

## Article

# Periodontal Decontamination Induced by Light and Not by Heat: Comparison between Oxygen High Level Laser Therapy (OHLLT) and LANAP

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**Abstract:** In periodontology, lasers have been suggested for the nonsurgical treatment of chronic periodontitis. Many wavelengths were tested, but unfortunately, most parts were not efficient. An Nd:YAG laser was applied in a specific protocol named the laser-assisted new attachment procedure (LANAP). LANAP seems to facilitate the refurbishment of new tissues from supporting structures of the periodontium, wherein the unhealthy surface of the roots exhibits pristine attachments in human beings. Photodynamic therapy (PDT) was investigated too. The aim of our study is to show the effects of oxygen high-level laser therapy (OHLLT) in removing all bacterial deposits on the root or implant surface by means of mechanical instrumentation and laser irradiation compared to LANAP and to nonsurgical debridement of periodontal pockets. At 7 days post-treatment, a real-time PCR test had similar results on the OHLLT and LANAP groups. After 9 months, all periodontal pockets were treated successfully, not showing significant differences in the clinical results between OHLLT and LANAP and with a decrease in the plaque index, bleeding on probing and probing depth compared with the nonsurgical debridement group.

**Keywords:** periodontitis; photodynamic therapy; diode laser; Nd:YAG laser; hydrogen peroxide; decontamination; periodontal bacteria



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## 1. Introduction

Periodontitis and peri-implantitis are pathologies that share the same microflora in periodontal lesions (pockets) and develop damage consisting of the loss of supporting bone tissue. The new classification of periodontal disease identifies the importance of bacterial contamination combined with the patient's individual susceptibility to get periodontitis.

A high percentage of failures [1] after nonsurgical or surgical periodontal or peri-implant instrumentation [2,3] is attributable to an incomplete detoxification of the root-implant surface as well as that of the adjacent soft tissues due to the presence of aggressive bacterial aggregations and red and orange Socransky complexes, such as *Porphyromonas gingivalis*, *Treponema denticola*, *Bacteroides forsythus*, *Fusobacterium nucleatum* and *Peptostreptococcus micros* [4–15]. The literature has shown that, in the presence of these bacteria, at one year, even under strict professional control, the recolonization of the periodontal pockets was total, either with nonsurgical therapy protocols or with regenerative surgery [1,2]. Therefore, the only solutions for advanced periodontitis involve the systematic use of combinations of antibiotics (against the indications of the WHO (World Health Organization), which recommends the reduction of the administration of these drugs to reduce the phenomenon of antibiotic resistance).

In recent years, many studies have been conducted to apply protocols that use lasers. Among the various types of lasers, those belonging to the family of penetrating lasers (between 600 nm and 1100 nm in wavelength) seem to be particularly suitable if properly parameterized.

### 1.1. Laser in Periodontics

Lasers have been investigated in periodontology since the 1980s. The numerous articles available in the literature suggested at the time that possibilities for decontamination by laser radiation could be found [16,17].

Furthermore, the Nd: YAG laser, interacting with endogenous chromophores (such as melanin and hemoglobin), would provoke the vaporization of the granulation tissue in a selective way, associated with reduced bleeding thanks to the hemostatic effect [6,7].

The laser-assisted new attachment procedure (LANAP) protocol applies this principle and finds comforting evidence in the literature [6–10]. However, the increase, albeit localized, of the temperature in the periodontal pockets exposes the patient's tissues to the risk of tissue alteration, which can lead to temporary consequences, such as the onset of pain and post-treatment oedema, or permanent consequences, such as the loss of periodontal tissue damaged by the thermal insult.

To overcome the post-operative consequences and reduce the risks associated with the use of the LANAP technique, which is operator-dependent and highly influenced by the experience of the clinician with high risk of overheating of the periodontal tissues, borrowing from oncology and even periodontology, the therapeutic protocols of photodynamic therapy were used, being understood as the inactivation of bacteria and micro-organisms induced by light and not by heat, which requires a photosensitizer or chromophore capable of linking to the bacterial membrane, delivering singlet oxygen, released thanks to the laser energy applied in the periodontal pocket, thanks to the oxygen transported by the chromophore [11–14,18,19].

