CLINICAL AND HISTOPATHOLOGICAL COMPARISON BETWEEN NON-SURGICAL PERIODONTAL THERAPY AND LASER CURETTAGE IN AGGRESSIVE PERIODONTITIS PATIENTS

Ahmed Shawkat Hashem* and Sherif Farouk Elgayar**

ABSTRACT

Background: In patients with aggressive periodontitis, scaling and root planing is usually combined with the use of systemic antibiotics. However, the effectiveness of these antibiotics over time was questioned. Diode laser has been introduced as an adjunctive treatment modality in the treatment of periodontal disease.

Objective: The aim of this study is to compare Laser debridement with non-surgical periodontal therapy for the treatment of aggressive periodontitis.

Subjects and methods: Probing depth and clinical attachment loss were measured in six sites for two teeth in all 10 aggressive periodontitis patients selected for the study. Also, Gingival index was recorded.

Scaling and root planing was performed on one tooth, and the other was treated using Diode Laser therapy. All measurements were carried at baseline and four weeks after completion of the therapy for both modalities. Moreover, gingival samples were taken from all sites before and after periodontal therapy.

Results: Statistically significant reduction was observed in mean gingival scores in both groups after intervention, and the Laser group showed more statistical significance than curettage group. However, there was no statistical significant difference between both groups concerning probing depth measurements. Again, no significant difference was observed between groups regarding clinical attachment loss improvement. Histopathological results showed less inflammatory infiltrate in cases treated with Laser therapy.

Conclusion: Clinical and histopathological investigations proved that Laser therapy is more effective in reducing inflammation in periodontitis patients. Thus, Laser application should be adjuncted to all periodontal procedures in order to improve clinical outcomes.

KEYWORDS: Aggressive Periodontitis, Diode Laser, Non-surgical, SRP.

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INTRODUCTION

Periodontitis is an inflammatory response characterized by gingival inflammation, loss of supporting periodontal tissues, and alveolar bone destruction. Although the main causative factor of periodontal disease is the microbial plaque, the rate of progression and degree of severity of such disease are mainly determined by the host immune response, which differs among different individuals.

Periodontal disease represents a chronic inflammatory pathology of infectious origin that affects periodontal tissues. The interactions between host and bacteria determine the nature of the resulting disease such as other infectious conditions.

Immunohistochemical markers of cellular activation have indicated that T and B-lymphocytes in crevicular fluid and gingival tissues are active in periodontal disease. These cells are playing a major role in the host local immune response. It is also well known its functional potential to release cytokines and antibodies (via plasmacyte differentiation) when activated, such as the anti-inflammatory cytokine IL-10, which induces the expression of tissue inhibitors of metalloproteinases and osteoprotegerin, the respective inhibitors of MMPs and RANKL systems. It is therefore thought to attenuate disease severity.

Active B-lymphocytes express surface molecules, such as TNF, in periodontal lesions. The expression of these cellular activation markers is variable according to the bacterial species that stimulated B cell activation in gingivitis and periodontitis.

Tumor necrosis factor-alpha (TNF-α) seems to dominate the pathology in periodontal diseases. In addition, the balance of metalloproteinases and their inhibitors determines the destruction of extracellular matrix in both conditions. In both settings, TNF-α contributes to the up-regulation of osteoclastogenesis and to the down regulation of osteoblastogenesis.

Aggressive periodontitis is a rapidly progressing form of periodontal disease that occurs in otherwise clinically healthy individuals. It is confirmed that, compared with patients with chronic periodontitis, patients with aggressive periodontitis show a more rapid epithelial attachment loss and bone destruction that occurs in younger patients. The patient’s age when attachment loss is detected is often an important factor used by clinicians to diagnose aggressive periodontitis.

When considering treatment options and goals, managing aggressive periodontitis is more or less similar to chronic periodontitis. However, the severity of bone loss relative to the young age of patients in aggressive periodontitis suggests more aggressive treatment modalities in order to arrest periodontal destruction, and retain as many teeth as possible.

There is conflicting opinions regarding treatment modalities for aggressive periodontitis, as there are no definite procedures or standards for reaching complete cure. Many studies have recommended different therapeutic options including surgical and non-surgical debridement, disinfectants and antibiotics for the treatment of such disease.

