International Journal of Drug Research and Dental Science
Volume 3 Issue 2 (Page: 1-34), 2021 ISSN: 2582-0826

"Comparative Evaluation of Clinical Changes and Microbial Flora Associated with Usage of Mouth Washes Containing Green Tea, Chlorhexidine (0.2%) and Essential Oils in Patients Undergoing Orthodontic Therapy"

Krishna Priya. B[1], Anil Melath[2], Anjali K[3], Mohammed Feroz T. P[4], Subair. K[5], Nanditha Chandran[6]

[1]Post-Graduate, Department of Periodontics, Mahe Institute of Dental Sciences and Hospital, Chalakkara, Kerala, India.

[2]Professor and HOD, Department of Periodontics, Mahe Institute of Dental Sciences and Hospital, Chalakkara, Kerala, India.

[3]Assistant Professor, Department of Microbiology, Mahe Institute of Dental Sciences and Hospital, Kerala, India.

[4,5]Reader, Department of Periodontics, Mahe Institute of Dental Sciences and Hospital, Chalakkara, Kerala, India.

[6]Senior Lecture, Department of Periodontics, Mahe Institute of Dental Sciences and Hospital, Chalakkara, Kerala, India.

Original Article

Address for Correspondence Author
Dr. Krishna Priya. B; Post-Graduate, Department of Periodontics, Mahe Institute of Dental Sciences and Hospital, Chalakkara, Kerala, India.
E-mail: krishnapb1993@gmail.com
Crossref doi: https://doi.org/10.36437/ijdrd.2021.3.2.A

ABSTRACT

Introduction: Periodontal disease results from a complex interplay between the subgingival biofilm and the host immune-inflammatory events that develop in the gingival and periodontal tissues in response to the challenge presented by the bacteria 1 an increasing number of people all over the world are turning to nature by using natural herbal products in both prophylaxis and treatment of different diseases. Plants are the source of more than 25% of prescription and over-the-counter preparations and the potential of natural agents for oral prophylaxis should therefore be considered.

Objectives: To evaluate the clinical changes before and after the use of Green tea, Chlorhexidine (0.2%), and Essential oil mouthwashes in patients undergoing orthodontic therapy. To evaluate the antimicrobial properties of the three mouthwashes in patients undergoing orthodontic therapy. To compare the clinical changes and antimicrobial properties of the three mouthwashes in patients undergoing orthodontic therapy.

Methodology: Sixty patients of age group 13-35 yrs with minimal crowding were selected for this study. Subjects with minimal crowding were selected based on the Little's Irregularity Index score range up to 3. The gingival health status and presence of bleeding on probing were assessed by Gingival index by Loe and Silness (1963) and Papillary bleeding index (PBI) by Muhleman and Saxer (1975) 64 after 2 weeks of

International Journal of Drug Research and Dental Science
Volume 3 Issue 2 (Page: 1-34), 2021 ISSN: 2582-0826

placement of orthodontic brackets. Plaque samples were collected and sent for microbiological analysis to estimate colony-forming units. The subjects were randomly allocated to one of the following groups. Each group consisted of 20 subjects.

Group I: Green tea mouth wash (10ml to be rinsed for 30 seconds twice daily)
Group II: Chlorhexidine mouth wash (0.2%) (10ml to be rinsed for 30 seconds twice daily)
Group III: Essential oil mouth wash (10ml to be rinsed for 30 seconds twice daily)

After the 14th day and 21st day, the subjects were recalled and the gingival and bleeding indices were recorded. Supragingival plaque sample was collected using a sterile jaquette scaler for microbiological analysis.
Results: When the gingival index was compared on the baseline, 14th, and 21st day between the three groups, it was observed that the chlorhexidine group showed a decrease in the gingival index followed by green tea mouthwash and essential oil mouthwash. When the papillary bleeding index was compared between the three groups on baseline, 14th day and 21st day it was observed that chlorhexidine shows the least papillary bleeding index followed by green tea mouth wash and essential oil mouthwash. A comparison of the total CFU of various mouthwashes in this study revealed that 0.2% CHX mouthwash was the most effective in reducing the total microbial colony count (33.96% reduction in total bacterial colony count from baseline to 14th day and 51.42% reduction in total bacterial colony count from 14th day to 21st day), followed by green tea mouth wash (15.8% reduction in total bacterial colony count from baseline to 14th day and 27.37% reduction in total bacterial colony count from 14th day to 21st day) and essential oil mouth wash (6.39% reduction in total bacterial colony count from baseline to 14th day and 2.6% reduction in total bacterial colony count from 14th day to 21st day).
Conclusion: It was concluded that all three mouthwashes in this study are equally effective by showing significant clinical and microbiological changes after the usage in patients undergoing orthodontic therapy. Chlorhexidine mouthwashes showed better clinical and microbiological changes after the usage followed by green tea mouthwashes and essential oil mouthwashes. Green tea mouthwash can be recommended as a safe anti-inflammatory, antimicrobial mouthwash to control gingival inflammation and to maintain good oral hygiene during orthodontic treatment.
Keywords: Chlorhexidine, Essential Oil, Green Tea, Mouthwash, Supragingival Plaque.

