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Comparative Evaluation of Anesthetic and Antimicrobial efficacy of 10% Propolis hydrogel with Scaling and Root planing in Generalized Chronic Periodontitis - A Clinico-Microbiological, Split Mouth Study

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Abstract

Background: Chronic periodontitis is a common inflammatory disease leading to the destruction of periodontal tissues. While scaling and root planing (SRP) remains the cornerstone of treatment, adjunctive agents can enhance therapeutic outcomes. Propolis, a natural bioactive compound, possesses significant antimicrobial and anesthetic properties. This split-mouth study was conducted to evaluate the efficacy of 10% Propolis hydrogel as an adjunct to SRP in improving clinical parameters and reducing microbial load in generalized chronic periodontitis. Methodology: 15 patients with generalized chronic periodontitis having pocket depth ≥ 4mm were selected. Maxillary and mandibular arch of each patient was randomly divided to receive either of Placebo gel (Group A) or 10% Propolis Hydrogel (Group B) followed by SRP and application of gel in periodontal pockets. Clinical parameters, VAS score were assessed and quantitative analysis of sub-gingival plaque samples for aerobic and anaerobic bacterial growths using colony count method at baseline and after 3 months was performed. Results: Intragroup comparison shows statistically significant results in all the clinical and microbiological parameters. Intergroup comparison showed statistically significant amount of reduction in pockets and Colony Forming Unit (CFU/ml) of aerobic as well as anaerobic bacteria in Group B when compared to Group A. Significant results were also found in Group B in terms of VAS score (assessing dentinal sensitivity) in both the groups. Conclusion: The adjunctive use of 10% propolis hydrogel with SRP provides enhanced anesthetic and antimicrobial benefits in the treatment of generalized chronic periodontitis, suggesting its potential as a valuable adjunct to mechanical therapy.

Keywords: Propolis, Local drug delivery, Chronic periodontitis, Anesthetic action, Colony forming units.

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INTRODUCTION

Periodontitis is an inflammatory disease that targets the supporting tissues of the teeth, initiated by specific microorganisms or complex microbial communities. It leads to the progressive destruction of the periodontal ligament and alveolar bone, frequently resulting in pocket formation or gum recession. The primary objective of Phase I periodontal therapy is to minimize tooth-associated biofilms and their toxic by-products such as endotoxins, antigens, enzymes, and other irritants that contribute to tissue inflammation and damage [1,2].

A crucial aspect of managing periodontal disease involves eliminating plaque biofilm through Scaling and Root Planing (SRP). While mechanical therapy effectively decreases the subgingival microbial load, it cannot completely eliminate all microorganisms residing deep within the connective tissues- these persistent pathogens are primarily responsible for ongoing tissue damage [3]. Systemic antibiotics are used to reduce the population of subgingival bacteria; however, their use is often associated with various side effects [4].

Various local drug delivery systems are used in the treatment of periodontal diseases, allowing direct application within the oral cavity. These systems include fibers, strips, films, microparticles, gels, and nanoparticles, all formulated to deliver antimicrobial agents precisely to the affected sites. However, each of these agents can have side effects. Research is ongoing into the use of natural ingredients to mitigate these adverse effects [5].

During SRP, patients often complain of hypersensitivity, characterized by a short, sharp pain in response to external stimuli. Hence, adjunctive therapy should be explored to enhance the treatment outcomes and to

improve the patient comfort during the therapy. For decades, researchers have extensively investigated potential solutions for hypersensitivity, resulting in the introduction of numerous desensitizing agents along the way. Furthermore, fluorides, chewing gums, lasers, dentin adhesive sealers, and protein precipitators have also been tested; however, none of these options has provided complete relief from hypersensitivity [6,7].

Recently, special attention has been provided to natural therapeutic agents. Propolis is a natural resinous compound gathered by honeybees from different plant sources. It contains a complex mixture of polyphenols, flavonoids, and other bioactive compounds that exhibit antimicrobial, anti-inflammatory, anesthetic and tissue reparative quality. It make propolis a promising adjunct in non-surgical periodontal therapy, by reducing microbial colonization and modulating pain responses[8]. Propolis has also demonstrated effectiveness in reducing dentin hypersensitivity, as evidenced by findings from multiple in vitro and in vivo studies [9,10].

