OVIDIUS UNIVERSITY OF CONSTANȚA

DOCTORAL SCHOOL OF MEDICINE

RESEARCH DOMAIN DENTAL MEDICINE

Summary of the doctoral Thesis

Study regarding the optimization of the periodontal treatments using dental lasers

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CONTENT

Note: In the summary of the PhD thesis are presented the results of the personal experimental research, the general conclusions and a selective bibliography. The same numbering was maintained for chapters, tables and figures as in the PhD thesis.

The present PhD thesis contains 89 figures, 30 tables, 11 graphs and 288 bibliographic references, maintaining the numbering from the thesis.

PERSONAL CONTRIBUTIONS

Chapter 5. GENERAL OBJECTIVES OF RESEARCH. GENERAL METHODOLOGY

5.1 General objectives

The main objective of this research was to evaluate the bactericidal effect of hydroxyl radicals generated by photolysis of H2O2 made by diode laser 940 nm on 9

periodontal pathogenic bacterial species and combining two laser wavelengths in periodontal treatments in patients with chronic periodontitis.

The second objective was the optimization of periodontal treatments using two laser wavelengths: Er,Cr: YSGG (2780 nm) and diode (940 nm) and the development of a treatment protocol for chronic periodontitis.

The effects were examined in three research segments, based on the benefits of minimal invasive laser therapy, in order and / or to cure periodontal diseases and to improve the quality of patients' life.

Although, there are insufficient published studies in this field, this research aims to provide a better understanding of non-surgical laser assisted periodontal treatments.

5.2 Material and method

Patients were selected based on the following inclusion criteria:

- Patients with good overall health and no comorbid conditions;
- Age between 32-70 years;
- Sound or with superficial to moderate carious lesions;
- Patients who have demonstrated the ability and willingness to perform the indicated periodontal therapy;
- At least 16 teeth present, distributed in the four quadrant;
- With a periodontal pocket depth (PPD) of at least 4 mm per quadrant;
- With clinically and radiologically evidenced bone resorption;
- With bleeding on probing in all four quadrants;

The exclusion criteria were represented by:

- Patients who are in active periodontal treatment;
- Patients who have undergone periodontal treatment during the last 12 months;
- Patients who have had antibiotic therapy (systemic or local) over the last 6 months;
- Smokers
- Systemic conditions that may alter the therapeutic outcome (type I and type II diabetes mellitus, immune deficiency, HBV, HCV, cancer, haematological disorders, epilepsy, etc.);
- Pregnancy, lactation;
- Inability or refusal to follow the study protocol.

5.2.1. Clinical and paraclinical assessment of patients:

Initial examination was performed one week prior to periodontal treatment and included clinical and radiological examination, respectively microbiological sampling.

Clinical data and measurements determined in this study were as follows: Patient's gender and age, Medical History, Intraoral Photographs, Digital Intraoral Dental Radiographs. Periodontal clinical indexes were: the degree of furcation involvement and tooth mobility, periodontal probing depth (PPD), the clinical attachment level (CAL), bleeding on probing (BOP). They were performed with the CP15 periodontal probe (Medesy Ltd., Maniago, Italy) in six sites per tooth, performed by the same examiner.

All clinical measurements were evaluated at baseline and at follow-up, which were performed at 3 and 6 months post operatory.

According to the initial findings, teeth were selected, one in each quadrant, teeth showing PPD ≥ 5 mm depth and BOP (+). The teeth with the largest periodontal probing depth in each quadrant were selected as testing sites.

Teeth with fixed prosthetic restorations (crowns or bridges), teeth with furcation involvement and secondary molars were excluded.

5.2.2. Microbiological assessment

The microbiological assay was performed by means of real-time PCR technique using the PET Deluxe Diagnostic Set (MIP Pharma GmbH, Blieskastel-Niederwürzbach, Germany), which allows to take an individually or pool sampling.

For pre- and postoperative assessment of subgingival biofilm, it was decided to perform an individual sampling from each periodontal pockets, in order to dissect parodontopathogenic agents.