Singlet oxygen, an unstable molecule present in nature (in the northern lights, for example) with a very low half-life in the order of less than a microsecond, is released in the periodontal pockets thanks to the interaction with the laser's energy in a high quantity due to the higher frequency of the laser being used (more than 6000 Hz).

However, the literature has shown how the proposed photodynamic therapy techniques, which use low-power penetrating lasers below 0.8 W (often proposed in continuous emission and without power peaks above 2 W) combined with a dye, such as Toluidine blue, phenothiazine chloride or Indochina, are insufficient to eradicate the bacteria responsible for periodontal disease and give, in the long term, results comparable to nonsurgical periodontal therapy [16]. Even in these tests, the results were not convincing [20].

To overcome these limitations, researchers have therefore developed protocols with diode lasers suitable for using high peak powers with low average powers below 0.8 W and an insuperable limit for not having a thermal increase of the tissues exceeding 42 degrees [21–28]. In this way, the laser can be operated in the periodontal pockets without an appreciable thermal rise, if not a slight vasodilation, which will be represented by light post-treatment bleeding [29–31].

Furthermore, photodynamic therapy protocols without dye have been proposed, which combine high-power lasers with solutions with high oxygen contents (above 2%) with the aim of releasing singlet oxygen in the periodontal pockets (oxygen high-level laser therapy (OHLLT) protocol or photodynamic therapy without dye).

Singlet oxygen can break down the bacterial membranes of microorganisms belonging to the red and orange Socransky complex, the main culprits of periodontal disease.

Few studies demonstrated the efficacy of oxygen high-level laser therapy (OHLLT) in the treatment of chronic periodontitis by means of a clinical and microbiological evaluation carried out with real-time PCR [25–29,31]. OHLLT combines the use of diode lasers (Wiser, Lambda) and a solution based on hydrogen peroxide, the Sioxil<sup>®</sup> solution, composed of stabilized hydrogen peroxide at 10 volumes (3%) in which cytotoxic residues are eliminated

and glycerophosphate is added, which favors cellular biostimulation of fibroblasts and keratinocytes [25,29–31]. After treatment, significant decreases in bacteria were observed for each species and for the total number of bacteria, including *Aggregatibacter actinomycetemcomitans* and *Porphyromonas gingivalis*, demonstrating that the OHLLT protocol is effective on periodontitis. The results showed that stabilized hydrogen peroxide (Sioxil® solution) performed better in the regeneration test than ordinary hydrogen peroxide at 10 vol. 3% while the biocidal activity remains comparable.

### 1.2. Aim of the Study

The aim of this research is to compare the two techniques, LANAP and OHLLT, for the nonsurgical treatment of periodontal disease, evaluating the microbiological efficacy (using real-time PCR) of the two techniques, compared to the conventional, periodontal nonsurgical protocol (control group).

## 2. Materials and Methods

### 2.1. Study Sample

This study evaluated the effectiveness of laser therapy combined with two different types of protocols. Thirty patients aged between 18 and 65 (18 men and 12 women) were recruited. Patients were treated between February 2019 and January 2020 at a private practice in Bergamo, Italy, and all of them signed the appropriate informed consent.

The present pilot study was approved by the ethics committee of the School of Medicine and Surgery at Milano Bicocca University (protocol n. 11/17), derived from the approval of the Italian National Institute of Health (ISS, Rome, Italy) prot. 30/07/2007-0040488, and it was conducted in observance of the Declaration of Helsinki.

All patients met the inclusion criteria described below. The study population was randomly divided into 3 groups of 10 subjects each:

- GROUP A: patients treated following the LANAP protocol;
- GROUP B: patients treated following the OHLLT protocol;
- GROUP C: patients treated exclusively with scaling and root planing (control group).

From each patient with periodontal disease, a triple sampling of subgingival plaque was performed in 2 distinct sites of the oral cavity.

The first sampling was carried out before the application of the protocols, the second happened 1 week after the end of the treatments, and the third was 9 months after the end of the treatments.

The operative procedure for subgingival plaque removal was carried out through the following steps: removal of supragingival plaque, isolation of the operative field, sampling with a sterile paper cone, insertion in the appropriate tubes, fast and careful shipping by a courier and delivery of the sample to the laboratory in good time.

