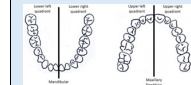


# Evaluating the Clinical and Microbiological Impact of Laser Therapy as an Adjunct to Non-Surgical Treatment for Chronic Periodontitis in High-Risk Groups

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**Abstract:** Type 2 Diabetes Mellitus (T2DM) and smoking are significant risk factors for periodontitis, complicating treatment outcomes. Despite the growing popularity of adjunctive laser therapy (ALT) combined with scaling and root planing (SRP), its long-term benefits for managing chronic periodontitis and its effects on microbial profiles across different risk groups remain poorly understood. This study compared treatment outcomes and microbiome composition in patients with these risk factors who received either SRP alone or SRP combined with ALT. We utilized next-generation sequencing of the 16S rRNA gene to analyze subgingival plaque samples collected at baseline and at 1, 3, 6, 9, and 12 months. QIIME 2 was employed to assess microbial community compositions and evaluate treatment efficacy through alpha- and beta-diversity, as well as taxonomy analysis. Our findings indicate that ALT can enhance the effectiveness of SRP, particularly in diabetic and smoker patients. Specifically, ALT led to more substantial reductions in pocket depth and the prevalence of key pathogenic genera, such as *Prevotella*, *Peptidiphaga*, and *Treponema*, which are associated with more severe periodontal conditions. These results suggest that ALT may improve treatment outcomes and microbial profiles in high-risk groups, thereby enhancing the overall management of periodontitis, especially in cases with deeper periodontal pockets.

## Background:

- Periodontal disease is a prevalent inflammatory condition that causes gingival tissue destruction and tooth loss, with its occurrence increasing with age.
- The aim of periodontal treatment is to eliminate pathogenic bacterial deposits, including plaque, biofilm, and calculus, through mechanical removal, thereby establishing a healthy and maintainable oral environment that reduces the risk of further infection and disease progression.
- Key pathogens, including *Porphyromonas gingivalis*, *Tannerella forsythia*, and *Prevotella denticola*, significantly exacerbate the condition, especially in deep periodontal pockets.

## Risk Factors:

- Type 2 Diabetes Mellitus (T2DM) increases susceptibility to periodontitis by promoting microbiome dysbiosis, with about one-third of diabetics suffering from severe periodontitis.
- Smoking further aggravates periodontal disease and complicates treatment outcomes.

## Treatment Options:

- Scaling and Root Planing (SRP) is a common treatment but often fails to completely eliminate pathogens, particularly in complex root structures.
- Adjunctive Laser Therapy, such as the Er,Cr:Las, has shown potential for enhancing treatment outcomes, although its effectiveness in high-risk groups remains insufficiently researched.

## Methods:

**Data Collection:** Fourteen patients (four with T2DM, four smokers, and six normoglycemic non-smokers) with stage II to IV periodontitis were recruited for a split-mouth study at the Postgraduate Periodontics and Implant Surgery Clinic at UCLA.



**Data Preparation:** DNA was extracted from the samples using the Epicenter MasterPure kit (Epicenter, Madison, WI). It was then sent to UCLA's Technology Center for Genomics and Bioinformatics for library preparation and sequencing.



## Analysis:

- Sequence quality control and feature table construction
- Denoised Single-End Sequences
- Comparison to the Human Oral Microbiome 16S rRNA Database to determine genus-level microbial community composition of SRP+ALT vs. SRP only sites.
- Beta-diversity (Weighted UniFrac) and principal coordinate analyses were calculated in QIIME2

## Results:

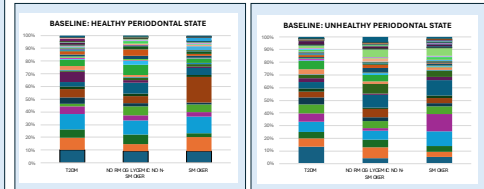


Figure 1: Genus-level taxonomic classification analysis showing the subgingival pathogenic bacterial profiles of patients at a healthy periodontal state and those with an unhealthy periodontal state.