To our knowledge, role of Propolis in treating sensitivity during SRP along with its antimicrobial nature as local drug delivery agent have been sparsely investigated. So, the aim of this study was to evaluate and compare the Anesthetic and Antimicrobial efficacy of 10% propolis hydrogel along with SRP in patients with Generalized Chronic Periodontitis.

MATERIALS AND METHODS

This split-mouth, randomized, parallel-group, placebo-controlled clinical trial was conducted on patients at the Outpatient Department of Periodontology and Oral Implantology, I.T.S Centre for Dental Studies and Research, Muradnagar, Ghaziabad, U.P. Ethical approval was obtained from the Institutional Ethics Committee (Protocol No. ITSCDSR/IIEC/RP/2024/003), and the study was registered with the Clinical Trial Registry of India (CTRI Registration No. CTRI/2024/06/069686). The trial was carried out in accordance with the Declaration of Helsinki and all relevant ethical guidelines.

A total of 15 patients with age group ranging from 25 to 60 years enrolled in this study. The maxillary and mandibular arch of each patient was randomly divided to receive either of Placebo gel (Group A) or 10% Propolis Hydrogel (Group B) followed by SRP and application of gel in periodontal pockets. Systemically healthy Patients with Generalized chronic periodontitis with probing depth ≥4 mm, providing positive informed consent and good compliance were included in this study. Exclusion criteria comprised Patients with dental caries, undergoing orthodontic treatment, history of periodontal treatment, using desensitizing toothpaste and those who are allergic to honey bee products.

STANDARDIZED PARAMETERS-

Clinical Parameters- Plaque index (PI), Gingival Index (GI), Pocket probing depth (PPD) and Clinical attachment level (CAL) at Baseline and after 3 months in both the groups. Visual Analog Scale (VAS) Score for assessing dentinal sensititvity on the same day between the groups.

Microbiological Parameters- Quantitative assessment of subgingival plaque samples for aerobic and anaerobic bacterial growth was performed by measuring Colony Forming Units (CFU/ml) at baseline and after 3 months in both groups.

10%PROPOLIS HYDROGEL PREPARATION

The procedure was carried out at ITS college of Pharmacy, Ghaziabad. Wax based raw Indian Propolis (Figure 1) was purchased from bee keeping house, Department of Horticulture, Aligarh (UP).

FOR 10% PROPOLIS EXTRACT

According to *Krell et al* [11], it was created after mixing 10gm of propolis (in powder form) in 90 ml of ethanol. The mixture was left in dark and periodically mixed every day for 2 weeks. It was then filtered using Whatman filter paper (Figure 2).

FOR 10% PROPOLIS HYDROGEL

Potassium sorbate was first dissolved in purified water at 50°C. Carbopol 940 and Sodium CMC were weighed, dispersed in purified water at the same temperature, and mixed thoroughly. The propolis extract, dispersed in PEG 400, was then added to the gel base, and the pH was neutralized by adding an appropriate amount of triethanolamine (Table 1). After formulation of gel, it was preserved in a dark, cool and dry place (Figure 3).

PROCEDURE

After a comprehensive case history and obtaining written informed consent, patients meeting all inclusion and exclusion criteria were enrolled in the study (Table 2). Before SRP, all clinical parameters (PI, GI, PPD, CAL) were recorded. Probing depth measurements at sites with PPD ≥ 4 mm were recorded using a UNC-15 periodontal probe (Figure- 4 & 8).

For microbiological sample collection, the sites were first superficially cleaned with cotton pellets and the supragingival area was dried using a stream of air. Subgingival plaque samples were then obtained from the deepest pocket using a sterilized gingival curette. Samples were aseptically placed in sterile Eppendorf tubes with Phosphate Buffer Saline (PBS) and stored at -20 °C for analysis.

***STEP-1 (Application of gel on tooth surface)* -** Placebo gel was applied in Group A (Figure 5) and in Group B, 10% Propolis gel was applied (Figure 6). 1 minute after application, full mouth SRP was done in all the groups. VAS score was assessed for Dentinal sensitivity on same day in Group A and Group B.