Each plaque sample arrived in the laboratory accompanied by a patient card containing information about the patient's age, sex, date of recruitment, tooth number and sampling site where the sample was taken, along with the depth of the periodontal pocket (PPD) and any presence of bleeding on probing or pus at the time of sampling. The primers and probe oligonucleotides were designed based on 16S rRNA gene sequences of the Human Oral Microbiome Database (HOMD 16S rRNA RefSeq Version 10.1), totaling 845 entries. All the sequences were aligned in order to find either a consensus sequence or less conserved spots. Three real-time polymerase chain reaction (RT-PCR) runs were performed for each sample. The first reaction quantified the total amount of bacteria using two degenerate primers and a single probe matching a highly conserved sequence of the 16S ribosomal RNA gene. The second reaction detected and quantified the three red complex bacteria (i.e., *P. gingivalis*, *T. forsythia* and *T. denticola*) in a multiplex RT-PCR. The third reaction detected and quantified *Aggregatibacter actinomycetemcomitans*, *Fusobacterium nucleatum*, *Echinococcus corrodens* and *Campylobacter rectus*. The oligonucleotide concentrations and RT-PCR conditions were optimized to ensure sensitivity, specificity and no inhibitions in case of unbalanced target amounts. Absolute quantification assays

were performed using the Applied Biosystems 7500 Sequence Detection System. The amplification profile was initiated by a 10 min incubation period at 95 °C to activate the polymerase, followed by a two-step amplification of 15 s at 95 °C and 60 s at 57 °C for 40 cycles. All these experiments were performed including nontemplate controls to exclude reagent contamination. Plasmids containing synthetic DNA target sequences (Eurofin MWG Operon, Ebersberg, Germany) were used as a standard for the quantitative analysis. Standard curves for each target were constructed in a triplex reaction by using a mix of the same number of plasmids in serial dilutions ranging from 10<sup>1</sup> to 10<sup>7</sup> copies. There was a linear relationship between the threshold cycle values plotted against the log of the copy number over the entire range of dilutions (data not shown). The copy numbers for individual plasmid preparations were estimated using the Thermo NanoDrop spectrophotometer. The absolute quantification of the total bacterial genome copies in the samples allowed for the calculation of a relative amount of red complex species. To prevent sample and polymerase chain reaction contamination, plasmid purification and handling were performed in a separate laboratory with dedicated pipettes [28].

## 2.2. Inclusion and Exclusion Criteria

The inclusion criteria were the following: signing informed consent, male and female sexes between the ages of 18 and 65, moderate or severe periodontitis (PPD > 4 mm in more than 3 sites), compliance of the patient to complete the study, presence of bone resorption greater than 4 mm in at least 30% of the sites examined and good general health.

The exclusion criteria were the following: pregnancy, liver disease, collagen disease, osteoporosis, recent surgery, chronic inflammatory disease, thyroid disease, diabetes, a medical condition or medical history that required antibiotic prophylaxis prior to treatment, use of anti-inflammatory drugs or antibiotics within the past 6 months, hard smokers (more than 10 cigarettes per day), and periodontal surgical treatment in the last 12 months.

## 2.3. Operational Protocol

The selected patients received information and instructions, both oral and written, on the modality and purpose of the study.

Patients were given the opportunity to ask questions for further clarification.

The duration of the study was a total of 9 months.

In the first session, all the patients recruited to the study had the following data collected: anamnestic data, radiographic and orthopantomography status, intra- and extraoral photographic collection, charting and intra- and extraoral examination.

The visit also included, through the use of periodontal probes (PC-PUNC and NABERS), the detection of the following information and the following periodontal biometric parameters: plaque index (IP), depth of the periodontal pocket on probing (PPD), clinical attack level (CAL), gingival recession (REC), bleeding on probing (BOP), mobility of dental elements (MOB), involvement of furcation and the presence of exudate. All patients agreed with the following maintenance home hygiene protocol: sonic brush with vertical movement (Broxo), interdental brushes and oral irrigators (Broxo) at least two times every day.

The second session was the beginning of the nonsurgical periodontal therapy.