Introduction

Periodontal disease results from a complex interplay between the subgingival biofilm and the host immune-inflammatory events that develop in the gingival and periodontal tissues in response to the challenge presented by the bacteria.[1] Plaque plays an important role in the development of gingivitis when in contact with the gingival tissues and therefore, plaque control represents the cornerstone of good oral hygiene practices. The tools that are commonly used in mechanical supragingival plaque control are the toothbrush (Manual or electric, floss, wood sticks, and interdental brushes). Despite the availability of these various oral hygiene devices, even the most meticulous patient will not completely remove all plaque.[2]

High gingivitis prevalence is commonly found in orthodontic patients.[3] The presence of brackets, bands, and other accessories as well as composite resin restorations and cements used to bond them, facilitate biofilm build-up and hinder its removal by patients, favoring enamel demineralization and gingivitis and promoting qualitative and quantitative changes in the oral microbiota.[4]

Orthodontic appliances also reduce the effect of brushing on plaque and salivary flow. Increased levels of Streptococcus mutans and lactobacilli are detected in the oral cavity after bonding of orthodontic attachments, furthermore metallic brackets have been found to make specific changes in the oral environment, such as a decrease in pH and affinity of bacteria to a metallic surface because of electrostatic reactions.[5] Difficulties to achieve ideal mechanical plaque control have led many researches to search for other methods such as chemical agents, which could contribute to daily biofilm control, removal or inhibition of plaque formation on tooth surfaces. Current evidence shows that when chemical agents are used as adjuncts to brushing and flossing, they can promote additional advantages compared to mechanical control alone as regards and safety of these products.[6]

Many of the chemical antiplaque agents in various formulations have been tried as an adjunct to mechanical measures for improving oral health. These antiplaque agents in various formulations have been tried as an adjunct to mechanical measures for improving oral health. These antiplaque agents can be delivered in the form of mouthwashes, dentifrice, chewing gums, gels, and chips. Mouthwashes, a safe and effective delivery system for antimicrobials can play an important role in plaque reduction.[7] Out of antiplaque agents, chlorhexidine is considered a gold standard agent for its clinical efficacy in chemical plaque control.[8] It has broad antibacterial activity, with very low toxicity and a strong affinity for epithelial tissues and mucous membranes. Besides its antiplaque effect, chlorhexidine is substantive, thus reducing levels of micro-organisms in saliva up to 90% for several hours.[9]

The ability of essential oil mouthwash to reduce plaque can be attributed to its broad antimicrobial range, its ability to penetrate plaque and kill the bacteria inside the biofilm. However, the alcohol content of essential oil rinses and their unpleasant taste is unacceptable to some patients. Thus none of these chemical agents is without shortcoming. So the search for an ideal and safe antiplaque agent continues.[10]

An increasing number of people all over the world are turning to nature by using natural herbal products in both prophylaxis and treatment of different diseases. Plants are the source of more than 25% of prescription and over-the-counter preparations and the potential of natural agents for oral prophylaxis should therefore be considered. Green tea is one of these, green tea is made solely with the leaves of camellia Sinensis that had undergone minimal oxidation during processing. The most abundant components in green tea are polyphenols, in particular flavonoids such as catechins, catechin gallates, and proanthocyanidins.

Studies conducted in the past have shown that the green tea polyphenolic catechins can inhibit the growth of a wide range of gram-positive and gram-negative bacterial species with moderate potency`.[11] A modest inverse association between the intake of green tea and periodontal disease has also been shown and the authors suggested that the application of concentrated green tea components, such as catechin may have a more beneficial effect on the periodontal conditions.[12] Though the literature shows the biological activities of green tea, much research has not to be done regarding its use as an antiplaque agent. Therefore the present study was performed to evaluate and compare the clinical changes and microbial flora associated with the usage of mouthwashes containing green tea, chlorhexidine (0.2%), and essential oils in patients undergoing orthodontic therapy.

Aim and Objectives

Aim of the Study: To evaluate and compare the antimicrobial potential of mouth washes containing Green tea, Chlorhexidine (0.2%) and Essential oil in patients undergoing orthodontic therapy.

Objectives of the Study

- To evaluate the clinical changes before and after the use of Green tea, Chlorhexidine (0.2%), and Essential oil mouthwashes in patients undergoing orthodontic therapy.
- To evaluate the antimicrobial properties of the three mouthwashes in patients undergoing orthodontic therapy.
- To compare the clinical changes and antimicrobial properties of the three mouthwashes in patients undergoing orthodontic therapy.

