewal of the air–water spray, simultaneous cooling of the tissue surface occurs by convection. Based on these characteristics, it is theoretically possible to stabilize surface temperatures (Spencer et al. 1996). The water irrigation also reduces the clogging of the probe with debris, thereby preventing a buildup of areas of excessive heat (Qadri et al. 2010b). Scientific evidence supporting the use of this Nd:YAG laser brand featuring water and air cooling has, until recently, only been published as abstracts (Lioubavina et al. 1997, Jensen et al. 2003). Two recent papers describing the short-term and the long-term effect of a single laser application in supplement to scaling and root planning showed a positive effect in favour of this laser (Qadri et al. 2010a, b) whereas another study did not find such a superior clinical effect (Jensen et al. 2010). A recent ''in vitro'' study showed that 15 s of this Nd:YAG laser use was effective for total killing of various periodontal pathogens (Kranendonk et al. 2010).

The purpose of this study was (1) to test whether the use of the Nd:YAG laser with water and air coolant adjunctive to SRP results in greater clinical improvements than ultrasonic scaling alone, (2) to investigate the reduction in the number of subgingival microorganisms directly after subgingival SRP with or without adjunctive Nd:YAG laser treatment and (3) to evaluate post-operative experiences and patient comfort with regard to the treatments provided.

Material and Methods

Ethical aspects

The study protocol was approved by the Medical Ethics Committee of the Academic Medical Center in Amsterdam (MEC #05/278). All voluntary participants were informed of the outline, purpose and duration of the study and signed an ''informed consent'' form.

Study population

For the present study, 19 patients (11♂, 8♀) were enrolled from March 2006 to February 2007. All patients had been referred by their general dentists to a clinic specializing in periodontal therapy. The following inclusion criteria were used: healthy, non-institutionalized patients; at least 30 years of age; a minimum of five natural teeth in every quadrant; clinical diagnosis before active periodontal treatment; moderate-to-severe generalized periodontitis characterized by the presence of $\geqslant 1$ site per quadrant with pocket depth > 6 mm and inter-proximal attachment loss of $\geqslant 3$ mm, presence of bleeding on pocket probing (BOPP) and radiographic evidence of alveolar bone loss; and systemically healthy. Exclusion criteria were professional periodontal therapy before enrollment in the study; antibiotics use for any purpose within 3 months before entering the study; and dental personnel.

Clinical assessments

The following measurements were performed before the initial therapy appointment and after the 3-month evaluation period.

- Probing pocket depth (PPD) using a manual probe (PCPUNC 15 mm probe, Hu-Friedy® Hu-Friedy Inc., Leimen, Germany);
- BOPP (Van der Velden 1979);
- Plaque index (Silness & Löe 1964, Danser et al. 2003).

All clinical measurements were taken at six sites (mesio-buccal, buccal, disto-buccal, mesio-lingual, lingual and disto-lingual) of each tooth and were rounded off to the nearest millimetre. All clinical measurements were performed by the same investigator, who was blinded to the treatment (W. H. S.). Access to the data of former assessments was not allowed during the course of the study.

Clinical procedure

This study was an examiner-blind, randomized, controlled 3-month clinical trial using a split-mouth design with a treatment protocol similar to Henskens et al. (1996) and Winkel et al. (2001). After establishing eligibility to enter the study and submitting written approval, patients were scheduled for the first session. A medical history form, including smoking habits and history, was filled out. A second investigator performed all treatments (A. A. K.). Local anaesthetics were provided during SRP using ultrasonic and hand instruments and laser treatment. Treatment was performed in two sessions approximately 1