In group A, treated with the LANAP protocol, a microbiological sampling of the periodontal pockets was made first. Then, a supragingival scaling using high-frequency ultrasound (Satelec) was made. Then, an Nd: YAG laser was used in the periodontal pockets with the following settings: 4.0 W, 200 mj, a duty cycle of 100 µs and a frequency of 20 Hz. Scaling and subgingival root planning with Gracey's curettes were made before the use of the laser to remove granulation tissue, with settings of 4.0 W, 200 mj, a duty cycle of 650 µs and a frequency of 20 Hz.

In group B, treated with the OHLLET protocol, a microbiological sampling of the periodontal pockets was made first. Then a supra and subgingival scaling with high-frequency ultrasound was made with a solution of Betadine diluted at 10% (Figure 1). An

airflow session with erythritol was performed (Figure 2), and then diode laser treatment with a Sioxyl<sup>®</sup> solution was performed, irrigating the periodontal pockets with the Sioxyl<sup>®</sup> solution (Figure 3), leaving the solution for at least 2 min and irradiating with a Wiser diode laser with a peak power of 2.5 W, T-On of 20 microns, T-Off of 80 microns, average power of 0.5 W, duty cycle of 100 microns and frequency of 10 KHz (Figure 4).



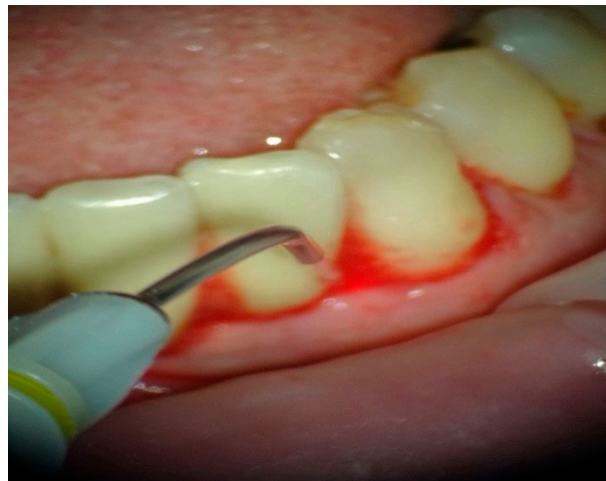
**Figure 1.** High-frequency ultrasound with betadine diluted to 1/10.



**Figure 2.** Airflow with erythritol, a low-abrasive powder.



**Figure 3.** Irrigation with a Sioxyl solution.



**Figure 4.** Treatment with a Wisier diode laser with a peak power of 2.5 W, T-On of 20 microns, T-Off of 80 microns, average power of 0.5 W, duty cycle of 100 microns and frequency of 10 KHz.

In the control group C, first, microbiological sampling (T1) of the periodontal pockets was conducted. Then, a supragingival scaling with high-frequency ultrasounds and a subgingival scaling and root planning in all periodontal pockets with high-frequency ultrasounds and Gracey's curettes were performed.

The third session (T2) was conducted 7 days after the second session.

In group A, which was treated with the LANAP protocol, the second microbiological sampling (T2) of the periodontal pockets was performed upon rinsing with gauze soaked in chlorhexidine.

In group B, which was treated with the OHLLET protocol, the second microbiological sampling (T2) of the periodontal pockets was performed. After that, a new diode laser treatment + Sioxil<sup>®</sup>, as in the second session, was conducted upon irrigating the periodontal pockets with the Sioxil<sup>®</sup> solution (Figure 3), leaving the solution for at least 2 min and irradiating with a Wisier diode laser with a peak power of 2.5 W, T-On of 20 microns, T-Off of 80 microns, average power of 0.5 W, duty cycle of 100 microns and frequency of 10 KHz. In the control group C, the second microbiological sampling of the periodontal pockets was performed. The fourth session (T3) was conducted 9 months after the second session.

#### 2.4. Statistical Analysis

The data were collected and entered on the Statistical Package for Social Sciences (SPSS) version 11.5. The results are thus expressed as proportions using appropriate Tables 1 and 2 and Figures 5–8.

