

Comparative evaluation of 5% *Moringa oleifera* and 0.2% Chlorhexidine subgingival irrigation as a supplement to non-surgical pocket therapy in Stage I and Stage II Periodontitis: A Clinical study

Reshmaa Rajendran¹, Shanmugapriya Ramamurthy^{2*}, Arunmozhi Ulaganathan³, Sheeja Varghese⁴, Kadhiresan Rathinasamy⁵, Saranya Varadarajan⁶

¹Department of Periodontics, Sri Venkateshwara Dental College & Hospital, The Tamilnadu Dr.M.G.R Medical University

²Professor, Department of Periodontics, PhD Scholar, Saveetha Dental College & Hospital, Saveetha Institute of Medicine and Technical Sciences (SIMATS), Saveetha University, Chennai 77; Sri Venkateshwara Dental College & Hospital, The Tamilnadu Dr.M.G.R Medical University

³Professor & Head- Department of Periodontics, Sri Venkateshwara Dental College & Hospital, The Tamilnadu Dr.M.G.R Medical University,

⁴Professor & Head - Department of Periodontics, Saveetha Dental College & Hospital; Saveetha Institute of Medicine and Technical Sciences (SIMATS), Saveetha University, Chennai 77.

⁴Kadhiresan Rathinasamy- Professor - Department of Periodontics, Sri Venkateshwara Dental College & Hospital, The Tamilnadu Dr.M.G.R Medical University

⁵Department of Oral Pathology, Sri Venkateshwara Dental College & Hospital, The Tamilnadu Dr.M.G.R Medical University

⁶Department(s) and institution(s), Department of Periodontics, Saveetha Institute of Medicine and Technical Sciences (SIMATS), Saveetha University, Chennai 77; Sri Venkateshwara Dental College & Hospital, The Tamilnadu Dr.M.G.R Medical University

ABSTRACT

Background: Although scaling and root planing (SRP) are the gold standard in nonsurgical periodontal treatment, it does not ensure complete eradication of all putative periodontal pathogens. So, subgingival irrigation is performed in addition to non-surgical debridement for removing periodontal pathogens by both mechanical and chemical action. The goal of the present study was to assess and compare the clinical efficacy of two episodes of subgingival irrigation using 0.2 percent chlorhexidine and 5% moringa oleifera combined with SRP.

Materials and Methods: Totally 15 patients with less than or equal to 4 mm of attachment loss as seen in Stage I and Stage II Periodontitis were recruited. From each patient, 2 periodontal pockets that are not adjacent and have a probing depth of minimum 6 mm were selected. Each patient's two deep periodontal pockets were given irrigation treatments, one with CHX digluconate 0.2 percent (control group) and the other with *Moringa oleifera* 5 percent (test group) in two episodes. Pre-irrigation (day 0), days 7, and 28 post-irrigation measurements of the plaque index (PI), gingival index (GI), probing pocket depth (PD), and clinical attachment level (CAL) were recorded.

Results: Following irrigation, the mean PI and GI scores decreased for both groups, with the moringa group experiencing a statistically significant greater decrease than the chlorhexidine group. The *Moringa* group had a greater reduction in PD and CAL than the CHX group.

Conclusion: The clinical outcomes of this investigation imply that subgingival irrigation with 5 percent *Moringa oleifera* may be an effective adjunctive treatment for patients with chronic periodontitis.

Corresponding Author: drshanpriya@gmail.com

How to cite this article: Rajendran R, Ramamurthy S, Ulaganathan A, Varghese S, Rathinasamy K, Varadarajan S. Comparative evaluation of 5% *Moringa oleifera* and 0.2% Chlorhexidine subgingival irrigation as a supplement to non-surgical pocket therapy in Stage I and Stage II Periodontitis: A Clinical study. Journal of Complementary Medicine Research, Vol. 13, No. 4, 2022 (pp. 77-81).

