

Original Research

Comparative Evaluation of Chlorhexidine, Diode Laser, and Photodynamic Therapy as Adjuncts to Scaling and Root Planing in Orthodontic Patients with Gingival Enlargement: A Split-Mouth Clinical and Microbiological Study

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INTRODUCTION

Orthodontia and Periodontia possess a symbiotic interrelationship. Orthodontic mechano-therapeutic treatment induces local irritation, increases plaque retention, alters the oral microflora^{1,2} causing localized or generalized inflammatory gingival tissue reactions^{3,4} and tissue growth.^{5,6} Gingival enlargement results in the formation of pseudo pockets which act as a reservoir for bacterial colonization, mainly the *Aggregatibacter actinomycetemcomitans* (Aa) and Black pigmented Red Complex bacteria. (*P. gingivalis*, *T. denticola*, *T. forsythia*)⁷ (BPB). The pseudo pockets present no evident clinical attachment loss but resolve on treating the gingival hyperplasia. If left untreated, this condition may further contribute to the development of periodontal disease.⁸ The treatment strategies include various non-surgical and surgical periodontal therapies. Scaling and root planing (SRP) is considered the “gold standard non-surgical therapy” for treating periodontal diseases.¹⁰ Clinically, SRP decreases probing pocket depth and enables attachment gain.¹¹ However, unsatisfactory outcomes arise due to poor access to the base of deep periodontal pockets in patients with fixed orthodontic appliances. Adjunctive treatment strategies using antimicrobial irrigation in conjunction with SRP have evolved based on the assumption that bacteria left behind during mechanical debridement could be

eliminated by application of an antimicrobial solution deep into the pocket.¹²

Low-level laser therapy (LLLT) has both localised and generalised anti-inflammatory effects.¹³ LLLT modulates the inflammatory processes triggered during orthodontic treatment, maintaining periodontal health.¹⁴

Photodynamic therapy (PDT) uses photosensitizing chemicals with light of an appropriate wavelength. PDT has cytotoxic effects on microorganisms by liberating free radicals and singlet oxygen species, thereby reducing the inflammation.¹⁵

Although there are various modalities for the treatment of gingival inflammation and enlargement associated with orthodontic therapy, literature provides limited evidence for comparing the clinico-microbiological efficacy of 0.2% Chlorhexidine subgingival irrigation, Low-level laser therapy (LLLT), and Photodynamic therapy (PDT) for the treatment of gingival inflammation or enlargement due to orthodontic treatment. This study assessed the outcome of all four modalities and compared their treatment efficacy.

OBJECTIVES

The purpose of this study is to find out how well the following treatment methods work in managing gingival enlargement in Orthodontic patients:

- SRP alone.
- SRP + 0.2% Chlorhexidine.
- SRP + Diode Laser.
- SRP+ Photodynamic Therapy using Indocyanine green as a photosensitizer.
- To compare the results of the above four groups.

MATERIALS & METHOD

GENESIS & PROCUREMENT OF DATA - Out of total 70 patients, 20 patients aged 14 to 27 were randomly chosen (11 men and 9 women). The patients had gingival inflammation and enlargement while they were getting fixed orthodontic treatments. Patients were selected and quadrants were randomly split into four groups. The study was described and written permission was asked beforehand. In this study, four groups of patients were given different treatments: scaling and root planing (SRP), SRP plus 0.2% chlorhexidine subgingival irrigation, SRP plus laser therapy with a Diode Laser at 810nm wavelength, and SRP plus photodynamic therapy (PDT) with indocyanine green photosensitizer once a month for six months. At the beginning, three months and six months later, clinical and microbiological parameters were checked on all cases. Indices included were Gingival Index (GI), the Plaque Index (PI), the Sulcus Bleeding Index (SBI), the Probing Pocket Depth (PPD), and the Gingival Overgrowth Index (GOI).

Criteria for subject grouping

This was a split-mouth study design wherein four quadrants from each patient were randomly allotted to one of four groups.

