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Efficacy of Spirulina Mouthrinse in Comparison to Chlorhexidine and Chlorine Dioxide in Patients Undergoing Orthodontic Treatment.

Masha Antony, Sai sri Soury, Dr.Murali Venkata Rama Mohan Kodali, P.L.Ravishankar

SRM Kattankulathur Dental College and Hospital,Chengalpattu,Tamil Nadu
SRM Kattankulathur Dental College and Hospital,Chengalpattu,Tamil Nadu
Associate Professor,College of Dentistry,King Faisal University, Al Ahsa-Kingdom of Saudi Arabia
SRM Kattankulathur Dental College and Hospital,Chengalpattu,Tamil Nadu

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ABSTRACT

Objective: The aim of the study was to determine the effectiveness of spirulina herbal mouthwash againstchlorhexidine and chlorine dioxide in preventing gingivitis among patients receiving fixed orthodontic treatment.

Methods: This comparative study was carried out with 30 patients undergoing fixed orthodontic treatment between the age group of 18 to 35 years. Patients were divided into 3groups -Group 1 patients used spirulina mouthwash, Group 2 used Chlorhexidine (CHX) mouthwash and Group 3 used chlorine dioxide mouthwash ,all of which were used for one minute twice daily. Plaque index and gingival index were recorded at 1st day, 30th day and 90th day intervals.

Results: All three groups showed a statistically significant reduction in mean gingival and plaque scores after 30 and 90 days. Significant difference noted in the mean gingival index score at 3months follow with highest mean gingival index score in the CHX group (0.378 ± 0.1873)and least mean score in chlorine dioxide group (0.295±0.139). At 1 month follow up CHX showed significant lower mean plaque index score(0.237±0.14) than spirulina and chlorine dioxide .

Conclusion: Spirulina mouthwashes seem to be effective in managing gingivitis in patients undergoing fixed orthodontic treatment. However, further well-designed and adequately powered clinical trials are necessary.

Key words- Chlorhexidine, Chlorine dioxide , Gingivitis, Gingival index , Plaque index, Spirulina.

I.INTRODUCTION:

Gingival inflammation is more common in patients receiving fixed orthodontic treatment (OT) because the bulkiness of fixed orthodontic appliances creates a niche for plaque accumulation .For orthodontic patients, mechanical plaque removal can be difficult due to which , different methods have been tried to reduce plaque formation , prevent gingivitis, and maintain good oral health**.**Orthodontic appliances shield plaque from the cleaning effects of brushing and chewing, allowing the plaque to remain undisturbed and potentially cause gingivitis, gingival hyperplasia, and periodontitis. [8]These conditions can hinder with orthodontic outcomes by causing separation of connective tissue and inhibiting remodeling. Improved antimicrobial treatments are desperately needed, according to a recent assessment of the literature, to avoid orthodontic treatment problems caused by biofilms. More precisely, it has been suggested that antimicrobial chemotherapeutic drugs such as Chlorhexidine (CHX) has been used in addition to the conventional oral hygiene regimen.[3] Although, extended use of these mouthwash has been linked to adverse effects such as burning sensations, hypersensitivity reactions, taste disturbance and changes in the colour of the tooth. However, due to reported adverse effects associated with CHX consumption, there is currently an increasing inclination towards the use of herbal mouthwashest[1].Herbal mouthwashes include antibacterial and anti-inflammatory qualities, this is another possible strategy for the maintenance of oral health in patients undergoing orthodontic treatment.

Spirulina, a photosynthetic cyanobacterium, possesses notable biological activity and is rich in a variety of essential and non-essential macro and micronutrients, making it an excellent nutritional supplement. Phycocyanin, a primary component of Spirulina, is present as a complex blend of trimers and hexamers. Additionally, Spirulina exhibits antioxidant properties, contributing to its potent anti-inflammatory effects. Recently, increasing evidence has highlighted the impact of oxidative stress (OS) caused by free radicals on periodontal

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tissues. While Spirulina has been evaluated in dentistry for its antioxidant properties in the treatment of conditions like oral submucous fibrosis and leukoplakia, its potential for reducing plaque and gingivitis has not been thoroughly investigated.[7]Thus, this is the first study of its kind assessing the effectiveness of Spirulina mouthwash on the reduction of dental plaque and gingivitis. With this background, The aim of this study is to compare the efficacy of spirulina mouthwash with Chlorhexidine(CHX) and chlorine dioxide on the fixed orthodontic patients.

