ORIGINAL ARTICLE

Effect of Tea Tree Oil in Chronic Periodontitis Patients: A Clinical and Microbiological Study

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Abstract

Aims and objectives: Tea tree oil (TTO) is known to have antimicrobial, antifungal, antiviral, antioxidant, and anti-inflammatory effect. Periodontitis, chronic inflammation of the supporting tissues of the teeth leading to loss of the periodontal ligament and bone, results in disruption of the balance between periodontopathic bacteria and the host response to these microorganisms. We tried to evaluate the effects of TTO alone and in combination with scaling and root planning (SRP) in a triple blind, randomized controlled clinical trial of volunteers with chronic periodontitis.

Materials and methods: Thirty systemically healthy, chronic periodontitis patients were included. The study period was 21 days and the "split-mouth" design was used. Tea tree oil films were inserted in the selected pockets on day 0, removed, and reinserted on day 7. Statistical analysis was done for comparisons of clinical parameters [plaque index (PI), gingival bleeding index (GBI), probing pocket depth (PPD), clinical attachment level (CAL), and microbiological levels of the pathogens *Porphyromonas gingivalis* and *Prevotella intermedia*].

Results: At day 21, the PI and GBI were significantly reduced by all treatment modalities. When ranked, the amount of PI and GBI reduction by the different treatments was SRP + TTO, SRP, TTO. For PPD and CAL, the best result was obtained with the SRP + TTO.

Conclusion: Tea tree oil could serve as a useful adjunct or alternative to periodontal treatment.

Clinical significance: Chronic periodontitis is the most prevailing dental problem in the elderly people. Tea tree oil can be most effectively used as a local drug delivery in those affected patients.

Keywords: Clinical attachment level, Local drug delivery, Periodontitis, Probing pocket depth, Randomized clinical trial, Tea tree oil.

CODS Journal of Dentistry (2019): 10.5005/jp-journals-10063-0052

Introduction

Periodontitis, a local chronic inflammation in the supporting tissues of the teeth leading to progressive loss of the periodontal ligament and bone, is believed to result from disruption of the homeostatic balance between pathogenic bacteria and the host response to these microorganisms. Pocket formation results from the accumulation of bacterial plaque on tooth surface in close proximity to the gingival sulcus. The microbial products cause tissue destruction leading to pocket formation, connective tissue attachment loss, and alveolar bone resorption.[1] An understanding of the etiopathogenesis of periodontitis has provided the clinicians and researchers with a plenty of diagnostic tools and technique that has broadened the treatment options.

Periodontal therapy aims at controlling the progression of disease and resolving the inflammation.[2] Clinical trials have reported that scaling and root planning (SRP) along with plaque control regimens are effective treatment modalities for arresting periodontitis but the presence of certain factors can limit the clinical and microbiological response.[3] Systemic and local administration of antimicrobials would not only enhance a treatment protocol but also serve as adjuncts to mechanical therapy.[4]

Usage of several systemic antibiotics for periodontal therapy to suppress or eliminate residual periodontal pathogens serves as an adjunct to the conventional mechanical therapy.[5] The local administration of antimicrobials can provide a solution to complications of systemic antimicrobials such as resistance, toxicity, sensitivity, growth of opportunistic infection, and interaction with other medications.[6,7] Local drug delivery provides 100-fold higher therapeutic doses of the agent in subgingival areas which can penetrate into the periodontal tissues providing a dual effect on pocket microorganisms as well as on pathogens invading the tissue resulting in enhanced clinical results.[8,9]

Interest has been gathering momentum with researchers becoming more interested in alternative therapy utilizing natural products.[10] For centuries, natural products have been a major source of drugs and many pharmaceuticals in use today are derived from natural products.[11] The antibacterial and anti-inflammatory effects of oriental medicines have been evaluated for their influence on periodontal disease and periodontal tissue regeneration.[12] Tea tree oil (TTO) derived from the paper bark tea tree[13] is a widely looked on product, and it has a broad-spectrum antimicrobial, antifungal, antiviral, antioxidant, and anti-inflammatory effect.[14–19] The effect of the local application of TTO on periodontal diseases has been shown to be useful in few studies.[13] Elgendy et al. reported the significant improvement of plaque index (PI), gingival bleeding index (GBI),

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How to cite this article: Siddabasappa KG, Vandana L. Effect of Tea Tree Oil in Chronic Periodontitis Patients: A Clinical and Microbiological Study. CODS J Dent 2019;11(2):26–31.

