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RESEARCH ARTICLE

COMPARATIVE EVALUATION OF CLINICAL AND MICROBIOLOGICAL EFFICACY OF 2% LEMONGRASS GEL VERSUS 1% CHLORHEXIDINE GEL AS A LOCAL DRUG DELIVERY SYSTEM IN THE TREATMENT OF CHRONIC PERIODONTITIS: A RANDOMIZED CONTROLLED CLINICAL TRIAL.

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Abstract

Background: research has revealed various medicinal plants offering new choice of antimicrobial therapy. *Cymbopogon citratus* (lemongrass) is a popular medicinal plant, at concentration of $\leq 2\%$; it inhibits several microorganisms including periodontal pathogens.

Aim: to compare and evaluate, clinically and microbiologically, the efficacy of 2% lemongrass gel versus 1% chlorhexidine gel as a local drug delivery agent in patients with chronic periodontitis after mechanical periodontal therapy.

Materials and methods: 45 systemically healthy patients with chronic periodontitis were included in the study. All the patients underwent scaling and root planing (SRP), patients were recalled after 1 month. Those having persistent probing pocket depth ≥ 5 mm were selected for the study and were equally divided into 3 groups of 15 patients each. Group I received lemongrass gel, group II received chlorhexidine (CHX) gel and group III received placebo gel as a local drug delivery agent. Clinical parameters like gingival index (GI), plaque index (PI) and probing pocket depth (PPD), clinical attachment level (CAL) were evaluated at baseline and 3 months follow-up. Microbial analysis was done by using subgingival plaque samples before and after treatment.

Results: when lemongrass results compared with chx gel, the results were statistically insignificant ($p > 0.05$) and when compared with placebo result where significant.

Conclusion: the LDD containing 2% lemongrass gel can be effectively used in the treatment of chronic periodontal pockets and is as effective as CHX gel in the treatment of chronic periodontitis.

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Introduction:-

Accumulation of plaque leads to periodontitis which is widely accepted to be caused by bacteria. If left untreated it leads to destruction of supporting tissues, resorption of alveolar process and loss of periodontal ligament attachment^[2,15]. Mechanical debridement is essential in such areas. Incomplete debridement in deep or moderate pockets may result in the failure of periodontal treatment due to the retention of bacterial plaque after therapy^[3, 13]. Hence, it seems reasonable to combine mechanical periodontal therapy with the use of chemotherapeutic agents.

Local drug delivery (LDD) concept was developed by Max Goodson in 1979. LDD limits the drug to its target site and limits the problems associated with systemic therapy (such as toxicity, interaction, bacterial resistance)^[7, 8]. As an adjunct to mechanical treatment various drugs have been used^[5]. Use of antibiotics systemically or locally along with non-surgical therapy eliminates or suppresses the periodontal pathogens^[10]. However unwanted side effects and resistance of microorganisms to antibiotics have altered the general perception of the capabilities of these antimicrobial agents. Phytosciences (phytoscience or phytotherapy is study of the use of extracts of natural origin as medicine or health promoting agent) has revealed various medicinal plants possessing antimicrobial activity with fewer side effects and reduced toxicity.

Cymbopogon citratus, Staf:-

(Lemongrass) is a popular medicinal plant used for treating different diseases. Its chemical components like phenol and flavonoid substances were reported to show many biological properties such as antioxidant, anti-inflammatory, and antimutagenic properties^[4]. It has many antimicrobial properties against both gram-positive and gram-negative organisms such as anti-amoebic, antifungal, and antibacterial^[14].

Chlorhexidine is one agent, which has been extensively studied, in the periodontal research. Chlorhexidine is one of the most effective topical antimicrobial agents. Friedman and Golomb^[17], proved its effectiveness in reducing the periodontal probing depth, clinical attachment loss and bleeding on probing.

Therefore the aim of our study was to compare, clinically and microbiologically, the efficacy of lemongrass gel with chlorhexidine gel and placebo gel in patients with chronic periodontitis.

Materials And Method:-**Source of data:-**

This single centre, three group parallel designed study was conducted from November 2017 to February 2018. The subjects enrolled in this study were selected from outpatient department of periodontology. Written informed consent was taken from all the patients for participation in the study.

Sample size calculation:-

The sample size was calculated for α error fixed at 5% with a power of 80%. Based on this calculation, the minimum sample size required in each group was 15 subjects. Subjects were enrolled in three groups.

Patient selection:-

All the patients with chronic periodontitis underwent full mouth supra and subgingival scaling and root planing. Local anaesthesia was used if required. Each patient was given oral hygiene instruction. Patients having periodontal pockets with probing depth ≥ 5 mm at recall visit were selected for the study. Total of 45 subjects having mild to moderate periodontitis from both the sexes with age ranging from 18- 60 years, willing to participate in the study, were included in the study.

