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Comparative evaluation of efficacy of punica granatum(pomegranate) and chlorhexidine mouth rinse in reducing gingivitis - A randomized controlled trial.

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Abstract

Introduction: Periodontal diseases are chronic bacterial infections that lead to gingival inflammation, periodontal tissue destruction, and alveolar bone loss.Acting as powerful allies in the fight against periodontal disease, natural compounds can help safeguard against lethal age-related diseases that emanate from our mouths.

Punicagranatum (pomegranate) shrub has been used as an astringent, haemostatic, antidiabetic, antihelmintic, and also for diarrhea and dysentery.

AIM: To compare the efficacy of Punicagranatum (Pomegranate) and chlorhexidine mouth rinses on the reduction of gingivitis.

Materials and methods: Thirty subjects with the age between 18- 25 years old were selected and randomly divided into two groups: Group 1 - Pomegranate mouthwash and Group 2 – 0.2% chlorhexidine mouthwash. Punicagranatum mouth wash was prepared using raw Punicagranatum peel. Patients were instructed to use the prescribed mouthwash for 15 days. Clinical evaluation was undertaken using the gingival index, the plaque index, and bleeding on probing at baseline, and 15 days.

Results: The clinical study observed significant improveMent in gingival status and plaque status in both the sites ($P<0.05$).Subjectsusing Punicagranatum mouthwash showed significant improveMent in bleeding and gingivitis score and plaque score as compared with chlorhexidine.

Conclusion: Punicagranatum mouthwash is beneficial in improving gingival status due to its profound styptic action, with sufficient reduction in plaque scores.

Keywords: Anti-bacterial, Herbal mouthwash, Pomegranate, Gingivitis.

Introduction

Gingivitis is inflammation of the gums and is a non-destructive periodontal disease. The most common form of gingivitis is due to bacterial biofilms (plaque) which are adherent to tooth surfaces and hence known as plaque-induced gingivitis. In the absence of treatment, gingivitis may progress to periodontitis, which is a destructive form of periodontal disease. Bacterial plaque initiates the body's immune response which causes destruction of the gingival tissues, which leads to the destruction of the periodontal tissue. Plaque gets trapped in small gaps between the teeth and accumulates the bacteria in them, which produce proteolytic enzymes and toxins which causes an inflammatory reaction in the gingiva.[1] Gingivitis is an inflammatory response of the gingival tissues to dental plaque that can be treated by mechanical or chemical methods.[2] Mechanical methods such as tooth brushing and dental flossing treat gingivitis by removing dental plaque. Chemical methods such as, rinsing with chlorhexidine (CHX), treat gingivitis by inhibiting dental plaque.[3]

Another method for treating gingivitis is to use an anti-microbial mouth rinse. Antimicrobial mouth rinses treat gingivitis by inhibiting the formation of dental plaque or controlling the deleterious bacterial by-products.

Although numerous studies have been conducted on various chemical agents for controlling dental plaque, CHX has been referred to as the gold standard because of its substantivity. A gold standard is a positive control against which the effectiveness of other alternative agents is compared. Substantivity is the ability of a drug or agent to be absorbed and then released in active form hours after the initial dose. CHX possesses substantivity because it can be found in an active form in saliva and on oral surfaces 5 and 12 hours, respectively.[4]

In addition to the mechanical control of dental biofilms, it is possible to use chemical control, and chlorhexidine is the gold standard for this purpose. Chlorhexidine is a Di cationic compound that is able to join anionic compounds, such as phosphate and carboxyl radicals from the tooth surface and salivary glycoproteins. Its action damages the cytoplasmic membrane, leading to bacterial cell lysis. Additionally, chlorhexidine has a retention capacity and remains in oral tissues for a prolonged time, exhibiting a high substantivity. Despite its potential benefits, chlorhexidine displays some disadvantages after extensive use, such as taste alterations and teeth and oral tissue pigmentation.[5]

