Evaluation of Propolis Gel in Two Different Polymeric Systems as an Adjunctive Aid to Non-Surgical Therapy in the Management of Stage III Grade B Periodontitis: A Randomized Clinical Trial

Abeer S. Gawish,[A] Mai S. Attia,[A,B] Dina A. Osman,[C] Mohamed Bahrawy,[D] Marwa M. Tawfeq[E]

[A]Department of Periodontology, Oral Diagnosis, and Oral Radiology, Faculty of Oral and Dental Medicine (Girls Branch), Al-Azhar University, Cairo, Egypt.

[B]Department of Periodontology, Oral Diagnosis, and Oral Radiology, Faculty of Oral and Dental Medicine, Misr International University, Cairo, Egypt.

[C]Department of Pharmaceutics and Industrial Pharmacy, Faculty of Pharmacy (Girls Branch), Al-Azhar University, Cairo, Egypt.

[D]Department of Oral Medicine, Periodontology, and Oral Diagnosis, Faculty of Oral and Dental Medicine, Ain Shams University, Cairo, Egypt.

[E]Department of Oral Medicine, Periodontology, and Oral Diagnosis, Faculty of Oral and Dental Medicine, Beni-Suef University, Beni-Suef, Egypt.

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Abstract

Background: The goal of this study was to clinically evaluate the effect of propolis gel in different polymeric systems as an adjunct to non-surgical therapy in the management of periodontitis patients. **Methods:** A total of 30 patients with stage III grade B periodontitis were divided into the following three groups: group I patients, who received propolis in a chitosan polymer gel with non-surgical therapy, group II patients, who received propolis in a polyox polymer gel with non-surgical therapy, and group III patients who served as a control treated with non-surgical therapy only. Clinical parameters were assessed at baseline, one month, and three months. **Results:** At three months, the mean gingival index (GI) of groups I and II was the same (0.6 ±0.52), and there was no change in the mean GI in group III. There was a reduction in the mean probing depth (PD) in group I (4.80 ±0.63) and group II (4.90 ±0.74) at the end of the study. The greatest percent gain in clinical attachment level (CAL) was noted in group II (17.26 ±6.71) followed by group I (5.93 ±9.87), whereas the least percent decrease was noted in group III (3.67 ±7.77). **Conclusion:** The adjunctive use of propolis in a polyox polymer with non-surgical therapy demonstrated superior clinical results over the use of propolis in a chitosan polymer in periodontitis patients.

Keywords: Periodontitis; propolis gel; polyox; chitosan; non-surgical periodontal therapy

Introduction

Periodontitis is a disease with an inflammatory nature.[1] The microbiota in subgingival plaque is the initiator of periodontitis which is influenced by the genetic predisposition that modulates the host inflammatory reaction to pathogenic bacteria.[2] The American Academy of Periodontology and the European Federation of Periodontology in 2018 updated the classification scheme to the current understanding of periodontal and periimplant diseases and conditions. The new classification categorized periodontal diseases and conditions into three categories: firstly periodontal health and gingival diseases and conditions, secondly periodontitis, and thirdly other conditions

affecting the periodontium.[3] The goal of the new classification of periodontal diseases was to establish clearly defined clinical entities using a variety of criteria that could facilitate diagnosis, prevention, and treatment.[4] The classification also characterized periodontal health and gingival inflammation in a reduced periodontium after completion of successful treatment of a patient with periodontitis.[5] It also reorganized the broad spectrum of non-plaque induced gingival diseases and conditions based on primary etiology.[6]

The first recommended approach to the control of periodontal infections is non-surgical periodontal therapy, consisting of scaling and root planing (SRP), which is the cornerstone of periodontal therapy.[7] The primary objective of SRP is to restore gingival health by eliminating or reducing putative pathogens and shifting the microbial flora to a more favorable environment to achieve a stable periodontal condition.[8,9]

In recent years, the use of herbal products has increased in the form of local drug delivery because of the relatively safe nature of herbal extracts. Many herbal extracts, such as aloe vera, green tea, and curcumin, provide promising results for the treatment of periodontitis.[10] Propolis is a natural remedy that gained attention over a long period of time with several beneficial effects as an anti-inflammatory, antimicrobial, antioxidant, and immunomodulatory substance.[11]