CD Overholser et al,.(1990) Conducted a double-blinded controlled clinical study in 128 healthy adults subjected to determine the efficacy of 2 mouth rinses, Listerine, and peridex in reducing the supragingival dental plaque and gingivitis. Following screening examination for entry levels of existing gingivitis, the plaque, baseline gingival and plaque area indices, extrinsic tooth stains, supragingival calculus, bleeding, and soft tissue conditions were recorded and the results showed that Listerine patients did not develop a significant level of stain or supragingival calculus at 6 months and peridex patients developed significant levels of extrinsic strains and supragingival calculus and the peridex was more effective than Listerine in the control of plaque.[13]

GB Anderson et al,.(1997) compared the short-term clinical effect of 0.12% CHX gluconate and placebo mouth rinses in 30 adolescents undergoing orthodontic treatment. Plaque index, gingival index, retention index, discoloration index, and probing depth were determined and the results showed that the use of CHX in addition to regular oral hygiene habits was effective in reducing plaque and gingivitis in adolescents undergoing orthodontic treatment.[14]

Hiraswa M et al,.(2002)- Did a study to determine the usefulness of green tea catechin for the improvement of periodontal disease. The minimum inhibitory concentration (MIC) and bactericidal activity of green tea catechin against black- pigmented gram–ve anaerobic rods (BPR) were measured. Hydroxypropyl cellulose strips containing green tea catechin as a slow-release local delivery system were applied in pockets in patients once a week of study. The clinical, enzymatic, and microbiological effects of the catechin were determined. Green tea catechin showed a bactericidal effect against black BPR and the combined use of mechanical treatment and the application of green tea catechin using a slow-release local delivery system was effective in improving periodontal status.[15]

Okamotom et al,.(2004)- Conducted a study to examine the effects of catechins and their derivatives on the activities of Arg-gingipain (Rgp) and Lys-gingipain (Kgp) in porphyromonas gingivalis. catechin derivatives which include (-) epigallocatechin gallate, (-) epicatechin gallate (-) gallocatechin gallate, (-) catechin gallate, significantly inhibited the Rgp activity. The 50% inhibitory concentrations (IC5OS) of these catechin derivatives for Rgp ranged from 3-5 micron while (-) epigallocatechin and (-) gallocatechin moderately inhibited Rgp activity (IC5OS, 20 microns) (-) epicatechin, (+) catechin, and gallic acid was not effective, with IC5OS greater than 300micron. These findings suggested that green tea catechins may have the potential to reduce periodontal breakdown resulting from the potent proteinase activity of P.gingivalis.[16]

Negestani et al,.(2007)- Studied the anti-streptococcal effects of black tea extract were evaluated and compared with those of green tea. in vitro evaluation of antibacterial effects was determined using high-performance liquid chromatography to compare their major polyphenol profiles. Different concentrations of the extracts or gallic acids, the abundant phenolic compound found in black tea, were used for bacterial

sensitivity tests in both pour plate and disc diffusion methods and results showed a synergetic effect of black and green tea against the streptococcus pyogenes.[17]

Yagakshi K et al,.(2008)- Determined the effect of Hop polyphenols (HPP) on water-insoluble glucan (NIG), which is a major component of dental plaque along with micro-organisms, and the effect of HPP containing tablets on the growth of dental plaque. The effects of HPP on streptococcus mutans MT 8148 were determined. HPP concentrations employed in this study were 0% (as HPP control), 0.001%, 0.01%, 0.1%, and 0.5% and the results showed by after 24 hr incubation, 0.5% of HPP significantly reduced the growth of S. mutans compared to control ($P<0.01$) and it was concluded that HPP tablets might be a significant means of delivery HPP onto tooth surfaces to prevent dental plaque formation.[18]

Tuteka et al.,(2008)- Conducted a study to determine the effectiveness of an essential oil mouth rinse in improving oral health in orthodontic patients. Patients within their first 6 months of orthodontic treatment were assigned either to the brushing and flossing or brushing, flossing, and use of Listerine. Measurements were recorded for bleeding, gingival, and plaque indices that provided baseline values, Subsequent measurements were taken at 3months (T2), and 6 months (T3), and the results showed that the use of Listerine mouthwashes can reduce the amount of plaque and gingivitis in patients undergoing orthodontic treatment.[19]

AS Hassani et al,.(2012)- did a study to evaluate the efficacy of semi-fermented and non-fermented Camellia Sinensis extracts (Black and green tea) against S. mutans ATCC 25175, S. mitis ATCC9616, and S. sanguis ATCC 10556. The minimum inhibitory concentration of both extracts was assessed by well diffusion and broth dilution methods and biofilm inhibitory concentration was observed on glass slides under phase contrast microscope and colony counts from glass beads. the results showed that the semi-fermented and non-fermented Camellia Sinensis extracts were able to prevent the growth of oral streptococci.[20]

Pandis et al,.(2010)- Studied to investigate the effect of bracket type (conventional and self-ligating) on the levels of S. mutans and total bacterial counts in the whole saliva of orthodontic patients and results showed the level of S. mutans in the whole saliva of orthodontically treated patients do not seem to be significantly different between conventional and self-ligating brackets.[21]

