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ORIGINAL RESEARCH

USE OF LOW LEVEL LASER THERAPY IN OLD PERIODONTAL PATIENTS: A RANDOMIZED CLINICAL TRIAL

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ABSTRACT

Purpose: The present clinical investigation aimed to evaluate the effect of low-level laser treatment on the clinical parameters and checking levels of inducible nitric oxide synthase in individuals with stage II periodontitis across various age groups.

Materials and methods: The current investigation has been carried out on 40 locations in 40 patients. Randomly selected locations were chosen from each studied case, and divided into four groups: group A consisted of 10 sites in elderly patients who underwent scaling combined with root planning exclusively. Group B included 10 sites in young patients who also got scaling and root planning exclusively, group C 10 sites in old age who received diode laser therapy, and group D: 10 sites in young age group received low-level laser (diode laser). The clinical examination used the following clinical parameters: clinical attachment level, plaque index, and Gingival index. An Enzyme-Linked Immunosorbent Assay study was conducted on iNOS.

Results: Baseline and 3-month postoperative groups were compared and showed significant differences in iNOS levels which was influenced by the treatment in the young age group (nonsurgical and laser), but showed no significant difference in the old age group on both types of treatment (nonsurgical and laser).

Conclusion: This clinical investigation showed that whereas non-surgical techniques in conjunction with low-level laser treatment were unsuccessful in the elderly population, it was helpful in the young group with stage II periodontitis.

Keywords: Diode laser, Inducible nitric oxide synthase, Inflammageing, Old age, Periodontal disease

INTRODUCTION

Periodontal diseases refer to inflammatory disorders that damage the tooth-supporting tissues. These conditions originate from the accumulation of bacterial biofilms, which provoke an immune response from the host. As gingival connective tissue above the gum line deteriorates, further destruction of periodontal ligament fibers may compromise the bone that anchors the teeth. Clinically, this manifests as a loss of attachment at the cemento-enamel junction and observable gum inflammation, including redness, swelling, and bleeding upon probing. Additional signs include periodontal pocket formation, gingival recession, involvement of furcation areas, and

radiographically evident bone loss around teeth [1].

In 1976, Page and Schroeder proposed a classification of periodontal diseases based on histologic and clinical indicators of inflammation, changes in gingival appearance, and bleeding upon probing. Their system outlined a progression from early lesions to advanced stages marked by significant attachment loss. Later, during the 1989 World Workshop in Clinical Periodontics, periodontitis was further categorized according to disease progression [2].

A more recent classification model, introduced in 2017, incorporates contemporary scientific knowledge and consists of a staging and grading system. Staging reflects the severity and complexity of periodontitis based on the extent of clinical attachment loss and radiographic bone destruction, while grading evaluates the rate of disease progression and the likelihood of future worsening, considering factors such as systemic health and smoking status. The use of existing clinical data sets is essential for validating and refining this classification scheme [3].

Aging is a natural biological process defined by a gradual decline in physical capabilities and increased vulnerability to chronic diseases, such as cardiovascular conditions, cancer, and neurodegenerative disorders. Research has identified several hallmarks of aging, including telomere shortening, genomic instability, epigenetic modifications, mitochondrial dysfunction, altered nutrient sensing, stem cell exhaustion, cellular senescence, and disrupted intercellular communication. These processes are linked to persistent systemic inflammation, though the precise timing and interactions remain under investigation [4].

Inflammaging describes the age-associated rise in baseline inflammation that results from reduced resilience to stress and cumulative exposure to antigenic triggers over time. This chronic pro-inflammatory state is believed to stem from age-related dysregulation of the immune system, which compromises its ability to maintain homeostasis. While certain innate immune mechanisms remain relatively robust in older adults, adaptive immunity declines more markedly, linking inflammaging with the phenomenon known as immunosenescence [5].

Immunosenescence refers to the age-related deterioration of immune function, which diminishes the body's capacity to respond to new infections and maintain immune surveillance. Changes in lymphoid tissues and immune cell populations contribute to a greater risk of infections, autoimmunity, cancer, and neurodegenerative diseases in elderly populations [6].

Nitric oxide (NO) is a reactive signaling molecule synthesized from L-arginine by nitric oxide synthase (NOS) enzymes. Inducible nitric oxide synthase (iNOS) is a form of NOS that produces high concentrations of NO during inflammation. While iNOS-derived NO helps to eliminate pathogens in acute inflammatory responses, its sustained expression can contribute to bone degradation in severe inflammatory conditions. Studies have shown that periodontal pathogens such as *Porphyromonas gingivalis* and *Aggregatibacter actinomycetemcomitans* can upregulate iNOS expression, thereby promoting the progression of periodontitis [7].