All types of propolis have antimicrobial activity despite the difference in composition. Some studies suggested that propolis constituents interfere with the division of bacterial cells through the formation of pseudomulticellular forms, cytoplasm disorganization, protein synthesis inhibition, and cell lysis.[12,13] Substances that are identified in propolis, such as caffeic acid, ferulic acid, pinobanksin, and benzyl ester, act on the bacterial membrane or cell wall causing functional and structural damage, and also inhibit bacterial DNA-dependent RNA polymerases. Moreover, rutin, quercetin, and naringenin increase the permeability of the inner bacterial membrane, thereby nullifying its potential by decreasing ATP production, and interfering with membrane transport and cell mobility.[14,15]

Many studies have pointed out that the anti-inflammatory properties of propolis are due to the presence of various active flavonoids.[16] These flavonoids have been shown to inhibit the activity of cyclooxygenase (COX) and lipoxygenase (LOX), thereby reducing the levels of prostaglandin E (PGE).[17] Moreover, caffeic acid inhibits the synthesis of arachidonic acid and suppresses the enzymatic activity of COX-1 and COX-2.[18] In addition, caffeic acid phenyl ester (CAPE) is a potent inhibitor of nuclear factor kappa B (NFκB) activation and the enzymatic activity of myeloperoxidase and tyrosine kinase.[19,20,21] CAPE has immunosuppressive activity in human T cells, and inhibits the early and late events of T cell activation and the consequent release of cytokines such as IL-2.[22] Furthermore, flavonoids can act on the nonspecific immune response by activating macrophages, inducing the release of hydrogen peroxide, and inhibiting the production of nitric oxide in a dose-dependent manner.[23]

Mucoadhesive polymers used in local delivery systems act as stabilizers by enhancing the mechanical support of the drug in order to increase the drug's ability to be released.[24] One of the mucoadhesive polymers is polyox, a hydrophilic, flowable, polymer with the chemical formula (-O-CH2-CH2-). It is prepared by polymerization of ethylene oxide using a catalyst.[25] Moreover, polyox is a water-soluble polymer with low levels of toxicity that is completely and rapidly eliminated from the body.[26] Another commonly used polymer is chitosan, a natural polymer obtained by alkaline deacetylation of chitin, which is a white, hard, inelastic mucopolysaccharide that is the supporting material of crustaceans and insects.[27] Chitosan exhibits properties including permeation enhancement, in situ gelling, and releasing at a constant rate which suggests that it is a good polymer for the continuous release of drugs.[28] Chitosan has been studied for its applications not only in drug delivery, but also as a biomaterial for tissue regeneration and for its antibacterial and anti-inflammatory properties.[29]

Although the literature has not gone into great depth with regards to the anti-inflammatory effects of propolis in periodontal disease, the effects of various mucoadhesive polymers on the effectiveness of propolis are under investigation. Thus, this study aimed to investigate the clinical effectiveness of propolis gel in different polymeric systems (polyox and chitosan) as an adjunct to non-surgical therapy in the management of periodontitis patients.

Materials and Methods

This study was performed on 30 patients selected from the outpatient clinic of the Department of Oral Medicine, Periodontology, and Oral Diagnosis of the Faculty of Dental Medicine for Girls, Al-Azhar University in Cairo, Egypt. The study population consisted of age-matched patients. A written consent was obtained for each participant who agreed to participate voluntarily prior to the start of the study. The research design was approved by the Research Ethics Committee of the Faculty of Dental Medicine, Al-Azhar University, with approval number REC-ME-21-04.

