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ORIGINAL ARTICLE

A thermosensitive gel with an active hyaluronic acid ingredient that contains an octenidine preservation system as an adjunct to scaling and root planning: a randomized prospective clinical study

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Abstract

Objectives To evaluate the adjunctive effect of a thermosensitive gel formulation that contains 0.8% oligo hyaluronic acid (HA) combined with a preservation system of octenidine HCl 0.625% and phenoxyethanol to scaling and root planing (SRP) as compared with SRP alone in the treatment of residual pockets of patients with stage 3 periodontitis.

Materials and methods Thirty-four patients (21 males and 13 females) aged 29–78 years (51.3 ± 13.1) with stage 3 periodontitis were recruited to participate in the present split-mouth study. None of the patients has been previously treated for periodontitis. Plaque index (PI), probing depth (PD), clinical attachment level (CAL), and bleeding on probing (BOP) were evaluated at baseline and at 3 and 6 months post treatment. Full-mouth SRP was performed in all residual pockets ≥ 5 mm. Treatment was performed by means of ultrasonic and hand instruments and lasted 45–60 min. The gel was applied subgingivally in the test sites immediately after SRP (baseline) and 1 month later. The paired *t*-test for two means was applied to test the statistical significance of the change from baseline within each arm and determine the difference between groups. The level of significance was set at 0.05 for all tests.

Results Mean PD reductions between baseline and 3 and 6 months were 1.98 mm and 2.79 mm for the test and 1.22 mm and 1.50 mm for the control group, respectively. Comparisons between the test and control groups revealed that SRP + gel yielded statistically significantly higher PD reductions compared to SRP alone ($p < 0.0001$). Compared to baseline, CAL and BOP values improved statistically significantly in both groups, although the test group presented statistically significantly higher CAL gains and BOP reductions than the control group ($P < 0.0001$).

Conclusion In residual pockets of stage 3 periodontitis patients, the local application of a thermosensitive gel with an active HA ingredient and a preservation system of octenidine HCl 0.625% in conjunction with SRP may additionally improve the clinical outcomes obtained with SRP alone.

Clinical relevance A novel HA and octenidine containing thermosensitive gel effectively improved the clinical parameters in stage 3 periodontitis patients over a 6-month period.

Keywords Dental biofilm · Hyaluronic acid · Octenidine HCl · Stage 3 periodontitis · Thermosensitive gel

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Introduction

Periodontal disease is caused by subgingival bacterial species that adversely affects the host immune system that creates and maintains unmitigated inflammation in gingival and periodontal tissues [1, 2]. The development of periodontitis is accompanied by profound shifts in the composition of subgingival different gram-negative species [3]. Among the enriched species are the classically described red-complex triad consisting of *Treponema denticola*, *Porphyromonas gingivalis*, and *Tanneralla forsythia* [3–5]. The *P. gingivalis* has been found to have a central effect on the oral microbial population during periodontal disease by triggering a state of dysbiosis and inflammation ultimately leading to bone loss [3, 4]. Periodontal treatment is based, first and upmost, on complete removal of subgingival biofilms and associated calculus deposits by scaling and root planing (SRP) [6, 7]. This can be performed in the traditional quadrant-wise or by full mouth delivery within 24 h [6, 7]. The choice of treatment delivery is based on patients' preferences and practical considerations such as general medical status, tolerance for chair time, or the need for repeated sessions of oral hygiene instructions [6, 7].

The subgingival instrumentation must be accompanied by supragingival dental plaque removal by the patient [6, 7]. It may also comprise adjunctive use of antimicrobials, applied locally or systemically [6, 7]. There is a consensus that thorough subgingival debridement needs to be performed prior to adjunctive antimicrobial therapy in order to disrupt the subgingival biofilm structure, because bacteria embedded in biofilms can be up to 1000 times more tolerant toward antimicrobials than their planktonic counterparts [8, 9]. Local application of a targeted antibiotic or antiseptic treatment as adjunctive therapy may ensure a high concentration of the treatment material in the pocket and minimize the reliance on patient compliance that is needed when toothpaste/gels or mouthwashes are used [10]. Different local antibiotics and anti-infective materials, such as tetracyclines [11–13] metronidazole [14] and chlorhexidine [15, 16] have shown beneficial effects as an adjunct to mechanical treatment. The efficacy of locally applied anti-infective treatment relies on its substantivity, i.e., the sustained release of the antimicrobials for at least 24 h in the periodontal pocket [17]; therefore, the irrigation of antiseptic materials into the periodontal pocket seems to have no effect on PD [10].

Another possible approach for local adjuncts to SRP is the application of immune regulatory materials such as viscous gels that contain long-chain hyaluronic acid [HA]. HA is a nonsulfated glycosaminoglycan that is a major component of the extracellular matrix [18]. In human periodontal ligament cells, fibroblast growth factor 2 regulates the production of HA [19]. High levels of glycosaminoglycan are detectable in the gingival crevicular fluid of periodontitis patients [20]. These amounts of glycosaminoglycan and HA are reduced after periodontal therapy [21]. The molecular size of gingival proteoglycans and glycosaminoglycans is lower in patients with early-onset periodontitis than in healthy individuals, suggesting degradation of the molecules [21]. Hyaluronan and hyaluronan-binding proteins can play a role in mitigating inflammation, assisting in healing after tissue injury, and initiating repair through the regulation of inflammatory cell recruitment, the release of inflammatory cytokines, and cell migration [17, 22]. Additionally, HA stimulates the growth of osteoprogenitor lines which are essential for bone regeneration [23]. Because of the anti-inflammatory properties of HA, [24, 25, 26] its medical application as a treatment option for osteoartritis [27], urinary incontinence in woman [28] and as a soft tissue filler [29] has been discussed.