Adwall et al,.(2011)- Conducted a study to assess the possible protective properties of green tea on oral health. The researchers used the following measurements: streptococcus mutans count in saliva and plaque. salivary and plaque pH values, gingival bleeding index (GBI). The above-mentioned measurements were applied to a sample consists of 25 subjects before and after rinsing with green tea for 5 min and the results of this showed a marked reduction in gingival bleeding and inhibition of salivary and plaque streptococcus mutans growth by preserving the plaque pH towards neutrality.[22]

Faira et al,.(2011)- Done a study to compare the antimicrobial effect of mouthwashes containing calendula Officinalis, camellia sinesis, and 0.12% CHX digluconate on the adhesive of micro-organisms suture materials after extraction of unerupted third molars. 18 patients with unerupted maxillary third molars indicated for extraction were selected. The patients were subjected to extracted on left tooth instructed not to use any type of antiseptic solution at the site of surgery (control group). and the results showed a reduced number of micro-organisms adherent to the sutures compared to the control group. A significant difference was observed in the 0.12% CHX gluconate group.[23]

Moghber et al,.(2011)- done a study to know the effect of green tea on the prevention of mouth bacterial infection, halitosis, and plaque formation on teeth. A comparative study was conducted on a green tea mouthwash containing 1% tannin with 10% ethanol, alcohol-free mouthwash, and a green tea herbal mouthwash with a CHX 0.2% and the results showed that green tea mouthwash could reduce the aerobic mouth bacterial load and may prevent plaque formation on teeth and come over halitosis due to infection of the bacteria.[24]

MH Tehrani et al,.(2011) Conducted a study to compare the effect of sodium fluoride and green tea mouth rinses on the level of salivary Streptococcus mutans and lactobacilli in children, and the results showed that green tea mouth rinses significantly reduce the colony number of S. mutans and lactobacillus which is comparable with sodium fluoride mouth rinse.[25]

Kuduva P et al,.(2011)- Evaluated the adjunctive use of locally delivery green tea catechin with scaling and root planning as compared to scaling and root planing alone in the management of chronic periodontitis a clinico-microbiological study was done by assessing the plaque index, gingival index, probing pocket depth at baseline and 21 days and for microbiological analysis at baseline, 1 week and 21 days and the results showed inter-comparison of the plaque and gingival index for test and control groups at 21 days was not significant with $p>0.05$, whereas the probing depth at 21 days was significant with $p<0.001$, inter-comparison between microbial results demonstrated a considerable reduction of the occurrence of AA, Prevotella intermedia, Fusobacterium and Capnocytophaga.[26]

Xu et al,.(2012)- Conducted a study to know the effect of Epigallocatechin gallate (EGCG)on the sucrose-dependent initial attachment of S. mutans UA159 a chemically defined medium was monitored over 4 hrs using a chamber side mode. The effects of EGCG on the aggregation and gtf B, C, D gene expression of S. mutas UA159 were also examined and results found that EGCG exhibited dose-depended inhibition of the initial attachment of S mutans UA159.[28]

Eckley et al,.(2012)- Done a study to determine the benzoyl-DL- arginine- naphthylamide test scores of periodontopathogenic bacteria including that of red-complex bacteria, p .gingivalis, T. denticola, and Tanneralla forsythia, as well as organism morphotypes, probing depth, plaque score in patients undergoing orthodontic treatment and the results showed a significant increase in plaque score, probing depth, and benzoyl –DL- arginine –naphthylamide scores were found at each interval after placement of orthodontic appliances. However, the levels returned to baseline after the removal of the appliances.[29]

N Jenabian et al,.(2012)- conducted a study to determine the effect of Camellia sinesis (green tea) mouthwash on plaque-induced gingivitis- A single-blinded randomized controlled trial was done in patients with chronic generalized plaque-induced gingivitis. Patients received either 5ml of green tea, 5% two times per day, or normal saline with the same dosage. gingival index, plaque index, and bleeding index were recorded at baseline and five consecutive weeks, and the results showed a significant improvement was observed in all periodontal indices during the study. The total amount of improvement was higher in the mouthwash group.[30]

Balappanavar et al,.(2013)- Evaluated and compared the effectiveness of 0.05% tea, 2% neem, and 0.2% chlorhexidine mouthwashes on 30 healthy human volunteers. plaque accumulation and gingival condition were recorded using plaque index and gingival index. oral hygiene was assessed by simplified oral hygiene index (OHIS), salivary PH was assessed by indikrom Ph strip and the results showed a mean plaque and

gingival scores were reduced over the three week trial period for the experimental and control group. Anti-plaque effectiveness was observed in all groups. neem and green tea showed comparative effectiveness on gingival better than CHX.[31]