INTRODUCTION

Periodontitis is a common infectious disease of the tooth supporting structure which is long standing in nature. The disease is microbiological in origin with extensive molecular pathways of host microbial interaction. The resultant inflammation causes progressive destruction of the attachment apparatus of the teeth. The most frequently isolated oral pathogens from patients with periodontitis are *Aggregatibacter actinomycetemcomitans* and *Porphyromonas gingivalis*. Production of proinflammatory cytokines occurs as a result of the host's defence mechanism against these bacteria. ¹ As a result, the host-microbe interaction results in the production of tissue-destructing enzymes, which in turn damages the periodontium.

KEYWORDS:

Chlorhexidin, Herbal extract, *Moringa oleifera*; subgingival irrigation.

ARTICLE HISTORY:

Received : Apr 20, 2022

Accepted : Jun 19, 2022

Published: Sep 18, 2022

DOI:

10.5455/jcmr.2022.13.04.16

Currently non-surgical periodontal therapy comprising of scaling and root planing is recommended to reduce the microbial load in biofilm. However, complete elimination of such pathogenic microorganisms always questionable owing to difficulty in instrumentation of the deep periodontal pockets, furcation regions. Hence host modulation with anti-inflammatory and antimicrobial were introduced.² Systemic antimicrobial therapy is associated with microbial resistance, poor tissue penetration, inadequate concentration at the desired site. Hence to overcome these limitations, local drug delivery systems were introduced in 1966 by Goodson et al.³ Local drug delivery has several advantages over systemic route like improved compliance, sustained action, increased microbial tolerance.⁴ In the treatment of periodontal disease, periodontal pocket irrigation, one of the local drug delivery methods, is utilised as a lavage to flush the microorganisms from the periodontal pocket.⁵

The mechanical effect of oral irrigation, helps to flush away any remaining biofilm in the pocket, and the antibacterial effect of the agent, lowers the bacterial count in the pocket, that contribute to the maintenance of dental hygiene. Additionally, it has been shown to have greater levels of penetration into the crevicular area, aiding in the removal of bacterial toxins and the disruption of unattached plaque in the subgingival environment.⁶ Many studies have reported that oral irrigation caused reduction in bleeding on probing (BOP), probing pocket depth (PPD) and microbial count in patients with chronic periodontitis. Various irrigation agents like chlorhexidine, hydrogen peroxide, boric acid, povidone iodine and stannous fluoride have been explored for management for periodontitis.

Despite several known agents, the search for herbal extract for oral irrigation continue owing to the diverse phototherapeutic effects and ease of availability. Hence herbal drugs have been widely accepted worldwide, since several decades in terms of both medicinal and economic implications. Herbal extracts of Neem, Curcumin, Pomegranate, Tulsi and many more have been researched extensively in nonsurgical management of periodontitis. They are widely used for their anti-microbial, anti-inflammatory, anti-fungal, anti-oxidant, anti-viral and analgesic effect.⁸ Due to their significant natural activity, active safety margin, easy availability, nativity, and lower cost, they are preferred to conventional medications. A recent systematic analysis conducted by Moro et al. in 2018 revealed that local phytotherapeutic administration in conjunction with periodontal therapy significantly reduces PPD, and the majority of the patients showed clinical attachment level gain. In addition to SRP, local phytotherapy application may enhance clinical attachment level gain and PPD reduction.⁹

Native to India, *Moringa oleifera* Lam is found throughout the country. There is a substantial amount of research on moringa and its therapeutic benefits. It is included into a standard diet and has anti-tumor, antipyretic, antispasmodic, diuretic, antiulcer, hypotensive, hypolipidemic, hepatoprotective, antifungal and antibacterial activity. The moringa tree is also identified as miracle tree, tree for live, wonder tree and amazing tree because each part of the *Moringa* plant, starting from the leaves, fruit, seeds, flowers, bark to the roots, have tremendous benefits on human health.¹⁰ Research on the effectiveness of several parts of *Moringa oleifera* (MO) extracts in oral healthcare is scarce. In particular the data

on clinical applications of *Moringa* in the management of Periodontitis is sparse. Considering the antioxidant, anti-inflammatory, anti-microbial effect of *Moringa oleifera* Lam. leaves, this study was taken up to investigate any possible benefits of *Moringa oleifera* Lam. in the management of periodontal disease. The objective of this study was to assess and compare the efficacy of two episodes of subgingival irrigation with 5% *M. oleifera* lam with 0.2% chlorhexidine in addition to scaling and root planning in patients with chronic periodontitis.