In dentistry, a few studies have shown a positive additive effect of HA to mechanical treatment in patients with gingivitis [30, 31]. Additionally, when HA application was combined with nonsurgical treatment for periodontitis, it has been associated with additive PD reduction, CAL gain, and BOP reduction [32–34]. Nevertheless, other studies found no additive effect of HA to SRP after the subgingival application of 0.2% HA gel for 6 weeks in chronic periodontitis patients [35]. One possible reason for the modest clinical effect of HA in periodontal patients is its formulation, which is a thick, viscous gel that is difficult to insert subgingivally.

Octenidine HCl is used as a preservative in many formulations due to its antibacterial properties. It is found in several medical devices and cosmetic products, including mouthwashes. Octenidine HCl at concentrations of 0.1–2.0% was found to be effective against gram-positive and gram-negative bacteria and yeasts [36, 37].

A novel nonviscous thermosensitive gel with 0.8% active HA combined with octenidine HCl 0.625% as a preservative and phenoxyethanol (Pocket-X Gel, Prudentix, i.e., "treatment gel") may influence the microbiota of periodontal pockets. Thus, it may prevent bacterial recolonization after SRP and improve the wound healing process. The product may be useful during the initial preparation and maintenance treatment of periodontal patients and as an adjunct to the mechanical treatment of peri-implantitis. Until now, no studies have evaluated this novel nonviscous thermosensitive gel in the treatment of periodontal pockets of stage 3 periodontitis patients [38].

Therefore, the aim of the present study was to determine the efficacy and monitor the safety of the treatment gel for

improving the gingival healing of deep periodontal pockets when used in conjunction with SRP. The material tested in the present study is a thermosensitive liquid that transforms into a gel at body temperature; therefore, this formulation could solve the drawback of viscous HA gels and present superior clinical efficacy.

Material and methods

This was a prospective, randomized, split-mouth clinical study. Patients were referred for periodontal treatment between February 2018 and April 2019 and were diagnosed with stage 3 periodontitis according to the 2017 World Workshop classification [38]. The treatment was provided in private periodontal practices by two periodontists (AH, RK) with more than 20 years of experience.

The study was approved by the Institutional Review Board of Mayanei-Hayeshua Medical Center, Bnei Braq (0034–17-MHMC), and was further approved by the Israeli Ministry of Health. Prior to commencing the study, the patients were informed of the benefits and risks associated with the study and gave their written consent.

Inclusion criteria

The patients were included in the study if they fulfilled the following:

- They were generally healthy and exhibited no known allergies.
- They had not received antibiotic therapy within the last 6 months and had not undergone previous periodontal treatment, besides random maintenance treatments by oral hygienists.
- All participants had ≥ 20 existing teeth (wisdom teeth excluded).
- They each had 2 or more contralateral quadrants with a minimum of 4 qualifying pockets of ≥ 5 mm that bled on probing.
- At least 2 of the pockets were ≥ 7 mm.
- All of the patients underwent full-mouth parallel periapical X-rays demonstrating bone loss extending to the mid-third of the root and beyond.

Exclusion criteria

- Pregnancy (suspected or uncertain) or nursing.
- Heavy smoking (more than 10 cigarettes/day).
- Chronic diseases such as uncontrolled diabetes mellitus (HbA1c ≥ 7.0%) or rheumatoid arthritis.
- Aggressive periodontitis (clinical patterns of rapid progression and/or early onset disease or molar/incisor pattern).
- History of radiotherapy or chemotherapy.
- Mucosal immune diseases.
- Mental disorders.
- Parafunctional habits such as bruxism.
- Plaque index (PI) [39] ≥ 1.5 at the baseline visit.
- Subjects with removable partial dentures.
- Patients undergoing orthodontic therapy.
- Sites next to recent extraction sockets.
- Teeth showing endodontic-periodontic lesions.

In this study, we used a novel nonviscous thermosensitive formulation containing 0.8% HA combined with 0.625% octenidine HCl as a preservative. The thermosensitive excipient used in the formula was Poloxamer 407, which was added in finely titrated amounts to mildly solidify from low viscosity to higher viscosity when it contacts the gingiva at a temperature of 37 °C and thus remain in the periodontal pocket instead of leaking out.

The nonviscous HA formulation (at 20 °C and a shear rate of 100 s^{-1}, the viscosity is approximately 430 cP) transforms to a viscous gel at 37 °C (at 37 °C and a shear rate of 1 s^{-1}, the viscosity is approximately 187,000 cP) upon contact with the warm gingival boundaries of the periodontal pocket (Prudentix patent no. IL250852A).

Treatment procedures

The schedule of the evaluation, treatment, and data collection at each time point is outlined in Table 1. Measurements of periodontal parameters were performed at the following visits: preliminary, baseline, and 3 and 6 months.

A preliminary visit was performed 2 weeks before the baseline visit to establish eligibility to participate in the study. During this preliminary visit, a thorough evaluation was carried out, including a full-mouth periodontal chart, occlusal analysis, and radiological examinations by means of full-mouth periapical X-rays. Supragingival and gross scaling were carried out, and oral hygiene instructions (OHIs) were provided. Probing was performed using a 1-mm periodontal probe (UNC 15, Hue- Hu-Friedy, Chicago, IL, USA).