- GROUP 1 - SRP alone
- GROUP 2 - SRP + 0.2% Chlorhexidine Mouthwash (HEXIDINE®0.2% Chlorhexidine Mouthwash B.P.)
- GROUP 3 - SRP + LASER (Diode Laser, 810nm, AMD Picasso®)
- GROUP 4 -SRP + PDT (Indocyanine green-Aurogreen®, Aurolab, Madurai, India) The patients were recalled for follow-up, up to six months at one-month intervals.

Inclusion criteria

1. Patients aged between 14 and 27 years.
2. Patient undergoing orthodontic treatment with gingival inflammation and enlargement in all four quadrants.
3. Patients without any systemic illness.
4. Patients without any history of known drug allergy, especially to indocyanine green.
5. Non-smokers.
6. A patient with a gingival overgrowth index ≥ 1 in each quadrant.

Exclusion criteria

1. Patients on drugs that induce gingival enlargement, e.g., calcium channel blockers, anticonvulsants, or immunosuppressants.
2. Patients with lingual orthodontic appliances.
3. Pregnant or lactating women.
4. Female patients on hormonal therapy.
5. Patients who are not competent to give consent.
6. Relapsed periodontal surgery patients who do not follow proper oral hygiene practices.

Treatment method

Scaling And Root Planing –

After a thorough clinical examination, all participants received detailed instructions about oral hygiene and underwent the following conventional therapy:

- Dental plaque and supragingival calculus removal – Scaling

- Subgingival Calculus removal – Root planing Subgingival Irrigation With 0.2% Chlorhexidine –

Isolating and drying one quadrant with air blow and cotton rolls the mesiobuccal, distobuccal, mesiolingual and distolingual areas of the teeth were irrigated with a solution of 0.2% Chlorhexidine digluconate (HEXIDINE®, 0.2% Chlorhexidine Mouthwash BP) as part of the treatment. Each of the four sites had subgingival irrigation for five seconds at a flow rate of twenty millilitres per minute using a five-millilitre clean syringe with a blunt tip.

Laser Therapy –

One of the quadrants got low-level laser treatment after regular therapy. Therapy with laser light was done once a month. It was used to shine an 810 nm AMD Picasso diode LASER on the soft tissue wall of the periodontal pocket. The laser had a continuous wave output power 0.1W and a 300µm optical fibre tip. Irradiation was done in touch mode to remove the pocket epithelium around the teeth. To keep damage to a minimum, the tip was moved in a sweeping zigzag pattern from the tip to the crown at a steady speed of 1 mm/s. The fibre end was checked for a carbonized hot tip at each treatment.

Photodynamic Therapy –

The photodynamic therapy was applied using 810 nm diode lasers at a power of 0.1W in continuous mode with 1mg/ml of Indocyanine green (ICG) as a photosensitizer. [Aurogreen®, Aurolab, Madurai, India]. Freshly prepared photosensitizer was prepared by adding 25 mg sterile lyophilized powder with 5 ml sterile water provided by the manufacturer, resulting in a 0.5% solution. The solution was filtered using a 0.2-micron syringe filter and drawn into a sterile syringe. Periodontal pockets were filled with the dye for 3 minutes from the bottom using a blunt cannula. Areas were isolated with cotton rolls and high vacuum suction, and excess photosensitizer was removed by distilled water irrigation before the therapy.

Clinical Parameters Used:

1. Plaque index (PI) - Sillness and Loe in (1964)
2. Gingival index (GI) - Loe and Sillness J (1963)

3. Sulcus Bleeding Index (SBI) - Muhlemann and Sons (1971)
4. Probing Pocket depth (PPD) - measured using a UNC-15 probe from the gingival margin to the pocket base.
5. Gingival overgrowth index (GOI) - Angelopoulos & Goaz (1972)

Plaque sampling for microbial analysis: Post superficial scaling the area was separated and plaque samples were taken from the deepest spots with clean universal curettes (Hu-Friedy, USA). The samples were put in sterile jars with 0.5 ml of Reduced Transport Fluid (RTF) and sent to the lab at first month, third month and sixth month.