Management of periodontal disease typically aims to reduce pathogenic bacteria through both nonsurgical and surgical therapies to prevent further tissue destruction. Procedures like scaling and root planing are effective at substantially lowering subgingival microbial loads and shifting biofilm composition toward less pathogenic bacterial communities. However, treatment outcomes may be limited by difficult-to-access areas, microbial resistance, virulence factors, or compromised host responses [8].

Adjunctive laser therapy has emerged as an innovative treatment modality for periodontal disease. Laser application can thermally and photodynamically disrupt periodontal pathogens, and its greater tissue penetration enhances bactericidal and detoxification effects, making it a promising complement to conventional periodontal therapy [9].

Diode lasers selectively target chromophores such as melanin, hemoglobin, and pigmented bacteria, leading to the destruction of diseased tissue while sparing healthy structures. When inserted into periodontal pockets, the laser tip removes the epithelial lining and inflammatory infiltrates and delivers low-level radiation to surrounding tissues. This process promotes healing through photobiomodulation by increasing blood flow, reducing swelling, and alleviating pain [10].

The aim of this study is to assess the impact of low-level laser therapy, when used alongside nonsurgical treatment, on salivary iNOS levels in

patients with chronic periodontitis across different age groups. Salivary iNOS will serve as a biomarker for oxidative stress, inflammaging, and immunosenescence in periodontal tissues [11].

MATERIALS AND METHODS

A controlled clinical trial was done on a sample of 40 participants who had been diagnosed with periodontitis. The patients contained in this research were chosen in sequential order from those who were referred to the Department.

Patients aged between above 70 years who were diagnosed with stage II, grade II periodontitis were included in the study. All participants provided written informed consent prior to enrollment. Eligibility criteria required participants to be systemically healthy and diagnosed with stage II periodontitis, with clinical attachment loss of 3–4 mm, radiographic bone loss ranging from 15% to 33%, no tooth loss attributable to periodontitis, and a maximum probing depth of less than 5 mm, predominantly associated with horizontal bone loss. Exclusion criteria comprised smokers, pregnant women, individuals who had received antibiotics within the preceding three months, those undergoing systemic drug therapy, and patients who had received any periodontal treatment within six months prior to the study.

The participants were randomly assigned into four equal groups. Group A consisted of 10 older patients who served as the control group and received nonsurgical periodontal therapy only. Group B included 10 younger patients who also acted as a control group and were treated exclusively with nonsurgical periodontal therapy. Group C comprised 10 older patients forming the study group, who received nonsurgical periodontal therapy in combination with low-level laser therapy. Group D included 10 younger patients in the study group and were treated with nonsurgical periodontal therapy supplemented by low-level laser therapy.

Study procedures:

Periodontal Therapy

All patients received phase I periodontal therapy, which included scaling and root planing performed both supra-gingivally and sub-gingivally, along with oral hygiene instructions. These procedures were carried out using a combination of ultrasonic and manual instruments. Clinical evaluations were performed at four time points: baseline, 4 weeks post-treatment, 3 months, and 6 months. The assessed clinical parameters included the Clinical Attachment Level (CAL).

Clinical Attachment Level (CAL)

Clinical attachment level was defined as the distance between the base of the periodontal pocket and the cemento-enamel junction. Measurements were recorded at the same sites used for probing depth assessment.

Laser Application

Laser disinfection was performed using a 300-µm non-initiated fiber tip directed into the periodontal pockets.

Salivary Sample Collection

Approximately 2 mL of unstimulated saliva was collected from each participant using the Draining method.

Biochemical Evaluation

Salivary samples were collected at two time points: baseline and after three months. The concentration of inducible nitric oxide synthase (iNOS) was determined using an western blot (protein detection).

RESULTS

Clinical evaluation outcomes Following their completion of the research, all participating studied cases underwent follow-up evaluations. The mean and standard deviation values of the values had been displayed. The ShapiroeWilk test of normalcy was used to examine the data for normalcy. ShapiroeWilk findings showed that the data had been regularly distributed (parametric data), hence one-way ANOVA was used for the pairwise comparison, followed by the Post Hoc test.

A significant threshold of P less than or equal to 0.05 had been established. The statistical pro- gramme SPSS had been used to conduct the analysis (version 25, IBM Colorado, USA).