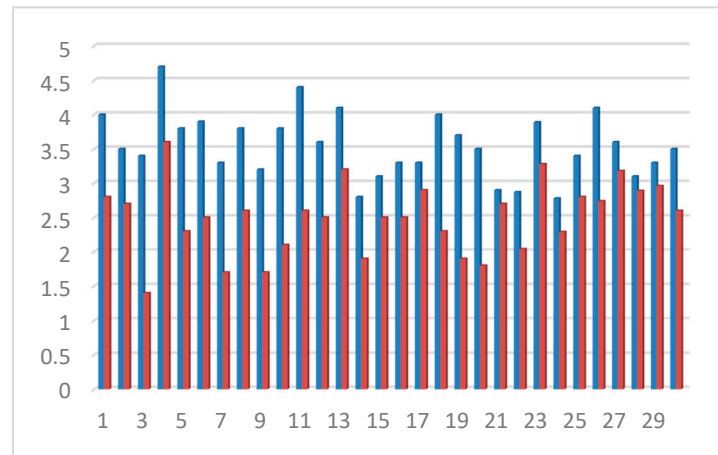
### 3. Results

Patients from 1 to 10 were included in group A, from 11 to 20 were in group B and from 21 to 30 were in group C. All clinical parameters showed improvement, with a decrease of the plaque index (total average decrease of 70.43%; group A average decrease of 70.7%, group B average decrease of 79.8% and group C average decrease of 60.8%) (Figure 5), bleeding on probing (total average decrease of 80.27%; group A average decrease of 97.0%, group B average decrease of 95.0% and group C average decrease of 48.8%) (Figure 6) and probing depth (total average decrease of 1.83 mm; group A average decrease of 2.14 mm, group B average decrease of 2.36 mm and group C average decrease of 0.99 mm) (Figure 7).

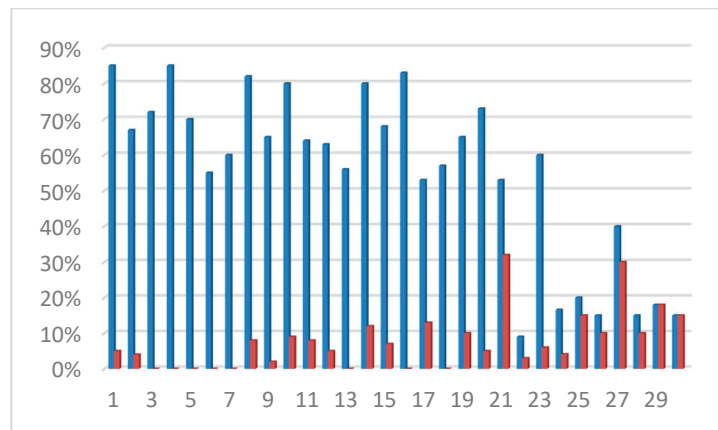
The results obtained from the study that evaluated the microbiological profile of 30 patients aged between 18 and 65 recruited and treated with three different types of protocols are summarized in the following paragraphs with the aid of Figures 5–8 and Tables 1 and 2.

In particular, an average with the respective microbiological analysis was reported for each group examined (group A: patients treated following the LANAP protocol; group B:

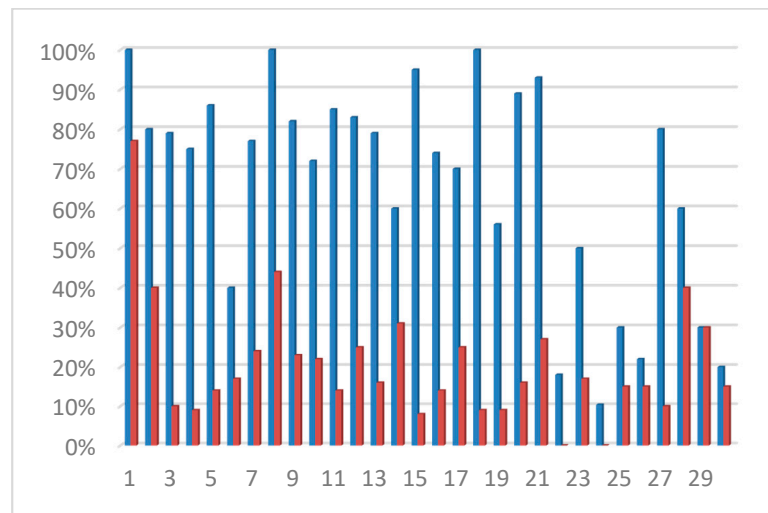
patients treated following the OHLLT protocol; and group C: patients treated exclusively with scaling and root planing) (Figure 5). The first sampling was carried out before the application of the protocols (T1), the second was 1 week after the end of the treatments (T2), and the third was 9 months after the end of the treatments (T3). At T2, 1 week after the beginning of protocols, the variation of the total amount of bacteria was calculated. Group A decreased by 80.3%, group B decreased by 81.4%, and group C decreased by 46.4%. At T3, 9 months after the end of the treatments, the variation of the total amount of bacteria was calculated. Group A decreased by 59.1%, group B decreased by 70.9%, and group C decreased by 8.9%. The statistical analyses are represented in Tables 1 and 2.