II. MATERIALS AND METHODS :

The study comprised thirty patients undergoing orthodontic treatment at the Department of Orthodontics and Dentofacial Orthopaedics, SRM Kattankulathur Dental College & Hospital, Potheri, Tamilnadu. This study was approved by Medical Ethical and Methodological Committees of SRMIST. (SRMIEC-ST0324-977). Patients of age group 18 to 35 years with atleast 20 natural teeth, and a healthy state of periodontium were included. Patients who are suffering from any systemic disorders like hypertension, diabetes mellitus, liver diseases, epilepsy, convulsions or fainting spells and any history of allergy or hypersensitivity reactions to mouth washes have been excluded from the study. The nature of this study was explained to all patients and signed consent was taken before inclusion. Patients who met all criteria for entering the study were then randomized by using the sealed envelopes method into:

Group 1 (n = 10) was given Spirulina mouthwash
Group 2 (n = 10) was given Chlorhexidine mouthwash.(Hexidine ,ICPA Health Products Limited)
Group 3 (n = 10) was given Chlorine dioxide mouthwash (Freshchlor, Group Pharmaceuticals Ltd**)**Fig.1

Mouthrinses were randomly allocated among the groups and all groups received thorough scaling and follow the same oral hygiene instruction and were directed to use 10 ml of mouthrinse for 1 min . Patients were instructed to follow the instructions for complete study period. Clinical indices namely PI, GI were measured and recorded on a chart at baseline, 30 days, and after 90 days.

Preparation of Spirulina mouthwash:

[5][16] Spirulina mouthwash was prepared by dissolving 10 mg of spirulina extract (Heilen biopharma Pvt.Ltd, Gujarat, India) in 100ml of distilled water and sweetening agent(sorbitol),o.oo5% of flavouring agent (pepperemint oil) and pH was adjusted to 4.

FIGURE1- Spirulina organic powder, Chlorhexidine, Chlorine dioxide

Statistical Analysis:

The data were analyzed utilizing IBM SPSS version 20 software (IBM SPSS, IBM Corp., Armonk, NY, USA). Analysis involved one-way analysis of variance with Turkey's post hoc tests and repeated measures analyses of variance. Bar charts and line diagrams were employed for presenting the data.

III RESULTS:

Table 1 shows the comparison of gingival index scores between the study groups at various time points.No significant differences were noted between the groups in the mean gingival index scores at baseline and the 1 month follow-up time points. Significant differences were noted in the mean gingival index scores at the 3 months' follow-up . In the post hoc tests for multiple pair wise comparisons, Spirulina demonstrated no significant differences with either of the Chlorine dioxide and Chlorhexidine groups, however, a significant difference was noted between the latter two.Fig.2

Table 2 reveals the comparison of plaque index scores among the study groups at various time intervals. No significant differences were noted between the groups in the mean scores at baseline. Significant differences were noted in the mean plaque index scores at the 1 month and 3 months' follow-ups. Multiple pair wise comparisons at the 1 month follow-up demonstrated that Chlorhexidine had significantly lower mean plaque index scores than the Spirulina

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and Chlorine dioxide groups, while the latter two showed comparable plaque index scores. At the 3 months' follow-up, no significant differences were noted for Spirulina group with the other two groups however, Chlorine dioxide group had importantly higher mean plaque index scores than the Chlorhexidine group in pairwise comparisons.Fig.3

Table 3 demonstrates the intra group comparison of gingival index scores in each of the study groups between various points. Post-hoc analysis showed that the mean change in gingival index score from baseline to 1 month and 3 months follow-ups were statistically significant, but the mean change from 1 month to 3 months was not significant. In the Chlorhexidine and Spirulina groups, all pairwise comparisons were found to be statistically significant.

Table 4 shows the intra group comparison of plaque index scores in each of the study groups between various time points.Post-hoc analysis showed that the mean change in gingival index score from baseline to 1 month and 3 months follow-ups were statistically significant, but the mean change from 1 month to 3 months was not significant. In the Chlorhexidine and Spirulina groups, all pairwise comparisons were found to be statistically significant.