Using the real time PCR technique, the laboratory was able to identify the total number of germs, and the qualitative and quantitative of the following parodontopathogenic bacterial species: *A. actinomycetemcomitans (Aa.), P. gingivalis (Pg.), T. forsythia (Tf.), T. denticola (Td.), F. nucleatum (Fn.), P. intermedia (Pi.), P. micros (Pm.), E. nodatum (En.), C. gingivalis (Cg.).*

In this research, Er,Cr: YSGG 2780 nm laser (WaterLase Iplus, Biolase, Irvine, USA) with the Gold handpiece (in contact mode) and the 940 nm diode laser (Epic 10, Biolase, Irvine, USA) were used.

CHAPTER 6. EVALUATION OF THE ANTIMICROBIAL EFFECT OF 940 nm DIODE LASER BASED ON PHOTOLYSIS OF H_2O_2 IN THE TREATMENT OF PERIODONTAL DISEASE

6.1. Hypothesis / Objectives

The working hypothesis of this research segment started from the premise of the benefits of minimally invasive laser therapy, applied according to current medical possibilities, as well as the bactericidal and regenerative possibilities of the 940 nm laser wavelength in periodontal treatment.

The aim of the study was to identify and to present an alternative procedure based on laser assisted disinfection in periodontal treatments.

The objective of this study was to evaluate the bactericidal effect of hydroxyl radicals generated by photolysis of H_2O_2 with diode laser 940 nm, on 9 periodontal pathogens and also to evaluate the clinical outcomes in patients with periodontal disease.

6.3. Material and method

For this study, 30 patients aged between 34 to 66 years (48.23 ± 8.63, median ± SD) were selected, diagnosed with moderate to severe periodontal disease. Five patients were excluded. A total of 25 patients, aged 35-62 years (47.8 ± 8, median ± SD), participated until the end of the study.

Based on the randomization method, each patient quadrant was assigned to one of the four treatment groups as follows: Group 1: SRP as monotherapy; Group 2: 3% H_2O_2 as monotherapy; Group 3: diode laser (λ = 940 nm) as monotherapy; Group 4: Photolysis of 3% H_2O_2 with diode laser (λ = 940 nm).

The test sites of each patient were treated by the four treatment procedures previously mentioned in order to compare the effects of therapies applied within the same patient.

6.4. Results

Table 6.V. PD and CAL results

Variables		PD (mm)		CAL (mm)	
		Initial	*3 months postop*	*Initial*	*3 months postop*
Group 1 SRP	Median	6	4	8	6
	25 - 75%	5.5 – 7	3 – 5	7 – 9	5.5 - 7
	p Value	0.000		0.000	
Group 2 H_2O_2	Median	6	4	8	8
	25 - 75%	5 – 7	4 - 6	7 – 9.5	7 - 9
	p Value	0.001		0.384	
Group 3 diode 940 nm	Median	6	4	7	6
	25 - 75%	5 – 7	4 – 7	7 – 9.5	5.5 – 8.5
	p Value	0.002		0.006	
Group 4 H_2O_2 + Diode 940 nm	Median	6	3	8	5
	25 - 75%	5 – 7	3 – 4	6.5 – 9	4 – 7
	p Value	0.000		0.000	