week apart. During each session, teeth in two contra-lateral quadrants were SRP using a piezoelectric ultrasonic unit (Piezon Master, EMS, Nyon, Switzerland) at a moderate setting and with the appropriate tips for initial therapy (A, P, PS, PL1–5, EMS). In addition, where deemed appropriate by the dental professional, hand instruments were used (204SD, 12/13 11/14 Hu-Friedy® Hu-Friedy Inc.). Depending on the randomization immediately thereafter, all pockets $\geqslant$4 mm were additionally treated with the Nd:YAG laser immediate following SRP or no additional treatment was provided. The non-laser treated contra-lateral teeth became controls. Randomization was based on a predetermined computer-generated set of random numbers that were obtained via http://www.random.org. The primary investigator and study coordinator (G. A. W.) was responsible for concealing the allocation. Sealed envelopes were prepared that stated which quadrants would receive additional laser treatment. These envelopes were opened only after SRP was finished. Following instrumentation, all supra-gingival surfaces were polished with a rubber cup and point in combination with an abrasive paste (*Tri-Fluor-O-Clean*, KerrHawe, Bioggio, Switzerland). The time necessary for treatment was recorded after every session. In addition, patients received instruction in personal oral hygiene procedures. After approximately 6 weeks, the level of oral hygiene was evaluated using an erythrosine stain. No other treatment except individual oral hygiene instructions was provided. Subjects were asked to continue their oral hygiene procedures, including both brushing and inter-dental cleaning, in adherence to the given instructions. At the end of the study period (3 months), all clinical measurements were recorded again. Figure 1 shows the flow diagram illustrating the passage of participants through this clinical trial.

Laser treatment

A solid-state crystal Nd:YAG laser (Genius Periodontal A/S, Copenhagen, Denmark) was used as additional therapy in the randomly allocated quadrants after SRP (SRP+Nd:YAG). The details for settings of this water-cooled Nd:YAG laser are shown in Table 1. The epithelium lining the inner pocket wall was dampened and the pocket was disinfected using the laser. The fibre tip was held with light pressure in contact with the tissue and parallelly aligned to the tooth. The ''perio'' setting of the laser was used adjusting power and cooling to allow a smooth instrumentation. The round flexible 0.6 mm laser fibre (0.2826 mm^2) emerging from the handpiece tip (see Fig. 2) was adjusted in length to correspond to the periodontal pocket probe charting. Small horizontal excursions of about 2 mm along the gingival margin were made, penetrating no deeper into the pocket than the probing depth. The laser was applied for no longer than 60 s per site (The tooth was divided into four sites; mb, ml, db, dl). Remnants of gingival tissue were removed using a manual curette. All laser procedures were performed with protective eyewear on the patient, dentist and assistant. At the decision of the operator, the fibre tip was cleaned when visible debris was attached to ensure its optical properties. The used laser fibre tip was cleaved and discarded. The laser fibre and handpiece were then cleaned. The handpiece was sterilized using an autoclave. Figure 2 shows the fibre tip and the handpiece tip. A mixture of air and water was sprayed over the fibre tip originating from the tip handle circumferential around the fibre.

Table 1. Nd:YAG laser parameters and range in the ''perio'' setting

Wavelength	–	1064
Power*	Range 1–12 W	6
Water*	Range 1–12	5
Air*	Range 1–12	5
Frequency	Range 10–100 Hz	50
Pulse duration	Range 100–800 μs	250
Pulse energy	Range 400–800 mJ	400
Energy density	J/cm^2	142†

*Display settings.
†One has to understand that the energy density J/cm^2 was calculated. However, due to the uncertainty about the actual light-emitting surface and the total area of tissue irradiated one has to interpret this with caution.