Exclusion criteria:-

The exclusion criteria for the patients were:

1. Patients with systemic diseases known to influence the periodontal disease;
2. Tobacco users in any form, alcohol use;
3. Regular use of mouthwash/other chemical plaque control agents;
4. Chronic use of antimicrobial, anti-inflammatory drugs and medication within 3 months prior to the study;
5. Patients who had undergone periodontal therapy within 6 months prior to the study; and
6. Pregnant females and lactating mothers.
7. Patients not willing to participate in study.

Inclusion criteria:-

Systemically healthy subjects having isolated periodontal pockets with a moderate probing depth were included in the study.

Informed consent:-

All potential participants were explained about the need and design of the study. Only those subjects who agreed to participate were enrolled in the study after obtaining their written informed consent.

Study design:-

This was a randomized controlled clinical trial. After the initial oral examination of all the patients, the sites were selected and assigned randomly into following group (by lottery method).

Group I: 15 sites treated with SRP and 2% lemongrass gel.

Group II: 15 sites treated with SRP and chlorhexidine.

Group III: 15 sites treated with SRP and placebo gel.

A single clinician (AK) provided treatment to three groups, and all pre- and post-treatment clinical parameters were recorded by another examiner (KS) who was masked to the type of treatment received by subjects.

Plaque sample collection:-

Sample sites were isolated using cotton rolls and supragingival plaque was removed to avoid sub-gingival plaque contamination. Sub-gingival plaque was collected using sterile absorbable paper points, which were inserted into gingival sulcus for 25 to 30 seconds and this paper points with collected plaque samples were diluted in 5ml saline after that 2-3 drops from diluted sample were transferred on blood agar plates for 48 hours and CFUs were analysed.

Microbial analysis:-

Blood agar plates were used to conduct the microbial analysis. Blood agar was used because it is general purpose, non selective and enriched medium that promotes growth of microorganism. Aerobic and Anerobic (using gaspack sachet) microbial analysis was performed.

Lemongrass Gel preparation:-

Gel was prepared by soaking appropriate quantity of carbopol in water for a period of 2 hrs. Triethanolamine (TEA) was used to neutralize Carbopol. Then 2% lemongrass essential oil was dissolved in appropriate and pre-weighed amounts of propylene glycol and ethanol. The solvent blend was transferred to carbopol container and agitated for an additional 20 mins. The dispersion was then allowed to hydrate and swell for 60 mins, and finally the pH was adjusted with 98% TEA until the desired pH value was approximately reached^[12].

Gel Placement:-

The gel was administered by means of a syringe with a bent, blunt-end needle (fig.1.1). The needle was carefully inserted into the periodontal pocket and the gel was applied in the test sites in a gentle probing manner, attempting to fill the full extent of the periodontal pockets. The gel was applied up to the gingival margin and the excess gel was removed with sterile gauze. After placement of the gel patients were instructed to follow strict oral hygiene protocol but were discouraged from using any interdental cleaning aids for 1 week. They were also asked not to chew hard or sticky foods at the gel placement sites. Patient's oral hygiene status was reassessed at 1 week interval

All patients were recalled for follow-up measurements at 1 and 3 months intervals.

Clinical analysis:-

Clinical parameters like gingival index, plaque index, probing pocket depth, clinical attachment level were evaluated at baseline and at 3 months.

Statistical analysis:-

Statistical analysis of the data was performed by using SPSS software. Continuous variables (PI, GI, PD, CAL) were expressed as the mean \pm standard deviation (SD). Tukey Kramer multiple comparison test was used for intergroup comparison. Statistical significance was defined as $P < 0.05$

Results:-

A total of 45 patients completed the study. Clinical and microbial parameters were evaluated in all the patients at baseline and 3 months. No adverse reaction or discomfort was observed in any subject during the study. Healing was uneventful. All subjects tolerated the drug, without any post application complications.

Mean plaque index, gingival index, probing pocket depth, clinical attachment level in all groups at different time intervals are shown in Table 1.a. Mean aerobic colony forming units and anaerobic colony forming units in all groups at different time intervals are shown in Table 1.b There was no significant difference between Groups I, II, and III with respect to plaque index, gingival index, aerobic colony forming units, and anaerobic colony forming units at baseline. There was a gradual decrease in plaque index and gingival index, probing pocket depth, gain in clinical attachment level scores in 3 months of interval, respectively in all the three groups (Tables 1). However reduction in plaque index, gingival index, probing pocket depth was more in lemongrass group (fig.1) and chlorhexidine group. The results clearly demonstrated scaling + lemongrass gel is as effective as chlorhexidine gel compared to placebo group. There were minor reductions seen in clinical and microbial parameters in placebo group.

Inter group comparison in various groups (Table 2) from baseline to 3 months showed a statistically no significant (>0.05) reduction in Group I and Group II comparison; while results were significant (<0.05) when group I and group II were compared with group III.