Bacterial culturing: Media for *Aa* Tryptic-Soy-Bacitracin-Vancomycinagar was made with tryptic soy agar and 10% serum. Yeast extracts (1 mg/ml), bacitracin (75 µg/ml), and vancomycin (5 µg/ml) were also added. To culture anaerobic BPBs, blood agar base with 5% rabbit blood, 5µg/ml Haemin, 0.5µg/ml Menadione and 40µg/ml Kanamycin was mixed to make the medium. Bacteria with black pigments, like *Porphyromonas gingivalis* and *Prevotella intermedia*, were found on blood agar as black colonies with β-hemolysis. *Aa* was found on TSBV agar as small, clear, white colonies.

Calculated amount of distilled water was mixed with the exact amount of each component of growth medium and then poured into petri dishes in a chamber with laminar air flow. The samples were ultrasonically mixed for one minute and poured on culture plates. The plates were then left to cool down. They were kept at 37°C for 72 hours in an anaerobic jar in a 5% CO₂ atmosphere.

RESULTS

Statistical analysis

It was coded and put into SPSS software (Version 23) for statistical analysis. Mean and standard deviation were examples of descriptive statistics. One-way ANOVA and Post hoc Tukey's test were used for inferential statistics. At a 95% confidence level, the significance level was set at 0.05.

Inter-group comparison

Plaque index –Inter-group comparison shows a reduction in PI values in all the groups compared to baseline, yet the values are not statistically significant.

Gingival index- Inter-group comparison shows significant changes from baseline to 3rd and 6th months.

Probing pocket depth – Inter-group comparison shows non-significant statistical changes between the treatment groups at the baseline. Moreover, in the 3rd month, group-4 showed statistically significant changes with a mean value of 0.36500±0.11298. and in the 6th month, group-3 and group-4 showed high

significance with a mean difference of 0.37500±0.10523, 0.59000±0.10523, respectively.

Sulcus Bleeding Index– Inter-group comparison shows non-significant statistical changes in the baseline between the treatment groups. In the 3rd and 6th month groups, group-3 & group-4 showed statistically significant changes with a mean value of 0.33000±0.28754, 0.62000±0.28754 & 0.29000±0.27708, and 0.73000±0.27708, respectively.

Gingival overgrowth index- Inter-group comparison shows statistically non-significant changes between the treatment groups in the baseline and 3rd month. In the 6th month, group 4 showed statistically significant changes with a mean value of 0.42000±0.14622.

Microbial CFU - A decrease in the microbial CFU from sites of group 1 to group 4, with mean differences of 22.34>40.2>50.13>64.33, was observed for *Aa*. Similarly, a decrease in the microbial CFU for BPB in samples from sites of group 1 to group 4 had mean differences of 28.47>35.06>44.67>47.87. This showed the increasing benefits of simple laser irradiation as an adjunct to scaling and root planing and the effectiveness of PDT over SRP and Laser irradiation.

Intra-Group Comparison

Plaque index: When PI readings are compared within groups, they all show statistically significant reduction compared to the baseline.

Gingival index: Study shows significant changes from the beginning to the third and sixth months. Upon observation there aren't any significant changes in the initial measurements. Group 4 showed statistically significant changes in the third month, with a mean value of 0.36500±0.11298. In the sixth month, there was a significant difference between groups 3 and 4, with a mean difference of 0.37500±0.10523 for group 3 and 0.59000±0.10523 for group 4.

Probing pocket depth- Statistically significant reduction noted in all comparative periods i.e. from baseline to third month and sixth month.

Sulcus Bleeding Index - When compared across groups, there were no statistically significant differences in the starting values for the treatment groups. In the third and sixth months, there were statistically significant changes in groups 3 and 4. Their mean values were 0.33000±0.28754, 0.62000±0.28754, and 0.29000±0.27708, 0.73000±0.27708, respectively.