Morozumi et al,.(2013)- Conducted a study to detect the effect of essential oils in combination with subgingival ultrasonic instrumentation and mouth rinsing on chronic periodontitis patients. The subgingival plaque was collected from a total of 90 pockets across all subjects subsequently, subgingival ultrasonic instrumentation was performed by using EO or saline for 7 days. Subgingival plaques were sampled. Periodontopathogenic bacteria were qualified using the modified invader plus assay. The total bacterial count in shallow pockets (probing depth (PPD) =4-5mm) was significantly reduced in both saline ($P<0.05$). The total bacterial count ($p<0.05$) and P. gingivalis ($P<0.01$) and T. forsythia ($P<0.05$) count in deep pockets (PPD≥6mm) were significantly reduced in the EO group.[32]

Rarsamarnasmosmaug et al,.(2013)- Conducted a study to determine the effects of green tea mouthwash on oral malodor, plaque, and gingival inflammation. gingivitis patients who had over 80 parts per billion of volatile sulfur in morning breath were randomly assigned into green tea or placebo mouth wash group and the results showed that green tea mouthwash significantly reduce the volatile sulphur level in gingivitis subjects after rinsing for 4 weeks.[33]

A Araghizadrh et al,.(2013) done a study to determine the in vitro inhibitory activity of green tea extract on some clinically isolated cariogenic and periopathogenic bacteria. 20 strains of each S. mutans, A.A, P. gingivalis, P. intermedia were isolated from carious teeth and periodontal pockets of patients with dental caries and periodontal diseases. Green tea extract was prepared by aquous extraction method and diluted from 50-1.50mg/ml. standard techniques of agar disc diffusion and broth microdilution assays were applied for qualitative and quantitative determinators of antibacterial activity of green tea extract on each isolates and the results showed that the minimal inhibitory concentration of green tea extract for S. mutans was 3.28±0.7mg/dl and A. A, P. gingivalis, and P. intermedia.[34]

Sankanaka S et al,.(2014)- Investigated the effect of polyphenolic compounds isolated from green tea (camellia sinesis) on the growth and adherence of Porphyromonas gingivalis into human buccal epithelial cells. Green tea polyphenols, especially (-)-epigallocatechin gallate (EGcg) which is a dominant component of tea polyphenols completely inhibit the growth and adherence of P. gingivalis on to the buccal epithelial cells at concentration 250-500µg/ml.[35]

Santos et al,.(2014)- Conducted a study to determine the use of 0.2% CHX as mouthwash would decrease air contamination caused by aerosolized sodium bicarbonate during dental prophylaxis, and results showed that 0.12% CHX mouthwash significantly reduces the contamination caused by aerosolized sodium bicarbonate during dental prophylaxis in the orthodontic clinic.[36]

R Shahakbari et al,.(2014)- did a study to evaluate the effectiveness of green tea mouthwash in controlling the pain and trismus associated with acute pericoronitis in comparison to CHX mouthwash and the results showed that the green tea mouth rinse could be an approximate and effective choice for the control of pain and trismus in acute pericoronitis.[37]

Sriparna et al,.(2015)- Done a study to evaluate and compare the effects of commercially available green tea mouthwash with Listerine and chlorhexidine mouthwash in gingivitis patients. The plaque index was

recorded at baseline followed by scaling, and at the 14th and 21st day postoperatively and the results demonstrated that green tea to be equally effective in reducing the periodontal indices as chlorhexidine. A significant reduction was seen in plaque, gingival and bleeding indices in all the groups.[38]

Priya et al,.(2015)- Conducted a study to compare the efficacy of the mouthwash containing green tea and chlorhexidine in the management of dental plaque-induced gingivitis, 30 patients who participated in this study were divided randomly into two groups, each group of 15 patients was prescribed with either chlorhexidine or green tea mouthwash. Plaque index, gingival index were recorded at baseline, 15 days, and 1 month and the results showed a significant decrease in plaque index, gingival index, and bleeding index in both the groups.[39]

Quintas V et al,.(2015)- Evaluated the in situ antiplaque effects after 4 days using 2 commercial antimicrobial agents in short term on undisturbed plaque-like biofilm. A crossover randomized clinical trial on 15 oral and systemically healthy volunteers between 20-30 years who were randomly and sequentially allocated in the same group performed 3 interactions in different randomized sequences. The participants were on an appliance in 3 different rinsing periods doing mouthwashes twice a day with essential oils, 0.2%CHX, or sterile water (negative control). At the end of each 15 day mouth wash period, samples were removed from staining. samples were analyzed using a confocal laser scanning microscope and the results showed that the essential oils and 0.2% CHX were significantly more effective than the sterile water at reducing bacterial vitality, thickness, and covering grade by the biofilm.[40]

Ardakani et al,.(2015)- Investigated in vitro and in vivo antibacterial effects of 3 mouthwashes on supragingival plaque microbiota. The three mouthwash under the study were 0.2% CHX, Listerine K, Persica (PM), water was used as a negative control. Plaque samples are collected and zone of inhibition was measured after incubation for 24 hrs and CFUS were counted and the results showed that 0.2% CHX inhibited the growth of bacteria to an average diameter 18-38mm, while Listerine R, PM, and water caused no inhibition of bacterial growth around the discs after 24 hrs.[41]