Propolis extract with 80% ethanol was prepared by leaving the sample to macerate in the dark for 72 hours at room temperature, and was then filtered with Whatman No.4 filter paper. The filtrate was subsequently evaporated at 50 °C using a rotary evaporator.[30] Chitosan (Fulka, Switzerland), carbopol 934 (Delta pharma, Egypt), polyox 1105 (Aldrich, Germany), and polyox LEO 205 (Aldrich, Germany) were used to obtain the preparation of the gel formulation. The amount of concentrated extract in gel base was (4% W/V) and the ratio of propolis to polymer was 1:1.[31] Propolis (4% W/V), sodium chloride (El-Nasr Pharmaceutical Co., Egypt) (0.9% W/V), benzalkonium chloride (Delta Pharma, Egypt) (0.01% W/V), and mucoadhesive polymer were dissolved in distilled water by agitation (stirred continuously) at room temperature, and were left overnight to allow the polymers to swell and hydrate. The resulting dispersion was kept at 4 °C until a clear solution was formed. Sodium chloride (0.9% W/V) was used for isotonicity adjustment and benzalkonium chloride (0.01% W/V) was added as a preservative.[32] In the case of formulations containing chitosan, chitosan was dissolved in 1 ml of 0.1 N acetic acid and was then prepared as previously mentioned.[33] In the case of prepared formulae containing carbopol 934, triethanolamine solution (Delta Pharma, Egypt) (0.01% W/V) was used to adjust the pH within the physiological oral pH range.[34] In polyox containing formulae the weighted amount of polyox was dissolved in distilled water and was then prepared as previously mentioned.[35] The prepared gels were placed in glass vials and stored in a refrigerator at a temperature of 4 °C to 8 °C until further evaluation.[32]

Patients were diagnosed with stage III grade B periodontitis according to Papapanou et al.[36] Inclusion criteria included a clinical attachment loss (CAL) of ≥5 mm with radiographic bone loss extending to the middle third of the root and beyond, ≤4 teeth lost due to periodontitis, probing depths (PDs) ≥6 mm, vertical bone loss ≥3 mm, class II or III furcation involvement, and moderate ridge defects. Patients had Grade B periodontitis describing a moderate rate of progression with indirect evidence of 0.25% to 1% bone loss, destruction commensurate with biofilm deposits, and a diagnosis of diabetes mellitus with HbA1c levels <7%. Patients were free from any other systemic conditions that affect the periodontium or interfere with periodontal treatment according to the modified Cornell Medical Index.[37] Smokers and patients taking medication that may affect soft and hard tissue healing, pregnant and lactating mothers, and patients who underwent periodontal surgery or antimicrobial therapy in the six months prior to our study were all excluded.

A total of 30 patients with stage III grade B periodontitis were divided into three groups with reference to propolis administration, the type of mucoadhesive polymer used, and non-surgical periodontal therapy. Group I patients received propolis in chitosan polymer gel with non-surgical therapy, group II patients received propolis in a polyox polymer gel with non-surgical therapy, and group III patients served as a control treated with non-surgical therapy only.

Following clinical examination, the proposed nature of the study was explained. Each patient was asked to select an envelope from several opaque sealed envelopes after fulfillment of the inclusion criteria and signing the informed consent to be enrolled in the study. Each envelope contained a group to which the selected patient was allocated. All patients received full mouth SRP using an ultrasonic scaler and hand instruments under local anesthesia to minimize pain. Patients were given detailed instructions on self-performed plaque control measures and were instructed not to use any form of chemical plaque control.

Two periodontal sites were selected for each patient who fulfilled the inclusion criteria. Each patient's periodontal status was evaluated by measuring plaque index (PI), gingival index (GI), PD, and CAL.[38,39,40] Measurements were taken by a blinded examiner using a graduated periodontal probe (Williams probe) and recorded to the nearest millimeter. The deepest PD was selected for each tooth per periodontal site. Measurements were recorded at baseline, one month, and three months. The mucoadhesive gel was applied after complete isolation using a blunt syringe inserted into the selected pockets for groups I and II after SRP.

Results

This study was conducted on 30 patients, including 16 males and 14 females with an age range from 36 to 48 years. All 30 patients completed treatment and had no adverse reactions to therapy.

Table 1 illustrates the statistical analysis between all groups regarding mean PI. There was a non-significant difference between all groups at baseline and one month, and a statistically significant difference at three months (p = 0.049). At baseline, the highest mean value was recorded in group II (2 ±0) followed by group III (1.90 ±0.32) and group I (1.70 ±0.48), respectively. At 3 months, the greatest decrease in mean plaque value was recorded in group II (0.60 ±0.52) followed by group I (0.90 ±0.32), and there was no significant change in group III from one month to three months.