Source of support: Nil

Conflict of interest: None

probing pocket depth (PPD), clinical attachment level (CAL), and pentraxin-3 level in the gingival crevicular fluid in the SRP and TTO gel group as compared to SRP alone at the end of 1, 3, 6, and 9 months.[20] The microbiological evaluation was not done in the study.

A MEDLINE search provided extensive data related to medical uses ranging from using it in wound dressing to vaginal candidiasis. However, the literature search using keywords periodontitis, TTO, and microbial test revealed limited data. Therefore, here an attempt has been to study anti-inflammatory and antibacterial effects of TTO clinically and microbiologically on chronic periodontitis patients.

Materials and Methods

The patients recruited in the study were new referrals to the outpatient Department of Periodontics, College of Dental Sciences, Davangere, Karnataka, India, and were used according to a protocol that satisfied the ethical standards of the institutional review board of College of Dental Sciences and Rajiv Gandhi University of Health Sciences, Karnataka, India. Patients diagnosed as suffering from chronic periodontitis were included in this study.

The patients were to meet the following inclusion criteria as patients diagnosed as suffering from chronic periodontitis as clinically evidenced by gingivitis together with mild-to-moderate periodontal pockets (5–7 mm) clinically and radiographic evidence of bone loss. The exclusion criteria were patients had ongoing antibiotic treatment or any systemic disease, patients who were pregnant, lactating, smokers, alcoholic, or who had undergone any surgical or nonsurgical therapy within 6 months prior to the start of the study. All patients gave informed consent.

A total of 30 patients were distributed between control group ($n = 15$) and test group ($n = 15$) (Table 1).

Study Methods

Study Outline

This was a randomized, triple blind, split-mouth design. Patients of both sexes within the age group of 35–50 years were eligible for inclusion. After an initial screening visit for recruitment, baseline measurements were recorded and samples taken. In the test group, TTO films were inserted in selected pockets in one side of mouth, and in the contralateral side, TTO films and SRP were performed. In the control group, SRP on one side and no treatment was done on contralateral side. Tea tree oil film was inserted at day 0 and day 7.

Clinical and microbiological parameters were recorded in at least 6–7 sites with 5–7 mm pocket depth at baseline (day 0) on day 21. The clinical parameters recorded were PI, GBI, PPD, and CAL. PPD and CAL were measured. All parameters were assessed by a single clinical investigator experienced with the index systems. SRP was performed using ultrasonic (Cavitron† BOBCAT PRO, DENTSPLY; Power-240AC 50/60Hz 80VA) and hand instruments (Universal Gracey Curettes, 2R/2L and 4R/4L Hufriedy). After SRP, the patients were instructed to perform regular oral hygiene habits, i.e., twice daily brushing by "roll-on technique" for a minimum of 2 min, using a regular toothpaste (STOLIN-R† Dr Reddy's Lab) and regular toothbrushes (Stim †toothbrush DentAids Pvt. Ltd.), which were provided to each subject.

The supragingival plaque was removed with the sample site being isolated with sterile cotton rolls and air-dried. A sterile curette was introduced to the base of the pocket, and the plaque was removed and dispensed in separate vials containing transport media, viz., thioglycolate broth with hemin and vitamin k (transport 8 medium) and sealed tightly to avoid contamination. Samples were processed within 2 days of collection. The sample was mixed thoroughly, and 5 mL aliquots were inoculated using a sterile loop onto Petri plates with the following mediums: kanamycin blood agar for *Porphyromonas gingivalis*, and kanamycin and vancomycin blood agar for *Prevotella intermedia*. The samples were then vortexed at 3,000*g* for 1 minutes to break the plaque and to obtain uniform dispersal of organisms. The plates were kept under anaerobic conditions. The plates were incubated for a minimum of 72 hours. The identity of these organisms was further confirmed by subjecting them to a series of biochemical reactions, which included indole production, nitrate reduction, and sugar fermentation tests. The tubes were incubated for 48 hours in a modified gas-pak anaerobic jar, and the fermentation reactions were noted by a change in the color of the indicator to yellow Indole, and nitrate reduction tests were performed by spot disk method using commercially available reagents (Hi-Media).