***STEP-2 (Local Delivery of gel in pockets)* -** The gel was administered using a blunt tip. The tip was carefully inserted to the base of the pocket, and the medication was slowly injected while gradually withdrawing the tip toward the gingival margin. Any excess gel was gently removed with sterile cotton (Figure 7).

Following the treatment, patients were instructed to maintain their regular oral hygiene practices. After 7 days, all patients were recalled and assessed for any adverse clinical signs. Follow-up visits were scheduled monthly. At the 3 month evaluation, clinical parameters (PI, GI, PPD, CAL) were assessed, and subgingival plaque samples were collected. All measurements at the designated sites were performed using a UNC-15 periodontal probe (Figure 4 & 8).

The bacterial load was assessed using a digital colony counter.

Statistical Analysis

The data were organized in an Excel spreadsheet and analyzed using the Statistical Package for the Social Sciences (SPSS 16, Inc., Chicago, IL, USA). Mean differences between groups were evaluated using the Independent t-test for normally distributed data and the Mann-Whitney U test for non-normally distributed data. Intra-group comparisons were performed using the Paired t-test for normally distributed data and the Wilcoxon signed-rank test for data that were not normally distributed.

The power analysis was done by Software G*Power version 3.1.9.7. The level of significance and confidence interval was 5% and 95% respectively, i.e. $p < 0.05$.

RESULTS

This literature review analyzes findings from 14 studies summarized in a The mean plaque index at baseline for Group A and Group B was 2.17 ± 0.29 and 2.20 ± 0.32, which significantly reduced to 1.14 ± 0.52 and 1.16 ± 0.37 after 3 months respectively. On intergroup comparison, the PI values showed statistically insignificant differences between two groups at baseline and 3 months (Table 3). The mean gingival index at baseline for Group A and Group B was 2.15 ± 0.35 and 2.10 ± 0.35, which significantly reduced to 1.34 ± 0.25 and 1.24 ± 0.42 after 3 months respectively. On intergroup comparison, the GI values were statistically insignificant between two groups at baseline and 3 months (Table 4). The mean pocket probing depth at baseline for Group A and Group B was 4.28 ± 0.10 and 4.34 ± 0.21, which significantly reduced to 2.90± 0.16 and 2.20 ± 0.29 after 3 months respectively. On intergroup comparison, Group B showed statistically significant reduction in PPD values when compared to Group A at 3 months (Table 5). The mean clinical attachment level at baseline for Group A and Group B was 4.28 ± 0.10 and 4.34 ± 0.21, which significantly reduced to 2.90± 0.16 and 2.20 ± 0.29 after 3 months respectively. On intergroup comparison, Group B showed statistically significant gain in CAL values when compared to Group A at 3 months (Table 6). On intergroup comparison, the mean VAS score for DH in Group B was 2.53±0.99 that was significantly less than that in Group A with mean score of 7.27± 1.22 (Table 7). The mean CFU/ml (×104) of aerobic bacteria at baseline for Group A and Group B was 186 ±176 and 227 ± 160, which significantly reduced to 9.81 ± 3.76 and 5.06 ± 1.75 after 3 months respectively. The mean CFU/ml (×104) of anaerobic bacteria cultured in anaerobic jar at baseline for Group A and Group B was 9.12 ± 6.83 and 10.8 ± 13.5, significantly reduced to 5.6 ± 4.43 and 2.2 ± 2.72 after 3 months respectively. On intergroup comparison, Group B showed statistically significant reduction in aerobic as well as anaerobic bacterial growth when compared to Group A at 3 months.(Figure 9 &10) (Table 8 & 9).