MATERIALS AND METHOD

Study design

This is a double blinded, split mouth, randomized controlled clinical study conducted in Department of Periodontology in a college in Tamil Nadu. The study was performed after the ethical approval from the Institutional Ethics Committee (IEC/ SVDCH/000286).

Sample Size

The G power analysis programme version 3.1 was used to calculate the sample size. The power of the study was set to 80% and the sample size estimated is 30 totally.

Patient Selection

Based on the following criteria, 15 patients with chronic periodontitis were selected:

Inclusion Criteria

1. Study participants of age between 25 and 65 years with minimum of 20 teeth.
2. Patients with mild to moderate periodontitis according to Armitage Criteria (1999) were selected. In every participant 2 sites each with periodontal pocket with a probing pocket depth of 5-6mm with bleeding on probing and clinical attachment loss 3-4mm with crestal bone loss seen radiographically was selected.

Exclusion Criteria:

1. Patients with serious, uncontrolled medical disorders.
2. Patients underwent SRP procedures within the immediate past six months.
3. Patients took antibiotics within the past six months.
4. Patients allergic to CHX.
5. Patients who are pregnant and lactating mothers.

Two weeks before commencement of the study, the selected patients were briefed about the study and were asked to sign an informed consent to certify their willingness to participate in the study. After receiving oral hygiene instructions patients underwent Scaling and Root planning. Following 14 days of SRP (T0) assessment of the baseline clinical parameters done. The indices and clinical parameters recorded were :

1. Plaque index (PI) (Sillness and Loe, 1964)
2. Gingival index (GI) (Loe and Sillness, 1963)
3. Probing depth (PPD)
4. Clinical attachment level (CAL)

With the use of the UNC-15 graded periodontal probe, clinical parameters were measured.

Randomization

The chosen sites were divided into two groups and randomly assigned by lottery using computer-generated numbers to each group. Each group received one of the two irrigants, which were delivered to the investigator in a concealed envelope and were designated as follows:

CONTROL GROUP: 0.2% Chlorhexidine (n=15)

TEST GROUP: 5% *Moringa oleifera* Lam (n=15)

For control group commercially available CHX was used without dilution.

Preparation of *M. oleifera* solution

In our previous study we determined the in-vitro antioxidant, anti-inflammatory effect of aqueous and ethanolic extract of *Moringa oleifera* Lam leaves and observed that aqueous extract exhibited better antioxidant and anti-inflammatory property. Hence the present study was conducted with 5% MO lam aqueous extract. Briefly the leaves were collected and cleaned with distilled water. The leaves were shade dried for a week and powdered. This powder was weighed and dissolved in 100ml distilled water and macerated and shaken for 24 hours in an orbital shaker. The extract was filtered and boiled after 24hours. The extract was concentrated to 10ml and stored.

Irrigation Protocol

With cotton rolls used to isolate the selected site, subgingival irrigation was carried out using an insulin syringe (1 ml, 0.4 mm, 28 gauge) containing the specified solution. In order to prevent further injury to the surrounding tissues and to ensure that the irrigant flowed to the most apical area of the plaque, the tip of the syringe was positioned approximately 1mm coronal to the bottom of the pocket as previously measured. For each selected sites, irrigation was done using 10ml of irrigant under slow, continuous pressure for around 60 seconds. Aspiration of extra irrigant solution was constant. On day 0 and then again on day 7, irrigation was carried out twice. The oral hygiene instructions were reinforced after the irrigation protocol. After then, the irrigation solution was not administered again. Patients were followed up on the 21st and 30th day after irrigation. On the thirtieth day evaluation of the clinical parameters at the selected sites was done.

For each study group, the samples were used to estimate the means and standard deviations. The Student paired t-test was used to compare the mean values. The P-value was computed using a Student's t-test. P<0.01 was regarded as the level of significance in the current study.