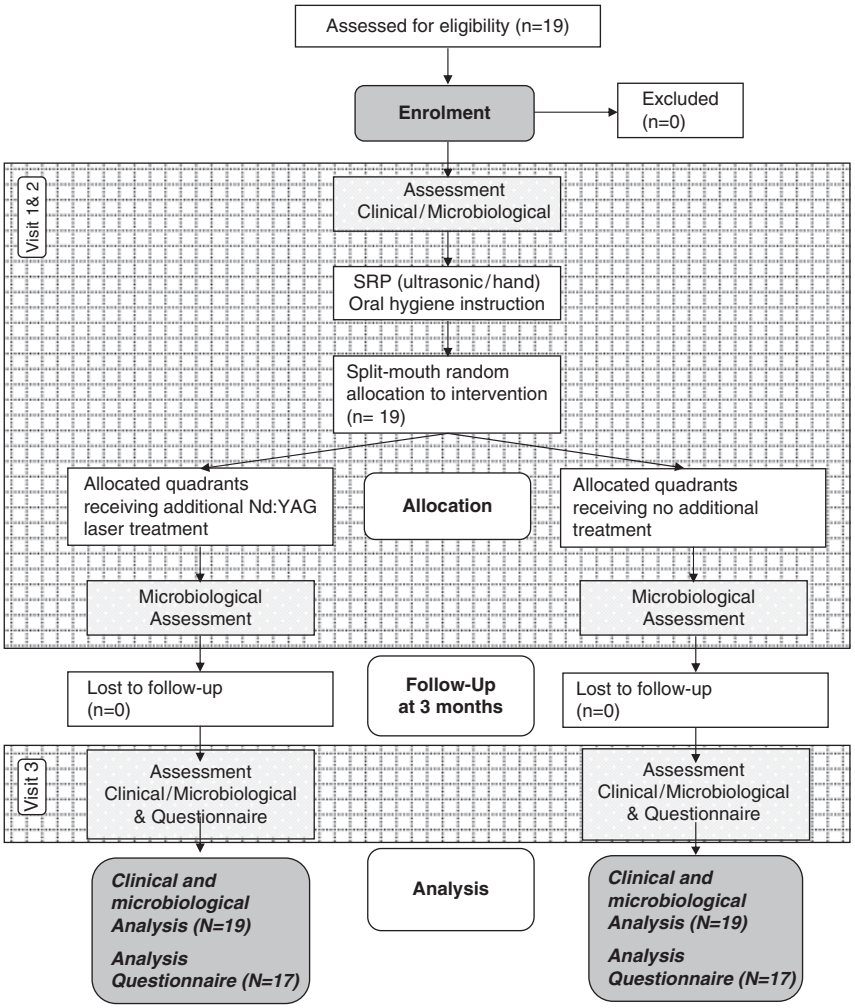

Fig. 1. Flowchart depicting subject enrollment and measurements.

Fig. 2. Nd:YAG laser fibre tip emerging out of handpiece tip.

Microbiological procedures

Sampling

The deepest inter-proximal site in each quadrant with BOPP was selected for microbiological sampling (Mombelli et al. 1991). In each quadrant, one pocket was sampled by means of two paper points. Next, samples were pooled for either the quadrants that received SRP alone or those that were treated by SRP+Nd:YAG. Selected sites were sampled at pre-instrumentation, immediately post-instrumentation and 3 months after initial treatment. Sites were subjected to careful removal of supragingival plaque deposits with a scaler. To avoid salivary contamination, the selected area was isolated with cotton rolls and gently air-dried. Before bacterial sampling, a periodontal probe (PCPUNC 15 mm probe, Hu-Friedy®) was inserted in the approximal pocket along the axis of the tooth until definite resistance was met. Two endodontic paper points (size 40#, Johnson & Johnson, Windsor, NJ, USA) were inserted for 10 s each into the pocket along the probe, with care taken not to fold or to push them into another area (Rhemrev et al. 2006). The paper points from the selected sites were collected in 1.8 ml of reduced transport fluid (RTF) (Syed & Loesche 1972).