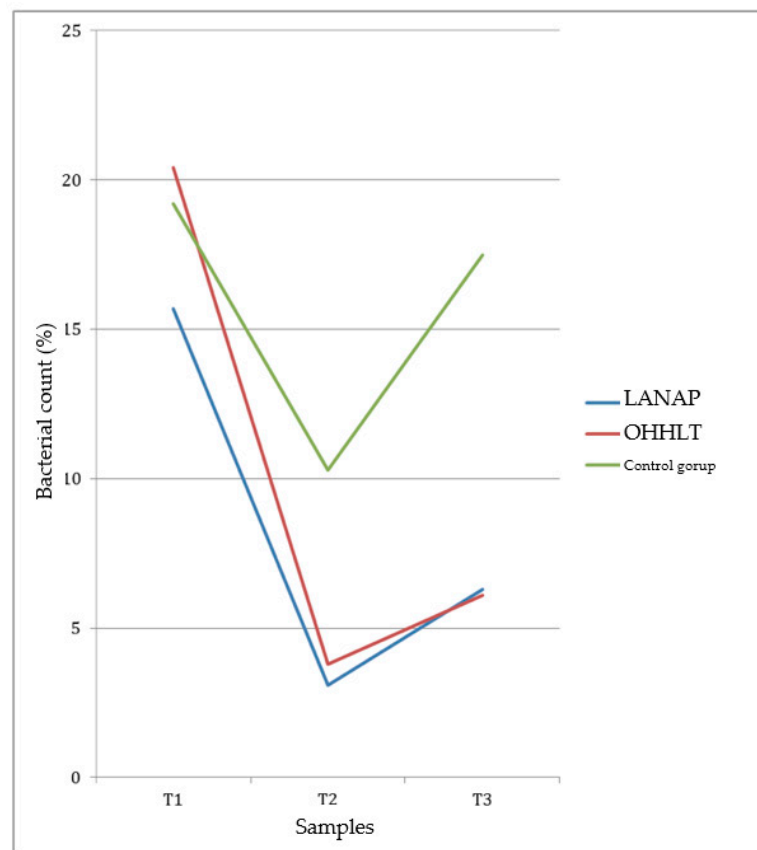
**Figure 5.** Average of the plaque index values before (T1) and after treatment (T3). Patients from 1 to 10 were included in group A, from 11 to 20 were in group B and from 21 to 30 were in group C. The total average decrease was 70.43%, with group A’s average decrease being 70.7%, group B’s average decrease being 79.8% and group C’s average decrease being 60.8%.



**Figure 6.** Average of the bleeding on probing index values before (T1) and after treatment (T3). Patients from 1 to 10 were included in group A, from 11 to 20 were in group B and from 21 to 30 were in group C. The total average decrease was 80.27%, with group A’s average decrease being 97.0%, group B’s average decrease being 95.0% and group C’s average decrease being 48.8%.



**Figure 7.** Average probing depth in millimeters before (T1) and after treatment (T3). Patients from 1 to 10 were included in group A, from 11 to 20 were in group B and from 21 to 30 were in group C. The total average was 1.83 mm. Group A's average decrease was 2.14 mm, group B's average decrease was 2.36 mm, and group C's average decrease was 0.99 mm.