Dehgani et al,.(2015)- Conducted a study to investigate the effects of combined mouth rinses containing CHX and NaF on clinical oral hygiene parameters and plaque bacterial level. A double-blind clinical study was conducted in 60 fixed orthodontic patients group 1 combined CHX/NAF, group 2-CHX 0.06%, group 3 NaF0.05%, group 4-placebo. Bleeding index, modified gingival index, plaque index was determined at baseline, after 3 weeks of rinsing. the supragingival plaque was obtained and bacterial colony count is determined and the results showed that CHX and the CHX/NaF induced significantly more changes than NaF and placebo and in microbiological measurement, except placebo other mouth rinses reduced total bacterial count.[42]

Hambire et al,.(2015)- Compared the antiplaque efficacy of 0.5% Camellia sinesis extract, 0.05% NaF, 0.2%CHX gluconate mouth wash in children. subjects were randomly assigned in to 3 groups. group A -0.2% CHX gluconate, group B 0.05% NaF, group C 0.05% camellia sinesis extract. Plaque accumulation and gingival condition were recorded using plaque index and gingival index. oral hygiene was assessed by simplified OHIS, salivary PH was also assessed and the results showed more effectiveness of 0.5% camellia sinesis when compared to 0.05% NaF and 0.2%chlorhexidine gluconate mouth rinses.[43]

Sargolzair et al,.(2015)- Conducted a study to compare green tea -aloe vera mouth wash and CHX 0.2% on gingival indices. A total of 60 patients with periodontal disease were randomly allocated into one of the

three double-blind groups, 20 in each, to receive the following treatment (1) 0.2% CHX, (2) Green tea – aloe vera, (3) distilled water. Plaque and gingival index were evaluated on the day of the beginning of the experiment and 14 days postoperative and the results showed that green tea – aloe vera mouthwash improve periodontal health status.[44]

S saint et al,.(2015)- Done a study to assess the efficacy of a mouthwash containing 2% green tea as compared to placebo mouthwash for the control of plaque and gingivitis and the results showed that green tea mouthwash was effective in the reduction of plaque and gingivitis scores.[45]

Thomas et al,.(2015)- Compared the antibacterial efficacy of CHX (0.2%), NaF (0.05%), fluoride with essential oils (0.05%), alum (0.02%), green tea, and garlic with lime mouth rinses against Streptococcus mutans, lactobacilli, and Candida Albicans. The three microbes were isolated from the saliva samples collected from children with severe early childhood caries. The zone of minimum inhibition was assessed using the agar diffusion method and the results showed that against S. mutans and lactobacilli, CHX mouth rinse was found to be most effective as compared to other mouth rinses, but against C. Albicans garlic with lime, mouth rinse was found to be more effective than other mouth rinses.[46]

Jain et al,.(2016)- Compared the antimicrobial efficacy of multi herbal mouth rinse with essential oil-based, fluoride-containing, and 0.2% CHX digluconate mouth rinse against streptococcus mutans, and the results showed that there was not a statistically significant difference in the distribution of baseline data groups, but a reduction in the S. mutans colony count on multi-herbal mouth rinse in comparison with the other mouth rinses had statistically significant value s except fluoride mouth rinse till 1-week post rinsing.[47]

Thomas et al,.(2016)- Done a study to evaluate and compare the antimicrobial efficacy of 0.5% green tea and 0.2% CHX mouth rinses against Streptococcus mutans, lactobacilli, and Candida albicans, and the results showed a statistically significant fall in colony count was found with both the mouth rinses streptococcus mutans and lactobacilli but not against Candida Albicans. Against Streptococcus mutans, green tea mouth rinse was found to be significantly better than CHX mouth rinse against lactobacilli, CHX mouth rinse was significantly better than green tea mouth rinse.[48]

Arab et al,.(2016)- Evaluated the changes in saliva properties and oral microbial flora in patients undergoing fixed orthodontic treatment. Selective media, sabouraud dextrose agar, mitis salivarius agar, and rogosa agar were used for isolation of candida Albicans, streptococcus mutans, and lactobacillus acidophilus, respectively after 12 and 15 weeks of commercially fixed orthodontic treatment, the total colony count of candida Albicans, streptococcus mutans, lactobacillus acidophilus showed a significant increase. The salivary PH decreased using orthodontic treatment while the salivary flow did not change significantly.[49]

Kim et al,.(2016)- Done a study to detect the initial changes in salivary levels of periodontal pathogens after orthodontic treatment with fixed appliances the simplified OHIS, plaque index, gingival index were obtained from central incisor, lateral incisor, and first molars of both arches. Whole saliva and periodontal parameters were obtained at the following 4-time points. immediately before debonding (T1), 1 week after debonidng (T2), 5 weeks after debonding (T3), 13 weeks after debonding (T4). Repeated measures analysis of variance was used to determine salivary bacterial level and periodontal parameters among 4-time pings after quantifying salivary levels of Aa, Fucobacterium nucleatum, Pg, Pi, Tannerala forsythia, and total bacteria using PCR and the results showed that all the parameters are significantly decreased immediately after

debonding (T2). The salivary levels of the bacteria and Pg were decreased at T3 and higher levels of Aa and F. nucleatum were present at saliva after debonding.[50]