Culture

Samples were cultured for microbiological analysis within 12 h. Samples were vortexed for 30 s and 10-fold serially diluted in RTF; 0.1 ml of each dilution was plated on 5% horse blood agar plates (Oxoid No. 2, Basingstoke, UK) supplemented with haemin (5 mg/l) and menadione (1 mg/l) for determination of the total anaerobic bacterial counts and specific periodontal pathogens. Samples were subsequently plated on trypticase soy serum–bacitracin–vancomycin plates (TSBV) for isolation and counting of *Aggregatibacter actinomycetemcomitans* (Slots 1982). TSBV plates were incubated in air with 5% CO_2 at 37°C for 3 days; blood agar plates were incubated for 14 days at 37°C in 80% N_2, 10% CO_2 and 10% H_2. Presence and proportions of the putative periodontal pathogens *Porphyromonas gingivalis*, *Prevotella intermedia*, *Tannerella forsythia*, *Fusobacterium nucleatum, Parvimonas micra* and *Campylobacter recta* were determined on the anaerobic blood agar plates (Van Winkelhoff et al. 1985). Identification of the selected bacterial species was based on Gram staining, cell and colony morphology, air tolerance, production of catalase and a number of biochemical reactions (Van Winkelhoff et al. 1986). *A. actinomycetemcomitans* was identified on the basis of its characteristic colony morphology (star-like inner structure), a positive catalase reaction with 3% hydrogen peroxide and a set of specific enzymes. Total colony-forming units (TCFUs) were estimated on the horse blood agar plates and expressed as the number of viable counts per millilitre of transport medium.

Questionnaire

Immediately after treatment, seven questionnaire forms were provided to each subject, one for immediate post-operative evaluation and one for each day of the following 6 days. Patients were asked to fill out the questionnaire at the end of each day. A visual analogue scale was used to assess patients' perception of pain, sensitivity discomfort, swelling and bleeding during and after treatment. This scale ranged from 0 to 10. Subjects marked a point on a 10-cm-long uncalibrated line with the negative extreme response (0) on the left end and the positive extreme response (10) at the right end. Additionally, the numbers of analgesic tablets taken were assessed.

Statistical analysis

Primary response variables were pocket depth and BOPP. For clinical measurements, a patient-level response variable was calculated for each parameter by computing the mean scores per patient at baseline and after therapy. The % of pockets $\geqslant 4$ mm was enumerated. Furthermore, for pocket probing measurements, an overall mean value of treated sites initially measuring $\geqslant 4$ mm was calculated. Parametric and non-parametric tests were performed where appropriate. Analyses were performed by ''intention to treat''. p values < 0.05 were accepted as significant. For probing depth reduction, the present design was able to discern a difference of 0.5 between therapies with a standard deviation of 0.7 and a power of $\geqslant 80\%$. Questionnaires were evaluated using either parametric tests comparing outcomes or VAS scales concerning the two treatments. The statistical analysis was performed by an investigator (N. A. M. R.), who was blinded to the randomization.

Results

Clinical findings

For the present study, 19 untreated periodontitis patients were enrolled from March 2006 to February 2007. In total, 11 males and eight females with a mean age of 45.3 (± 8.67) years (range: 34–62 years) were selected. Ten of the subjects were smokers, three were former smokers and six had never smoked. The subjects were selected from those consulting the Clinic for Periodontology in Utrecht, the Netherlands for treatment of periodontal disease. All enrolled patients completed the 3-month study. At baseline, both contra-lateral quadrants (SRP+Nd:YAG *versus* SRP) were found to be balanced with respect to the clinical parameters.

The average SRP instrumentation time per quadrant was 33.89 (± 5.16) min. The extra time needed for the adjunctive use of the laser was 8.47 (± 4.38) min. per quadrant. Table 2 shows the means (SD) of all clinical parameters at baseline and end, comparing SRP+Nd:YAG laser *versus* SRP. After 3 months, all parameters were improved significantly compared with baseline for both regimens. No statistically significant differences for any of the investigated parameters were found at the baseline and the end-trial between the two treatment modalities. No adverse effects of laser treatment were observed or reported by the patients.

Microbiological findings

The results of the effects of instrumentation on the total anaerobic counts of the subgingival microflora during the

Table 2. Means (SD) of all clinical parameters during the study for both treatment modalities