To remedy this problem, several herbal mouth rinse are available. Herbal drugs have long era of use and good patient tolerance as well as better public acceptance. Herbal drugs are renewable source which is our only hope for sustainable supplies of cheaper medicines for the worlds growing population. Availability of medicinal plants is easy in developing countries like India having rich agro-climatic, cultural and ethnic biodiversity. Throughout the world herbal medicine has provided many of the useful and vast variety of drugs to the modern medical science.[6] Mouth rinses are widely used to provide oral hygiene by degrading the plaque which is responsible for gingivitis. Several substances are used for their effectiveness on removing plaque which includes bisbiguanides, essential oils, enzymes, and herbal extracts. Certain substances produce side effects which limits their regular use. Hence, herbs can be used which does not have any side effects.[1]

Punicagranatum (Pomegranate) is currently finding important applications in the field of dental health. Clinical studies have shown that this popular antioxidant superstar attacks the causes of tooth decay at the biochemical level, with remarkable vigour. When used regularly in combination with toothpaste that has been reinforced with bioactive botanical extracts,pomegranate containing mouthwash may fight dental plaque and tartar formation by inhibiting the activities of the micro-organisms that cause plaque. Additionally, pomegranate compounds possess anti-inflammatory properties that may help soothe irritated tissues. Fascinating research shows that pomegranate extract suppresses the ability of these micro-organisms to adhere to the surface of the tooth. The trick is to inhibit a common species of Streptococcus, preventing it from producing chemicals that create favourable conditions for fungi and other micro-organisms to thrive. Plaque may involve four or more different microorganisms combining forces to colonize the surface of the teeth. Remarkably, nature's own pomegranate fights the organisms' ability to adhere by interfering with production of the very chemicals the bacteria use as "glue".[7]

Hence with this background, this present study was aimed to verify the efficacy of the use of 0.5% Punicagranatummouthrinse on the reduction of the supragingival dental biofilm.

Materials and methods

The study was conducted in a dental college at Madurai. A randomized, parallel arm, controlled trial study was carried out among 30 dental college students who were 18 – 25 years old in Madurai. Ethical clearance was obtained from institutional review board. Informed consent was obtained from the study participants before the study.

Participants who are 18 years and older and willing to participate in the study were included.Students with systemic disease, under anti-microbial therapy and allergic to chlorhexidine, as well as smokers, and individuals presenting periodontitis, i.e., pocket depths of 5 mm or more in more than 2 sites in the mouth were excluded from the trail.

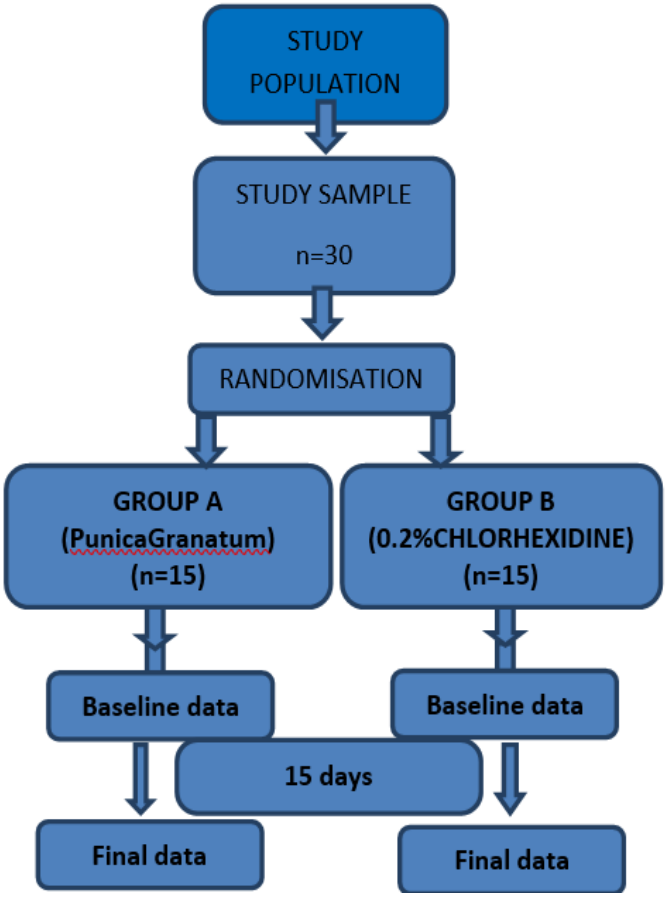

Figure 1: Study design - parallel arm, randomized controlled trial with two groups.