Tea Tree Oil Film Preparation

Dental films of TTO were prepared using polymers by casting method. Ethyl cellulose (800 mg) was weighed and placed in 5 mL of dichloromethane. The contents were stirred on a magnetic stirrer for 15 minutes, and 5 mL of chloroform was added to the above mixture 3 drops of tea tree oil (65 mg) was added. A glass mold of size 5×3 cm^2 was placed over a flat surface, and using a funnel, the drug polymer mixture was poured into the mold and allowed for drying, kept closed using an inverted plastic funnel for 24 hours.

The film was removed and packed in wax paper and stored in desiccator. The films were sterilized using UV irradiation and were handled using a sterilized instrument.

Clinical Measurements and Sampling

In each patient, at least 6–7 sites with pocket depths of 5–7 mm or greater were selected, wherever possible one in each quadrant. At each site, PI, GBI, PPD, and CAL were recorded at baseline (before SRP) 21 days, 3 months, 6 months, and 9 months.

After the clinical measurements were recorded, a subgingival pooled plaque sample was taken from each site using separate sterile curettes. Samples were dispensed in separate vials containing thioglycolate broth.

Statistical Analysis of Data

For clinical parameters, intragroup comparisons were made by analysis of variance and intergroup comparison by Tukey's *post hoc* analysis. For microbiological parameters, nonparametric methods were used for analysis as microbes were not normally distributed;

Table 1: Distribution of treatment groups

Group I (control group)			*Group II (test group)*		
N	*No treatment*	*SRP*	*N*	*TTO Film*	*SRP + TTO Films*
Patients	Quadrants	Quadrants	Patients	Quadrants	Quadrants
15	A	B	15	C	D

N, Number; TTO, Tea Tree Oil; SRP, Scaling and Root Planing

the Wilcoxon's signed rank test was used for intragroup comparison and the Mann–Whitney test for intergroup comparison. For all tests, a *p* value of less than 0.05 was considered statistically significant. Results are expressed as mean SD and proportions as percentages.

Results

This study included 120 quadrants from 30 systemically healthy periodontitis patients.

At baseline, all the clinical and microbiological parameters showed no significant difference between the various groups (Fig. 1).

Within the groups of no treatment, SRP, TTO, and TTO, the maximum reduction of PI was obtained for the combination of the SRP + TTO (0.87), which was statistically significant ($p < 0.000$) when compared to no treatment, SRP, and TTO groups (Fig. 2).

The GBI reduction was obtained with the combination of SRP + TTO (96.67%) as compared with other treatment modalities. An interesting observation was that TTO alone (94.7%) demonstrated a highly significant ($p < 0.000$) GBI reduction as compared to SRP alone (39.43%).

In the untreated quadrants, the PPD and CAL showed slight changes after the TTO treatment; however, these changes were not significant either over time or between groups. After 21 days, SRP alone significantly reduced PPD from 6.05 to 5.67 mm. The maximum PPD reduction was observed in SRP+TTO treatment from 6.07 to 5.26 mm, which was statistically significant ($p < 0.000$), whereas CAL showed improvement in the TTO group from 4.99 to 4.66 mm, whereas in combination of SRP + TTO, CAL showed improvement from 5.07 to 4.32 mm, which was statistically significant ($p < 0.000$) (Fig. 2).

MIC value of TTO for various microorganisms has been determined. MIC value of *Porphyromonas gingivalis* is 0.11–0.25 and *Prevotella intermedia* is 0.003–0.1. The concentration of TTO (65 mg) used in the current study met these values.