At the baseline visit, after repeated full -mouth periodontal chart, randomization was performed using a predetermined computer- generated randomization scheme (Excel, using the index function, random between x to y patients regarding left or right side) to receive the test treatment (SRP+gel) or to serve as SRP controls. At least 4 interproximal tests and 4 control sites were chosen in each subject. Only sites presenting PD>= 5mm and positive bleeding on probing (BOP) with no suppuration at baseline visit examination were included in the study; these sites had to

Table 1 Schematic outline of the study

Treatment time	Preliminary visit	Baseline visit/s	1 month	3 months	6 months
Screen	x				
Admission criteria	x				
Informed consent	x				
Demographics	x				
Medical history	x				
Periapical parallel X-ray status	x				
Clinical photographs	x				
Periodontal history	x				
Periodontal examination	x	x		x	x
Gel application		x	x		
Oral hygiene instruction	x	x	x		
Supragingival and gross scaling	x				
SRP		x			
Adverse events/illness check		x	x	x	x

be located in contralateral quadrants of the same jaw and on the same tooth side (i.e., mesiobuccal vs. mesiobuccal) and had to have a difference of PD<=1 mm between them. The patients were not informed of the locations where the treatment gel was applied. SRP of the entire mouth, including all target sites, was performed using ultrasonic tips (2007–2021, Osada Los Angeles, CA USA 90,034) and hand instruments (Gracey curettes, Hu-Friedy, Chicago, IL, USA). For most patients (31/34), 2 sessions were performed; the number of sessions depended on the amount of calculus and the severity and extension of residual pockets. Each session lasted 45–60 min. The jaw quadrants were treated consequently. The control sites received SRP alone, whereas the test sites were treated with SRP and the administration of the treatment gel (Prudentix Pocket-X® gel-7110604 Lod, Israel) to all matched mesial and distal pockets with a probing depth of ≥ 5 mm. The gel was administered with the plastic cannula of a syringe inserted to its maximum depth. The gel was slowly released as the cannula was moved in a coronal direction from the bottom of the pocket until it was visible at the gingival margin. The treatment gel was applied after the pocket was gently dried using an air syringe and isolated with cotton rolls. The treatment gel was applied to all test pockets at the end of the first or second treatment (when two sessions were performed) to reduce potential crossover to control sites. The patients were instructed to refrain from eating for 2 h after treatment and from drinking hot beverages for 4 h. Gentle brushing was allowed, but interproximal cleaning was not recommended for the first 2 days. The treatment gel was packaged in the form of preloaded 1-ml syringes with a blunt cannula or a brush-type canula.

The patients were instructed to practice daily plaque control using the Bass technique or the modified Bass technique with soft- or medium-bristled brushes (PARO Esro AG Dorfstrasse 143 Kilchberg, Swiss) and interproximal brushes matched to the embrasure space (TePe Munhygienprodukter Bronsåldersgatan 76 Malmö, Sweden) or toothpicks (PARO Esro AG Dorfstrasse 143 Kilchberg, Swiss).

A third visit was performed 1 month after the baseline visits. At this visit, meticulous OHIs were reinforced, and the treatment gel was reapplied to all test sites after subgingival plaque was removed.

At 3 and 6 months after the baseline visit, a periodontal examination was performed, and treatment outcomes were evaluated.

Table 1 shows the diagnostic and treatment procedures performed at each visit.

Standardization and uniformity of examinations

Two examiners (AH, RK) performed the SRP procedures, provided the OHIs, and recorded the clinical data. The Polson criteria [40] were applied to ensure that all clinical procedures were standardized according to the study protocol and to minimize intra- and interexaminer variability across time points. Briefly, as part of the standardization exercise, each of the two examiners measured the same periodontal pocket depth (PPD) of ≥ 5 mm and clinical attachment level (CAL) at 10 sites in 5 patients. Interproximal probing was performed by tilting the probe to reach the deepest value underneath the contact area/point (tilting was more pronounced in molars than in premolars and centrals). For both examiners and all 50 sites, the mean accepted difference between the 1st and 2nd measurements was 1 mm for both parameters. According to those criteria,

the observed interexaminer agreement (within 1 mm) was 87% for PPD and 88% for CAL. The assessors were unaware of the treatment being performed as the periodontist who performed the treatment and applied the gel kept clinical records of the patients, and the other periodontist, who was not aware of the location of the test sites, performed the clinical recordings.

Clinical outcome measures

- PI: the amount of visible unstained plaque thickness along the gingival margin (scored from 0—no plaque to 3—heavy plaque accumulations at the gingival margin and interdental spaces filled with plaque) measured on four sites per tooth (mesial, midfacial, distal and palatal/lingual) at the soft tissue margin [39].
- BOP: a dichotomous recording of the absence or presence of bleeding of the gingival pocket. BOP was considered positive if bleeding occurred within 10 s after gentle intracrevicular probing [41].
- PPD*: measured using light probing force (approximately 25 g) with a 1-mm periodontal probe at six sites around each tooth (mesiobuccal, midbuccal, distobuccal, mesiolingual, distolingual/palatal, midlingual/palatal). A full mouth periodontal chart was done at preliminary visit for screening and periodontal diagnosis. The values included for mean CAL and PPD calculations were only those of treated sites (test) and their contralateral matched sites (control).
- Recession level: distance from the cementoenamel junction (CEJ) to the free gingival margin (FGM).
- CAL*: distance in millimeters from the CEJ to the bottom of the probable periodontal pocket [distance from the FGM to the CEJ + PPD)].
- Mobility grade: 0: no detectable movement when force is applied other than what is considered normal (physiologic). 1: greater than normal (physiologic) mobility. 2: movement of up to 1 mm in the buccolingual direction. 3: movement of more than 1 mm in the buccolingual direction combined with the ability to depress the tooth.
- Furcation level: class I: horizontal loss of periodontal support not exceeding one-third of the width of the tooth. Class II: horizontal loss of periodontal support exceeding one-third of the width of the tooth but not encompassing the total width of the furcation area. Class III: thorough horizontal destruction of the periodontal tissues in the furcation area.