Mechanical debridement comprising scaling, root planing, and curettage; is the most common treatment strategy performed in aggressive periodontitis. It has been reported that mechanical debridement when combined with antibiotic therapy is more effective for treating periodontal disease than when scaling and root planing is the only treatment modality. Systemic antibiotics are given in order to decrease number of pathogenic bacteria and help establishing a health-associated biofilm. However, many factors should be considered when using antibiotics, such as adverse effects, bacterial resistance, and patient’s compliance.
Traditional scaling, root planing (SRP) and regular oral hygiene practice by the patient have been shown to be effective for the reduction of inflammation and probing depths as well as restoring clinical attachment levels, however challenging situations associated with deeper pockets, root morphology and difficult access areas decrease the rates of healing following nonsurgical periodontal therapy\(^\text{17}\). Adjuncts such as antimicrobials and lasers have been advocated to overcome these limitations. Lasers have been increasing in popularity in dental hygiene practice that may be used in the treatment of periodontitis as a mono-therapy or as an adjunct to SRP during initial periodontal therapy, surgery, or periodontal maintenance therapy\(^\text{18}\).

Laser therapy is indicated for sulcular debridement, also known as gingival curettage, and for bactericidal activity within the periodontal pocket. Unlike other therapeutic procedures used by dentists, there is no standard accepted protocol for the use of lasers. As a general rule, the performance of a given laser is governed by its absorption, or depth of penetration into the tissues, which depends on the wavelength. Diode laser is deeply penetrating whereas CO2 laser penetrates superficially\(^\text{19}\).

Based on the systematic review and meta-analysis by Slot et al, the adjunctive use of the most commonly employed diode laser (809 to 980 nm) as an adjunct to traditional mechanical debridement of periodontal therapy in patients with periodontitis is questionable\(^\text{19}\).

According to the literature, there is no difference between the treatment concepts used for treating chronic periodontitis or aggressive periodontitis. Although the effect of nonsurgical treatment on chronic periodontitis is well documented\(^\text{20}\), its effect on aggressive periodontitis is much less clear. As such, there is still need to clarify the effect of nonsurgical therapy alone as a treatment for aggressive periodontitis. The current study aimed to assess the effect of non-surgical periodontal therapy and compare it with Laser debridement for the treatment of aggressive periodontitis both clinically and histologically.

**SUBJECTS AND METHODS**

This study was conducted on 10 female patients diagnosed with generalized aggressive periodontitis who were selected from different private practice clinics. The selection of the subjects was based on the classification of periodontal disease by the international workshop for classification of periodontal diseases and conditions in 1999\(^\text{21}\).

Patients were recruited to participate in this study after a screening visit that included full mouth periodontal evaluation and panoramic radiographic examination in order to confirm the diagnosis. All subjects were female patients with age range of 22 – 40 years.

**Inclusion criteria:**
- Free from any systemic diseases.
- Free from oral diseases other than periodontitis.
- Non-smokers.
- Good general health according to medical history and no allergies to local anaesthetics.
- Presence of at least 1 hopeless tooth in each side with attachment loss more than 10 mm.

**Exclusion criteria:**
- Subgingival instrumentation within 6 months prior to the baseline examination.
- History of antibiotic therapy within 6 months prior to the start of the study.
- On-going drug therapy that might affect the patient’s clinical response.
- Pregnant women.

The patients were informed about the purpose and the method of the study, and their signed consents to participate were obtained. The study was
in compliance with the rules set by the International Conference on Harmonisation of good clinical practice guidelines, and the Declaration of Helsinki.

In each patient one hopeless tooth was selected to be treated by mechanical debridement using periodontal curettes, while another hopeless tooth was designated to be treated by laser therapy using Diode laser device.

For all patients, measurements of probing depth (PD) and clinical attachment loss (CAL) were made at six sites (mesiobuccal, buccal, distobuccal, mesiolingual/palatal, palatal/lingual, and distolingual/palatal) for each tooth, using a manual periodontal probe (William’s periodontal probe designed with 1, 2, 3, 5, 7, 9, and 10 mm calibrations). Gingival index (GI) was recorded at four sites (mesiobuccal, buccal, distobuccal, and palatal/lingual). Periodontal examination was carried out by a single trained periodontist.