Effect of time interval and age of patients, on the CAL under the same group type (Intra-group Comparison)

1.1 Non-Surgical group

1.1.1 Effect of time interval on the CAL under the same patient age and same group

Non-Surgical Young: the mean of the CAL was (3.53±0.36 mm) at baseline, (2.6±0.35 mm) after 4 weeks, (2±0.24 mm) after 3 months, and (1.8±0.3 mm) after 6 months, while in the **Non-Surgical Old:** the mean of the CAL was (3.77±0.19 mm) at baseline, (2.77±0.33 mm) after 4 weeks, (2.23±0.15 mm) after 3 months, and (2.1±0.15 mm) after 6 months. So *For both subgroups (young and old patients):* According to the Tukey Post Hoc test, there was a significant difference between baseline and other time intervals, as well as between 4 weeks and other time intervals; however, there was no significant difference between 3 months and 6 months. According to the ANOVA test, the overall P-value for intragroup comparison was statistically highly significant (P-value < 0.001). This significant was attributed to the difference between baseline (the highest mean of CAL) and 6 months (the lowest mean of CAL).

1.1.2 Effect of patient age on the CAL at the same time and in the same group

For all time intervals: Although the CAL mean for the old patient was higher than that for the young patient, the difference between the old and young subgroups was not statistically significant.

1.2 Laser group

1.2.1 Effect of time interval on the CAL under the same patient age and same group

Non-Surgical Young: the mean of the CAL was (3.77±0.28 mm) at baseline, (2.5±0.33 mm) after 4 weeks, (1.77±0.4 mm) after 3 months, and (1.6±0.37 mm) after 6 months.

Non-Surgical Old: the mean of the CAL was (3.8±0.27 mm) at baseline, (2.47±0.46 mm) after 4 weeks, (1.9±0.62 mm) after 3 months, and (1.73±0.37 mm) after 6 months.

For both subgroups (young and old patients): Like the Non-surgical group, according to the Tukey Post Hoc test, there was a significant difference between baseline and other time intervals, as well as between 4 weeks and other time intervals; however, there was no significant difference between 3 months and 6

months. According to the ANOVA test, the overall P-value for intragroup comparison was statistically highly significant (P-value < 0.001). This significant was attributed to the difference between baseline (the highest mean of CAL) and 6 months (the lowest mean of CAL).

1.2.2 Effect of patient age on the CAL at the same time and in the same group

For all time intervals: Although the CAL mean for the old patient was higher than that for the young patient, the difference between the old and young subgroups was not statistically significant.

Table 1. Mean ± SD and intra-group comparison of clinical attachment loss (mm) between the twotime intervals (Before and After) and the two ages (Young and Old) for both groups.

	Non-Surgical			Laser		
	Young	**Old**	P-value*	**Young**	**Old**	P-value*
Baseline	3.53±0.36[a]	3.77±0.19[a]	**0.237[NS]**	3.77±0.28[a]	3.8±0.27[a]	**0.853[NS]**
4 Weeks	2.6±0.35[b]	2.77±0.33[b]	**0.455[NS]**	2.5±0.33[b]	2.47±0.46[b]	**0.899[NS]**
3 Months	2±0.24[c]	2.23±0.15[c]	**0.098[NS]**	1.77±0.4[c]	1.9±0.62[c]	**0.697[NS]**
6 Months	1.8±0.3[c]	2.1±0.15[c]	**0.079[NS]**	1.6±0.37[c]	1.73±0.37[c]	**0.580[NS]**
P-value**	**< 0.001[HS]**	**< 0.001[HS]**		**< 0.001[HS]**	**< 0.001[HS]**	

-* *P-value for Intra-group comparison (Young vs. Old) from Ina*

-Test.