RESULTS

The number of subjects with Periodontitis included were 15 in this split mouth designed study. The sites were randomly allocated into the test (15 MO group) and control group (15 CHX). All the patients in the study were able to complete the follow up period. This investigation had no dropouts.

Mean Plaque index

The mean plaque index (PI) in CHX group reduced from 2.09 \pm 0.23 to 1.48 \pm 0.39 in one month. Similarly in MO group, the mean plaque index decreased from 2.13 \pm 0.31 to 1.11 \pm 0.41. (TABLE -1) On comparing one month post treatment values of CHX and MO group, the decrease in the mean plaque index score in MO group was observed to be significantly greater in comparison with CHX group.

Mean Gingival index

The mean gingival index (GI) in CHX group decreased from 2.06 \pm 0.32 to 1.49 \pm 0.41 (GRAPH 1) and in MO group there was a significant decrease in gingival index from 2.18 \pm 0.36 to 1.49 \pm 0.33. The decrease in the mean gingival index score in MO group after a month was greater than the CHX group. The results were significant

Mean PPD

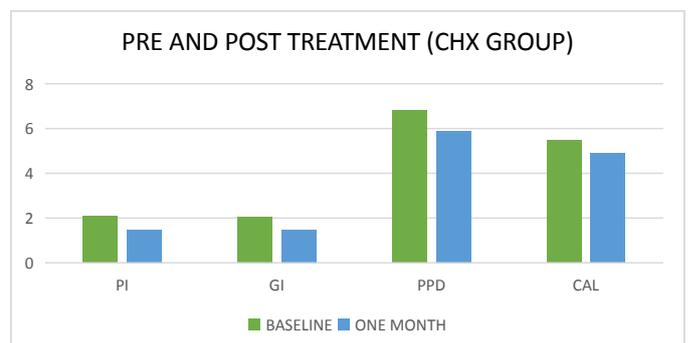
There was a significant decrease in the mean probing depth (PPD) values for both the groups from baseline to one month. In CHX group, the mean PPD reduction was from 6.8 \pm 0.93 to 5.9 \pm 0.87 and in MO group, the PPD reduction was from 6.8 \pm 0.9 to 5.6 \pm 0.67 (graph 2). However, there was no significant difference in decrease of this parameter between the groups.

Mean CAL

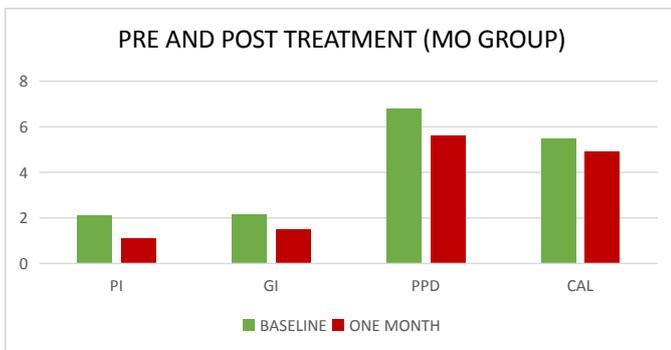
There was a mild decrease in the mean clinical attachment level (CAL) from day 0 to day 30 in both the groups. The mean CAL values in CHX group changed from 5.47 \pm 0.71 to 4.93 \pm 0.59 and in MO group from 6.07 \pm 0.9 to 4.67 \pm 0.61. The inter-group comparison showed no statistically significant difference among both groups. (GRAPH-3)

Table 1: Comparison of Pre Treatment and Post Treatment Parameters of CHX Group and MO Group