**Periodontal therapy**

At the initial treatment, each patient was subjected to full mouth supra- and subgingival scaling using sonic scaler and also full mouth polishing was performed. Then, all patients received oral hygiene instructions to be followed. One week after the end of initial treatment, the tooth selected for manual curettage was manipulated using area-specific curettes (Hu-Friedy Co. Inc., Chicago, IL, USA) under 2% Lidocaine anaesthesia. Mechanical debridement was completed on two sessions over two weeks period. Concerning the other treatment modality, a Diode laser equipment (Quicklase Co, London, UK) with a wavelength of 810 nm, delivered by a 400-µm diameter fiber optic device was used for this trial. After infiltration of anaesthesia, the fiber optic was introduced in the periodontal pocket parallel to the long axis of the tooth, one-millimeter coronal to the base of the pocket. Sweeping motions were performed for 20 seconds using a power of 1.5 W on both the buccal and lingual sides of the tooth using the pulsed mode. Laser irradiation was performed over two sessions also in two weeks interval. All clinical parameters were re-evaluated four weeks after completion of both procedures in concerned teeth.

**Histopathological preparation of specimens**

A small part (2-3 mm) from the gingival sulcus was excised by a sterile blade to be used for histopathological examination. This was performed for every patient before and after periodontal treatment. Each specimen was fixed in formalin and embedded in paraffin. Tissue sections, 3-µm thick, were cut and mounted on glass silanized microscope slides (3-aminopropyltriethoxysilane; Sigma Chemical Co., St Louis, MO, USA). Immunohistochemistry was carried out using the streptavidin-biotin complex method. The sections were treated with primary antibody against TNF-α. Antigen retrieval was performed with Steamer during 15 min. The dilution used was 1:40 and sections were incubated for 60 min.

**Analysis of the Immunostained Cells**

Scores from 0 to “+++” were established to evaluate the immunostaining cellular density in the lamina propria of the specimens in accordance with the methodology, modified for this study. The following parameters were considered: Score 0 = negative expression, Score + = discrete number of immunopositive cells; Score ++ = moderate number of positive cells; Score +++ = large number of immunopositive cells. A descriptive analysis of the microscopic findings regarding localization (beneath and/or far from the epithelium) and distribution pattern of TNF-α positive cells in gingival connective tissue was performed.

**Statistical Analysis**

The mean and the standard deviation of the variables PD, CAL, GI, from the experimental sites, were considered for the statistical analysis. Repeated measures analysis of variance (ANOVA) was used
to determine the differences between the averages of the groups. For intra-groups comparisons, the groups were evaluated using the ANOVA and post hoc Tukey’s analysis, p values <0.05 were considered statistically significant.

RESULTS

All patients completed the course of the study. No complications, such as pain or infection, were detected in this study. All clinical measurements were carried out for all patients before, and four weeks after completion of the designated procedure in each group.

Gingival index scores

Mean gingival index scores were measured before and after intervention in each group, statistically significant reduction was observed in mean gingival scores in both groups after intervention (Table 1).

When comparing the Laser group and the curettage group, Tukey’s post hoc analysis revealed the presence of statistically significant difference between the groups after intervention for the favor of the Laser group. (Table 2)

Also, this table confirmed that the reduction in mean gingival index scores within both groups was statistically significant.

TABLE 1:

<table>
<thead>
<tr>
<th>Group</th>
<th>Mean±SD</th>
<th>ANOVA</th>
<th></th>
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</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>F-value</td>
<td>P-value</td>
<td></td>
</tr>
<tr>
<td>Before curettage</td>
<td>2.25±0.70</td>
<td>4.746</td>
<td>0.008</td>
<td></td>
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<tr>
<td>After curettage</td>
<td>1.12±0.99</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Before Laser therapy</td>
<td>2±0.75</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Before Laser therapy</td>
<td>1±0.75</td>
<td></td>
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</tr>
</tbody>
</table>

When comparing the Laser group and the curettage group, Tukey’s post hoc analysis revealed the presence of statistically significant difference between the groups after intervention for the favor of the Laser group. (Table 2)

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TABLE 2:

<table>
<thead>
<tr>
<th>Multiple Comparisons</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dependent Variable: gingival index</td>
</tr>
<tr>
<td>Tukey HSD</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>(I) grp</th>
<th>(J) grp</th>
<th>Mean Difference (I-J)</th>
<th>Std. Error</th>
<th>Sig.</th>
<th>95% Confidence Interval</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Lower Bound</td>
<td>Upper Bound</td>
<td></td>
</tr>
<tr>
<td>cur-b</td>
<td>cur-a</td>
<td>1.12500*</td>
<td>.40505</td>
<td>.045</td>
<td>.0191</td>
</tr>
<tr>
<td></td>
<td>las-b</td>
<td>.25000</td>
<td>.40505</td>
<td>.926</td>
<td>-.8559</td>
</tr>
<tr>
<td></td>
<td>las-a</td>
<td>1.25000*</td>
<td>.40505</td>
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<td>.1441</td>
</tr>
<tr>
<td>cur-a</td>
<td>cur-b</td>
<td>-1.12500*</td>
<td>.40505</td>
<td>.045</td>
<td>-2.2309</td>
</tr>
<tr>
<td></td>
<td>las-b</td>
<td>-.87500</td>
<td>.40505</td>
<td>.159</td>
<td>-1.9809</td>
</tr>
<tr>
<td></td>
<td>las-a</td>
<td>.12500</td>
<td>.40505</td>
<td>.990</td>
<td>-.9809</td>
</tr>
<tr>
<td>las-b</td>
<td>cur-b</td>
<td>-.25000</td>
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<td></td>
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</table>

* The mean difference is significant at the 0.05 level.
**Probing depth**

Mean probing depth scores were measured before and after intervention in both Laser and curettage groups. Again, statistically significant reduction in mean probing depth was found after intervention in both groups. (Table 3)

<table>
<thead>
<tr>
<th>Group</th>
<th>Mean±SD</th>
<th>ANOVA</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>F-value</td>
</tr>
<tr>
<td>Before curettage</td>
<td>9.1±0.35</td>
<td>7.661</td>
</tr>
<tr>
<td>After curettage</td>
<td>7.8±1.12</td>
<td></td>
</tr>
<tr>
<td>Before Laser therapy</td>
<td>9.2±0.46</td>
<td></td>
</tr>
<tr>
<td>Before Laser therapy</td>
<td>8.1±0.64</td>
<td></td>
</tr>
</tbody>
</table>

Interpretation of the following table revealed there is significant difference between the groups and reduction in mean probing depth scores after intervention in both the groups were statistically significant. Probing depth scores were more statistically significant in the Laser group as evidenced by multiple comparisons. (Table 4)

**Attachment loss**

Mean attachment loss was measured before and after interventions in each group, analysis of these calculations showed statistically significant reduction in attachment loss in both groups after intervention. However, Tukey's post hoc analysis revealed there is no significant difference between the groups. (Table 5)

**Table 3:**

<table>
<thead>
<tr>
<th>Group</th>
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<tbody>
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</table>

**Table 4:**

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<thead>
<tr>
<th>Dependent Variable: probing depth</th>
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</tr>
</thead>
<tbody>
<tr>
<td>Multiple Comparisons</td>
<td></td>
</tr>
<tr>
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<tr>
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<tr>
<td>las-b</td>
<td>las-a</td>
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<td>las-b</td>
<td>cur-a</td>
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<td>las-b</td>
<td>cur-a</td>
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<tr>
<td>las-a</td>
<td>cur-a</td>
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<tr>
<td>las-a</td>
<td>las-b</td>
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</tbody>
</table>

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TABLE 5:

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</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>F-value</td>
</tr>
<tr>
<td>Before curettage</td>
<td>7.37±0.51</td>
<td>3.175</td>
</tr>
<tr>
<td>After curettage</td>
<td>6.87±0.35</td>
<td></td>
</tr>
<tr>
<td>Before Laser therapy</td>
<td>7.50±0.53</td>
<td></td>
</tr>
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</table>

Various outcome measurements and comparisons between Laser group and curettage group are illustrated in figures 1 & 2.

Histopathological results either through H&E show that there are less inflammatory infiltrate in cases treated by laser Figure (3, 4) and positive immunostain expression with anti-TNF-α for macrophages tended to cluster around blood capillaries often infiltrating and disrupting vascular endothelium. It is suggested that this degree of TNF-α production probably contributes significantly to the pathogenesis periodontitis, by impairing the integrity of epithelial and endothelial membranes Figure (5,6)

**Histopathologic statistical results**

The mean of immune-positive TNF-α cells scores were measured before and after intervention in each group, statistically significant reduction was observed in mean immune-positive TNF-α cells scores for the group treated by laser as shown in figure 7 and table 5

Fig. (1)

Fig. (2)

Fig. (3) Photomicrograph for aggressive lymphocytic inflammatory infiltration in connective tissue in specimen treated by curettage (H&E X200)

Fig. (4) Photomicrograph for moderate lymphocytic inflammatory infiltration in connective tissue in specimen treated by laser (H&E X200)
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DISCUSSION

This study was designed in order to investigate and compare the clinical and inflammatory effects of the use of Diode laser versus scaling and root planning for the treatment of generalized aggressive periodontitis. A split-mouth design was used in this study, which has the advantage of controlling individual variations between subjects. However, this kind of design could be influenced by variations in disease patterns from one side of the mouth to the other, thus this point should be considered during subjects’ selection.