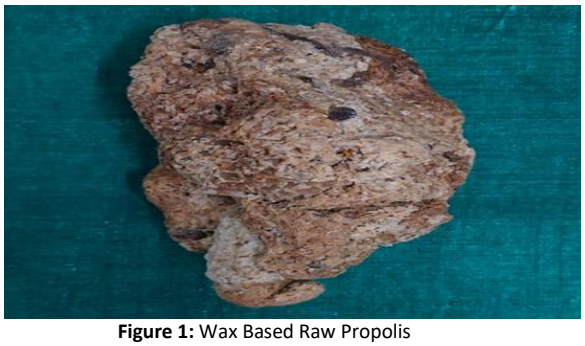

Figure 1: Wax Based Raw Propolis

Figure 2: 10% Propolis Extract

Figure 3: 10% Propolis Hydrogel

Figure 4: Pre and Post PPD in Group A

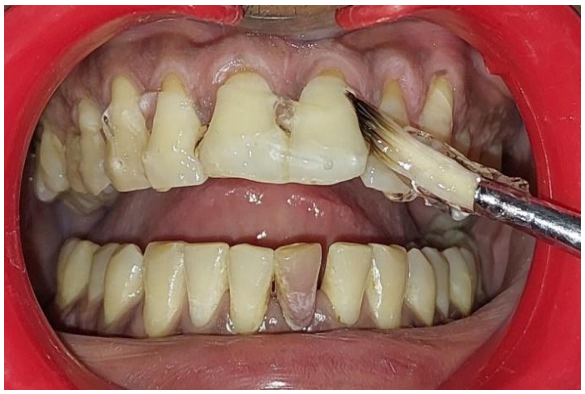

Figure 5: Application of Placebo gel

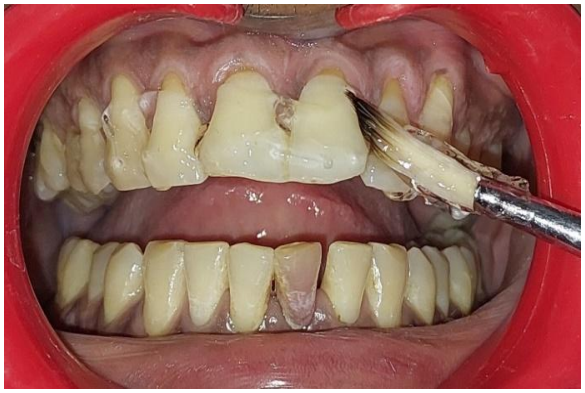

Figure 5: Application of Placebo gel

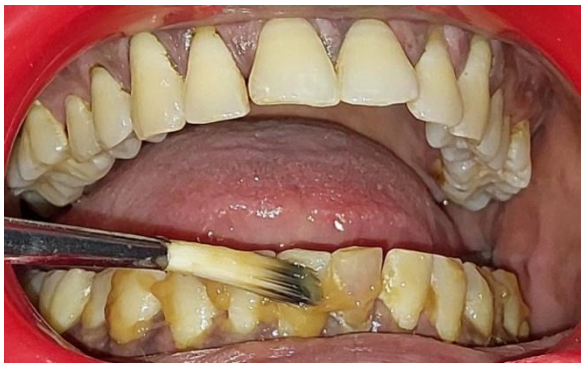

Figure 6: Application of Test (10% Propolis Hydrogel) gel

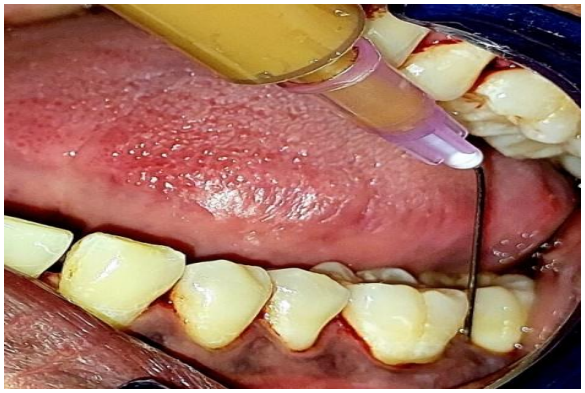

Figure 7: Local Delivery of 10% Propolis gel

Figure 8: Pre and Post PPD in Group B

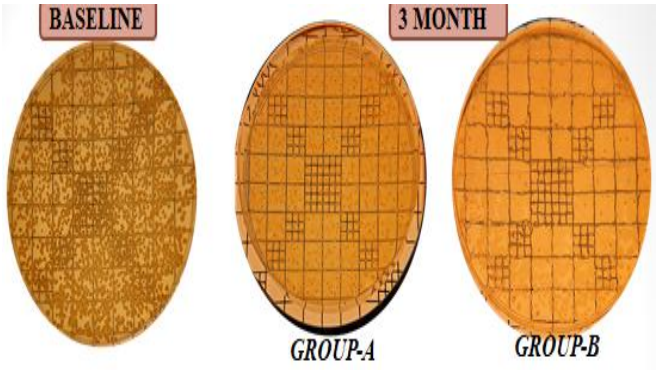

Figure 9: CFU/ml of Aerobic Bacteria

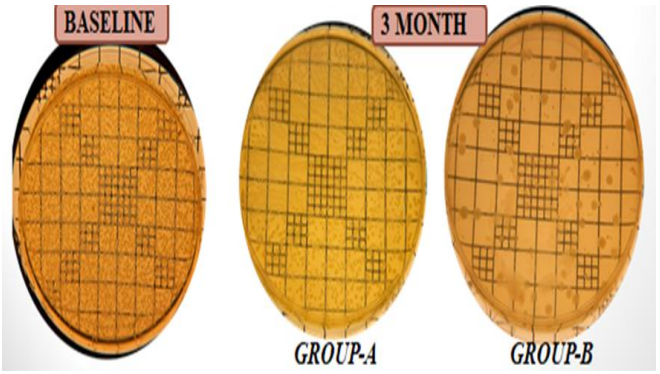

Figure 10: CFU/ml of Anaerobic Bacteria

Table 1: Composition of Propolis Hydrogel

INGREDIENTS	QUANTITIY
Carbopol 940	1gm
Sodium CMC	3gm
Potassium sorbate	0.2gm
Polyethylene glycol 400	13gm
Triethanolamine	3 drops
Distilled water	100ml