Table 6.IV. Qualitative analysis of microbiological variables

Variables	Group 1 SRP		Group 2 H_2O_2		Group 3 Diode 940 nm		Group 4: H_2O_2 + Diode 940 nm	
	No. Bacteria %	*p*	*No. Bacteria %*	*p*	*No. Bacteria %*	*p*	*No. Bacteria %*	*p*
Total Number of Germs (NTG)								
Preop	25 (100%)	-	25 (100%)	-	25 (100%)	-	25 (100%)	-
1 month postop	25 (100%)		25 (100%)		25 (100%)		25 (100%)	
Aggregatibacter actinomycetemcomitans (A.a.)								
Preop	0 (0%)	<0.05	0 (0%)	<0.05	0 (0%)	-	2 (8%)	<0.05
1 month postop	3 (12%)		3 (12%)		0 (0%)		0 (0%)	
Porphyromonas gingivalis (P.g.)								
Preop	22 (88%)	0.10	23 (92%)	0.063	22 (88%)	0.061	20 (80%)	0.001
1 month postop	21 (84%)		18 (72%)		17 (68%)		3 (12%)	
Treponema denticola (T.d.)								
Preop	25 (100%)	>0.05	25 (100%)	>0.05	25 (100%)	>0.05	25 (100%)	<0.05
1 month postop	19 (76%)		23 (92%)		24 (96%)		2 (8%)	
Tannerella forsythia (T.f.)								
Preop	25 (100%)	<0.05	24 (96%)	0.021	24 (96%)	0.031	24 (96%)	<0.05
1 month postop	3 (12%)		16 (64%)		18 (72%)		0 (0%)	
Prevotella intermedia (P.i)								
Preop	18 (72%)	0.754	20 (80%)	0.008	21 (84%)	0.375	20 (80%)	<0.001
1 month postop	16 (64%)		12 (48%)		18 (72%)		6 (24%)	
Peptostreptococcus micros (P.m.)								
Preop	22 (88%)	0.687	24 (96%)	0.063	25 (100%)	>0.05	24 (96%)	<0.001
1 month postop	20 (80%)		19 (76%)		23 (92%)		4 (16%)	
Fusobacterium nucleatum (F.n.)								
Preop	10 (40%)	0.581	8 (32%)	0.791	6 (24%)	0.039	13 (52%)	0.001
1 month postop	13 (52%)		10 (40%)		13 (52%)		2 (8%)	
Eubacterium nodatum (E.n.)								
Preop	13 (52%)	<0.001	12 (48%)	0.003	13 (52%)	0.002	11 (44%)	<0.05
1 month postop	1 (4%)		1 (4%)		3 (12%)		0 (0%)	
Capnocytophaga gingivalis (C.g.)								
Preop	14 (56%)	0.012	18 (72%)	0.219	22 (88%)	0.996	17 (68%)	1
1 month postop	23 (92%)		22 (88%)		21 (84%)		17 (68%)	

Table 6.VI. BoP results

Variables		BoP (%)	
		Initial	***3 months postop***
Group 1 (SRP)	+ Values (%)	24 (96%)	18 (52%)
	p Values	0.003	
Group 2 (H_2O_2)	+ Values (%)	23 (92%)	18 (72%)
	p Values	0.063	
Group 3 (diode 940 nm)	+ Values (%)	24 (96%)	15 (60%)
	p Values	0.012	
Group 4 (H_2O_2 + Diode 940 nm)	+ Values (%)	24 (96%)	2 (8%)
	p Values	< 0.001	

Table 6.VII. Comparison between the investigated groups on the identified bacterial species and the periodontal clinical parameters, pre - and postoperative.

	p Values											
	Group 1 vs. Group 2		**Group 1 vs. Group 3**		**Group 1 vs. Group 4**		**Group 2 vs. Group 3**		**Group 2 vs. Group 4**		**Group 3 vs. Group 4**	
	Initial	*Postop*	*Initial*	*Postop*	*Initial*	*Postop*	*Initial*	*Postop*	*Initial*	*Postop*	*Initial*	*Postop*
NTG	0.485	0.554	0.691	0.771	0.372	0.002	0.221	0.229	0.168	0.000	0.854	0.005
A.a.	1.000	0.932	1.000	0.077	0.153	0.077	1.000	0.077	0.153	0.077	0.153	1.000
P.g.	0.861	0.992	0.749	0.992	0.923	0.000	0.985	0.658	0.478	0.000	0.382	0.000
T.d.	0.248	0.472	0.684	0.035	0.362	0.000	0.461	0.123	0.048	0.000	0.218	0.000
T.f.	0.388	0.000	0.923	0.000	0.641	0.077	0.221	0.484	0.168	0.000	0.684	0.000
P.i.	0.300	0.252	0.915	0.906	0.674	0.001	0.350	0.192	0.280	0.034	0.733	0.000
P.m.	0.541	0.453	0.461	0.177	0.607	0.000	0.177	0.992	0.295	0.000	0.907	0.000
F.n.	0.668	0.245	0.337	0.673	0.258	0.000	0.524	0.428	0.150	0.005	0.047	0.000
E.n.	0.597	0.977	0.943	0.312	0.859	0.317	0.568	0.274	0.816	0.317	0.794	0.077
C.g.	0.389	0.992	0.024	0.683	0.209	0.006	0.152	0.734	0.595	0.031	0.696	0.084
PPD	0.731	0.000	0.606	0.040	0.686	0.328	0.903	0.372	0.943	0.000	0.976	0.005
CAL	0.641	0.000	0.350	0.478	0.401	0.700	0.594	0.026	0.607	0.000	0.889	0.008