As showed in many studies, scaling and root planing (SRP) decrease the clinical parameters of periodontitis. The results of this study also confirmed this fact in patients with aggressive periodontitis.

It is generally accepted that the treatment of the aggressive form of periodontal diseases is more challenging to clinicians. Thus, a variety of local and systemic antimicrobial agents have been evaluated in adjunction with scaling and root planing in order to improve healing results. These studies were not able to confirm which antimicrobial agent; dose and duration of application would provide optimal clinical and antimicrobial effects in those patients. Moreover, bacterial resistance to antibiotics could be the cause of the lack of efficacy of such drugs in the treatment of periodontitis. The use of laser
Clinical and histopathological comparison between non-surgical 

has the advantage of not being restricted by such 
drawbacks as it is applied locally to the targeted 
area with no harmful effect to adjacent region. 
Regarding antimicrobial effect of Diode laser, some 
studies have demonstrated a reduction in number of 
pathogenic microorganisms in periodontal pockets 
treated with laser therapy, whereas, other studies 
have showed no significant decrease in number of 
pathogenic microorganisms in pockets treated with 
SRP and diode lasers compared to SRP treatment 
alone. These contradictions may be due to 
variations in the irradiation parameters used in these 

Another advantage of laser therapy is that it 
is more comfortable for the patient and less time 
consuming, as the fiber tip should be held in the 
periodontal pocket for only a short time. Also, 
no risk for tissue trauma is encountered like seen 
during mechanical debridement.

The results of this study were consistent with 
those of another study that compared also same 
techniques in a split-mouth design for aggressive 
periodontitis patients. Clinical parameters decreased 
after 3 months, although the results did not reveal 
significant differences between the two groups.

Virtually Tumor necrosis factor-alpha (TNF-α) 
as well all of the cytokines and inflammatory 
mediators having a great role in the pathology of 
periodontal diseases, TNF-α contributes to the up-
regulation of osteoclastogenesis and to the down-
regulation of osteoblastogenesis.

Photo-bio-modulator properties of the laser 
have great influence to decrease in number of the 
mast cells in groups treated with laser therapy is 
likely due to the that promotes analgesic and anti-
inflammatory effects and accelerates tissue repair. 
There are many different therapeutic actions 
of lasers on tissues, including increased local 
microcirculation, reduction of the number of 
inflammatory cells, inhibition of cyclooxygenase-2 
(COX-2) and pro-inflammatory cytokine synthesis, 

increase in collagen synthesis and stimulation of the 
proliferation of epithelial cells and fibroblasts

Difficulty in determining active disease and 
ongoing destruction in periodontal tissue by 
traditional diagnostic aids like probing depth and 
attachment loss has proved them to be inadequate 
in modern era of periodontal therapeutics. Search 
for a biomarker for periodontitis has resulted in 
researchers trying out and finding new molecules 
that can guide a clinician in many a decision 
regarding the patient’s condition.

Tumor necrosis factor-alpha (TNF-α) is a 
proinflammatory cytokine released by macrophages 
is known for its substantial role in periodontitis 
mediated bone loss.

The positive expression of TNF-α for macro-
phages tended to cluster around blood vessels, often 
infiltrating and disrupting vascular endothelium, 
explaining TNF-α role in the pathogenesis of both 
periodontitis, by impairing the integrity of epithelial 
and endothelial membranes, increasing inflamma-
tory cell recruitment, and by prothrombotic effects 
on the vascular endothelium.

Concerning two parameters of the study, 
which are the gingival index, and probing depth, 
statistically significant difference was observed in 
both groups after intervention, which was proved 
in another study, and there was also significant 
difference between the 2 groups in favor of the Laser 
group, which is correlated with the histopathological 
picture that showed less inflammation in Laser 
specimens.

Regarding the attachment loss, although 
there was statistically significant difference after 
treatment in both groups, there was no statistical 
significance detected between the groups. However, 
the reduction in attachment loss calculated regardless 
of its statistical significance, it was clinically 
non-significant as it was too little to improve the 
condition of the affected teeth.
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