Moreover, lemongrass gel when compared to chlorhexidine gel showed no significant difference in the clinical and microbial parameters (fig 2, 3) suggesting that both are equally effective in improving the clinical and microbial parameters.

Discussion:-

There are various locally delivered antimicrobial agents commercially available, the need for safe, effective, and economical agents has motivated the search of various natural extracts. Local drug delivery along with SRP appears to provide additional benefits in pocket depth reduction and gain clinical attachment level^[4].

Lemongrass gel at a concentration of $\leq 2\%$, inhibits the growth of several kinds of microorganisms including periodontal pathogens, especially the strains of *Actinomyces naeslundii* and *Porphyromonas gingivalis*, which were resistant to tetracycline hydrochloride^[17]

In the present study, the additive effect of 2% lemongrass gel was evaluated as an alternative to 1% chlorhexidine gel on clinical and microbiological parameters in patients with chronic periodontitis. The above agents were compared against a placebo gel and were used as local drug delivery agents. The results of this study demonstrated significant improvement in clinical and microbial parameters in the test groups, as compared with the control group (placebo gel). In this context, the present study evaluated the effectiveness of lemongrass gel in reducing plaque scores, gingival score, probing pocket depth, aerobic colony forming units, and anaerobic colony forming units, gain in clinical attachment level and was as effective as chlorhexidine gel which is considered as gold standard

In a previous study conducted by *Warad SB et al*, local drug delivery of lemongrass resulted in a reduction of gingiva index, probing depth and gain in clinical attachment level in the treatment of chronic periodontitis. Hence, this study aimed to compare the efficacy of lemongrass gel with that of chlorhexidine gel and placebo gel in patients with chronic periodontitis and to evaluate whether which of adjunctive local antimicrobial therapy would better reduce the number of sites harbouring specific subgingival periodontal pathogens in the treatment of chronic periodontitis. The results of present are consistent with the previous study done by *Warad SB et al* (2013), regarding the lemongrass essential oil gel^[18]. Effectiveness of lemongrass gel as an anti-inflammatory and antimicrobial agent against periodontal pathogens can be seen as reduction in inflammation, which might have prevented microbial recolonization of periodontal pockets. Antioxidant property of lemongrass promotes tissue healing for periodontal pockets.

Anand et al. (2011) conducted study to evaluate the efficacy of lemongrass oil mouthwash and its antioxidant property by estimating salivary and gingival crevicular fluid GCF superoxide dismutase levels before and after its administration. He found that Superoxide dismutase levels increased when compared with the initial values in the

groups, with reduction in gingivitis. It was implied that the lemongrass oil mouthwash may have an additive effect on the treatment outcome, when it is used along with scaling^[1].

Susanto et al. (2010)^[17] determined the salivary glutathione level of moderate gingivitis patients after they gargled with different concentrations like 0.5%, 1%, 2%, or 4% of lemongrass essential oil. Glutathione, also known as sulfhydryl glutathione (GSH), is one of the nonenzymatic antioxidants in the body found in every cell and plays an important role in protection against oxidative stress. Gargling with different concentrations of lemongrass essential oil increased the salivary GSH levels in moderate gingivitis patients, especially 2% and 4% lemongrass essential oil showed the same potency as hexetidine 0.1%. It was concluded that 2% lemongrass essential oil solution can accelerate the gingivitis healing process better than at other concentrations in protection against oxidative stress. Gargling with different concentrations of lemongrass essential oil increased the salivary GSH levels in moderate gingivitis patients, especially 2% and 4% lemongrass essential oil showed the same potency as hexetidine 0.1%. It was concluded that 2% lemongrass essential oil solution can accelerate the gingivitis healing process better than at other concentrations.

However results of the present study cannot directly compared to the previous studies conducted because of certain reasons like, lack of knowledge regarding the exact mechanism by which improvement in clinical parameters occurred, also difference in methodology and vehicle for delivery used. Thus, further randomized controlled clinical trials supported by biochemical and histological analysis and longitudinal studies with larger sample size needs to be conducted to give a conclusive evidence regarding the beneficial effect of lemongrass gel in the treatment of chronic periodontitis.

The limitation of the present study was that specific microbial analysis was not performed; sample size was small, no biochemical investigation was done to support antioxidant activity of lemongrass, no histological analysis was done to evaluate the healing mechanism, lack of standardization of probe and short duration to determine the efficacy of experimental drug.

Conclusion:-

Within the limitations of the present study 2%, *lemongrass* gel as an adjunct to SRP proved to be effective in the treatment of chronic periodontitis. Further long term, trials may are required to assess and establish the efficacy of lemongrass gel in the management of chronic periodontitis to affirm the observations of our study.