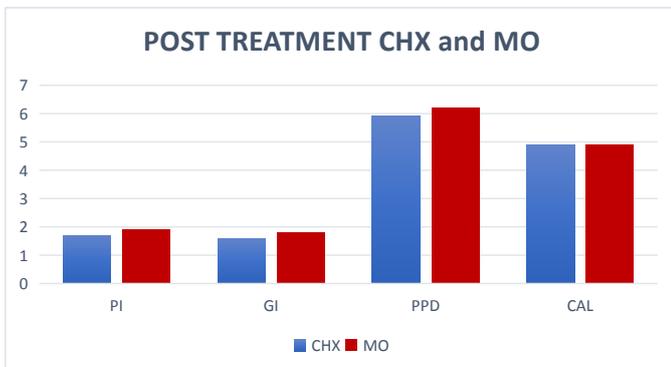
PARAMETERS (T1)	DURATION	CHLORHEXIDINE (CONTROL)	MORINGA OLEIFERA (TEST)
PI	BASELINE	2.09 \pm 0.23	2.13 \pm 0.31
	1 MONTH	1.48 \pm 0.39	1.11 \pm 0.41
GI	BASELINE	2.06 \pm 0.32	2.18 \pm 0.36
	1 MONTH	1.49 \pm 0.41	1.49 \pm 0.33
PPD	BASELINE	6.8 \pm 0.93	6.8 \pm 0.9
	1 MONTH	5.9 \pm 0.87	5.6 \pm 0.67
CAL	BASELINE	5.47 \pm 0.71	6.07 \pm 0.9
	1 MONTH	4.93 \pm 0.59	4.67 \pm 0.61



Graph 1: Pre (T0) and Post Treatment (T1) Comparison – CHX Group



Graph 2: Pre (T0) And Post Treatment (T1) Comparison – Mo Group



Graph 3: Post Treatment Comparison Of Chx And Mo Group

DISCUSSION:

Nonsurgical periodontal therapy is still regarded as the “gold standard” despite the fact that it has changed throughout time. The total elimination of supra- and subgingival microbial pathogens is the main goal of periodontal therapy. However, deep pockets are resistant to the traditional non surgical, closed debridement of plaque from subgingival root surfaces. Therefore, full or appropriate debridement may not always be achieved by professional maintenance therapy. As complete plaque removal by mechanical means was shown to be challenging in patients with pocket depths more than 5 mm, adjunctive techniques that may improve the effectiveness of mechanical measures were used. 2

One of the commonly employed treatment modalities in treating periodontitis is subgingival irrigation of periodontal pockets by antimicrobial and mechanical flushing of debris. 5 Several studies have used antibiotics, or herbals as irrigants which are used as an adjunct with conventional treatment. Studies have reported that agents like chlorhexidine, tetracycline, povidone iodine, boric acid etc have reported significant improvement in both the clinical and microbiological parameters of periodontitis. 6

PPD is one of the most vital tools to determine determining the extent of loss of periodontal tissues. PPD and CAL are both diagnostic and theranostic tools for periodontal disease. Therefore, we measured PPD and CAL before and after therapy in the current investigation.

Although chlorhexidine has been considered the gold standard it has a few side effects like staining of teeth, restoration and tongue when used for long duration of time. So alternative approach to this usual treatment in inhibition of subgingival bacteria is to use herbal agents. 13

M. oleifera is a medicinal plant which has been reported to have various pharmaceutical effects. Moringa also has found its application in dentistry It also used in the form of tooth paste and mouthwash for preventing dental caries owing to its anti-bacterial property of inhibiting *Streptococcus mutans*. Moringa leaf extract has antimicrobial activity against cariogenic bacteria and biofilms which are more resistant to antibiotic agents because of the glucan exopolymer matrix. This matrix inhibits the diffusion of the positive charged antibiotic agent in the biofilm. Studies have also showed positive results of Moringa as root canal irrigant which is shown to be successful against *E. faecalis*. 14 It also finds it application in the management of oral ulcers and as an adjuvant for healing of extraction sockets.