6.6. Conclusions

Within the limits of this study, the following conclusions can be drawn:

1. Photolysis induced by hydrogen peroxide using diode laser (λ = 940nm) provided an effective antimicrobial effect against major parodontopathogenic species due to hydroxyl radical formation, which could represent an additional procedure to conventional periodontal treatment.
2. The proposed non-surgical periodontal therapy by means of photolysis H_2O_2 with diode laser (λ = 940nm) showed clinical efficacy on all investigated periodontal parameters, results that were maintained up to three months postoperatively, even without the mandatory conventional periodontal therapy (SRP).
3. Laser-assisted antimicrobial therapy based on photolysis H_2O_2 offers a beneficial alternative to the use of antibiotics in periodontitis.
4. Clinical and microbiological changes due to sub gingival bacterial biofilm were significantly reduced in the groups where H_2O_2 photolysis with diode laser (λ = 940nm) was applied compared to the quadrants treated with SRP, H_2O_2 and the diode laser as monotherapy.

Chapter 7
ANTIMICROBIAL EFFECT OF HYDROGEN PEROXIDE PHOTOLYSIS USING 940 nm DIODE LASER AS AN ADJUNCT TO CONVENTIONAL PARODONTAL TREATMENT

7.1. Hypothesis / Objectives

The working hypothesis of this researched segment is the antimicrobial benefit of 3% hydrogen peroxide photolysis by diode laser (λ = 940 nm) as an adjunctive method to the classical non-surgical periodontal therapy.

The purpose of this research report is to compare the antimicrobial effects produced by the hydroxyl radical generation by photolysis H2O2 made with diode laser (λ = 940 nm) in combination with non-surgical periodontal classic therapy.

The objective/aim of this study was to quantify the bactericidal effect of hydroxyl radicals generated by photolysis of H2O2 with the 940 nm diode laser in combination with non-surgical periodontal conventional therapy on 9 periodontal pathogens and also to evaluate the clinical response in patients with periodontal diseases.

7.3. Material and method

In this study, there were selected 40 patients, aged 35 to 62 years, (47.60 ± 7.82, mean ± SD) diagnosed with moderate to severe periodontal disease. Based on the exclusion criteria, 2 were excluded. A total of 38 patients, aged 35-62 years (47.45 ± 7.82, median ± SD), participated until the end of the study.

Based on the randomization method, three quadrants were selected, and each quadrant was assigned to one of three treatment methods as follows: Group 1: SRP as monotherapy; Group 2: SRP associated with diode laser (λ = 940 nm); Group 3: SRP associated with H_2O_2 photolysis by diode laser (λ = 940 nm).

7.4. Results

Table 7.V. PD and CAL results

Variable		PD (mm)		CAL (mm)	
		Preop	**3 months postop**	**Preop**	**3 months postop**
Group 1 (SRP)	Median	6.5	4	8	6
	25 - 75%	6 – 7	3 – 5	7 – 9	5.75 - 7
	p Value	0.000		0.000	
Group 2 (SRP + diode 940 nm)	Median	6	4	8	6.5
	25 - 75%	5 – 7	3 – 5.25	7 – 10	5 – 8.25
	p Value	0.000		0.000	
Group 3 (SRP + H_2O_2 + diode 940 nm)	Median	6	3	8	5
	25 - 75%	5 – 8	2 – 4	7 – 10	4 – 6.25
	p Value	0.000		0.000	