All subjects ($N = 19$)	SRP+Nd:YAG			SRP			T-test† (p-value)	95% Confidence interval
	baseline	3 months	difference	baseline	3 months	difference		
Plaque index	1.40 (0.28)	1.06 (0.30)*	0.34 (0.33)	1.46 (0.26)	1.12 (0.26)*	0.34 (0.28)	0.947	(– 0.12; 0.11)
BOPP	1.57 (0.31)	0.81 (0.26)*	0.76 (0.25)	1.58 (0.29)	0.87 (0.26)*	0.71 (0.25)	0.435	(– 0.09; 0.19)
PD	4.19 (0.69)	3.54 (0.49)**	0.65 (0.43)	4.14 (0.58)	3.52 (0.43)**	0.62 (0.33)	0.617	(– 0.09; 0.16)
PD of sites ⩾4 mm	5.23 (0.63)	4.83 (0.39)**	0.40 (0.42)	5.22 (0.57)	4.68 (0.29)**	0.54 (0.40)	0.138	(– 0.32; 0.05)
% sites PD ⩾4 mm	28.02 (9.23)	18.68 (9.22)	9.34 (5.21)	28.27 (8.02)	19.67 (7.95)	8.60 (4.46)	0.450	(– 2.73; 1.26)

*Significantly different from baseline ($p < 0.05$, Wilcoxon's test).
**Significantly different from baseline ($p < 0.05$, paired t-test).
†T-test comparing incremental change from baseline – end for each treatment modality.

study are presented in Table 3. The mean total anaerobic counts from the selected sites, determined by culture, were not statistically different at any time between the two treatment modalities (Paired t-test). Immediately after instrumentation, both SRP+Nd:YAG and SRP selected sites showed significantly reduced TCFUs at 0.09×10^6 and 0.44×10^6/ml, respectively. However, at 3 months post-treatment, the mean TCFUs of the SRP+Nd:YAG and SRP selected sites had increased to 27.59×10^6/ml and 44.93×10^6/ml, respectively. The mean TCFUs 3 month post-treatment was not significantly different compared with pre-instrumentation for both treatment modalities.

Table 4 presents all subjects found to be positive for each of the analysed species, namely *A. actinomycetemcomitans*, *P. gingivalis*, *P. intermedia*, *T. forsythia*, *F. nucleatum* and *C. recta*, at pre-instrumentation, immediately post-instrumentation and at 3 months post-instrumentation. Immediately after instrumentation, all species showed a decreased prevalence. At 3 months post-instrumentation, there was a noticeable tendency towards relapse to baseline for *P. micra* and *F. nucleatum*. The presence of *A. actinomycetemcomitans* in the SRP+Nd:YAG group was no longer detected in culture immediately post-instrumentation or at the 3-month visit.

Questionnaires

Table 5a shows the questions and suggestions related to the two extremes. Table 5b shows the results of the questionnaire. Only 17 subjects returned the questionnaires. Repeated measure analysis between both treatment modalities showed only a significant difference for post-operative pain in favour of SRP. Post-operative experience of pain was more pronounced in the first 3 days for the SRP+Nd:YAG group. Table 6 shows the mean number of analgesics used by patients in each group per day. In the course of the day following treatment, the SRP+Nd:YAG group used 3 × more analgesics than the SRP group. No analgesics were used following either treatment after day 2.

Discussion

The purpose of this study was to test whether use of an Nd:YAG laser with water and air coolant after SRP results in a greater clinical improvement than SRP alone. The appointment protocol suggested by Raffetto (2004) was used, where the tooth and root surfaces were debrided first, followed by laser bacterial reduction and dampening/coagulation of the epithelial tissue. The results clearly show that both SRP and SRP+Nd:YAG treatment resulted in a decrease of all clinical parameters tested. However, the difference between responses to SRP and responses to SRP+Nd:YAG was small and not statistically significant after 3 months. These results are in support of a recent systematic review, which concluded that there is limited evidence to support the adjunctive use of a pulsed Nd:YAG laser as compared with conventional therapy alone (SRP, ultrasonics and/or hand instrumentation) in the initial treatment of patients with periodontitis (Slot et al. 2009). Schwarz et al. (2008) in their review also concluded that there is insufficient evidence to support the clinical application of the Nd:YAG laser. The present results now add to the evidence that the Nd:YAG has no adjunctive effect over SRP alone in initial periodontal treatment. This is also supported by the clinical and microbiological outcome of two other recent studies (Gómez et al. 2010, Jensen et al. 2010). The results of two papers describing short-term and long-term effects within the same patient population are, however, in conflict with this conclusion (Qadri et al. 2010a, b). The reason for this discrepancy is unclear. It might be attributable to differences in laser settings. Which in the Qadri et al. (2010a, b) studies were lower and set at 4 W. Their study was also restricted to mandibular teeth. Furthermore, it is striking that only in the test sites a reduction in plaque scores was observed whereas in control sites no such effect was found. This may have impacted clinical outcomes such as PPD reduction. In the present study, this was not the case where the improvement in plaque control was similar for both treatment modalities.