-** *Overall P-value for Intra-group comparison between the four-time intervals (ANOVA Test).*

- *Small letters for pairwise comparison between different time intervals (Tukey Post Hoc test) and the means with different superscripts are statistically significant different at P ≤ 0.05. - S= Statistically significant at P ≤ 0.05 - NS= Non-significant P < 0.05.*
- *HS= Highly significant at P ≤ 0.001*

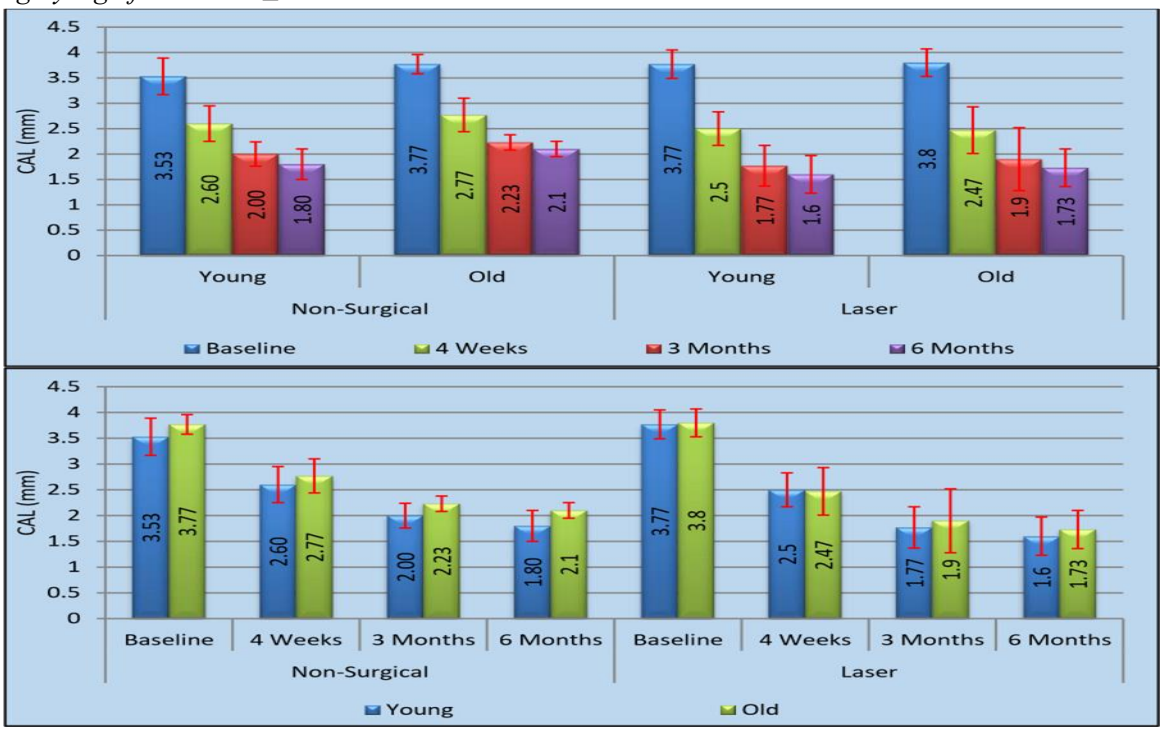

Figure 1. Bar chart representing Mean and SD of CAL at the two-time intervals (Before and After) and for the two ages (Young and Old) for both groups

DISCUSSION

Periodontal disease is initiated and sustained by a dysregulated host immune response to microbial plaque biofilms rather than by bacterial challenge alone. An exaggerated immune reaction leads to persistent inflammation, characterized by continuous release of inflammatory mediators such as cytokines and chemokines. This chronic inflammatory state contributes to excessive production of reactive oxygen and nitrogen species, thereby intensifying oxidative stress and impairing the antioxidant defense system within periodontal tissues [12,13].

Oxidative stress plays a pivotal role in the pathogenesis of periodontitis by promoting connective tissue breakdown, alveolar bone resorption, and cellular senescence. Prolonged exposure to oxidative damage accelerates tissue aging and apoptosis through mitochondrial dysfunction and sustained inflammatory signaling. Consequently, therapeutic strategies aimed at controlling periodontal disease should incorporate approaches that modulate oxidative stress and suppress excessive inflammatory mediator release [14–16].

Photobiomodulation therapy represents a non-invasive modality that exerts its biological effects through the absorption of light energy by cellular chromophores, triggering a cascade of photochemical and photobiological reactions. These reactions enhance mitochondrial activity, improve cellular metabolism, and regulate inflammatory signaling pathways. Several studies have demonstrated that low-level laser therapy can effectively reduce oxidative stress and downregulate inflammatory mediators, thereby supporting periodontal tissue repair and regeneration [17,18].

In the present study, variations in salivary iNOS levels following nonsurgical periodontal therapy were particularly evident among younger individuals. This observation may be attributed to the dual role of nitric oxide as both a protective antimicrobial agent and a mediator of tissue destruction. During early inflammatory responses, increased nitric oxide production may function as a host defense mechanism against microbial invasion. However, sustained nitric oxide activity has been shown to promote extracellular matrix degradation through inhibition of collagen

synthesis and stimulation of matrix metalloproteinase activity [19,20].