Table 1: Comparison of gingival index scores between the study groups at various time points

Time	Group	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		F value	P value
						Lower Bound	Upper Bound		
Baseline	Spirulina	10	0.952169	0.1346214	0 .0425710	0.855867	1.048471	0.949	0.4
	Chlorhexidine	10	0.836640	0.2509973	0.0793723	0.657087	1.016193		
	Chlorine dioxide	10	0.879140	0.1636875	0.0517625	0.762045	0.996235		
1 Month	Spirulina	10	0.532720	0.1246330	0.0394124	0.443563	0.621877	2.829	0.077
	Chlorhexidine	10	0.378870	0.1447344	0.0457690	0.275333	0.482407		
	Chlorine dioxide	10	0.532700	0.2172150	0.0686894	0.377314	0.688086		
3 Months	Spirulina	10	0.428990	0.1244278	0.0393475	0.339980	0.518000	8.765	0.001*
	Chlorhexidine	10	0.295770	0.1395113	0.0441173	0.195970	0.395570		
	Chlorine dioxide	10	0.578980	0.1837530	0.0581078	0.447531	0.710429		

One way analysis of variance; p≤0.05 considered statistically significant; * denotes significance; Groups with similar superscript presented with the mean had significant differences in Tukey's post hoc comparisons

Table 2: Comparison of plaque index scores between the study groups at various time points

Time	Group	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		F value	P value
						Lower Bound	Upper Bound		
Baseline	Spirulina	10	1.009970	0.1372080	0.0433890	0.911817	1.108123	3.08	0.058
	Chlorhexidine	10	0.883276	0.1664266	0.0526287	0.764222	1.002330		
	Chlorine dioxide	10	1.041639	0.1178527	0.0372683	0.957332	1.125946		
1 Month	Spirulina	10	0.56613	0.1629205	0.0515200	0.449590	0.682682	11.98	<0.001 *
	Chlorhexidine	10	0.2374	0.1403302	0.0443763	0.137074	0.337846		
	Chlorine dioxide	10	0.54997	0.1991493	0.0629765	0.407516	0.692442		
3 Months	Spirulina	10	0.399830	0.1042756	0.0329748	0.325236	0.474424	8.876	0.001*
	Chlorhexidine	10	0.24163	0.1072355	0.0339109	0.164927	0.318351		
	Chlorine dioxide	10	0.53334	0.2229315	0.0704971	0.373864	0.692816		

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One way analysis of variance; p≤0.05 considered statistically significant; * denotes significance; Groups with similar superscript presented with the mean had significant differences in Tukey's post hoc comparisons

Table 3: Intra group comparison of gingival index scores in each of the study groups between various time points

Group	Time	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		F value	P value
						Lower Bound	Upper Bound		
Spirulina	Baseline	10	0.9521	0.1346214	0.0425710	0.855867	1.048471	108.39	<0.001*
	1 month	10	0.5327	0.1246330	0.0394124	0.443563	0.621877		
	3 months	10	0.4289	0.1244278	0.0393475	0.339980	0.518000		
Chlorhexidine	Baseline	10	0.8366	0.2509973	0.0793723	0.657087	1.016193	31.73	<0.001*
	1 month	10	0.3788	0.1447344	0.0457690	0.275333	0.482407		
	3 months	10	0.2957	0.1395113	0.0441173	0.195970	0.395570		
Chlorine dioxide	Baseline	10	0.8791	0.1636875	0.0517625	0.762045	0.996235	32.6	<0.001*
	1 month	10	0.5327	0.2172150	0.0686894	0.377314	0.688086		
	3 months	10	0.5789	0.1837530	0.0581078	0.447531	0.710429		

Repeated measures analysis of variance; p≤0.05 considered statistically significant; * denotes significance; Groups with similar superscript presented with the mean had significant differences in Tukey's post hoc comparisons

Table 4: Intra group comparison of plaque index scores in each of the study groups between various time points

Group	Time	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		F value	P value
						Lower Bound	Upper Bound		
Spirulina	Baseline	10	1.0099	0.1372080	0.0433890	0.911817	1.108123	159.98	<0.001*
	1 month	10	0.56613	0.1629205	0.0515200	0.449590	0.682682		
	3 months	10	0.39983	0.1042756	0.0329748	0.325236	0.474424		
Chlorhexidine	Baseline	10	0.88327	0.1664266	0.0526287	0.764222	1.002330	160.43	<0.001*
	1 month	10	0.2374	0.1403302	0.0443763	0.137074	0.337846		
	3 months	10	0.24163	0.1072355	0.0339109	0.164927	0.318351		
Chlorine dioxide	Baseline	10	1.0416	0.1178527	0.0372683	0.957332	1.125946	43.25	<0.001*
	1 month	10	0.54997	0.1991493	0.0629765	0.407516	0.692442		
	3 months	10	0.53334	0.2229315	0.0704971	0.373864	0.692816		

Repeated measures analysis of variance; p≤0.05 considered statistically significant; * denotes significance; Groups with similar superscript presented with the mean had significant differences in Tukey's post hoc comparisons