There was no external control of laser parameters during the treatments within the present experiment design. Because this infrared radiation as well as the effects of laser tissue interactions are not visible, this implies that there was no control in order to ensure the correct working of the tested system. However, before the laser system was set-up for this study, it was serviced and tested to ensure that it worked according to the manufacturers specifications.

The ''classical'' Nd:YAG laser parameters used in periodontology are between 0.5 and 3 W (Ishikawa & Sculean 2007, Slot et al. 2009). The present study used a substantially higher level of 6 W (Table 1). In order to limit side effects with this higher power parameter the last provided an air–water coolant simultaneously with laser activation, which was directed over the tip. A substantial amount of the surface heat, generated by a laser, was therefore dissipated (Spencer et al. 1996).

Generally, subgingival debridement in combination with oral hygiene instruction by itself is an effective treatment modality (Badersten et al. 1981, 1984a, b, Pihlstrom et al. 1981). When

Table 3. Mean total CFU/ml ($10^6 \pm$ SD) during the study for both treatment modalities

$N = 19$	SRP+Nd:YAG	SRP	*T*-test (*p*-value)	95% Confidence interval
Pre-instrumentation	59.18 (81.89)	54.03 (83.63)	0.631	(– 27.27; 16.89)
Immediately post-instrumentation	0.09 (0.34)*	0.44 (1.37)*	0.287	(– 0.33; 1.05)
3 months post-instrumentation	27.59 (52.15)	44.93 (123.77)	0.576	(– 46.56; 81.24)

*Significantly different from pre-instrumentation ($p < 0.05$, paired *t*-test).

Table 4. Prevalence among subject of specific periodontal bacteria during the study for both treatment modalities

$N = 19$	Time of sampling	Periodontal bacteria						
		Aa	Pg	Pi	Tf	Pm	Fn	Cr
SRP+Nd:YAG	Pre-instrumentation	1	5	10	16	14	17	1
	Immediately post-instrumentation	–	3	3	4	8	7	1
	3 months post-instrumentation	–	1	6	10	16	17	–
SRP	Pre-instrumentation	–	8	7	19	18	17	2
	Immediately post-instrumentation	–	4	4	5	6	5	–
	3 months post-instrumentation	–	4	3	7	15	16	2

Aa, *Aggregatibacter actinomycetemcomitans*; Pg, *Porphyromonas gingivalis*; Pi, *Prevotella intermedia*; Tf, *Tannerella forsythia*; Fn, *Fusobacterium nucleatum*; Pm, *Parvimonas micra*; Cr, *Campylobacter recta*.

an effective treatment modality is used as a golden standard of comparison, it may be difficult to show any adjunctive effect in addition to the original treatment, as was the case with the Nd:YAG laser in the present study (Timmerman et al. 1996). The majority of the treated patients were (former) smokers. This may have had an impact on the clinical outcome. Although this was a split-mouth model, this risk factor may cause an underestimation of the magnitude of a potential clinical effect comparing test and control sites (Preber & Bergström 1986). On the other hand, because smoking is a risk factor, and many periodontal patients are (former) smokers (Van der Weijden et al. 2001), the outcomes of this study are applicable to periodontal practice.

Results of microbiological studies are highly dependent on the sampling procedure used. It has been shown that the composition of the microflora may change relative to the distance from the gingival margin (Listgarten 1976, Slots et al. 1979, Magnusson et al. 1984). Treatment causes periodontal tissues to tighten around the teeth (Beardmore 1963). As a consequence, it is more difficult to introduce a paper point to the bottom of a pocket at re-evaluation. To avoid sampling problems, a standardized sampling technique, described by Rhemrev et al. (2006), was used.