The microbiological parameter showed a maximum mean reduction in SRP + TTO group for *Porphyromonas gingivalis* (381.34) and *Prevotella intermedia* (197.33), which was statistically significant ($p < 0.003$; $p < 0.001$, respectively) (Figs 3 and 4).

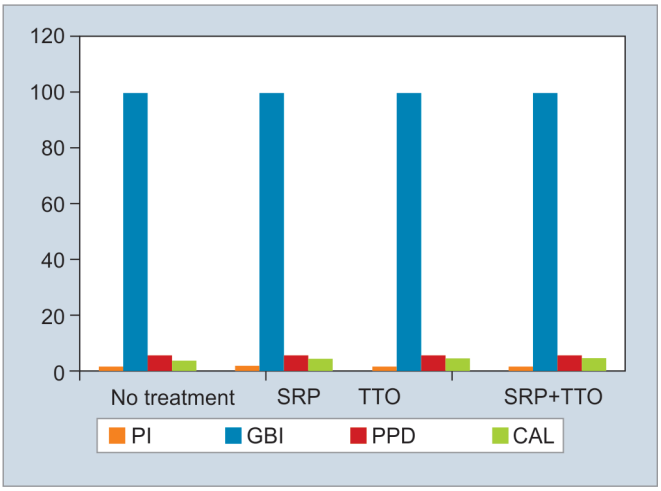

Fig. 1: Baseline parameters. SRP, Scaling and Root Planing; TTO, Tea Tree Oil; PI, Plaque Index; GBI, Gingival Bleeding Index; PPD, Probing Pocket Depth; CAL, Clinical Attachment Level

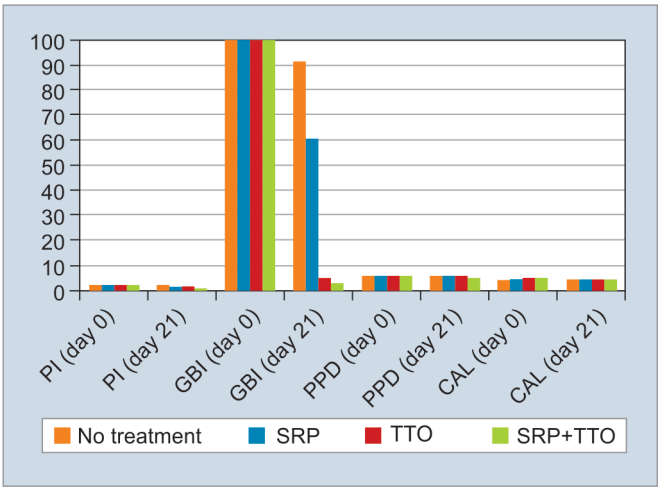

Fig. 2: Intergroup analysis (0–21 days). SRP, Scaling and Root Planing; TTO, Tea Tree Oil; PI, Plaque Index; GBI, Gingival Bleeding Index; PPD, Probing Pocket Depth; CAL, Clinical Attachment Level

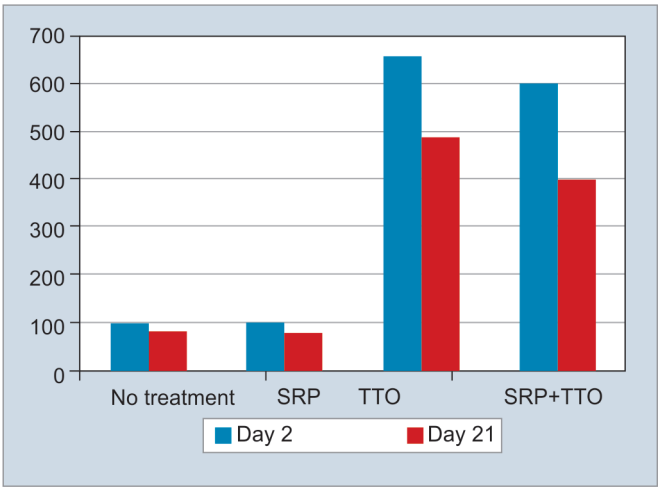

Fig. 3: Intergroup comparison of microbiologic values: *Porphyromonas gingivalis*. TTO, Tea Tree Oil; SRP, Scaling and Root Planing