Table 7.IV. Qualitative analysis of microbiological variables

Variable	Group 1 (SRP)		Group 2 (SRP + Diode)		Group 3 (SRP + H_2O_2 +Diode)	
	No. Bacteria (%)	*p*	No. Bacteria (%)	*p*	No. Bacteria (%)	*p*
Total Number of Germs (NTG)						
Preop	38 (100%)	-	38 (100%)	-	38 (100%)	-
1 month postop	38 (100%)		38 (100%)		38 (100%)	
A. actinomycetemcomitans (A.a.)						
Preop	0 (0%)	>0.05	0 (0%)	-	1 (2.7%)	>0.05
1 month postop	1 (2.7%)		0 (0%)		2 (5.3%)	
Porphyromonas gingivalis (P.g.)						
Preop	35 (92.1%)	<0.001	35 (92.1%)	<0.001	33 (86.8%)	<0.001
1 month postop	17 (44.7%)		20 (52.6%)		5 (13.2%)	
Treponema denticola (T.d.)						
Preop	35 (92.1%)	<0.001	36 (94.7%)	<0.001	36 (94.7%)	<0.001
1 month postop	18 (47.4%)		20 (52.6%)		3 (7.9%)	
Tannerella forsythia (T.f.)						
Preop	37 (97.4%)	<0.001	38 (100%)	<0.001	37 (97.4%)	<0.001
1 month postop	8 (21.1%)		15 (39.5%)		0 (0%)	
Prevotella intermedia (P.i)						
Preop	27 (71.1%)	0.118	30 (78.9%)	0.727	28 (73.7%)	<0.001
1 month postop	20 (52.6%)		28 (73.7%)		6 (15.8%)	
Peptostreptococcus micros (P.m.)						
Preop	36 (94.7%)	0.125	36 (94.7%)	0.065	38 (100%)	<0.001
1 month postop	31 (81.6%)		29 (76.3%)		7 (18.4%)	
Fusobacterium nucleatum (F.n.)						
Preop	17 (44.7%)	0.815	12 (31.6%)	0.549	17 (44.7%)	<0.001
1 month postop	19 (50%)		15 (39.5%)		5 (13.2%)	
Eubacterium nodatum (E.n.)						
Preop	22 (57.9%)	<0.001	18 (47.4%)	<0.001	16 (42.1%)	<0.001
1 month postop	1 (2.6%)		3 (7.9%)		38 (100%)	
Capnocytophaga gingivalis (C.g.)						
Preop	25 (65.8%)	0.057	28 (73.7%)	>0.1	24 (63.1%)	>0.1
1 month postop.	33 (86.8%)		29 (76.3%)		25 (65.7%)	

Table 7.VI. BoP (bleeding on probing) 3 months postoperatively

Groups		BoP (%)	
		Baseline	**3 months postop**
Group 1 (SRP)	+ Values (%)	37 (97.4%)	21 (55.3%)
	p Values	< 0.001	
Group 2 (SRP + Laser)	+ Values (%)	36 (94.7%)	19 (50%)
	p Values	< 0.001	
Group 3 (SRP + H_2O_2 + Laser)	+ Values (%)	37 (97.4%)	3 (7.9%)
	p Values	< 0.001	

Table 7.VII. Comparison between the investigated groups on the identified bacterial species and the periodontal clinical parameters, pre - and postoperative.

Bacteria	*p* Value					
	Group 1 vs. Group 2		Group 1 vs. Group 3		Group 2 vs. Group 3	
	Preop	Postop	Preop	Postop	Preop	Postop
NTG	0.226	0.540	0.880	0.000	0.448	0.000
A.a.	-	-	-	-	-	-
P.g.	0.823	0.415	0.526	0.001	0.282	0.000
T.d.	0.827	0.743	0.352	0.000	0.451	0.000
T.f.	0.629	0.095	0.747	0.003	0.336	0.000
P.i.	0.679	0.034	0.282	0.001	0.352	0.000
P.m.	0.294	0.437	0.771	0.000	0.483	0.000
F.n.	0.164	0.432	0.918	0.000	0.237	0.003
E.n.	0.333	0.314	0.477	0.317	0.964	0.079
C.g.	0.346	0.595	0.750	0.015	0.544	0.086
PPD	0.262	0.201	0.915	0.023	0.462	0.001
CAL	0.800	0.257	0.799	0.002	0.577	0.000

7.6. Conclusions

Based on the results from this study, we can conclude that:

1. Conventional periodontal therapy used as monotherapy fails to provide a stable and effective antimicrobial effect without the addition of a complementary procedure in the treatment of periodontal disease;
2. Combining mechanical therapy and photoactivation of hydrogen peroxide with 940 nm diode laser provides a safe and effective antimicrobial effect against the most important periodontal pathogenic agents;
3. The photolysis procedure of H_2O_2 with the 940 nm diode laser represents an adjunctive procedure to conventional periodontal treatment, without any absolute contraindications;
4. The proposed protocol may represent a new alternative in the treatment of periodontal disease, eliminating the need for local or general antibiotic administration;
5. Additional research is needed to explore the biochemical mechanisms of photoactivation of hydrogen peroxide with the 940nm wavelength and to perform further studies that investigate a larger number of patients on a longer period of time (> 1 year).