*Probing depths and CALs were rounded to the nearest 0.5 mm.

Statistical methods and analyses

The rationale for the sample size was based on the ability to demonstrate a difference in the change in pocket depth between the tested sites and the controls with an effect size of 0.5, 80% power, and a 5% significance level. All measured variables and derived parameters are listed individually and, if appropriate, tabulated with descriptive statistics.

For continuous variables, summary tables are provided, and the sample size, arithmetic mean, and standard deviation are provided by study arm.

The paired *t*-test for two means was applied to test the statistical significance of the change from baseline to the follow-up visits at 3 and 6 months for each study arm. It was also used to test the statistical significance of the difference in the changes between study arms (test vs. control sites) at baseline and at each visit.

The analysis was performed first by patient and then by smoking status.

All tests were two-tailed, and a p value of 5% or less was considered statistically significant.

The data were analyzed using SAS® version 9.4 (SAS Institute, Cary, NC).

Results

The study included thirty-four patients (21 males, 61.8%) aged 29–78 years (51.3 ± 13.1). The distribution of the treated teeth included in the study according to the jaw, molars/nonmolars, and tooth side (mesial/distal) is shown in Table 2. The baseline parameters (PI, PPD, recession depth, furcation, mobility, BOP) of the tests vs. controls are presented in Table 3. No differences were found between the groups. One patient dropped out after 3 months.

PI

The PI scores at baseline and at the 3- and 6-month follow-ups are shown in Table 4 and Fig. 1. There was a significant

Table 2 Distribution of the teeth according to the jaw, molars/nonmolars, and tooth side (mesial/distal)

		(Same for both treatment arms)
Jaw	Mandible	63 teeth
	Maxilla	109 teeth
Molars	Molars	79 teeth
	Nonmolars	93 teeth
Tooth side	Mesial	165 pockets
	Distal	172 pockets

Table 3 Baseline characteristics and comparison of the control and treatment arms

			Control		Treatment		Difference between treatment and control		
		N	Mean	SD	Mean	SD	Mean	SD	*P*-value, paired *t* test
Baseline visit	Parameter D pocket	34	7.76	1.07	7.58	1.01	−0.19	0.93	0.2445
	M pocket	34	7.17	1.03	7.22	1.07	0.04	0.70	0.7528
	Mean D and M pockets	34	7.46	0.98	7.39	0.91	−0.08	0.67	0.4950
	B recession	34	1.13	0.83	1.27	1.01	0.13	0.44	0.0906
	L recession	34	0.66	0.72	0.74	0.79	0.08	0.49	0.3657
	B furcation	32	0.98	0.79	0.96	0.77	−0.02	0.21	0.5699
	L furcation	32	0.64	0.69	0.54	0.66	−0.10	0.31	0.0735
	Mobility	34	0.13	0.30	0.12	0.28	−0.01	0.05	0.3600
	Attachment level D	34	8.88	1.56	8.49	1.71	−0.42	1.00	0.0208
	Attachment level M	34	8.30	1.55	8.49	1.71	0.18	0.72	0.1630
	Mean PI	34	1.19	0.35	1.23	0.40	0.04	0.14	0.1226
	Mean CAL	34	8.59	1.51	8.49	1.71	−0.13	0.78	0.3569
	BOP	34	46.84	7.71	48.07	7.19	1.23	7.77	0.3616

reduction in the PI from baseline to the 3- and 6-month visits for both the test ($P<0.01$) and control ($P<0.01$) sites. However, when the results of the test and control sites were compared, the test sites showed significantly better outcomes at 3 months ($P=0.0367$). At 6 months, a borderline significant difference ($p=0.0519$) was found in favor of the test sites.

PD

Lower PPD values compared with baseline were observed at both the test and control sites at the 3- and 6-month follow-up visits (Table 5; Fig. 2). For the test group, the PPD measurements at baseline and at 3 and 6 months were 7.39 ± 0.91 mm, 5.41 ± 0.70 mm, and 4.69 ± 0.55 mm, respectively ($P=0.001$) (Table 5). For the control group, the PPD values were 7.46 ± 0.98 mm, 6.24 ± 0.88 mm, and 5.96 ± 0.70 mm, respectively ($P=0.001$). While baseline values were similar for both study arms, the differences between the test and control sites at 3 and 6 months were highly statistically significant in favor of the test sites ($P<0.0001$).

The successful endpoint of "pocket closure" was defined as PPD ≤ 4 mm with no bleeding on probing measured 6 months after treatment. The proportions of sites in which this favorable outcome were achieved are presented in Table 6. Statistically significant better outcomes were found in the test compared to the control sites. This was demonstrated both in initially shallow pockets of 5–6 mm (73.1% vs. 27.1% pocket closure in the test and the control arm, respectively, $p<0.0001$), and in deeper pockets of ≥ 7 mm (11.8% vs. 0%, $p=0.0002$).