The sample size was estimated by using a previous study [8] and the number of participants required in each group is 15 for a significance level of 95% and power of 95%.

All examination was carried out by a trained and calibrated examiner. To assess intra- examiner calibration, the intra- class correlation coefficient (ICC) was calculated. The ICC was 0.92, indicating an excellent level of intra- examiner calibration.

Randomization was done to minimize "allocation bias". A box containing concealed envelopes was used for this purpose. Each envelope carried either code "A" or code "B". Participants who were selected to participate in the study were each asked to pick a concealed envelope from the box. Participants were allocated into Group 1 (n = 15, 0.2% Chlorhexidine gluconate mouthwash) and group 2(n=15, PunicaGranatum Mouthwash). All the Subjects were instructed to use 10 ml of their respective mouthwash twice daily for 15 days.

Commercially available 0.2% ChlorHexidine gluconate mouthwash (Rexidine, Batch number:RAQ7A63, Warren, Indoco) was used. Chlorhexidine was chosen as it is hailed as the "gold standard" mouthwash. Chlorhexidine exhibits both bactericidal and bacteriostatic effects depending on the concentration. However, the property of substantivity is a unique characteristic of this chemical plaque control agent.

Preparation of PunicaGranatum Mouth Rinse

The mouth rinse was prepared from whole fresh fruit peel that were cut into small pieces and blended with a mixture of ethanol and distilled water (1: 1, v/v). The material was filtered through several layers of gauze, and evaporated at 60°C to one-third of its original volume or until all the ethanol evaporated. The volume was restored with distilled water, and 1 ml sample was completely evaporated in the oven, in order to get the insoluble residues/ml (used to express the final hydroalcoholic concentration, which ranged between 50 and 60 mg/ml), which was further used to adjust the concentration of mouthwash.

Data collection

A special proforma was prepared to collect the required data. The proforma was prepared in English language. The first section of the proforma contained provision to record patient's demographic details such as Name, Age/Gender, Address and Phone Number. The second section contained recording charts to record gingival index[9] and plaque index.[10]

A single examiner, who was blinded to the treatment assignment, carried out all the clinical examination. The examination was conducted under artificial light using standardized instruments ADA specification type III examination. Gingival Index (GI) Modified Loe and Silness (1963)[10] and Plaque Index (PI). Modified Silness and Loe (1964)[11] indices were used in this study. The measurements were taken on four sites per tooth (mesial-buccal, buccal, distal-buccal and lingual) on all teeth except third molars and teeth with crown and bridge coverage. Index scores were averaged per tooth then added together and divided by number of teeth for the full mouth score.

Statistical analysis

The information collected regarding all the selected cases were recorded in a Master Chart. Data analysis was done with the help of computer using Statistical Package for Social Sciences (SPSS Inc., Chicago, IL, version 22 for Windows).Paired and unpaired 't' tests were used to test the significance of difference between quantitative variables. A 'p' value less than 0.05 is taken to denote significant relationship. Microsoft PowerPoint was used to prepare the graphs.

Results

A randomized controlled trial was conducted to compare the efficacy of Punicagranatum mouth rinse and chlorhexidine on the reduction of gingivitis. A total of 30 dental students were included in the study and who were divided into two groups. Socio-demographic data revealed that the mean age of the participants was 19.5 years. Majority of the participants were aged 18 years (43.3%). All participants were equally distributed among both genders. (Table 1)

Table 2 reveals that the comparative efficacy of mouth rinses in relation to plaque index scores. There is a statistically significant difference of plaque scores in all the two groups after using mouth rinse. Intergroup comparison reveals that statistically significant difference seen when punicagranatum mouth rinse is compared with control group

Table 3 reveals the comparative efficacy of mouth rinses in relation to gingival index scores. A statistically significant difference was observed between both groups after using mouth rinses.

Intergroup comparison reveals statisticallysignificant difference between PunicaGranatummouth rinse and chlorhexidine group.

Table 1: Socio demographic details of the study participants.