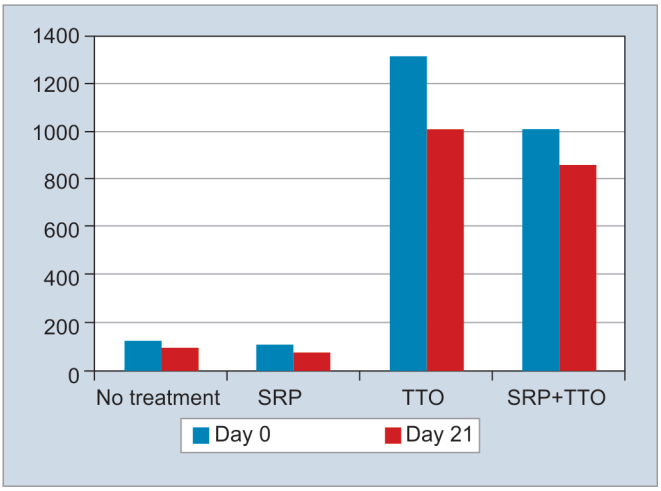

Fig. 4: Intergroup comparison of microbiologic values: *Prevotella intemedia*. TTO, Tea Tree Oil; SRP, Scaling and Root Planing

Table 2: Intergroup comparison of TTO and TTO + SRP groups at different intervals

Parameters	*TTO (mean ± SD)*	*TTO + SRP (mean ± SD)*	*t*	**p value*
PI at 90 days	1.27 ± 0.13	1.27 ± 0.18	0.00	> 0.05
PI at 180 days	1.10 ± 0.08	0.9 ± 0.26	3.62	< 0.01
PI at 270 days	0.83 ± 0.17	0.79 ± 0.19	0.82	> 0.05
GBI at 90 days	24.00 ± 9.10	26.00 ± 11.83	−0.51	> 0.05
GBI at 180 days	16.0 ± 7.38	12.0 ± 4.14	2.10	= 0.05
GBI at 270 days	10.67 ± 2.58	10.67 ± 2.58	0.00	> 0.05
PPD at 90 days	5.36 ± 0.87	5.21 ± 0.88	0.89	> 0.05
PPD at 180 days	5.36 ± 0.87	5.21 ± 0.88	0.96	> 0.05
PPD at 270 days	5.36 ± 0.87	5.21 ± 0.88	0.96	> 0.05
CAL at 90 days	4.36 ± 0.87	4.28 ± 0.77	0.56	> 0.05
CAL at 180 days	4.36 ± 0.87	4.25 ± 0.77	0.82	> 0.05
CAL at 270 days	4.36 ± 0.87	4.25 ± 0.77	0.82	> 0.05

SRP, Scaling and Root Planing; TTO, Tea Tree Oil; PI, Plaque Index; GBI, Gingival Bleeding Index; PPD, Probing Pocket Depth; CAL-Clinical Attachment Level

All patients tolerated the film well without any complications or adverse reactions to the TTO film.

The clinical parameters were evaluated from baseline to 90, 180, and 270 days. The long-term assessment of TTO and TTO + SRP groups up to 270 days demonstrated that the reduction of clinical parameter was similar (Table 2). On intragroup comparison from baseline to 90, 180, and 270 days, the various clinical parameters, PI, GBI, pocket probing depth (PPD) reduction, and CAL improvement, were statistically significant within TTO + SRP and TTO groups ($p < 0.001$). On intergroup comparison of TTO + SRP and TTO groups, there was no significant difference between the groups at various intervals from baseline.