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Figure 2: Comparison of gingival index scores between the study groups at various time points

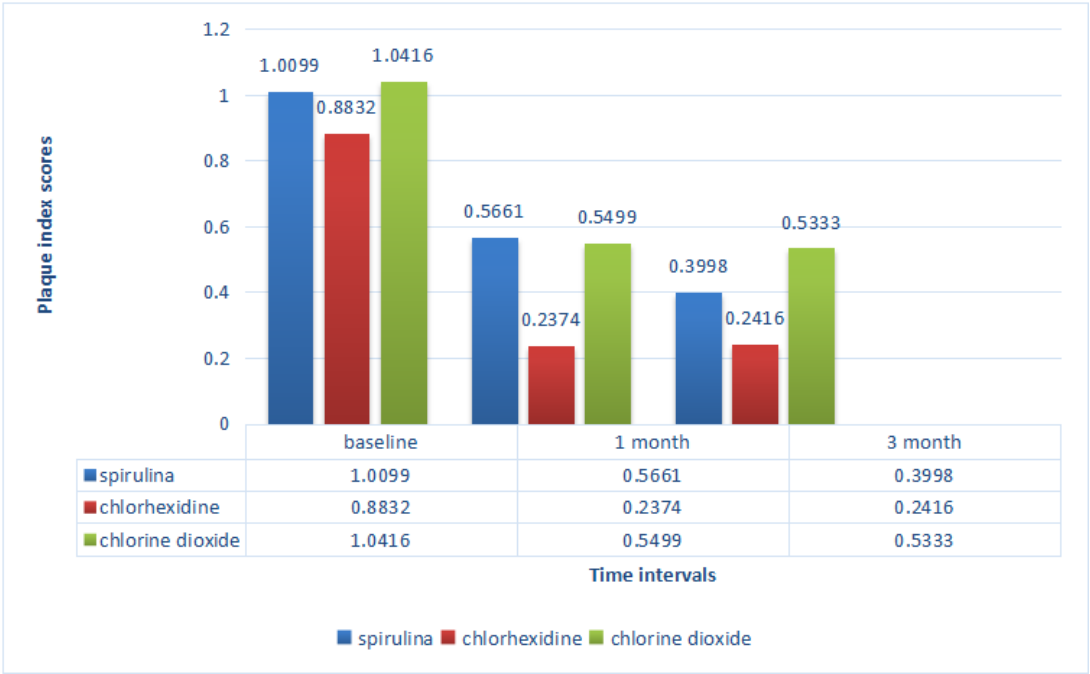

Figure 3: Comparison of plaque index scores between the study groups at various time points

IV.DISCUSSION

Preventing periodontal diseases and dental caries in orthodontic patients can be fulfilled through proper oral hygiene, which includes regular and thorough tooth brushing and rinsing with mouthwashes that contain antibacterial agents. The presence of brackets and arch wires hinders the effective removal of plaque and food debris. Consequently, dentists recommend the prolonged use of mouth rinses to prevent plaque buildup, gingivitis, and poor oral hygiene, which are common in patients undergoing orthodontic treatment. [6] [20] Chlorhexidine has shown the best clinical results in reducing plaque and preventing gingival inflammation, making it a standard mouthwash. However, its long-term use is limited due to numerous adverse effects. [10]cently, chlorine dioxide has emerged as a novel oral disinfectant. It has shown substantial results, demonstrating effective antibacterial properties in controlling gingivitis[10].

According to Gupta DA et al[4], naturally occurring phytochemicals isolated from medicinal plants used in traditional medicine can be effective

alternatives to conventional antibacterial agents. Many such medicinal plants and their derivatives are widely used for the prevention and management of diseases affecting the teeth and gums. Among these, Spirulina stands out due to its long history of medicinal use, dating back to biblical times, highlighting its significant medicinal value.

In this study, Spirulina was selected as a mouthwash due to its unique properties, including anti-inflammatory, immunomodulatory, antiviral, antibacterial, and antioxidant activities.[11] These properties are especially relevant for addressing the oxidative stress and inflammation induced by periodontitis, where endogenous gram-negative periodontal bacteria trigger a cascade of inflammatory responses in periodontal tissues, as noted by Hoseini and colleagues.[12] For the purpose of assessing clinical parameters in this study, a 3-month interval following non-surgical periodontal therapy has been selected for re-evaluation. [13] This is due to the fact that a period of three months is an appropriate amount of time to assess the effectiveness of nonsurgical periodontal therapy, which helps to fully resolve gingival inflammation and repair tissue. [5] Plaque index is used to evaluate patients' oral hygiene, while gingival index is used to evaluate the degree of gingival inflammation both before and after SRP . Phycocyanin (PC) and gamma-linolenic acid (GLA), two of Spirulina mouthwash's key ingredients, are responsible for the study's notable anti-inflammatory and anti-gingivitis effects. 1.3% of the dry weight of Spirulina is made up of natural blue pigment called C-PC, while the remaining 14% is made up of GLA. Upon reviewing the literature, it was discovered that no studies have been conducted on usage of Spirulina mouthwash till date to evaluate the same effect in vivo, hence the results of this study could not be compared with those of other studies.