Table 2: CONSORT Flow Chart

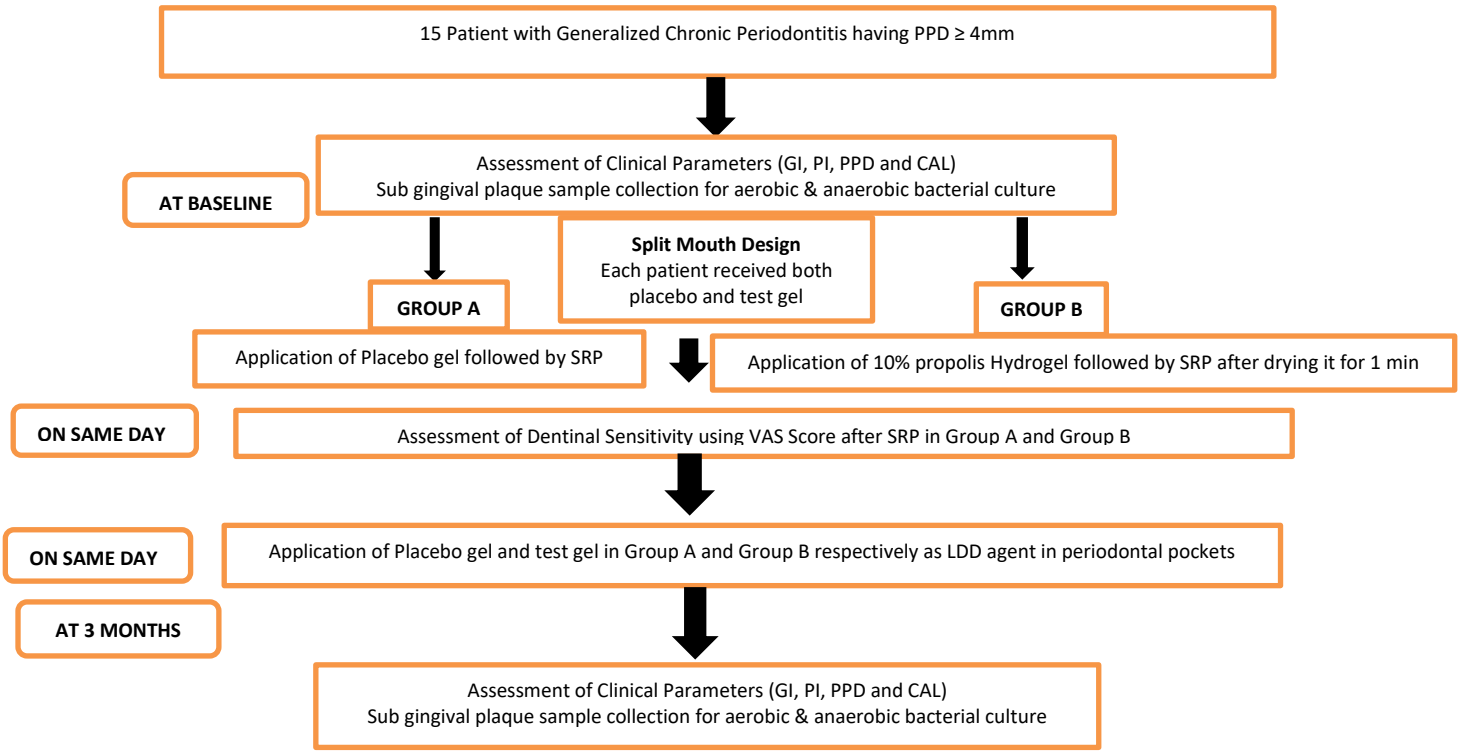

Table 3: Comparison of PLAQUE INDEX in Group A and B at baseline and after 3 months

VARIABLE		BASELINE		3 MONTH		p value (INTRA)
		MEAN	SD	MEAN	SD	
PLAQUE INDEX	*GROUP A*	*2.17*	*0.29*	*1.14*	*0.52*	*0.001**
	GROUP B	*2.20*	*0.32*	*1.16*	*0.37*	*0.001**
	p value (INTER)	*0.8*		*0.9*		

SD- Standard Deviation, P-Probability
*p Value is significant (<0.05)

Table 4: Comparison of GINGIVAL INDEX in Group A and B at baseline and after 3 months

VARIABLE		BASELINE		3 MONTH		p value (INTRA)
		MEAN	SD	MEAN	SD	
GINGIVAL INDEX	GROUP A	2.15	0.35	1.34	0.25	0.001*
	GROUP B	2.10	0.35	1.24	0.42	0.001*
	p value (INTER)	0.7		0.4		

SD- Standard Deviation, P-Probability
*p Value is significant (<0.05)

Table 5: Comparison of POCKET PROBING DEPTH in Group A and B at baseline and after 3 months

VARIABLE		BASELINE		3 MONTH		p value (INTRA)
		MEAN	SD	MEAN	SD	
POCKET PROBING DEPTH	GROUP A	4.28	0.10	2.90	0.16	0.001*
	GROUP B	4.34	0.21	2.20	0.29	0.001*
	p value (INTER)	0.3		0.001*		

SD- Standard Deviation, P-Probability
*p Value is significant (<0.05)

Table 6: Comparison of CLINICAL ATTACHMENT LEVEL in Group A and B at baseline and after 3 months

VARIABLE		BASELINE		3 MONTH		p value (INTRA)
		MEAN	SD	MEAN	SD	
CLINICAL ATTACHMENT LEVEL	GROUP A	4.28	0.10	2.90	0.16	0.001*
	GROUP B	4.34	0.21	2.20	0.29	0.001*
	p value (INTER)	0.3		0.001*		

SD- Standard Deviation, P-Probability
*p Value is significant (<0.05)