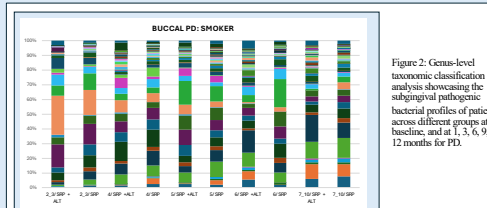
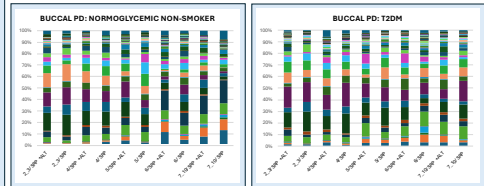


Figure 3: Genus-level taxonomic classification analysis showing the subgingival pathogenic bacterial profiles of patients across different groups at baseline, and at 1, 3, 6, 9, and 12 months for PD.

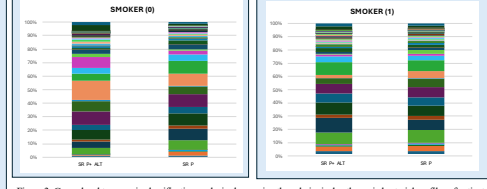
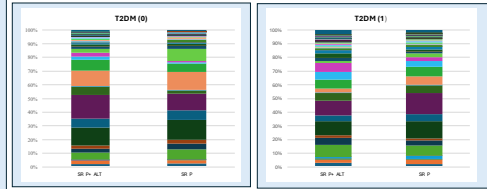
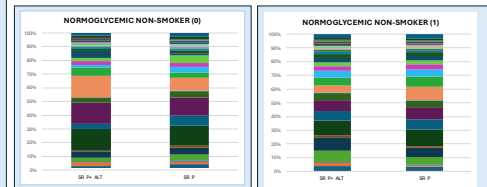


Figure 3: Genus-level taxonomic classification analysis showing the subgingival pathogenic bacterial profiles of patients across different groups at baseline, and at 1, 3, 6, 9, and 12 months for BOP.

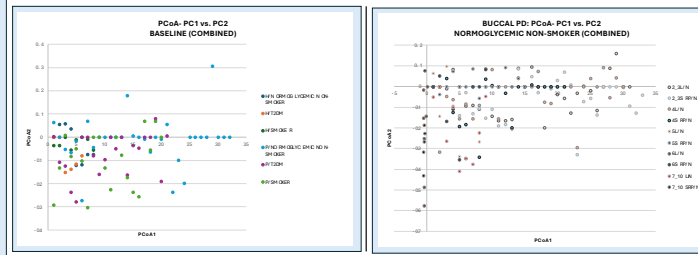


Figure 4: Beta-diversity analysis comparing healthy sites to diseased sites (referred to as "P") within the baseline group.

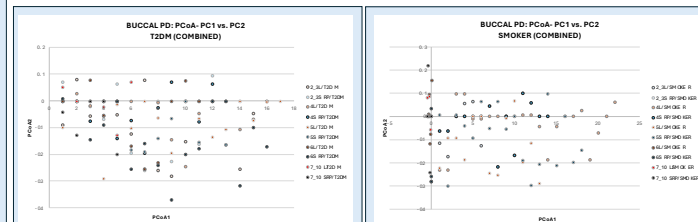


Figure 5: Beta-diversity analysis comparing buccal pocket depth across normoglycemic non-smokers, T2DM patients, and smokers.

- Porphyromonas
- Treponema
- Tannerella
- Fretibacterium
- Peptidiphaga
- Fusobacterium
- Prevotella
- Campylobacter
- Streptococcus
- Veillonella
- Actinomyces
- Capnocytophaga
- Eubacterium
- Rothia
- Corynebacterium
- Leptotrichia
- Neisseria
- Haemophilus
- Saccharibacteria, [FM2]-[G]-1
- Saccharibacteria, [FM2]-[G]-2
- Streptococcus
- Aggregatibacter
- Pseudoleptotrichia
- Akkermansia
- Veillonella
- Lautropia
- Cardobacterium
- Peptidiphaga
- Granulicatella
- Parvimonas
- Lachnospiraceae, [G]-1
- Gemmatimonadetes
- Gemmatimonadetes, [G]-2
- Ordiwalia
- Bacteroidetes, [G]-5
- Cabotella
- Saccharibacteria, [FM2]-[G]-5
- Bacteroidetes, [G]-3
- Stomatobacterium
- Peptostreptococcaceae, [G]-7
- Orbacterium
- Lachnospiraceae, [G]-3
- Bifidobacteria
- Abiotrophia
- Peptostreptococcaceae, [G]-6
- Absconditibacteria, [SR1]-[G]-1
- Erythrobacter
- Gramlibacteria, [GM2]-[G]-1
- Peptostreptococcus
- Lanceselabellia
- Veillonellaceae, [G]-1
- Weekseelaceae, [G]-1
- Ruminococcaceae, [G]-1
- Johnsonella
- Peptococcus
- Lachnospiraceae, [G]-8
- Solobacterium
- Mycoplasma
- Schaeffia
- Olsenella
- Clostridiaceae, [F]-[G]-1
- Arcobacter
- Other