CHAPTER 8. BACTERICIDAL EFFECT OF LASER ASSISTED PERIODONTAL THERAPY BY COMBINING ER,CR:YSGG 2780 NM AND H_2O_2 PHOTOLYSIS WITH 940 NM LASERS

8.1. Hypothesis / Objectives

The working hypothesis of this study is represented by the antimicrobial and regeneration capacity by combining two complementary laser wavelengths in moderate to severe periodontal disease.

The aim of this study is to develop a minimally-invasive, efficient and safe laser-assisted periodontal protocol by means of two laser wavelengths, like 2780 nm and 940 nm.

The objective of the study is to compare the clinical and microbiological results of conventional periodontal treatment (SRP) with and without adjunctive therapy represented by 940 nm and 2780 nm laser wavelengths in the management of periodontal disease.

8.3. Material and method

For this study, 53 patients aged 32 to 70 years (46.98 ± 8.4, mean ± SD) were selected, diagnosed with moderate to severe periodontal disease. A total of 50 patients, aged 32-70 years, participated until the end of the study (47.04 ± 8.64, median ± SD). Based on the randomization method, each patient's quadrant was assigned to one of the two treatment groups as follows: Group 1: SRP as monotherapy; Group 2: SRP + Er,Cr:YSGG laser + H_2O_2 photolysis with 940 nm diode laser, as combination therapy;

8.4. Results

Table 8.VI. Clinical evaluation of PD and CAL baseline and 3 months postop

Variable		Group 1 (SRP)			Group 2 (SRP + Er,Cr:YSGG + H_2O_2 + diode 940 nm)		
		Median	25 – 75%	*p* Value	Median	25 – 75%	*p* Value
PD (mm)	*Baseline*	6.5	6 – 7.5	0.000	6	5 – 7.25	0.000
	Postop (3 months)	4	3 – 5		2	1 - 3	
CAL (mm)	*Baseline*	8	7 – 9	0.000	9	8 - 10	0.000
	Postop (3 months)	7	6 – 7.25		4	3 - 6	

Table 8.VII. Clinical evaluation of BoP baseline and 3 months postop

Variable		Group 1 (SRP)		Group 2 (SRP + Er,Cr:YSGG + H_2O_2 + Diode 940 nm)	
		+ Values (%)	*p* Value	+ Values (%)	*p* Value
BoP (%)	*Baseline*	48 (96%)	< 0.001	48 (96%)	< 0.001
	Postop (3 months)	30 (60%)		3 (6%)	

Table 8.IV. Microbiological variables at baseline and 1 month postoperatively.