However its application as a local drug delivery agent in improving periodontal health has been less explored. Therefore, the present study aimed to compare the clinical efficiency of irrigation with Moringa as an adjunct with scaling and root planing as with CHX irrigation. The results of the present study showed that both chlorhexidine and MO caused significant reduction of PPD, and CAL. Gottumukkala et al. reported a greater reduction in PPD after 6 months following irrigation with Chlorhexidine along with SRP in comparison with SRP alone. In another study a pulsating irrigator with 0.2 percent CHX solution was used to irrigate periodontal pockets twice a day for 14 weeks after SRP. They also observed a significantly lower PPD reduction in the irrigant group in comparison with placebo solution. 11 MacAlpine et al., reported no significant differences in PPD levels between the CHX irrigation and SRP alone groups statistically. But in our study, there was a significant reduction in PPD values between both CHX and MO groups. Following irrigation of periodontal pockets with both chlorhexidine and saline, Krishna et al. discovered similar improvements in CAL value, which is consistent with the findings of our investigation. 12

There was a statistically significant difference between CHX and MO groups in terms of plaque index. According to Vinholis et al. 15 and Paolantonio et al. 16's study, which demonstrated statistically significant differences in plaque index between CHX and Curcumin, these findings are consistent. The reduction in probing pocket depth between the CHX and MO groups in the current investigation at the end of 21 days was not statistically significant; these findings are similar with those of Nayyar et al. 17 and Mizrak et al. 18 Due to curcumin's ability to suppress the inflammatory response, the gingival inflammation in the curcumin group significantly improved more than the control group. Similar anti-inflammatory effects were seen in our study which shows Moringa could inhibit cyclooxygenase-2 (COX-2), lipoxygenase, that reduces synthesis of the inflammatory cytokines like interleukin (IL) - 1, -2, -6, -8, and -12, tumor necrosis factor- alpha (TNF-a), and monocyte chemoattractant protein (MCP). 19

The improvement in clinical parameters following irrigation with Moringa could be attributed to its antimicrobial activity, anti-inflammatory and antioxidant effects as reported in our previous study. Both aqueous extract and ethanolic extract of moringa exhibited significant anti-inflammatory and anti-oxidant property at 50microgram per ml. Aqueous extract had higher anti-inflammatory and anti-oxidant property than ethanolic extract. So 5% aqueous moringa oleifera extract was used in this study. 10

The shortcomings of this study are only few subjects were enrolled and the effects of Moringa on specific periodontal pathogens were not assessed. Also, the follow up period of 4 weeks is not enough to determine the long-term effects. Future studies must therefore be conducted to determine whether moringa extract may be used as a supplement to mechanical instrumentation in a larger population with extended follow up time frame.

CONCLUSION

Due to its ability to dilute and remove bacterial toxins, hinder plaque development, and remove unattached plaque mass, subgingival irrigation provides a good basis for plaque control. They are therefore particularly helpful in preventing gingivitis, especially in individuals who are unable or unwilling to practise proper inter-proximal hygiene. With severe periodontitis, refractory periodontitis, and difficult-to-maintain locations such fixed prostheses, orthodontic appliances, and inaccessible embrasure spaces, this treatment option may be very helpful as a non-surgical option for medically compromised patients. As a result, using irrigation devices to keep deep pockets free of microorganisms would reduce the need for open flap debridement procedures. There may be new opportunities for nonsurgical periodontal therapy if the use of sub-gingival irrigations loaded with herbal counterparts are investigated for the treatment of mild-to-moderate and moderate-to-severe chronic periodontitis patients who are otherwise candidates for flap procedures.