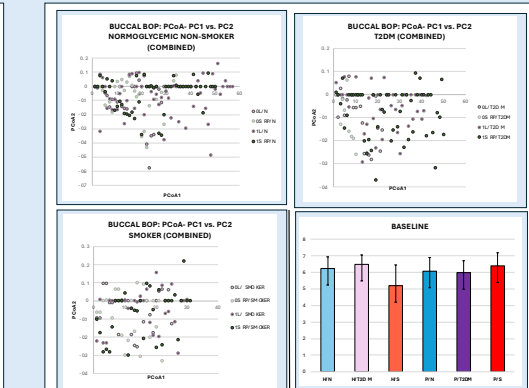


Figure 6: Beta-diversity analysis comparing buccal bleeding on probing across normoglycemic non-smokers, T2DM patients, and smokers.

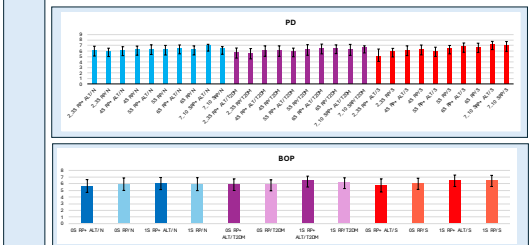


Figure 7: Alpha-diversity analysis comparing baseline, pocket depth, and bleeding on probing across normoglycemic non-smokers, T2DM patients, and smokers.

## Conclusion:

### Taxonomic Classification

**BOP:**

- Adjunctive Laser Therapy significantly reduces harmful genera in patients presenting with risk factors for periodontitis (diabetes or smoking) compared to patients without, suggesting better bacterial control in high-risk groups, especially diabetics.

### PD:

- Adjunctive Laser Therapy is more effective in deeper pockets (7-10 mm), particularly in reducing *Prevotella* in diabetic patients.
- In 5-6 mm pockets, laser therapy shows greater effectiveness against *Peptidiphaga* and *Prevotella* compared to SRP alone.

### Beta-Diversity

- **Baseline (Combined Group):**
  - Significant beta diversity observed across groups (normoglycemic non-smokers, T2DM patients, and smokers), with distinct outliers and variations indicating differences in microbial community composition.
- **Normoglycemic Non-Smokers (Buccal PD):**
  - Beta diversity is relatively lower, with clusters showing less variation, particularly in lower pocket depths. However, deeper pockets (7-10) demonstrate some distinct separation.
- **T2DM Patients (Buccal PD):**
  - Increased beta diversity compared to normoglycemic non-smokers, with more spread and less overlap among clusters, especially in deeper pocket depths (7-10).
- **Smokers (Buccal PD):**
  - Highest beta diversity observed, with distinct clustering and greater variation in both lower and deeper pocket depths, particularly in those treated with SRP.
- **Normoglycemic Non-Smokers (BOP):**
  - Moderate beta diversity with some distinct clusters, especially in BOP-positive sites.
- **T2DM Patients (BOP):**
  - Higher beta diversity with more spread-out clusters, indicating greater microbial variability related to BOP.
- **Smokers (BOP):**
  - High beta diversity with distinct clustering, showing significant microbial differences between BOP positive and negative sites.

### Alpha-Diversity

- **Baseline:**
  - Alpha diversity is generally consistent, slightly higher in HT2DM and PS groups.
- **PD:**
  - Alpha diversity decreases with deeper pockets, which is higher in smokers and T2DM patients with SRP + ALT treatment.
- **BOP:**
  - Higher alpha diversity in BOP-positive groups, particularly in T2DM and smokers, with SRP + ALT showing slight increases.

### Future Research:

- Generate a Microbiome Index
- Increased sample size for risk groups: Diabetes and Smokers
- Longitudinal analysis focusing on a Diabetic-Smoker risk group

### Acknowledgements:

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