Discussion

To date, the basic/initial treatment for periodontal patients in terms of SRP (cleaning of teeth; as commonly referred) has not been compared with the TTO effect in the reduction of clinical and microbiological parameters. Hence, an initial attempt has been made in this randomized, triple blind clinical trial to evaluate and compare the benefits of the TTO alone and SRP + TTO in the treatment of chronic periodontitis.[21]

The results of the current study are discussed below. In both the test and the control groups, PI[21] and GBI[22] were significantly reduced within each treatment group over 21 days, and thus, even the no-treatment group had a significant effect. However, such effects are known to occur as a simple consequence of the inclusion into the clinical trial, i.e., Hawthorne effect,[23] that they were instructed on proper brushing technique when the study started.

In the test group, the combination of SRP + TTO demonstrated a significant reduction of PI when compared to SRP and TTO effects individually; thus, the plaque reduction brought about by SRP was enhanced by the use of TTO. TTO and SRP alone were similarly efficacious in plaque reduction, i.e., no difference in mean plaque reduction was observed. Elgendy et al. reported no significant difference between SRP and SRP + TTO groups for plaque reduction.[20] Arweiler et al.[24] used TTO AS mouthwash on plaque formation which did not differ from the placebo mouthwash on any day from the study periods. Soukoulis and Hirsch[13] performed a double-blind, longitudinal study evaluating the effect of TTO gel (2.5%) as a brushing aid twice daily, wherein TTO did not reduce plaque scores.

The possible reason for the plaque reduction by TTO in the present study could be due to TTO strips which released the active agent *in situ*, unlike the TTO gel and mouthwash which lack the release of TTO over a certain period of time.

In the test group, the GBI percentage reduction was significant in both TTO alone and in combination with SRP. An interesting observation was that the TTO-alone group demonstrated a highly significant gingival bleeding reduction as compared to SRP alone which could be due to the anti-inflammatory and antibacterial effect of the TTO. TTO as an antimicrobial agent acts to disrupt the permeability barrier of microbial membrane structures.[24] These results are consistent with the study reported by Soukoulis and Hirsch;[13] in patients with severe gingivitis, TTO gel used twice daily as a dentifrice, which demonstrated a significant reduction of gingival inflammation and bleeding. In addition, gingival inflammation showed improvement due to the anti-inflammatory activity of TTO as reported in other studies.[18,25,26]

There was a significant improvement of PPD[27] and CAL[27] in the test group, which could be due to the significant reduction in GBI and PI. The maximum PPD reduction was observed in those receiving SRP + TTO treatment, and this reduction (0.81 mm) was more than the SRP-alone and the TTO-alone reduction (0.33 and 0.38 mm), respectively, suggesting a synergistic effect.[28] SRP-alone yielded 0.14 mm improvement and TTO yielded 0.33 mm improvement which in combination resulted in a CAL improvement of 0.75 mm. It should be noted that to our knowledge, this is the first time that TTO treatment has been shown to improve CAL. Clinical improvements, reduction in PPD, and gain in CAL seen the following extensive SRP which are due to the reduction of inflammation secondary to modification of the subgingival bacteria.[26] In addition, scaling procedure may also trigger a local and systemic host response that would aid in eliminating local infection and promote healing.[28]

The following are the possible mechanisms of TTO that are responsible for the treatment outcomes. In topical application, the components of TTO have lipophilic properties which facilitate its diffusion through the epithelium and readily absorbed with its anti-inflammatory property into the gingival connective tissues that serve to be a unique, nontoxic agent that would be as effective to the current range of chemotherapeutic periodontal treatment options;[13] in addition, TTO suppresses the monocyte production of inflammatory mediators and superoxide, and thereby may prevent tissue damage that may be seen in more chronic inflammatory states. The anti-inflammatory activity of TTO following the topical application could control inflammatory responses to foreign antigens and enable neutrophils to be fully active in an acute inflammatory response and eliminate foreign antigens, concealing monocyte inflammatory mediator and superoxide production and thereby preventing oxidative tissue damage that may be seen in chronic inflammatory states. Being a potential topical anti-inflammatory agent requires confirmation and thorough documentation of a reduction of inflammatory cells and evaluation of inflammatory biomarkers. The antimicrobial activity of TTO is already well-established. If its potential anti-inflammatory properties can be further explained, the acceptability of TTO for the treatment of chronic diseases like periodontal disease will be increased.[28] *In vitro* work over the last

decade has demonstrated that TTO affects a range of immune response, both *in vitro* and *in vivo*. For example, the water-soluble components of TTO can inhibit the lipopolysaccharide-induced production of the inflammatory mediator's tumor necrosis factor alpha (TNF-α), interleukin-1 (IL-1), and IL-10 by human peripheral blood monocytes by approximately 50% and that of prostaglandin E2 by about 30% after 40 hours.[29]