Table 1. Comparison of PI within the same group at different observation times

Time	Group	Mean	SD	Std. Error	95% Confidence Interval for Mean		Min.	Max.	p
					Lower Bound	Upper Bound			
Baseline	Group 1	1.70	0.48	0.15	1.35	2.05	1.00	2.00	0.142 ns
	Group 2	2.00	0.00	0.00	2.00	2.00	2.00	2.00	
	Group 3	1.90	0.32	0.10	1.67	2.13	1.00	2.00	
One Month	Group 1	1.00	0.00	0.00	1.00	1.00	1.00	1.00	1 ns
	Group 2	1.00	0.00	0.00	1.00	1.00	1.00	1.00	
	Group 3	1.00	0.00	0.00	1.00	1.00	1.00	1.00	
Three Months	Group 1	0.90[b]	0.32	0.10	0.67	1.13	0.00	1.00	0.049*
	Group 2	0.60[b]	0.52	0.16	0.23	0.97	0.00	1.00	
	Group 3	1.00[a]	0.00	0.00	1.00	1.00	1.00	1.00	

*Significant at $p < 0.05$; ns: non-significant; SD: standard deviation; Std. Error: standard error; Min.: minimum; Max.: maximum; different superscripts in the same column indicate a statistically significant change over time.

The statistical analysis between all groups regarding mean GI demonstrated that there was no statistically significant difference at baseline, one month, and three months. A reduction in GI at one month (1 ±0) compared to baseline (2 ±0) was shown in group II, and the same reduction was found in group III at one month (0.8 ±0.42) compared to baseline (1.8 ±0.42), followed by group I at one month (0.80 ±0.63) compared to baseline (1.50 ±0.71). At three months, the mean GI of groups I and II were the same (0.6 ±0.52) and there was no change in the mean GI of group III (Table 2).

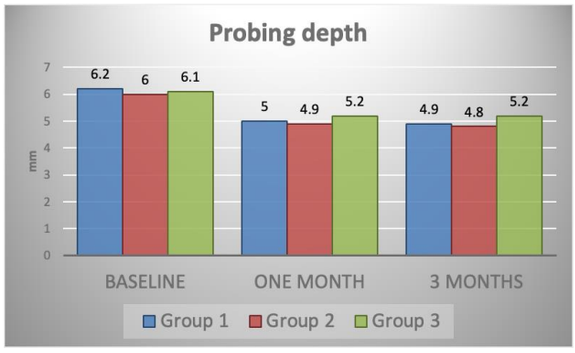
Table 2. Comparison of GI in different groups at different observation times

Time	Group	Mean	SD	Std. Error	95% Confidence Interval for Mean		Min.	Max.	*p*
					Lower Bound	Upper Bound			
Baseline	Group 1	1.50	0.71	0.22	.99	2.01	0.00	2.00	0.084ns
	Group 2	2.00	0.00	0.00	2.00	2.00	2.00	2.00	
	Group 3	1.80	0.42	0.13	1.50	2.10	1.00	2.00	
One Month	Group 1	0.80	0.63	0.20	.35	1.25	0.00	2.00	0.448ns
	Group 2	1.00	0.00	0.00	1.00	1.00	1.00	1.00	
	Group 3	0.80	0.42	0.13	.50	1.10	0.00	1.00	
Three Months	Group 1	0.60	0.52	0.16	.23	0.97	0.00	1.00	0.560ns
	Group 2	0.60	0.52	0.16	.23	0.97	0.00	1.00	
	Group 3	0.80	0.42	0.13	.50	1.10	0.00	1.00	

*Significant at $p < 0.05$; ns: non-significant; SD: standard deviation; Std. Error: standard error; Min.: minimum; Max.: maximum.

The statistical analysis between all groups regarding mean PD showed that there was a non-statistically significant difference at baseline, one month, and three months. The greatest reduction in PD at one month (5 ±0.67) was reported in group II compared to baseline (6.20 ±0.92). At three months, the mean PD for group III was the same as at one month (5.20 ±0.63), and the mean PD in group II was reduced to 4.80 ±0.63 compared to 4.90 ±0.74 in group I (Figure 1).