Relatively few studies have investigated the microbiological effect of subgingival scaling and root planing directly after completion of the procedure. This aspect was recently investigated by Rhemrev et al. (2006). They observed that mechanical cleaning itself has a limited effect in actually removing bacteria. In agreement with Rhemrev, the present ''in vivo'' effect does not support the ''in vitro'' effect as found previously by Kranendonk et al. (2010) where after 15 s of laser use total killing of perio pathogens was observed. In the present study, a significant reduction in CFU's was observed between pre- and immediately posttreatment. However, no difference in effect between SRP+Nd:YAG and SRP was established. Furthermore, at 3 months post-instrumentation, TCFUs values were not different between treatment and not different from baseline. This result is in agreement with previous studies, which have shown that re-colonization of the subgingival area by microorganisms may occur within 2–8 weeks of treatment (Mousques et al. 1980, Magnusson et al. 1984, Van Winkelhoff et al. 1988, Wade et al. 1992).

Results of the present study show that immediately post-instrumentation, there was a trend towards reduced prevalence of *P. gingivalis* as compared with pre-instrumentation, whereas Rhemrev et al. (2006) found that all patients positive for *P. gingivalis* remained culture positive immediately post-instrumentation. Three months post-instrumentation in the present study, a trend towards reduced prevalence of *P. gingivalis*, *P. intermedia* and *T. forsythia* was seen. Rhemrev et al. (2006) had already observed this shift in the composition of microflora at 2 weeks post-instrumentation. It seems feasible to suppose that such a shift lasts for at least 3 months after treatment, a finding in line with the observed clinical improvement in periodontal condition.

In each quadrant, one sample was taken using two paper points, and samples were pooled for either the quadrants that received SRP alone or those that were treated by means of SRP+Nd:YAG. Mombelli et al. (1991) evaluated the feasibility of detecting microorganisms using selected sites in order to indicate increased proportions in periodontitis patients. It was concluded that in some periodontitis patients, the outcome of a test depends greatly upon the number of samples taken and the strategy of site selection. Selection of the deepest pocket in each quadrant was the most efficient method of sampling. In the present study, samples were taken from the deepest pocket in each quadrant for the SRP+Nd:YAG and SRP sites. Whether a pooled sample of two sites is sufficient for assessment of the actual presence of a given microorganism remains a matter of discussion.

Following initial periodontal treatment using hand and ultrasonic instruments with or without the additional use of an Nd:YAG laser, a patient may experience some degree of pain and swelling in addition to post-operative sensitivity to warm and cold temperatures. Harris et al. (2004) performed a retrospective analysis of patients receiving laser sulcular debridement. The four clinicians reported anecdotally that patients seemed to experience less pain and discomfort and recover more rapidly when the laser was included in the treatment protocol than when it was excluded. It is theorized that this pain

Table 5a. Questions used in the questionnaire with extremes from the VAS score

Paraphrase	Complete question	With extremes	
		from (0)	to (10)
Bleeding	Did you experience any bleeding at the treated sites today?	''No'' bleeding	''Very much'' bleeding
Swelling	Did you experience any swelling in the mouth today?	''No'' swelling	''Very much'' swelling
Post-op pain	Did you experience any post-operative pain in the mouth today?	''No'' pain	''Very much'' pain
Sensitivity	Did you experience any post-operative experience of sensitivity to warm/cold today?	''No'' sensitivity	''Very much'' sensitivity

Table 5b. Mean (SD) VAS score response (0.0–10.0) to the questionnaire presented by regimen