In the present study, the mean CFU reduction of the two assessed pathogens was highest for the combination of SRP-TTO followed by TTO alone. In the present study at baseline, the 30 chronic generalized periodontitis patients demonstrated the presence of *Porphyromonas gingivalis* and *Prevotella intermedia*, in accordance with the findings of Socransky et al.[1]

The antimicrobial effect of TTO is reported to be on a wide range of oral gram positive and gram negative bacteria, yeasts, and fungi.[16] The most susceptible microorganisms were *Aggregatibacter actinomycetemcomitans, Fusobacterium nucleatum*, and *Porphyromonas gingivalis*, whereas *Streptococcus mutans* and *Prevotella intermedia* were the least susceptible ones *in vitro*. However, in the current *in vivo* study, TTO was effective in reducing PI. The long-term assessment of TTO and TTO + SRP groups up to 270 days demonstrated that the reduction of clinical parameters was similar (Table 2).

On intragroup comparison from baseline to 90, 180 and 270 days, the various clinical parameters, PI, GBI, PPD reduction, and CAL improvement, were statistically significant within the TTO + SRP and TTO groups ($p < 0.001$). On intergroup comparison of TTO + SRP and TTO groups, there was no significant difference between the groups at various intervals from baseline. This is suggestive of the beneficial effects of TTO in the nonsurgical treatment of periodontitis.

There are few published studies assessing the TTO's effect on plaque formation *in vivo* and as periodontal bacteria. From the existing *in vivo* studies,[24] direct comparison of the results is not possible because the TTO vehicle in those investigations was a mouthwash rather than a gel, and the study protocol was very different.

Minimum inhibitory concentrations ranged from 0.0293 to 1.25% for the TTO solution and 0.0082 to 1.25% for the TTO gel. The values for minimum bactericidal/fungicidal concentrations were in the range from 0.0521 to 2.5% for the TTO solution and from <0.0098 to 3.33% for the TTO gel.[13,24] Both for the chlorhexidin-digluconate solution and for PlakOut, the values for the minimal inhibitory concentration were between <0.0002% and 0.0125%, respectively.[30]

The MIC value of TTO for various microorganisms has been determined. MIC value of *Porphyromonas gingivalis* is 0.11–0.25 and *Prevotella intermedia* is 0.003–0.1. The concentration of TTO (65 mg) used in the current study met these values.

Within the limitation of this study, the present randomized controlled trial confirms the plaque inhibition, anti-inflammatory, and antimicrobial effects of TTO. Hence, the TTO is suggested as an additional herbal medicament to mechanical debridement during the nonsurgical therapy and during the maintenance phase of the periodontal treatment. Further randomized controlled clinical trials over longer periods along with biochemical parameter estimation are required to build up stronger evidence for TTO, supporting a periodontal treatment protocol. Further, easier forms of TTO formulation for local drug delivery needs to be considered.

Clinical Significance

Chronic periodontitis is the most prevailing dental problem in the elderly people. Tea tree oil can be most effectively used as a local drug delivery in those affected patients.

The antibacterial and anti-inflammatory activity of TTO provides better chance to treat periodontitis than single allopathic drug.

Acknowledgement

We acknowledge Dr J Thimmasetty, Professor and Head, Department of Pharmaceutics, Bapuji Pharmacy College and Dr Kishore G Bhat, Professor and Head, Department of Microbiology, Maratha Mandal's NGH Institute of Dental sciences and Research Centre for their sincere and timely help during the study.

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