The gingival overgrowth index- Inter-group comparison shows no statistically significant differences between the treatment groups at the start and after 3 months. A mean value of 0.42000±0.14622 was seen in group 4 in the sixth month, a statistically significant change.

Microbial CFU- Average changes in the number of from sites in group 1 to group 4 were found to be 22.34, 40.2, 50.13, and 64.33 for *Aa*. In the same way, the number of bacteria CFU for BPB decreased

in samples from sites in groups 1 through 4. The average differences were 28.47, 35.06, 44.67, and 47.87. This showed that simple laser irradiation

plusSRP is beneficial and PDT works better than SRP and laser irradiation.

Table I- Intergroup comparison

Parameters	Intervals	Site - 1	Site - 2	Site - 3	Site - 4	Diff b/w 1&2	Diff b/w 2&3	Diff b/w 3&4	Diff b/w 1&3	Diff b/w 1&4	Diff b/w 3&4
Plaque index (PI)	Baseline	1.9800	1.9600	1.8300	1.8900	0.020	0.130	0.060	0.150	0.090	0.060
	3 rd Month	1.5450	1.5400	1.3950	1.3650	0.005	0.145	0.030	0.150	0.18	0.030
	6 th Month	1.2400	1.2750	1.1100	1.0550	0.035	0.165	0.055	0.130	0.185	0.055
Gingival index (GI)	Baseline	2.0850	2.1350	2.4150	2.5250	0.050	0.280	0.110	0.330	0.440	0.110
	3 rd month	1.8750	1.7450	1.7800	1.5450	0.130	0.200	0.235	0.095	0.330	0.235
	6 th Month	1.5130	1.3800	1.3000	1.1250	0.133	0.080	0.175	0.213	0.388	0.175
Sulcus Bleeding Index (SBI)	Baseline	2.8430	2.8330	3.0650	3.0500	0.010	0.232	0.015	0.222	0.207	0.015
	3 rd month	2.4350	2.3500	2.3050	2.1450	0.085	0.045	0.16	0.130	0.29	0.160
	6 th Month	1.9800	1.8850	1.7250	1.4200	0.095	0.160	0.305	0.255	0.56	0.305
Probing Pocket depth (PPD)	Baseline	4.5850	4.3350	4.4550	4.4950	0.250	0.120	0.040	0.130	0.090	0.040
	3 rd month	4.1250	3.9600	3.8600	3.7600	0.165	0.100	0.100	0.265	0.365	0.100
	6 th Month	3.8900	3.7800	3.5150	3.3000	0.110	0.265	0.215	0.375	0.590	0.215
Gingival overgrowth index (GOi)	Baseline	1.6350	1.6400	1.7900	1.8150	0.005	0.150	0.025	0.155	0.180	0.025
	3 rd month	1.5600	1.5450	1.4800	1.3300	0.015	0.065	0.150	0.080	0.230	0.150
	6 th Month	1.2550	1.2150	1.0950	1.8350	0.040	0.120	0.740	0.160	0.580	0.740
BPB CFU	Baseline	132.67±15.81	128.13±10.89	133.27±11.48	127.27±11.82	4.540	5.140	6.000	-0.600	5.400	6.000
	3 rd month	110.93±13.49	105.73±7.70	106.93±8.79	99.93±10.38	5.200	1.200	7.000	4.000	11.000	7.000
	6 th Month	104.20±12.81	93.07±5.85	88.60±7.73	79.40±10.34	11.13	4.470	9.200	15.600	24.800	9.200
AA CFU	Baseline	143.07±14.85	142.87±10.78	144.53±11.82	146.00±11.63	0.200	1.660	1.470	1.460	2.930	1.470
	3 rd month	123.60±12.09	116.27±8.50	115.87±9.12	105.13±10.02	7.330	0.400	10.740	7.730	18.470	10.740
	6 th Month	120.73±8.52	102.67±10.50	94.40±8.79	81.67±9.58	18.060	8.270	12.730	26.330	39.060	12.730

Intergroup comparison showed statistically significant reduction of all clinical & microbiological parameters observed across all groups from baseline to 3rd month and 6th month.