**Figure 8.** Microbiological results of the three groups at the three collection moments.



**Table 1.** Periodontal and microbial parameters among the A + B groups and C group before treatment.

	Group A + B (n = 20)	Group C (n = 10)	p Value
	Mean ± SD	Mean ± SD	
<b>BoP</b>	1.92 ± 0.58	1.86 ± 0.66	<0.0018 *
<b>P.I.</b>	1 ± 0.23	1.84 ± 0.43	0.006
<b>P.D.</b>	4.28 ± 0.25	4.25 ± 0.21	0.281

BoP: bleeding on probing; P.I.: plaque index; and P.D.: periodontal depth. \* p value < 0.001 is statistically significant.

**Table 2.** Periodontal and microbial parameters among the A + B groups and C group after treatment.

	Group A + B (n = 20)	Group C (n = 10)	p Value
	Mean ± SD	Mean ± SD	
<b>BoP</b>	0.12 ± 0.05	1.35 ± 0.25	<0.0015 *
<b>P.I.</b>	0.12 ± 0.15	1.43 ± 0.52	0.004
<b>P.D.</b>	2.15 ± 0.21	3.21 ± 0.34	0.112

BoP: bleeding on probing; P.I.: plaque index; and P.D.: periodontal depth. \* p value < 0.001 is statistically significant.

#### 4. Discussion

The goal of our research was to evaluate the effectiveness of two different protocols (LANAP and OHLLT) that rely on the use of lasers to treat patients suffering from periodontitis through the microbiological analysis of real-time PCR.

Studies conducted in the literature showed the effectiveness of this highly specific and sensitive method, which allows the quantitative and specific detection of significant markers of periodontitis and peri-implantitis and the determination of the total bacterial load [32].

This appears to be extremely interesting considering the relevant literature data, in particular in relation to the efficacy of periodontal surgical and nonsurgical protocols, when periodontal pathogenic bacteria were present in the periodontal pockets or in the maxillary and mandibular alveoli.

The persistence of specific subgingival species, including *A. actinomycetemcomitans*, *P. gingivalis* and *T. denticola* (their ability to invade the subjacent periodontal tissues), has been associated with poor response to treatment by scaling and root planing. In effect, if *Actinobacillus actinomycetemcomitans*, *Porphyromonas gingivalis*, *Prevotella intermedia* and *Campylobacter rectus* are inside the pockets, the nonsurgical periodontal approach is not effective [1]. The same results with a regenerative surgical approach, as indicated by Heitz Mayfield and the Ergoperio Group [2], where the presence of a high bacterial load and specific pathogen complexes in deep periodontal and peri-implant pockets had a significant negative impact on the 1-year outcome of surgical or regenerative treatment.

If the presence of a red and orange Sokrasky's complex has a negative impact on the nonsurgical and surgical periodontal approaches, we suggest a real-time PCR for every time it is needed to treat an affected tooth.

In a recent meta-analysis, Swider et al. [27] evaluated the in vivo articles published in accordance with PRISMA guidelines [33] and the *Cochrane handbook for Systematic reviews of Intervention* [34] concerning the in vivo treatment of peri-implantitis with lasers protocols. The authors analyzed 49 articles, but just 7 were included due to the strict eligibility criteria of the quality assessment. The seven studies included were the following: Birang et al. [35], Caccianiga et al. [26], Persson et al. [36], Arisan et al. [37], Yoshino et al. [38], Bassetti et al. [39] and Dörtbudak et al. [40]. They concluded that "a high-power diode laser may have some effect on peri-implant pathogens causing peri-implantitis, whereas Er:YAG laser application shows no significant effect on oral bacteria in the long term. aPDT has the ability to reduce the total count of the different bacterial strains associated with peri-implantitis, e.g., *A. actinomycetemcomitans*, *P. gingivalis*, *P. intermedia*,

*T. denticola*, *T. forsythia*, *F. nucleatum*, and *C. rectus*". In effect, the conclusions of the authors were exactly the same as those published in 2016 from Caccianiga et al. [27].

The technologies based on real-time PCR allowed us to evaluate and compare the microbiological analysis before and after the treatment with the two protocols.

The results of our research take on greater significance if we divide the population under study into two groups:

- Test group (group A and group B);
- Control group (group C).