Table 7: Comparison of VAS SCORE for Dentinal Hypersensitivity in Group A and B immediately after treatment

VARIABLE		BASELINE		p Value (INTER)
		MEAN	SD	
VISUAL ANALOG SCALE	GROUP A	7.27	1.22	0.001*
	GROUP B	2.53	0.99	

SD- Standard Deviation, P-Probability
*p Value is significant (<0.05)

Table 8: Comparison of CFU/ML OF AEROBIC BACTERIA in Group A and B at baseline and after 3 months

VARIABLE		BASELINE		3 MONTH		p value (INTRA)
		MEAN	SD	MEAN	SD	
CFU/ml($\times 10^4$)Of AEROBIC BACTERIA	GROUP A	186	176	9.81	3.76	0.001*
	GROUP B	227	160	5.06	1.75	0.001*
	p value (INTER)	0.1		0.001*		

SD- Standard Deviation, P-Probability
*p Value is significant (<0.05)

Table 9: Comparison of CFU/ML OF ANAEROBIC BACTERIA in Group A and B at baseline and after 3 months

VARIABLE		BASELINE		3 MONTH		p value (INTRA)
		MEAN	SD	MEAN	SD	
CFU/ml ($\times 10^4$) Of ANAEROBIC BACTERIA	GROUP A	9.12	6.83	5.6	4.43	0.001*
	GROUP B	10.8	13.5	2.2	2.72	0.001*
	p value (INTER)	0.1		0.02*		

SD- Standard Deviation, P-Probability
*p Value is significant (<0.05)

DISCUSSION

Propolis, a natural resinous substance rich in flavonoids and phenolic compounds, exhibits potent antimicrobial, anti-inflammatory, and anesthetic properties. Despite propolis' well-known ability to heal wounds, its medicinal value has received limited attention in dentistry as an anesthetic agent. Therefore this study was conducted to assess the anesthetic as well as antimicrobial role of propolis in patients with chronic periodontitis.