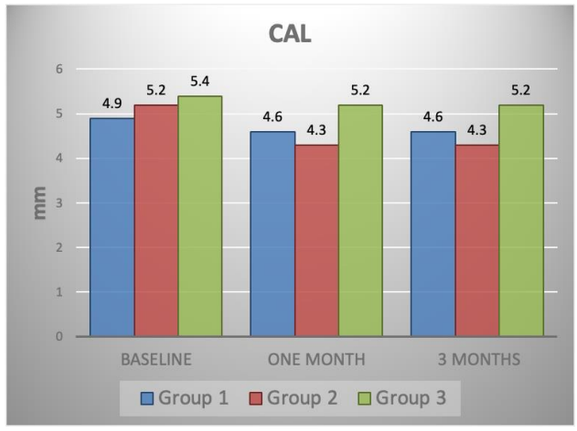
Figure 1.



Bar chart showing mean value of PD in different groups

The results of the mean CAL did not represent a statistically significant difference between all groups at baseline, one month, and three months follow-up. At baseline, the highest mean value was recorded in group III (5.40 ±0.70), followed by group II (5.20 ±0.92), then group I (4.90 ±0.99) (p = 0.452). At one month, the highest mean value was recorded in group III (5.20 ±0.79), followed by group I (4.60 ±0.97), then group II (4.30 ±0.82). At three months, statistical analysis revealed that the difference in the results of the mean values of CAL was not statistically significant (p = 0.77) (Figure 2).

Figure 2.



Bar chart showing mean value of CAL in different groups

Discussion

Periodontal disease can be treated successfully by non-surgical or surgical mechanical approaches to reduce tissue inflammation.[41] The recent development of alternative local delivery systems in the form of gels, films, pastes, strips, and fibers have provided the possibility of maintaining effective intra-pocket levels of antibacterial agents for extended periods of time.[42] Among the natural extracts used in dentistry is propolis, which is highly recommended for its various pharmacological benefits.[43]

Utilizing the property of bio-adhesion of certain polymers, which become adhesive upon hydration, could target any drug to facilitate release over extended periods of time.[44] One of these polymers is chitosan,

which acts as a promising matrix for controlled and sustained drug release.[45] The other polymer used in this study was polyox, a mucoadhesive polymer with properties ideally suited for controlled drug delivery vehicles.[46] Polyox exhibits film-forming and water retention properties, high water solubility, low toxicity, and high flow due to its silica content (≈1.5%). It also has high binding efficiency and can be cross-linked to form gels.[47] Thus, the clinical evaluation of propolis gel in two different polymeric systems with non-surgical periodontal therapy may provide a mechanism to manage periodontitis and enhance treatment outcomes.

Both polymeric forms improved clinical parameters which could be attributed not only to the SRP and the appropriate oral hygiene measures maintained by the patients, but also to the modulating effects of propolis on the periodontal tissues. Propolis has anti-inflammatory, antimicrobial, and antioxidant effects that enhance the healing of periodontal tissues.[11]

Probing depth reduction is a beneficial marker in monitoring periodontal disease since it produces an environment less favorable for the establishment of periodontopathic microorganisms. In the present study, groups I and II showed a reduction in PD at three months. These results are in agreement with studies carried out by Kirti et al. and de Andrade et al. who reported a similar reduction in PD with subgingival irrigation using propolis as an adjunct to non-surgical periodontal therapy.[48,49] The results of this study regarding the gain in CAL were also in parallel with a case-control study that evaluated the clinical efficacy of propolis in treating chronic periodontitis when delivered subgingivally.[50]

The ability of polyox to delay the release rate of soluble and insoluble drugs may lead to a significant improvement in clinical parameters in periodontal disease. This property, along with the ability of polyox to form hydrogels that quickly initiate and regulate the release of active ingredients, make polyox an ideal choice for time release formulations.[24]

In conclusion, the use of propolis in two polymeric forms as an adjunct to non-surgical periodontal therapy resulted in favorable clinical results in the treatment of stage III grade B periodontitis. The polyox polymer demonstrated superior results over other treatment modalities. This represents an important clinical advantage for patients with diabetes mellitus. Further controlled and prospective studies are needed to investigate the effects of propolis in polyox or chitosan polymers as adjunctive aids to the non-surgical approach of periodontal therapy in healthy and medically compromised patients utilizing different biological markers.

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Corresponding author:
Dr. Mohamed Bahrawy
Lecurer, Department of Oral Medicine,
Periodontology, and Oral Diagnosis
Faculty of Oral and Dental Medicine
Ain Shams University
Cairo, Egypt
E-mail: Bahrawy21@hotmail.com