Characteristic		**Percentage (n)**
Age	18 years	43.3% (n=13)
	19 years	36.7% (n= 11)
	20 years	13.3% (n= 4)
	21 years	6.7% (n=2)
Gender	Males	50% (n=15)
	Females	50% (n=15)

Table 2: Comparative Efficacy of mouth rinses in Relation to Plaque Index Scores.

Groups	Before(mean +SD)	After(mean + SD)	P value(Paired t test)
Punica granatum	0.90 + 0.35	0.540 + 0.23	0.00**
Chlorhexidine	1.01 + 0.12	0.66 + 0.15	0.004**
p value(Unpaired t test)	0.508	0.044**	

**p Value < 0.05 is statistically significant

Table 3: Comparative Efficacy of mouth rinses In Relation to Gingival Index Scores.

Groups	Before(mean + SD)	After(mean + SD)	P value(Paired t test)
Punica granatum	0.86+_ 0.22	0.45+_ 0.18	0.00**
Chlorhexidine	1.00+_ 0.30	0.56+_0.14	0.032**
p value(Unpaired t test)	0.227	0.038**	

**p value < 0.05 is statistically significant

Discussion

In the recent years, the use of plants with preventive and therapeutic effects contributes to health care needs. There are three main reasons to be interested in the treating and healing power of plant extract. First, pharmacological studies have demonstrated that many of plants are known to possess anti-microbial agents; second, people are becoming aware of the side effects associated with the over prescription of traditional antibiotics; third, time to time resistant micro-organisms against antibiotics are increasing.[12,13] Among these plants, punica granate has an important role in folk medicine.

Pomegranate is known as a rich source of pharmacological properties which have been evaluated due to anti- parasitic, anti-bacterial, anti-fungal, anti-proliferative, apoptotic and anti-cancer effects as well as

protection against herpes virus, inhibition of LDL oxidation and decrease in atheromatous plaque formation and reduction of systolic blood pressure.[12,14,15]Though around 6000 plants in India are used in herbal medicines, little research has been conducted on efficacy, safety, and properties of herbal products. Over the decades, very few studies have been conducted to show the clinical efficacy of Punicagranatum. In the present study, it was demonstrated that hydroalcoholic extract from pomegranate fruit exerted a significant reduction in clinical parameters.

Analysis of plaque index values suggests that both the mouthwashes were helpful in reducing it but chlorhexidine reduced plaque scores to lower extent as compared with pomegranate mouthwash group.[16]

These findings are in agreement with the studies Menezes et al. showed that after 1 minute mouth rinsing, more reduction in plaque was observed with Punicagranatum (84%) as compared with chlorHexidine (79%).[17]

Analysis of gingival index scores revealed that Punicagranatum was more efficient in reducing gingival score and bleeding on probing as well due to its strong styptic action. Similar results were reported by Hafajee et al.[16]

Contradicting our results, Analysis of plaque index values suggests that both the mouthwashes were helpful in reducing it but chlorhexidine reduced plaque scores to greater extent as compared with pomegranate mouthwash group. These findings are in agreement with the studies by Overholser et al.[18] and Haffajee et al.[16] A study by Salgado et al. in 2006 on 10% Punicagranatum gel does not support our finding as this gel was not efficient in preventing supra- gingival dental plaque formation and gingivitis. [19]

In an urge of looking for better antiplaque and anti-gingivitis agents with limited side effects as compared with chlorhexidine, various herbal products have been tried with fruitful results. Punicagranatum is a recent herbal product used in field of dentistry. So, more clinical and microbiological studies on a long-term basis are required to know the precise effectiveness of this product.

However, the limitation of the present short-term study was conducted with a small sample size on the age group between 18 – 25 years old individual. The present study recommended that further studies to be conducted to evaluate retention over a longer period of time and could be applied to other ages. Also, it recommended that Incorporation of Punicagranatum (Pomegranate) mouth rinse in routine hygiene practices will be Feasible and it has positive impact on gingival health.

Conclusion

Based on the results, the use of Punicagranatum (Pomegranate) mouth rinse produced additional reductions in the supra- gingival biofilm and gingivitis relative to the use of conventional chlorhexidine mouth rinse.

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