Nitric oxide is increasingly recognized as a central regulator in periodontal immuno-inflammatory pathways. Its overexpression has been associated with disease severity and progression, suggesting that targeted modulation of nitric oxide signaling could be beneficial in periodontal therapy. Emerging therapeutic approaches include selective inhibition of iNOS activity and the use of antioxidant compounds to neutralize reactive nitrogen species and limit peroxynitrite-mediated tissue damage [21].

Further studies should be conducted with a larger sample size to ensure the oxi-inflammaging theory.

DECLARATION

Conflicts of interest

The author declares that he has no conflict of interest

Ethical approval

The study was approved by the Institutional Ethics Committee and was conducted in accordance with the Declaration of the World Medical Association.

Informed consent

Informed consent was obtained from all individual participants included in the study.

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REFERENCES

1. Smith J, Brown L, Taylor R. Pathogenesis and clinical manifestations of periodontal diseases. *J Periodontol*. 2021;92(4):512–520.
2. Lee A, Johnson P. Evolution of periodontal disease classification systems. *Periodontol 2000*. 2020;82(1):7–21.
3. Garcia M, Hernandez F, Lopez R. The 2017 classification of periodontitis: Staging and grading concepts. *J Clin Periodontol*. 2022;49(3):230–238.
4. O'Connor D, Patel S. Biological hallmarks of aging and chronic inflammation. *Ageing Res Rev*. 2021;67:101311.
5. Torres C, Huang Y. Inflammaging and immune system dysregulation in aging populations. *Clin Immunol*. 2023;246:109214.
6. Martinez L, Singh N. Immunosenescence and age-related immune dysfunction. *Nat Rev Immunol*. 2021;21(4):215–230.
7. Kim J, Park S, Lee K. Role of inducible nitric oxide synthase in periodontal inflammation. *Mol Oral Microbiol*. 2022;37(2):79–88.
8. Wang H, Roberts M. Nonsurgical periodontal therapy: Microbial and clinical outcomes. *Int J Dent*. 2020;2020:8893412.
9. Chen Y, Lo E. Adjunctive laser therapy in periodontal treatment: A systematic review. *Lasers Med Sci*. 2021;36(3):493–504.
10. Patel R, Mehta V, Shah N. Diode laser applications in periodontal therapy and wound healing. *Photomed Laser Surg*. 2023;41(2):85–92.
11. Almeida P, Rivera C. Salivary biomarkers of oxidative stress and inflammaging in chronic periodontitis. *Arch Oral Biol*. 2024;154:105394.
12. Hajishengallis G. Immunomicrobial pathogenesis of periodontitis: Keystones, pathobionts, and host response. Trends Immunol. 2014;35(1):3–11
13. Kinane DF, Stathopoulou PG, Papapanou PN. Periodontal diseases. Nat Rev Dis Primers. 2017;3:17038.
14. Chapple IL, Matthews JB. The role of reactive oxygen and antioxidant species in periodontal tissue destruction. Periodontol 2000. 2007;43:160–232.
15. Sczepanik FSC, Grossi ML, Casati MZ, Goldberg M, Glogauer M, Fine N, et al. Periodontitis is an inflammatory disease of oxidative stress. J Clin Periodontol. 2020;47(3):305–316.
16. Bullon P, Newman HN, Battino M. Obesity, diabetes mellitus, and periodontal disease: Oxidative stress as a common factor linking these conditions. J Dent Res. 2014;93(7):574–579.
17. Hamblin MR. Mechanisms and applications of the anti-inflammatory effects of photobiomodulation. AIMS Biophys. 2017;4(3):337–361.
18. de Freitas LF, Hamblin MR. Proposed mechanisms of photobiomodulation or low-level light therapy. IEEE J Sel Top Quantum Electron. 2016;22(3):7000417.
19. Batista AC, Silva TA, Chun JH, Lara VS. Nitric oxide synthesis and severity of human periodontal disease. Oral Dis. 2002;8(5):254–260.
20. Kendall HK, Marshall RI, Bartold PM. Nitric oxide and tissue destruction. Oral Dis. 2001;7(1):2–10.
21. Preshaw PM, Taylor JJ. How has research into cytokine interactions impacted our understanding of periodontitis? J Clin Periodontol. 2011;38(Suppl 11):60–84.