Variabile	Grupul 1 (SRP)			Grupul 2 (SRP + Er,Cr:YSGG + H_2O_2 + Diode 940nm)		
	Mediana	25 - 75%	*p*	Mediana	25 - 75%	*p*
Numărul total de bacterii (NTG)						
Preoperator	12.5×10^6	$6.35 \times 10^6 - 28.75 \times 10^6$	0.420	20.5×10^6	$5.7 \times 10^6 - 61 \times 10^6$	0.000
1 lună postoperator	7×10^6	$1.725 \times 10^6 - 29 \times 10^6$		0.885×10^6	$0.11 \times 10^6 - 8.375 \times 10^6$	
Aggregatibacter actinomycetemcomitans (A.a.)						
Preoperator	0×10^6	$0 \times 10^6 - 0 \times 10^6$	-	0×10^6	$0 \times 10^6 - 0 \times 10^6$	-
1 lună postoperator	0×10^6	$0 \times 10^6 - 0 \times 10^6$		0×10^6	$0 \times 10^6 - 0 \times 10^6$	
Porphyromonas gingivalis (P.g.)						
Preoperator	0.059×10^6	$0.01425 \times 10^6 - 0.4125 \times 10^6$	0.001	0.29×10^6	$0.047 \times 10^6 - 0.635 \times 10^6$	0.000
1 lună postoperator	0.0355×10^6	$0.056 \times 10^6 - 0.1017 \times 10^6$		0×10^6	$0 \times 10^6 - 0 \times 10^6$	
Treponema denticola (T.d.)						
Preoperator	0.0495×10^6	$0.019 \times 10^6 - 0.1375 \times 10^6$	0.002	0.035×10^6	$0.016 \times 10^6 - 0.18 \times 10^6$	0.000
1 lună postoperator	0.029×10^6	$0.0085 \times 10^6 - 0.086 \times 10^6$		0.11×10^6	$0.0048 \times 10^6 - 0.086 \times 10^6$	
Tannerella forsythia (T.f.)						
Preoperator	0.026×10^6	$0.006 \times 10^6 - 0.0902 \times 10^6$	0.000	0.0285×10^6	$0.016 \times 10^6 - 0.2025 \times 10^6$	0.000
1 lună postoperator	0×10^6	$0 \times 10^6 - 0.0068 \times 10^6$		0×10^6	$0 \times 10^6 - 0 \times 10^6$	
Prevotella intermedia (P.i)						
Preoperator	0.0175×10^6	$0.0011 \times 10^6 - 0.1025 \times 10^6$	0.058	0.0068×10^6	$0.0012 \times 10^6 - 0.2375 \times 10^6$	0.000
1 lună postoperator	0.068×10^6	$0.0012 \times 10^6 - 0.2375 \times 10^6$		0×10^6	$0 \times 10^6 - 0.019 \times 10^6$	
Peptostreptococcus micros (P.m.)						
Preoperator	0.01×10^6	$0.0032 \times 10^6 - 0.0255 \times 10^6$	0.067	0.0097×10^6	$0.0023 \times 10^6 - 0.0322 \times 10^6$	0.000
1 lună postoperator	0.0051×10^6	$0.0008 \times 10^6 - 0.0122 \times 10^6$		0×10^6	$0 \times 10^6 - 0 \times 10^6$	
Fusobacterium nucleatum (F.n.)						
Preoperator	0.0001×10^6	$0 \times 10^6 - 0.0147 \times 10^6$	0.283	0×10^6	$0 \times 10^6 - 0.0068 \times 10^6$	0.000
1 lună postoperator	0.0004×10^6	$0 \times 106 - 0.0068 \times 10^6$		0×10^6	$0 \times 10^6 - 0 \times 10^6$	
Eubacterium nodatum (E.n.)						
Preoperator	0.0001×10^6	$0 \times 106 - 0.0011 \times 10^6$	0.000	0×10^6	$0 \times 10^6 - 0.0001 \times 10^6$	0.000
1 lună postoperator	0×10^6	$0 \times 10^6 - 0 \times 10^6$		0×10^6	$0 \times 10^6 - 0 \times 10^6$	
Capnocytophaga gingivalis (C.g.)						
Preoperator	0.0003×10^6	$0 \times 10^6 - 0.0029 \times 10^6$	0.000	0.0024×10^6	$0.0001 \times 10^6 - 0.0252 \times 10^6$	0.112
1 lună postoperator	0.0055×10^6	$0.0012 \times 10^6 - 0.0252 \times 10^6$		0.0015×10^6	$0 \times 10^6 - 0.006 \times 10^6$	

Table 8.VIII. Comparison between the investigated groups on the identified bacterial species and the periodontal clinical parameters, pre - and postoperative.

	p Value	
	Group1 vs. Group 2	
	Baseline	*Postop*
NTG	0.291	0.000
A.a.	-	-
P.g.	0.021	0.000
T.d.	0.107	0.000
T.f.	0.262	0.000
P.i.	0.145	0.000
P.m.	0.828	0.000
F.n.	0.283	0.000
E.n.	0.938	0.042
C.g.	0.024	0.002
PPD	0.700	0.000
CAL	0.067	0.000