HR Abdul Baqi et al,.(2016)- conducted a study to evaluate the Salvadora persica and green tea anti-plaque effect and the result showed that a combination of 0.2mg/ml green tea and 7-82mg/ml Salvadora persica C, aqueous extracts were found to exhibit synergistic antibacterial and anti adherence effects against primary plaque colonizers biofilm.[51]

L T et al,.(2017)- Compared the effect of curcumin (CMN) mouth rinse with chlorhexidine (CHX) mouth rinse on clinical parameters and reactive oxygen metabolites (ROM) levels in participants with chronic gingivitis. Thirty plaque-induced generalized chronic gingivitis participants were randomly assigned to three groups – Group I (control/saline), Group II (CHX), and Group III (CMN), respectively. Baseline plaque index (PI), gingival index (GI), and salivary ROM were estimated, and oral prophylaxis was done. The parameters were recorded and evaluated again at the end of 4 weeks and the results showed that the three groups have shown a significant reduction in PI, GI, and ROM levels at the end of 4 weeks. However, on the intragroup comparison, only the CMN group has shown a significant reduction in ROM levels at the end of 4 weeks ($P < 0.05$). CMN mouth rinse can be considered as an alternative antigingivitis agent to CHX because of its anti-inflammatory and antioxidant properties.[52]

Kaur et al.,(2017)- Compared the antiplaque efficacy of green tea catechin mouthwash with chlorhexidine gluconate mouthwash, a single-blinded cross-over study was conducted among 30 participants, plaque score was compared and the difference between the green tea catechin and chlorhexidine mouthwash was determined by t-test. The difference between the plaque scores was not statistically significant ($p>0.05$). The results showed that both the groups that are the green tea mouthwash and chlorhexidine mouthwash have comparable results in plaque reduction.[9]

Remya et al,.(2017)- Evaluated the efficacy of green tea, Listerine, and chlorhexidine mouthwash in plaque reduction among the 30 orthodontic patients undergone orthodontic therapy. Gingival status was assessed using sulcus bleeding index and plaque accumulation was assessed using Turesky- Glikmore- Glickman modification of Quigley Hein index, and the results showed that the mean gingival and plaque score was reduced in all three groups. However green tea mouthwash was estimated to have the highest mean difference from 2. 17±0.610 at baseline to 1.48±0.474 on the 15th day.[53]

Sharma et al,.(2017)- Studied the protective property of green tea on periodontal health and also any side effect of green tea prevails in terms of staining of teeth. A prospective randomized clinical intervention study was conducted in 840 patients with chronic periodontitis. Three dependent variables: probing depth (PD), clinical attachment loss (CAL), and bleeding on probing (BOP) were measured to reflect the periodontal diseases and the results showed a statistically significant reduction in PD, CAL. BOP following the introduction of green tea and concluded that green tea showed antioxidant, antimicrobial, and anti-collagenase activities on periodontal health.[54]

Kulkarni et al,.(2017)- Compared the antiplaque efficacy of alcohol-based mouthwash with essential oils and non–alcohol based CHX mouth rinse in 4 days plaque reformation. Two commercially available mouth rinses, one containing alcohol and one without alcohol were placed in an identical bottle and distributed to Group A and Group B patients. Whereas the group C patients were instructed to use warm saline mouth

rinse and the results showed that essential oils with an alcohol-based and CHX alcohol-free mouth rinse (0.2%) groups compared to normal saline showed a significant reduction in GI, PI score (P<0.001).[55]

Lenerato et al,.(2017)- Done a study to know the impact of mouth rinsing using CHX gluconate 0.2%with the number of plaque-causing bacterial colonies in fixed orthodontic users, and the results showed that the use of 0.2% CHX significantly affects the amount of plaque-causing bacterial colonies in fixed orthodontic users (P<0.05) with percentage drop 61.84%.[56]

G Radafshar et al,. (2017)- Done a study to explore the effect of Iranian green tea mouthwash containing 1% tannin on dental plaque and chronic gingivitis and the results showed that 1% tannin green tea mouth wash could be a safe and feasible adjunct to mechanical plaque control.[57]

Yadav et al,.(2018)- Evaluated and compare the efficacy of three different mouthwashes containing CHX, essential oils, and herbal extracts by using a pre-procedural rinsing agent in reducing the bacterial load of the aerosol produced by ultrasonic scalers. The aerosol was collected and the microbiological study was done from different operating positions and the results showed 0.2% CHX was found to be the most effective pre-procedural mouth rinse in reducing the bacterial load in the aerosol produced during ultrasonic scaling followed by essential oil and herbal mouthwash Respectively.[58]