Revealed in line with the efforts of the authors who, over the years, have investigated the qualitative and quantitative presence of bacteria involved in the onset and progression of periodontal and peri-implant disease, the data thus analyzed show the following:

- There is a correlation between the severity of periodontitis and the total quantity of the bacterial load inside the periodontal pockets [1,2,32];
- Actinomycetemcomitans, being a pathogen responsible for the aggressive forms in adolescent patients, was found in minimal concentrations or even observed to be absent, which was in line with the chosen sample (2% of AA in adult periodontitis) [32];
- Real-time PCR proved to be a decisive technique for the objective of our study, allowing us to qualitatively and quantitatively detect the main anaerobes involved in periodontal disease [31,32].

Subsequently, with the data obtained, by repeating the real-time PCR test, it was possible to determine the presence of any active sites after treatment and assist the patient in determining future recall intervals (follow-up).

In conclusion, it can be stated that in the sample groups (groups A, B and C) at times T1, T2 and T3, respectively, a higher relative proportion (%) of *P. gingivalis*, *T. forsythia* and *T. denticola* was found, as well as a minor amount of *F. nucleatum* ssp. and *P. intermedia* and an almost zero relative proportion of *A. actinomycetemcomitans*.

In particular, we found the following:

- At T2 (microbiological sampling performed one week after the end of treatment with the respective protocols), there was a reduction of the average bacterial load of 80.3% in the subjects belonging to group A, a reduction in the average bacterial load of 81.4% in the subjects belonging to group B and a reduction in the bacterial load of 46.4% in the subjects belonging to group C;
- At T3 (microbiological sampling 9 months after the end of treatment with the respective protocols), there was a reduction in the average bacterial load of 59.9% (compared with T1) in the subjects belonging to group A, a reduction in the average bacterial load of 70.1% (compared with T1) in the subjects belonging to group B and a reduction in the average bacterial load of 8.9% (compared with T1) in the subjects belonging to group C.

Therefore, it was shown that the use of the OHLLT protocol (group B) led to a greater reduction in the total bacterial count than the use of the LANAP protocol in the long term (group A).

In the research, in the nonsurgical periodontal treatment, the use of LANAP and OHLLT protocols confirmed, in a microbiological evaluation with real-time PCR, an effective ability to reduce the bacterial load present inside the periodontal pockets [8–11,21–28,30,31], compared with conventional debridement, made by conventional instruments [1,2].

In particular, for the clinical measurements, we considered and measured the plaque index, bleeding on probing and probing depth at the beginning of treatment (T1) and after 9 months (T3).

The plaque indexes, influenced by the same oral hygiene procedures in all groups, were equivalent in all groups. The bleeding on probing results at 9 months were equivalent for group A (97%) and group B (95%) and showed poor results for group C (48.8%). The probing depth measurements showed that, once again, the LANAP (−2.14 mm) and OHLLT

(−2.36 mm) protocols had the best results compared with the conventional nonsurgical periodontal approach (−0.99 mm).

On analyzing the BOP and PPD in the three groups, we could consider that the OHLLT and LANAP protocols had quite the same results at 9 months, with an important decrease in the BOP and an interesting reduction of the PPD. On the other hand, group C showed the worst results, and according to literature [1,2] and the red and orange complex is involved in the periodontal pockets.

## 5. Conclusions

At the conclusion of this study, after clinical and microbiological evaluations, it appears evident that both the OHLLT and LANAP protocols are efficient in the nonsurgical treatment of periodontal disease. It is necessary to consider, however, that the OHLLT protocol does not expose the patient to any thermal risk, which is present instead with the LANAP method. New studies should be made in the future to confirm these preliminary results about the efficacy of the LANAP and OHLLT protocols in nonsurgical or surgical treatments of periodontitis and peri-implantitis, even if our study confirmed the resistance of periodontal bacteria to the nonsurgical, conventional periodontal approach and their sensitivity to the OHLLT and LANAP protocols.

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**Informed Consent Statement:** Informed consent was obtained from all subjects involved in the study.

**Data Availability Statement:** PubMed, DOI, PMC.

**Conflicts of Interest:** The authors declare no conflict of interest.

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