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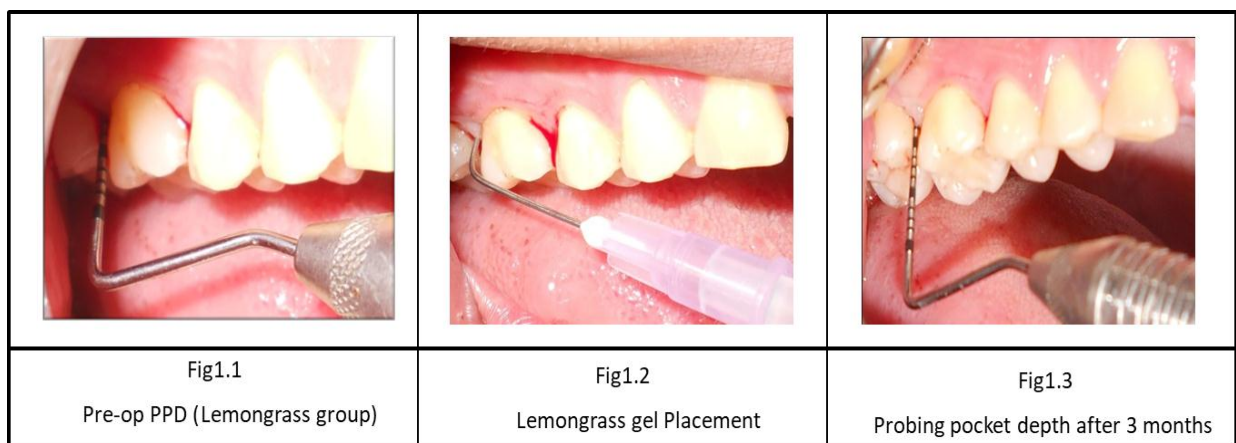


Figure 1






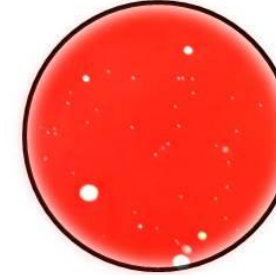
ANAEROBIC MICROBIAL ANALYSIS			
	Group I (L.G)	Group II (CHX)	Group III(Placebo)
B A S E L I N E			
3 M O N T H S			

Figure 2:-




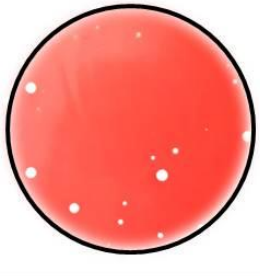
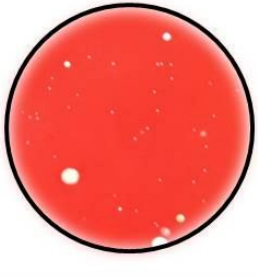

AEROBIC MICROBIAL ANALYSIS			
	Group I (L.G)	Group II (CHX)	Group III(Placebo)
B A S E L I N E			
3 M O N T H S			

Figure 3:-

Table I a:-Mean and standard deviation of Clinical parameters (PI, GI, and PPD, CAL) at various intervals

Parameters	Visit	Group I	Group II	Group III
PI	Baseline	2.26±0.17	1.91±0.58	2.3±0.3
	3 months	0.55±0.3	0.36±0.28	0.89±0.54
GI	Baseline	1.55±0.30	1.24±0.16	1.34±0.15
	3 months	0.44±0.2	0.46±0.28	0.46±0.28
PPD	Baseline	4.8±1.033	4.4±0.57	5.1±0.73
	3 months	2.9±0.87	2.8±0.788	5.1±0.73
CAL	Baseline	6.4±1.07	6.2±1.03	6.7±1.05
	3 months	3.9±0.87	3.8±0.78	5.0±1.17

P I- plaque index, GI-gingival index , and PPD-probing pocket depth, CAL-clinical attachment level

Table I b:-Mean and standard deviation of Aerobic and Anerobic CFUs at various intervals

Parameter	Visit	Group I	Group II	Group III
Aerobic CFU	Baseline	134±14.0	127±15.4	136±8.7
	3 months	122±7.5	117±8.2	132±55
Anerobic CFU	Baseline	90±8.16	81±8.16	89±8.7
	3 months	45±10.8	43±13.3	61±7.5

Table 2:-Inter Group Comparison (After 3 Months)

Parameters	Group I vs II	Group I vs III	Group II vs III
	P value	P value	P value
GI	>0.05	<0.05*	<0.05*
PI	>0.05	<0.05*	<0.05*
PPD	>0.05	<0.05*	<0.05*
CAL	>0.05	<0.05*	<0.05*
Aerobic CFU	>0.05	<0.05*	<0.001***
Anerobic CFU	>0.05	<0.01**	<0.01**

P<0.05* significant

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