[14][15]In the present study, the plaque and gingival indices showed a reduction at the end of 90 days in the Spirulina group, when compared to that of Chlorhexidine group which was not significant. Where in comparision with Chlorine dioxide the Spirulina proved to be effective This study indicates a noticeable reduction in plaque after each subsequent visit, with the most significantdecrease observed during the period from baseline to day 30 in all three groups.. A slight increase in the plaque and gingival score is seen in the next 90 days visit. In our study there was significant reduction in PI,GI at all evaluation periods after using CHX which is in the agreement with kripal et al and sherouk M et al.

All three groups experienced a statistically significant reduction in Pl score on both the 30days and 90 days follow up in the present study,whereas the mean GI scores at baseline and the one-month follow-up showed no significant changes between the groups.[17] However Chlorhexidine group showed the superior reduction followed by Spirulina and then chlorine dioxide groups.Gingival index of Spirulina group demonstrated no significant differences with either of the Chlorine dioxide and Chlorhexidine groups,but greater significant difference seen between CHX and Chlorine dioxide group which is in accordance with Paraskevas S et al who compared Chlorine dioxide mouthwash and Chlorhexidine mouthwash.

This study aims to determine the potential function of a natural product in gingival disease prevention and assess the product's effectiveness in comparison to CHX and chlorine dioxide. As demonstrated by the findings, mouthwashes work well against gingivitis and plaque prevention.[18] Nonetheless, when compared to mouthwash containing Chlorine dioxide, herbal mouthwash demonstrated superior outcomes in every clinical aspect evaluated which is in accordance with the study by Chandrahas et al., comparing a herbal mouthwash against a positive control and a placebo, the herbal mouthwash demonstrated a greater reduction in plaque and gingival index compared to the placebo group. However, it showed a reduction lesser than the positive control, although this difference was not statistically significant[19].Furthermore, the study conducted by Rahman et al. provides support for the efficacy of tea tree oil, an essential oil, as an anti-plaque agent when compared to Chlorhexidine.

[21]However, there was no significant difference between CHX and Spirulina at 3months follow up for both the gingival and plaque index indicating that these two treatments were similarly effective which coincides with the results of Rakhee sinha et al who compared the CHX and herbal mouthwashes. Based on the observed outcomes of this study, several recommendations can be made for clinical practice and further research on Spirulina mouthwash. Considering the similar efficacy of Spirulina mouthwash to that of other mouthwashes in reducing gingival and plaque index scores, it is suggested that it can be regarded as a viable supplementary treatment for chronic gingivitis in orthodontic braces patients who may experience adverse effects from Chlorhexidine or prefer natural alternatives. Increasing the sample size would enhance the statistical power of the study and offer a more comprehensive evaluation

of the mouthwashes' efficacy. Additionally, the long-term effectiveness and safety of regular use of these mouthwashes, particularly the herbal formulation, need further investigation.Our study is significant for being the first to compare the effectiveness of Spirulina mouthrinse with that of Chlorhexidine. The results indicate that Spirulina mouthwash may offer advantages similar to those of Chlorhexidine in some areas of oral health.

V.CONCLUSION:

According to the outcome of this study, it can be inferred that both Spirulina and Chlorine dioxide mouthwash showed similar beneficial effects and can be considered as a safe and effective herbal alternative to CHX as they demonstrated comparable reduction in the plaque buildup without any side effects. The active ingredients of Spirulina mouthwash containingtocopherols,beta carotene,phycocyanin and phenolic compounds helped in restraining plaque and gingivitis on both 30 th and 90 th days in patient receiving fixed orthodontic appliance therapy.It also has an array of antibacterial,anti-oxidant,anti-inflammatory and antifungal properties . Additional research should be conducted using larger sample sizes,different parameters and varying trial durations to confirm the effectiveness of this mouthrinse in preventing periodontal issues. This could pave the way for new avenues of research in oral healthcare.

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