At baseline, the evaluation of clinical and microbiological parameters in the current study revealed statistically non- significant differences between group A and group B indicating that there were no biases or confounding factors at the outset of the study.

In the present study, on intragroup comparisons, gingival index and plaque index showed statistically significant reduction in both Group A and Group B from baseline to 3 months. On intergroup comparison, the results were statistically non-significant amongst the groups. The results are in line with the study of *Farhood et al* [12] who stated that no significant difference in PI and GI was seen while comparing curcumin oral gel as an adjunct to SRP with SRP alone. The improvement in Gingival index and Plaque index scores in our study could be attributed to patient education initiatives taken at follow up visits.

In our study, both Group A and Group B showed significant mean reduction in Pocket probing depth and gain in clinical attachment level from baseline to 3 months. On intergroup comparison, Group B has shown significantly better results in terms of PPD and CAL when compared to Group A. Similar to our study, *Nakao et al* [13] evaluated the topical application of propolis, curry leaf, and minocycline in periodontal pockets and reported that propolis treatment significantly improved both PPD and CAL, while also reducing *P. gingivalis* levels in the GCF.

On intragroup comparison, there was significant reduction in aerobic as well as anaerobic bacterial counts in both the groups. Also on intergroup comparison, Propolis group showed statistically significant reduction compared to placebo group. This supports its broad-spectrum antimicrobial action for a period of 3 months. These results are similar to the study done by *Sanghani et al* [14] wherein the mean values of colony count/ml for *Porphyromonas gingivalis* (Pg), *Prevotella intermedia* (Pi) *and* Fusobacterium nucleatum (Fn) in test group (SRP + Propolis) showed significant reduction when compared with control group (SRP only). The antimicrobial action is attributed to cell wall disruption, enzyme inhibition and impaired microbial adhesion.

Furthermore, the significant reduction in Visual Analogue Scale (VAS) scores for dentinal hypersensitivity in the Propolis group indicates its mild anesthetic and anti-inflammatory effects, which likely enhanced patient comfort post-treatment. This is particularly relevant for individuals with high pain sensitivity or anxiety regarding dental procedures. Our study was also comparable with findings of *Askari et al* [15] where propolis extracts and dentin bonding agent were equally effective in relieving dentin hypersensitivity. Contrary to the results of our study *Alqahtani et al* [16] stated that propolis hydrogel was not superior to the APF and NaF desensitizing agents.

The present study demonstrated that the adjunctive use of 10% Propolis hydrogel with SRP significantly enhanced both clinical and microbiological outcomes in the management of generalized chronic periodontitis. Also, it showed promising results in terms of decrease in DH during SRP.

This split-mouth study design is effective in controlling inter-individual variability, allowing for a more accurate comparison of treatment effects. However, the limitations of this study included small sample size, chances of bacterial translocation, patients with a wide age range and without site specificity. Further research with different or various percentage of propolis in narrow range of patients are needed to assess the long-term effect of the dentin-tubule-occluding molecules associated with the newer agent, i.e., propolis and also to confirm its antimicrobial action with use of more sensitive microbiological analysis techniques like polymerase chain reaction.

CONCLUSION

Within the limitations of this study, these finding highlights Propolis as a promising natural adjunct in periodontal therapy. The use of propolis as local drug delivery adjunctive to SRP has shown better results when compared to SRP alone. Also it has shown significant anesthetic action adding to patient comfort. Hence, these results encourage the use of propolis as an anesthetic and antibacterial agent along with SRP. However, more research with larger sample size and long term follow up is required in this area to validate the findings of the study.

Conflicts of Interest

The author reports no conflicts of interest.

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