($N = 17$)	Day	0	1	2	3	4	5	6	p-value**
Bleeding	SRP+Nd:YAG	4.21 (3.49)	2.88 (2.88)	2.05 (2.32)	1.45 (1.57)	1.06 (1.38)	0.81 (1.55)	0.58 (1.13)	0.221
	SRP	2.98 (2.69)	2.10 (2.42)	1.08 (1.78)	1.05 (1.84)	0.72 (1.08)	0.32 (0.50)	0.30 (0.45)	
	p-value*	0.166	0.327	0.116	0.400	0.323	0.105	0.197	
Swelling	SRP+Nd:YAG	3.76 (2.97)	3.05 (2.80)	2.09 (2.02)	1.14 (1.30)	1.20 (1.90)	0.93 (1.82)	0.84 (1.71)	0.060
	SRP	2.01 (2.12)	1.15 (1.66)	0.61 (1.37)	0.84 (1.55)	0.56 (1.21)	0.48 (1.17)	0.33 (0.70)	
	p-value*	0.003	0.016	0.009	0.551	0.293	0.364	0.210	
Post-op pain	SRP+Nd:YAG	5.89 (2.64)	3.66 (2.95)	2.44 (2.73)	2.71 (2.89)	1.66 (1.79)	1.08 (1.66)	1.05 (1.45)	0.033
	SRP	4.63 (2.96)	1.72 (1.92)	1.04 (1.56)	1.03 (1.53)	0.85 (1.29)	0.67 (1.23)	0.55 (1.19)	
	p-value*	0.017	0.008	0.066	0.043	0.178	0.313	0.106	
Sensitivity	SRP+Nd:YAG	4.38 (3.77)	3.49 (3.55)	2.26 (2.97)	2.01 (2.95)	1.92 (2.95)	1.95 (3.24)	2.05 (3.16)	0.588
	SRP	3.07 (2.90)	2.12 (2.75)	2.36 (3.00)	1.86 (2.80)	1.65 (2.59)	1.71 (2.52)	1.01 (1.50)	
	p-value*	0.060	0.029	0.782	0.756	0.502	0.482	0.145	

**Repeated measure analysis.
*Paired t-test.

Table 6. Number of analgesic tablets taken following each treatment

Day	0		1		2	
$N = 17$	# subjects	# tablets	# subjects	# tablets	# subjects	# tablets
SRP+Nd:YAG	12	27	3	5	1	1
SRP	5	9	1	1	1	1

reduction may be due to the protein coagulum, which is formed on the wound surface and may act as a biological dressing. These anecdotal remarks have not, however, been scientifically validated (Rossmann 2002). In the present study, the post-operative pain as appears from the questionnaire was more pronounced in the SRP+Nd:YAG group. However, it should be emphasized that the patients were not masked with respect to the modality of treatment. This may have affected patients' judgements regarding the novel instrument. On the other hand, on day one the SRP+Nd:YAG group also used more analgesics, which corresponds with the complaint of post-operative pain.

Conclusion

The results of the present study indicate that SRP, with or without the adjunctive use of an Nd:YAG laser, result in a lowered subgingival bacterial load immediately post-instrumentation. In addition, the primary clinical parameters (BOPP, PPD) comparing baseline and end following both treatment modalities showed an improvement. However, at the 3-month evaluation, no additional clinical or microbiological advantage could be established for the water-cooled Nd:YAG laser.

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Clinical Relevance

Scientific rationale for the study: The Nd:YAG laser is capable of removing pocket lining epithelium and has a bactericidal effect, suppressing and eradicating putative periodontal pathogens from periodontal pockets. Investigators have proposed the use of the Nd:YAG laser as an adjunct to ultrasonic scaling and root planing. The cooled Nd:YAG laser allows for higher energy setting without adverse effects and has recently been shown to be effective in bacterial killing.

Principal findings: The results of the present study indicate that subgingival mechanical SRP, especially with the adjunctive use of an Nd:YAG laser, has the effect of lowering the total bacterial load immediately post-instrumentation. However, clinical improvement of periodontal status was found to be comparable with or without the adjunctive use of an Nd:YAG laser after initial treatment by SRP.

Practical implications: The results of this study are applicable to patients diagnosed with moderate-to-severe periodontitis who are willing to undergo treatment by a specialist. Because clinical results are not improved by adding laser treatment to conventional ''non-surgical'' periodontal therapy, the use of the Nd:YAG laser as an adjunct to debridement should be questioned.