CAL

Attachments values were significantly improved at both the test and control sites at 3 and 6 months (Table 6). The CAL values at baseline and at 3 and 6 months were 8.49 ± 1.71 mm, 6.22 ± 1.41 mm, and 5.22 ± 0.88 mm at the test sites and 8.59 ± 1.51 mm, 7.45 ± 1.47 mm, and 7.23 ± 1.47 mm at the control sites (Table 7; Fig. 3). While baseline values were similar for both study arms, the differences between the test and control sites at 3 and 6 months were highly statistically significant in favor of the test sites ($P<0.0001$).

BOP

The BOP scores at baseline and subsequent measurements are shown in Table 8 and Fig. 4. The baseline values were similar for the test and control sites. Both groups showed significantly reduced BOP scores at 3 months compared with the baseline values ($P<0.01$). Lower BOP scores were observed in the test group than in the control group at 3 and 6 months ($P<0.001$). The BOP values of the test group were 7.05 ± 6.67 and 3.03 ± 4.04 at 3 and 6 months, respectively. The values for the control group were 18.21 ± 14.17 and 17.66 ± 12.68 at 3 and 6 months, respectively (Table 8; Fig. 4).

Smoking

Nine patients (26.5%) were light smokers, and the remaining 25 (73.5%) were nonsmokers.

Table 4 Mean dental plaque accumulation (% [interquartile range]) on teeth treated with SRP with or without the adjunctive application of the treatment gel at baseline and 3 and 6 months post-treatment

	Visit												
	Baseline				3 months					6 months			
	N	Mean	95% CI lower bound	95% CI upper bound	*N*	Mean	95% CI lower bound	95% CI upper bound	*P*-value (from baseline)	*N*	Mean	95% CI lower bound	95% CI upper bound	*P*-value (from baseline)
Control	34	1.19	1.07	1.31	34	0.20	0.12	0.29	< 0.0001	33	0.13	0.07	0.20	< 0.0001
Treatment	34	1.23	1.09	1.37	34	0.13	0.08	0.18	< 0.0001	33	0.08	0.03	0.13	< 0.0001
P-value (between groups)	0.1226				0.0367					0.0519				

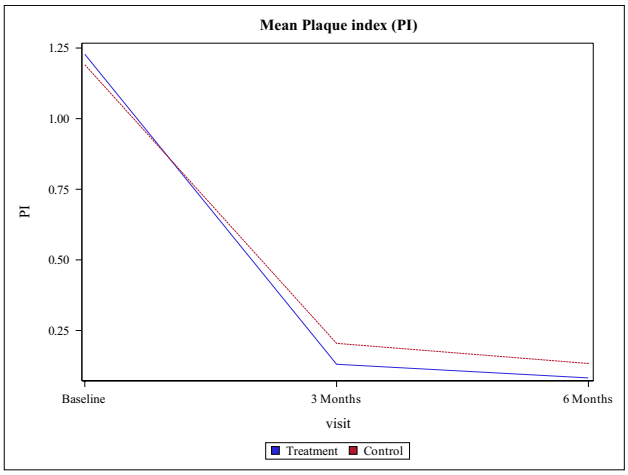

Fig. 1 Plaque index

The baseline values of CAL and PD were similar for the two groups and significantly improved in both study arms for both smokers and nonsmokers at 3 and 6 months (Table 9). No statistically significant difference was found between the smokers and nonsmokers at 3 and 6 months**.**

Safety

No adverse or allergic reactions to the treatment gel were observed**.**

Discussion

In the present study, the adjunctive application of a thermosensitive gel with an active HA ingredient had beneficial effects on periodontal health in patients undergoing SRP. The treatment gel/SRP protocol resulted in a statistically significant reduction in BOP and PD scores and an improved CAL gain at 3 and 6 months compared to treatment with SRP alone. These observations occurred in the absence of statistically significant differences in oral hygiene levels and baseline periodontal parameters between the groups.

While a statistically significant reduction in terms of PI was found in both the test and control groups, subgingival gel application had a minor additional effect on dental plaque formation. It is likely that the reduced plaque formation resulted from improved gingival health following SRP [42] combined with enhanced plaque control as a result of patient adherence to standard dental hygiene routine.

The goal of periodontal therapy is to obtain shallow probing pocket depth ("pocket closure") and absence of bleeding, indicating sufficient removal of biofilm/calculus and subsequent resolution of the inflammatory lesion [7].