Table II - Intragroup comparison

Parameters	Area	Baseline	Third Month	Sixth Month	F	Significance
Plaque index (PI)	Site 1	1.9800	1.5450	1.2400	10.491	0.000 (H.S)
	Site 2	1.9600	1.5400	1.2750	8.656	0.000 (H.S)
	Site 3	1.8350	1.3950	1.1100	13.962	0.000 (H.S)
	Site 4	1.8900	1.3650	1.3650	21.950	0.000 (H.S)

Gingival index (GI)	Site 1	2.0850	1.8750	1.5130	6.929	0.002 (H.S)
	Site 2	2.1350	1.7450	1.3800	14.272	0.000 (H.S)
	Site 3	2.4150	1.7800	1.3000	30.801	0.000 (H.S)
	Site 4	2.5250	1.5450	1.1250	45.952	0.000 (H.S)
Sulcus Bleeding Index (SBI)	Site 1	2.8430	2.4350	1.9800	3.836	0.027 (S)
	Site 2	2.8330	2.3500	1.8850	4.946	0.010 (S)
	Site 3	3.0650	2.3050	1.7250	10.474	0.000 (H.S)
	Site 4	3.0500	2.1450	1.4200	18.557	0.000 (H.S)
Probing Pocket depth (PPD)	Site 1	4.5850	4.1250	3.8900	16.042	0.000 (H.S)
	Site 2	4.3350	3.9600	3.7800	7.391	0.001 (H.S)
	Site 3	4.4550	3.8600	3.5150	41.689	0.000 (H.S)
	Site 4	4.4950	3.7600	3.3000	72.439	0.000 (H.S)
Gingival overgrowth index (GOi)	Site 1	1.6350	1.5600	1.2550	3.124	0.052 (N.S)
	Site 2	1.6400	1.5450	1.2150	4.176	0.020 (S)
	Site 3	1.7900	1.4800	1.0950	9.914	0.000 (H.S)
	Site 4	1.8150	1.3300	1.8350	19.618	0.000 (H.S)
BPB CFU	Site 1	133.67±15.81	111.93±13.49	105.20±12.81	22.58	0.050 (NS)
	Site 2	129.13±10.89	106.73±7.70	94.07±5.85	83.93	0.020 (S)
	Site 3	133.27±11.48	106.93±8.79	88.60±7.73	109.07	0.000 (H.S)
	Site 4	128.27±11.82	99.94±10.38	80.40±10.34	98.19	0.000 (H.S)
AA CFU	Site 1	143.07±14.85	126.67±12.09	120.35±8.52	1.65	0.051 (NS)
	Site 2	142.87 ±10.78	116.27±8.50	102.67±10.50	1.106	0.021 (S)
	Site 3	144.53±11.82	115.87±9.12	94.40±8.79	1.523	0.000 (HS)
	Site 4	146.00±11.63	105.13±10.02	81.67±9.58	1.303	0.000 (HS)

Intragroup comparison depicts statistically significant reduction of all clinical & microbiological parameters across all groups from baseline to 3rd month and 6th month.



Intra-oral pre-operative view



Recording initial probing pocket depth. Note the presence of BOP



Application of 0.2% Chlorhexidine mouthwash into periodontal pockets



Diode LASER application into periodontal pocket



Photosensitizer Indocyanin green inserted into periodontal pocket



Photodynamic therapy performed



Reduction of BOP & probing pocket depth after 3rd month.



Restoration of gingival health post debonding & SRP at 6th month.