REFERENCES

- Badersten, A., Nilvéus, R. and Egelberg, J. (1981), Effect of nonsurgical periodontal therapy. *Journal of Clinical Periodontology*, 8: 57-72. <https://doi.org/10.1111/j.1600-051X.1981.tb02024.x>
- Lang NP. Indications and rationale for non-surgical periodontal therapy. *International dental journal*. 1983 Jun 1;33(2):127-36.
- Goodson, J.M., Haffajee, A. and Socransky, S.S. (1979), Periodontal therapy by local delivery of tetracycline. *Journal of Clinical Periodontology*, 6: 83-92. <https://doi.org/10.1111/j.1600-051X.1979.tb02186.x>
- Arthur J. Bonito, Linda Lux and Kathleen N. Lohr. Impact of local adjuncts to scaling and root planing in periodontal disease therapy: A systematic review. *J Periodontol* 2005; 76:8, 1227-1236.
- Greenstein, G. (1987). Effects of subgingival irrigation on periodontal status. *Journal of Periodontology*, 58(12), 827-836.
- Wennström, J. L., Heijl, L., Dahlén, G., & Gröndahl, K. (1987). Periodic subgingival antimicrobial irrigation of periodontal pockets: (I). Clinical observations. *Journal of clinical periodontology*, 14(9), 541-550.
- Greenstein, G. (2005). Position paper: The role of supra-and subgingival irrigation in the treatment of periodontal diseases. *Journal of Periodontology*, 76(11), 2015-2027.
- Eid Abdelmagyd HA, Ram Shetty DS, Musa Musleh Al-Ahmari DM. Herbal medicine as adjunct in periodontal therapies- A review of clinical trials in past decade. *J Oral Biol Craniofac Res*. 2019 Jul-Sep;9(3):212-217. doi: 10.1016/j.jobcr.2019.05.001. Epub 2019 May 14. PMID: 31193290; PMCID: PMC6525324.
- Moro, MG, Silveira Souto, ML, Franco, GCN, Holzhausen, M, Pannuti, CM. Efficacy of local phytotherapy in the nonsurgical treatment of periodontal disease: A systematic review. *J Periodont Res*. 2018; 53: 288- 297. <https://doi.org/10.1111/jre.12525>
- Ramamurthy S, Thiagarajan K, Varghese S, et al. Assessing the In Vitro Antioxidant and Anti-inflammatory Activity of Moringa oleifera Crude Extract. *J Contemp Dent Pract* 2022;23(4):437-442.
- Gottumukkala SN, Koneru S, Mannem S, Mandalapu N. Effectiveness of sub gingival irrigation of an indigenous 1% curcumin solution on clinical and microbiological parameters in chronic periodontitis patients: A pilot randomized clinical trial. *Contemp Clin Dent* 2013;4:186-91
- MacAlpine, R., Magnusson, I., Kiger, R., Crigger, M., Garrett, S. and Egelberg, J. (1985), Antimicrobial irrigation of deep pockets to supplement oral hygiene instruction and root debridement. *Journal of Clinical Periodontology*, 12: 568-577. <https://doi.org/10.1111/j.1600-051X.1985.tb01390.x>
- Kumar, S. B. (2017). Chlorhexidine mouthwash-a review. *Journal of Pharmaceutical Sciences and Research*, 9(9), 1450.
- Yadav NR, Jain M, Sharma A, et al. Role of a miracle tree (*Moringa oleifera*) in healthcare. *J Evolution Med Dent Sci* 2021;10(21):1628-1632, DOI: 10.14260/jemds/2021/338
- Vinholis AH, Figueiredo LC, Marcantonio Júnior E, Marcantonio RA, Salvador SL, Goissis G. Subgingival utilization of a 1% chlorhexidine collagen gel for the treatment of periodontal pockets. A clinical and microbiological study. *Braz Dent J*. 2001;12(3):209-13. PMID: 11696921.
- Jelena Lecic, Sasa Cakic, Ognjenka Janjic Pavlovic, Ana Cicmil, OliveraVukotic, Vanja Petrovic & Smiljka Cicmil (2016): Different methods for subgingival application of chlorhexidine in the treatment of patients with chronic Periodontitis, *Acta Odontologica Scandinavica*
- Nayyar N, Dodwad V, Arora K. Comparative evaluation of 1% curcumin solution and 0.2% chlorhexidine irrigation as an adjunct to scaling and root planing in management of chronic periodontitis: A clinicmicrobiological study. *J Pharma Biomed Sci* 2012;14:1-7.
- Mizrak T, Guncu GN, Caglayan F, Balci TA, Akhtar GS, Ipek F. Effect of controlled release chlorhexidine chip on clinical and microbiological parameters and prostaglandins E2 levels in gingival crevicular fluid. *J Periodontol* 2006;77:437-43.
- Pistorius, A., Willershausen, B., Steinmeier, E. M., & Kreisler, M. (2003). Efficacy of subgingival irrigation using herbal extracts on gingival inflammation. *Journal of periodontology*, 74(5), 616-622.