Table 5 Mean PD in mm (95% confidence interval) for teeth treated with SRP with or without gel at baseline and 3 and 6 months posttreatment

	Visit														
	Baseline					3 months					6 months				
	N	Mean	SD	95% CI lower bound	95% CI upper bound	*N*	Mean	SD	95% CI lower bound	95% CI upper bound	*N*	Mean	SD	95% CI lower bound	95% CI upper bound
Control	34	7.46	0.98	7.12	7.81	34	6.24	0.88	5.93	6.54	33	5.96	0.70	5.71	6.21
Treatment	34	7.39	0.91	7.07	7.71	34	5.41	0.70	5.16	5.65	33	4.69	0.55	4.49	4.88
P-value (between groups)		0.4950					< 0.0001					< 0.0001			

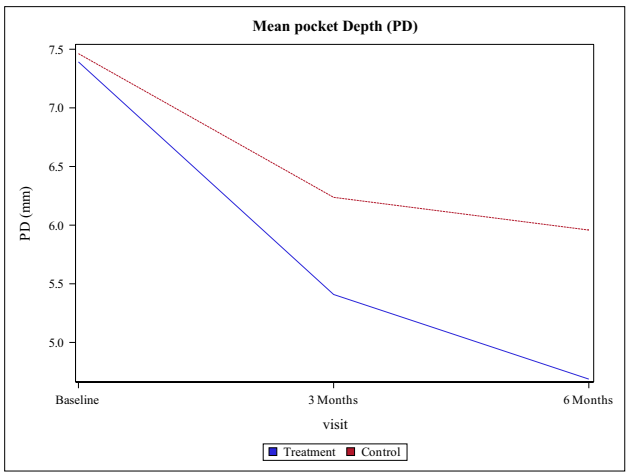

Fig. 2 Pocket depth

Table 6 Percentage of closed pockets* at 6 months divided by the initial pocket depth

Pocket depth at baseline	Closed pockets (healthy)		
	Treatment *N*/total (%)	Control *N* (%)	*P* value
Mean Pocket depth 5–6 mm	38/52 (73.1)	16/59 (27.1)	< 0.0001
Mean pocket depth ≥ 7 mm	14/119 (11.8)	0/112 (0)	0.0002
All	52/171 (30.4)	16/171 (9.4)	< 0.0001

*Closed pockets are defined as pocket depth ≤ 4 mm with no bleeding

PD

PD was the primary outcome variable used to evaluate the efficiency of the treatment gel compared to mechanical treatment only (SRP). We found PD reductions of 1.98 and 2.70 mm at the test sites 3 and 6 months, respectively, after treatment. It has been shown in a recent systematic review that, at shallow sites (4–6 mm), a mean reduction of PD of 1.5 mm can be expected at 6–8 months, while at deeper sites (≥ 7 mm), the mean PD reduction was 2.6 mm [7]. This PD reduction was consistent, irrespective of the choice of instrument (sonic/ultrasonic vs. hand), or mode of delivery (full-mouth vs. quadrant) [7]. These results are in accordance with the findings of Salvi et al. showing a PD reduction of 2–2.5 mm after SRP in sites exceeding 6 mm at baseline [43]. Thus, it has been suggested that proving the additive effect of a locally applied anti-infective treatment mandates a PD reduction exceeding these values. However, in our study, the PD reduction of deep sites in the SRP control group was only 1.5 at 6 months posttreatment. Therefore, the 6-month pocket reduction in the test group (2.7 mm) implies a significant clinical benefit of the adjunctive gel treatment.

Our findings of additional PD reductions of 0.83 mm and 1.28 mm at test sites 3 and 6 months after treatment, respectively, were similar to the results of studies using subgingival application of antiseptic agents [44, 45]. Kanoriya et al. found an extra PD reduction of 0.96- and 1.26-mm 3 and 6 months using a 0.75% boric acid gel compared to placebo gel [44], and Cosyn et al. reported an extra PPD reduction of 0.93 mm in initially deep sites (≥ 7 mm) 6 month after chlorhexidine varnish application in favor of test groups [45]. The present study positive results are also similar to previous reports regarding the subgingival application of HA as an adjunct to SRP in the treatment of chronic periodontitis [46–51]. In these studies, the application of 0.2% or 0.8% HA, 1–5 times resulted in an additive PD reduction of 0.20–0.96 mm compared to mechanical treatment only. However, other studies failed to demonstrate an additive effect of HA gels with SRP [35, 52–54].

The mean baseline PPD pockets in the present study were 7.46 mm and 7.39 mm in the control and treatment (test) groups, respectively. This may explain the superior results attained in our study compared to studies of HA in which the baseline PDs were smaller (2.71–6.8 mm) [34]. Our finding of PPD reductions at both the test and control sites is in accordance with previous reports

Table 7 Mean CAL in mm (95% confidence interval) for teeth treated with SRP with or without gel at baseline and 3 and 6 months posttreatment

	Visit														
	Baseline					3 months					6 months				
	N	Mean	SD	95% CI lower bound	95% CI upper bound	*N*	Mean	SD	95% CI lower bound	95% CI upper bound	*N*	Mean	SD	95% CI lower bound	95% CI upper bound
Control	34	8.59	1.51	8.06	9.12	34	7.45	1.47	6.93	7.96	33	7.23	1.47	6.71	7.75
Treatment	34	8.49	1.71	7.89	9.09	34	6.22	1.41	5.73	6.71	33	5.22	0.88	4.91	5.54
P-value (between groups)	0.3569					< 0.0001					< 0.0001				