Culture media sample showing *Aggregatibacter actinomycetemcomitans* colony



Culture media sample showing *Black pigmented Bacteria* colony

DISCUSSION

This study compared the clinical & microbiological efficacy of 0.2% Chlorhexidine, Diode laser, and Photodynamic therapy as an adjunct to SRP in treating gingival inflammation and enlargement in Orthodontic patients. Twenty subjects between 14 and 27 years were selected according to the inclusion criteria. All treatment modalities were performed every month for 6 months. Clinical parameters and subgingival plaque sampling for BPB and Aawere evaluated at baseline, 3rd month, and 6th month. Clinical parameters recorded were PI, GI, SBI, PPD & GOI. The study showed a statistically significant reduction of all clinical parameters for all groups. Scaling & root planing stabilizes most cases of gingival and periodontal diseases. This coincides with a study by Novokovic et al.¹⁸, where meticulous SRP reduces inflammation by reducing bacterial burden. The reduction of PPD might have resulted from complete re-epithelialization and healing by long junctional epithelium. Group 2 (SRP+CHX) showed significant changes from baseline to 6 months in all the clinical parameters. This is as per a study by SoH et al.¹⁹ which showed a significant reduction in PI, SBI, GI, and PPD in the CHX group (mean 1.2 mm). The most significant reductions in (supragingival) plaque and periodontal inflammation were initiated within one week of irrigation and continued until the treatment period's completion. The SRP+LASER group has shown a significant reduction in all clinical parameters from baseline to the 6th month, which was statistically significant. This is by the study by Mărtu et al,²⁰ where LLLT slowed down periodontal inflammation by lowering BOP, PD scores, and bacterial load. It has a bactericidal effect on periopathogenic bacteria, modulates the pain of orthodontic activation and causes faster recovery of the initial periodontal status. LLLT was combined with PDT as a synergistic treatment modality, in a study by Lui et al²¹, as PDT generates toxic free radicals that destroy localized microbial entities. To our knowledge, no studies have investigated all four treatment modalities (SRP, CHX, PDT, and LLLT) clinically and microbiologic cultures on long-term basis and nonetheless, their efficacy in treating the gingival inflammation of orthodontic patients. In this study, inter-group comparison in GI projected a statistically significant difference between Group-1 versus Group-3 and Group-4 without any significant change between Group-1 versus Group-2. This indicates that adjunctive use of PDT and LASER is better than SRP alone and SRP with CHX in reducing PI, GI, SBI, PPD, and GOI scores. These results stand parallel to several studies; however, with different methodologies, by Malgikar et al²² and R. Attin et al²³, whereby CHX, low-level lasers, and PDT have reduced PI, PD, GI, and SBI with resultant reduction of gingival inflammation. Qadri et al²⁴ found in their study that additional treatment with low-level lasers reduced inflammation of the periodontium and

probing pocket depth which is as per our study. In our study, from baseline to 6 months, there is a significant improvement in all the clinical parameters in all the groups. Still, when comparing the differences between the groups, the results were significant only with the PDT group. In the case of gingival enlargement or overgrowth from baseline to the 6th month, all three groups showed significant reductions, except group 1. Whereas comparing the groups, the SRP + PDT group showed a highly significant reduction in the gingival overgrowth. Irrespective of the results, a few studies state that SRP + PDT has no added advantage over SRP.^{25,26} These variations in results are challenging to interpret due to heterogeneity in study designs with various types of photosensitizers and different wavelengths of laser used. These results provide new data about repeated applications of PDT as highlighted by Gómez et al²⁷ which are essential in monitoring gingival health status in the orthodontic patient population. In the present study, the repeated treatment with SRP, CHX subgingival irrigation, LASER therapy, and PDT showed a remarkable positive effect over time.

CONCLUSION

Within limitations, the present short clinical & microbiological trial suggests that all four treatment strategies (SRP alone, SRP + 0.2% CHX, SRP + LASER & SRP + PDT) benefit patients under orthodontic treatment with gingival enlargement. Combining multiple applications of subgingival irrigation with 0.2% Chlorhexidine, LASER Therapy, and PDT with 810 nm Diode laser and ICG dye is more beneficial than SRP. Even though all modalities showed promising results, multiple applications of PDT showed better improvement in all clinical and microbiological parameters, followed by LASER therapy, 0.2% Chlorhexidine subgingival irrigation, and SRP. Further long-term longitudinal studies using different dyes and methodologies are necessary to elucidate their beneficial effects compared to conventional methods.

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