Table 8.V. Qualitative analysis of microbiological variables

Variable	Group 1 (SRP)		Group 2 (SRP + Er,Cr:YSGG + H_2O_2 + Dide 940 nm)	
	Nr. bacterii (%)	*p*	Nr. bacterii (%)	*p*
Total Number of Bacteria (NTG)				
Preop	50 (100%)	-	50 (100%)	-
1 month postop	50 (100%)		50 (100%)	
Aggregatibacter actinomycetemcomitans (A.a.)				
Preop	0 (0%)	-	4 (8%)	<0.005
1 month postop	0 (0%)		0 (0%)	
Porphyromonas gingivalis (P.g.)				
Preop	47 (94%)	0.016	44 (88%)	<0.001
1 month postop	40 (80%)		8 (16%)	
Treponema denticola (T.d.)				
Preop	48 (96%)	0.021	45 (90%)	<0.001
1 month postop	40 (80%)		4 (8%)	
Tannerella forsythia (T.f.)				
Preop	49 (98%)	<0.001	48 (96%)	<0.001
1 month postop	23 (46%)		0 (0%)	
Prevotella intermedia (P.i.)				
Preop	40 (80%)	0.118	40 (80%)	<0.001
1 month postop	33 (66%)		10 (20%)	
Peptostreptococcus micros (P.m.)				
Preop	49 (98%)	0.039	49 (98%)	0.065
1 month postop	42 (84%)		11 (22%)	
Fusobacterium nucleatum (F.n.)				
Preop	25 (50%)	0.523	19 (38%)	0.002
1 month postop	29 (58%)		6 (12%)	
Eubacterium nodatum (E.n.)				
Preop	26 (52%)	<0.001	24 (48%)	<0.001
1 month postop	4 (8%)		0 (0%)	
Capnocytophaga gingivalis (C.g.)				
Preop	33 (66%)	0.002	38 (76%)	0.383
1 month postop	46 (92%)		33 (66%)	

8.6. Conclusions

By combining two laser wavelengths (2780 nm and 940 nm) and the photolysis of H_2O_2 in periodontal treatment, we can benefit from a series of advantages in comparison to conventional therapy:

1. Improvement of all periodontal clinical indexes (PD, CAL, BOP);
2. Strong bactericidal effect on periodontal tissues improving all investigated clinical and paraclinical parameters (NTG, Pg., Td., Tf., Pi., Pm., Fn., En., Cg., PPD, CAL);
3. Hemostatic effect;

4. Compared to conventional techniques, the laser provides superior access to difficult anatomical areas such as furcation and distal teeth;
5. Comfortable treatment for patients by avoiding periodontal surgery (pockets over 6 mm) and expensive bone augmentation;
6. The use of this therapeutic method avoids the development of resistance to bacteria or to the allergic and toxic reactions induced by antibiotics and anti-inflammatory medication by eliminating them from the treatment plan,
7. Minimal thermal effects on root surfaces, keeping the teeth vital,
8. Possible regenerative effect induced by diode laser through biostimulation of periodontal tissue
9. Minimally-invasive and effective method with high acceptability from patients.

Chapter 9

GENERAL CONCLUSIONS

The present studies have highlighted a number of new aspects in the field of non-surgical periodontal therapy. Based on this results, I can conclude the following:

1. The results of this study underline once again that conventional periodontal therapy represented by SRP (manual and / or piezo-electric) used as monotherapy fails to provide an effective and stable long-term antimicrobial effect in the absence of a complementary procedure in the treatment of periodontal disease.
2. The proposed non-surgical periodontal therapy, represented by photolysis H_2O_2 with diode laser (λ = 940nm) demonstrated significant clinical and microbiological efficiency, even in the absence of association with mandatory conventinal therapy (SRP).
3. The results of this research support the superior antimicrobial efficiency of H_2O_2 photolysis with the 940 nm diode laser compared to 940 nm diode laser as a monotherapy and SRP + diode laser 940 nm as a mandatory therapy indicated by the literature.
4. In contrast to expensive dyes used in antimicrobial photodynamic therapy (aPDT), the use of 3% hydrogen peroxide solution offers a cheap and easy-to-procure alternative.

5. Laser-assisted antimicrobial therapy based on H_2O_2 photolysis provides a beneficial alternative to antibiotic prescription for both chronic and acute periodontal disease.
6. In the periodontal therapy, the combination of two laser wavelengths (2780 nm and 940 nm), the photolysis of H_2O_2 and SRP, offers an accumulation of superior benefits compared to classical procedures.
7. Applying the proposed final protocol presented in this research (SRP + photolysis H_2O_2 with λ 940 nm and Er,Cr: YSGG laser) shows a significant bactericidal effect compared to traditional periodontal therapy, significantly improving all clinical and microbiological indexes investigated in this research study.
8. This research results offer a simple and effective periodontal therapeutic alternative to diode laser users (λ = 940 nm) and those using the Er,Cr:YSGG laser (λ = 2780 nm) with stable long-term results.

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