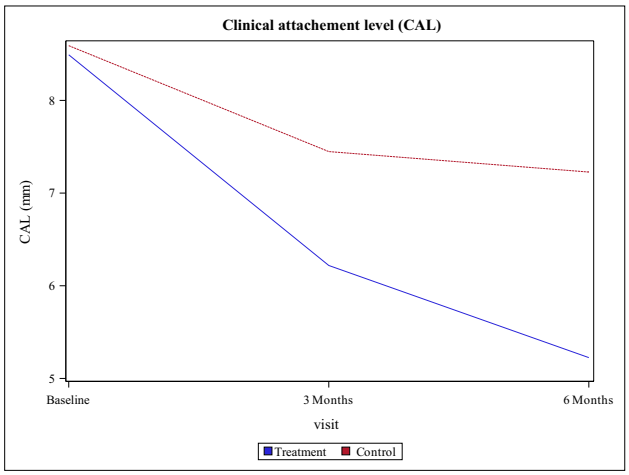

Fig. 3 Clinical attachment level

showing a greater effect of SRP on initially deeper PPDs [55, 56]. At the present study, 73.1% of the shallow test sites (5–6 mm), and only 11.8% of the deeper sites reached the successful treatment endpoint of "pockets closure" (pocket depth ≤ 4 mm with no bleeding). At the same time, in the control sites, the percentage was much lower, 27.1% and 0% for shallow and deep sites, respectively. Our results regarding the test shallow sites are similar to the findings of Wenstrom et al. [57], who performed a single session of full mouth ultrasonic debridement vs. quadrant SRP with hand instruments in chronic periodontitis patients and repeated the treatment 3 month later at sites with PPD ≥ 5 mm [57]. At 6 months, 86% of pocket were closed in the initially-shallow sites (5,6 mm). Yet, in the deeper sites (≥ 7), the authors' results were better than ours with 47% and 50% pocket closure using the ultrasonic debridement and quadrant SRP, respectively [57].

CAL gain

The CAL gain in our study was significantly greater at the test sites than at the control sites. Furthermore, the 2.27mm and 3.27mm CAL gains in the test sites at 3 and 6 months, respectively, observed in the present study were higher than the 0–1.34mm gains reported by other researchers using the HA/SRP protocol [34]. Similarly, our CAL results were better than those reported in a meta-analysis that showed an additive value of 0.4 mm for SRP plus chlorhexidine chips compared to SRP alone and additive values of 0.64 and

Table 8 Mean BOP (% of bleeding teeth per subject) for teeth treated with SRP with or without gel at baseline and 3 and 6 months posttreatment

	Visit														
	Baseline					3 months					6 months				
	N	Mean	SD	95% CI lower bound	95% CI upper bound	*N*	Mean	SD	95% CI lower bound	95% CI upper bound	*N*	Mean	SD	95% CI lower bound	95% CI upper bound
Control	34	46.84	7.71	44.14	49.53	34	18.21	14.17	13.27	23.16	33	17.66	12.68	13.16	22.15
Treatment	34	48.07	7.19	45.56	50.58	34	7.05	6.67	4.72	9.37	33	3.03	4.04	1.60	4.46
P-value (between groups)	0.3616					< 0.0001					< 0.0001				

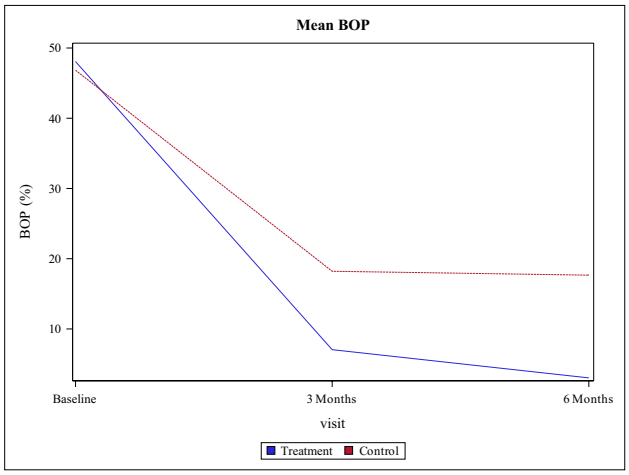

Fig. 4 Bleeding on probing

0.24 mm for doxycycline hyclate and minocycline microspheres, respectively [58].

Differences in baseline patient features, treatment protocols, observation intervals, disease severity, and measurements may explain the variations in outcomes between previous studies and the present study. Possible reasons for our beneficial results may be the baseline high CALs of 8.59 mm and 8.49 mm at the control and test sites, respectively. Additionally, we used a novel gel vehicle. It has been shown that the different effects of various topically applied antimicrobial compounds are largely related to their pharmacodynamics or to the vehicle that enables their sustained release. This effect was very clear when analyzing the results ofdifferent CHX formulations. The additive CAL gain obtained using a xanthan gel formula compared to SRP only was 0.90 mm at 6 months posttreatment [59]. This effect was better than the additive CAL gain of 0.56 mm found at 6 months using CHX-digluconate 2.5 mg (PerioChip) compared to SRP alone [15]. These differences could reflect the capacity of the vehicle to be preserved in the pocket and inhibit bacterial recolonization of the pocket for a prolonged period. Another possible explanation for our results is the repeated application of the treatment gel 1 month after the baseline visit, which could have enhanced the healing process.

BOP

We observed a statistically significant reduction in BOP scores following local subgingival application of the treatment gel formulation for both the test and the control sites, with statistically significant better results for the test sites. Our results are in accordance with studies that analyzed changes in BOP using treatments with locally delivered CHX formulationsas adjunctive to SRP [45, 60–62]. The substantivity of the gel in the pockets has not been tested. However, the positive additive results of the gel for up to 6 months after insertion imply that the material has a capacity for sustained activity. This is at variance with the results of a previous study demonstrating that a liquid form of subgingivally applied antiseptics did not significantly change PD, CAL, or BOP [63].

Hyaluronan has been shown to have bacteriostatic effects in vitro [64]. Octenidine HCl has been widely used since 1987, primarily in Europe, as an antiseptic during medical procedures, with no reported bacterial resistance [36, 37]. The main disadvantage in applying HA gels to periodontal pockets is their high viscosity (approximately 100,000 cP), a characteristic that may challenge the insertion of gels into V-shaped pockets. High-viscosity gels that are pushed into periodontal pockets could potentially trap air or may simply not fill the pocket, mainly due to the flow of gingival crevicular fluid, which is usually increased in inflamed gingiva

Table 9 Change from baseline in periodontal pocket depth (PPD) and clinical attachment level (CAL; mean of mesial and distal) for smokers and nonsmokers

	Time	PPD-control	PPD-test	Difference in PPD reduction-test vs. control	CAL gain-control	CAL gain-test	Difference in CAL gain-test vs. control
Nonsmokers	Baseline						
	3 months						
	$n=25$	-1.18 ± 0.56	-1.97 ± 0.52	-0.79 ± 0.70	1.14 ± 0.84	2.31 ± 1.04	1.17 ± 1.06
	6 months						
	*$n=24$	-1.47 ± 0.52	-2.61 ± 0.61	-1.14 ± 0.69	1.46 ± 0.80	3.23 ± 1.37	1.77 ± 1.33
Smokers	Baseline						
	3 months						
	$n=9$	-1.36 ± 0.33	-2.03 ± 0.41	-0.66 ± 0.40	1.06 ± 0.36	2.18 ± 0.63	1.12 ± 0.62
	6 months						
	$n=9$	-1.46 ± 0.68	-2.78 ± 0.60	-1.33 ± 0.52	0.96 ± 0.68	3.20 ± 0.82	2.24 ± 0.75

*One patient dropped out after the 3-month visit

compared to healthy gingiva [65]. The use of an HA formulation that is nonviscious at 20 °C and transforms to a viscous gel at 37 °C upon contact with the warm gingival boundaries of the periodontal pocket may overcome this drawback (Prudentix patent no. IL250852A). Nevertheless, liquid formulations that do not fill or solidify in the pocket tend to leak out rapidly within seconds, especially due to the high influx of crevicular fluid [65].

Quirynen [66] compared the local delivery of antiseptics vs. antibiotics and found only a modest benefit of controlled antibiotic delivery to subgingival sites. Considering the potential problems with selectivity of antimicrobial action and the possible development of resistant bacteria and adverse host reactions, topical antibiotic therapy seems to be a less desirable choice than the use of a broad-spectrum antiseptic agent with low potential for adverse reactions, such as the treatment gel applied in this study. Additionally, commercial topical antibiotic products tend to carry high financial costs. Antiseptics and preservatives have a considerably broader spectrum of activity and have multiple intracellular targets, which reduces the likelihood of resistance development [67]. Importantly, in the present study, similar beneficial results were found for smokers and non-smokers. This contrasts some but not all previous publications [68–72]. Our results are in accordance with reports showing similar favorable outcomes (CAL, BOP, PPD) after mechanical treatment in smokers versus non-smokers [70, 71]. Notably, in the present study, we included only light smokers (≤ 10 cigarettes per day). It has been previously reported that light smokers (< 20 cigarettes per day) have more favorable outcomes following SRP than heavy smokers (≥ 20 cigarettes per day) [72]. Additionally, it has been suggested that difficulty in eliminating subgingival pathogens in smokers is a possible factor leading to unfavorable outcomes following SRP [68, 69]. This may support the use of adjunctive antimicrobial therapy in smokers as done in our study.

This study has several strengths and limitations. A major strength is the professional administration of the treatment gel, which eliminated any possible influence of patients' compliance on the results. This contrasts with treatment protocols in which patients apply hyaluronan sprays or gels themselves. Moreover, the treatment was applied according to a split-mouth protocol rather than in parallel. The use of a sequential treatment protocol in which the gel is applied to all of the test sites at the end of the SRP treatment abolished the potential crossover effects of the treatment gel on the control sites.There was uniformity in the disease severity (stage 3 periodontitis), which was reflected by the similar baseline features of the test and control sites. The test and control locations were matched by teeth and sites. As several teeth were included for each patient, the total number of treated sites was high, which strengthened the results.

One limitation of the study is the absence of subgingival bacterial or GCF samples. Additionally, in the present study, we did not include a control group of patients treated with a placebo gel. Finally, the study did not include the use of acrylic stents to assure repeatable positioning of the periodontal probe. However, we sampled only mesial and distal sites, and the probe was placed at the distal/mesial transitional line angles and tilted toward the center of the buccal-lingual width under the contact point/area. As tilting angles are different in molars and central teeth, the use of stents was not relevant.

Conclusions

The treatment of periodontitis with SRP supplemented by the subgingival application of thermosensitive gel with an active HA ingredient and an octenidine preservation system achieved better results than SRP alone. The gel effectively increased improvements in clinical parameters in patients with stage 3 periodontitis over a 6-month period. PPD and CAL results were distinctly better when the treatment gel was used with SRP than when SRP was used alone. Similarly, BOP results were better when the gel was used. Further studies are required to investigate variations in administration protocols to optimize results and monitor substantivity using pocket samplings and bacterial tests.

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Declarations

Ethical approval The study was approved by the Institutional Review Board of Mayanei-Hayeshua Medical Center, Bnei Braq (0034–17-MHMC), and was registered in the NIH (NCT03358251).

Informed consent All patients signed an informed consent.

Conflict of interest The authors declare no competing interests.

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