Bareoso et al,.(2018)-conducted an in–vitro study to determine the inhibitory activity of a novel proprieting blend of green tea and black tea aqueous extracts on Streptococcus Mutans and the results showed a minimum inhibitory concentration (MIC)of 12.5gm/ml and a minimum bactericidal concentration (MBC) of 12.5gm/ml was established against S. mutans meaning that at concentrations of 12.5mg/ml and higher, the proprietary tea blend is effective against the growth of S .mutans.[59]

Karim et al,.(2019)- did a study to compare the tolerability of none- alcohol-containing green tea-based (NAGT) mouth rinse with CHX mouth rinse. 40 healthy subjects were instructed to use the mouthwashes twice daily for 4 weeks. Collected data included age, gender, smoking history, in addition to subjective assessment using a validated questionnaire. Intraoral clinical examination was completed at baseline and 2 weeks time points and results showed overall less tolerability of a non-alcohol containing green tea-based mouth rinse compared to CHX gluconate.[60]

Alim et al,. (2019)- Done a study to compare the antimicrobial efficacy of green tea (0.5%) mouth rinse to that of CHX (0.125%) against streptococcus mutans and lactobacilli species in 42 children who had early childhood caries were recruited in this study and randomly divided, using lottery method into 3 groups. group A-children using green tea mouth rinses, group B- children using CHX mouthwash, group C–children using coloured flavoured non–sweetened tape water and were instructed to rinse for 1 min using 5ml of respective mouthrinse 30 min after toothbrushing for 2 weeks. A volume of 2ml saliva samples was collected prior to the commencement of mouth rinsing and after 2 weeks rinsing data were collected and the results revealed both CHX and green tea mouth rinses showed a statistically significant fall in colony count of S. mutans and lactobacilli species.[61]

Materials and Methods

Source of Data

- Outpatients from the Department of Orthodontics, Mahe Institute of Dental Sciences and Hospital, who were undergoing orthodontic treatment were selected for the study.
- Patients are enrolled in the study after signing the informed consent.

Inclusion Criteria

- Subjects undergoing orthodontic treatment with fixed appliance of 0.22 slot MBT prescription.
- Patients with minimal crowding.
- Subjects with mild to moderate gingivitis.
- Patients with minimum 20 teeth.
- Systemically healthy patients.
- Patients who had not undergone any periodontal therapy for past 6 months.

Exclusion Criteria

- Pregnant and lactating women.
- History of smoking and alcohol consumption.
- Any history of antibiotics intake within last three months.
- History of allergy to chemical or any herbal products.

Methodology

Sixty patients of age group 13-35 yrs with minimal crowding were selected for this study. Subjects with minimal crowding were selected based on the Little's Irregularity Index score range up to 3.[62] The gingival health status and presence of bleeding on probing were assessed by Gingival index by Loe and Silness (1963)[63] and Papillary bleeding index (PBI) by Muhleman and Saxer (1975)[64] after 2 weeks of placement of orthodontic brackets. Plaque samples were collected and sent for microbiological analysis to estimate colony-forming units. The subjects received complete oral prophylaxis initially. The subjects were randomly allocated to one of the following groups. Each group consisted of 20 subjects.

Group I: Green tea mouth wash (Listerine Green tea) (10ml to be rinsed for 30 seconds twice daily)
Group II: Chlorhexidine mouth wash (0.2%) (Rexidine SRS) (10ml to be rinsed for 30 seconds twice daily)
Group III: Essential oil mouth wash (Listerine cavity fighter) (10ml to be rinsed for 30 seconds twice daily).

All participants were asked to use an orthodontic toothbrush during the study phase. After the 14th day and 21st day, the subjects were recalled and the gingival and bleeding indices were recorded. Supragingival plaque sample was collected using a sterile jaquette scaler for microbiological analysis.

Marginal Plaque Collection

Marginal plaque along the gingival margin was collected from all the teeth present. The site was isolated from saliva by applying cotton rolls and was gently dried with an airway syringe to avoid contamination. With the help of a sterile jaquette scaler, marginal plaque samples were collected and are immediately transported to the laboratory in brain heart infusion broth as the transport medium and further microbial analysis was done.

International Journal of Drug Research and Dental Science
Volume 3 Issue 2 (Page: 1-34), 2021 ISSN: 2582-0826

Figure 1 and 2: Armamentarium

Figure 3: Littles irregularity index done using mandibular cast and the dial caliper

International Journal of Drug Research and Dental Science
Volume 3 Issue 2 (Page: 1-34), 2021 ISSN: 2582-0826

Figure 4: Patients undergoing orthodontic treatment

Figure 5: Chlorhexidine (0.2%) mouthwash

Figure 6: Green tea mouthwash

International Journal of Drug Research and Dental Science
Volume 3 Issue 2 (Page: 1-34), 2021 ISSN: 2582-0826

Figure 7: Essential oil mouthwash

Figure 8 and 9: Sample collection

International Journal of Drug Research and Dental Science
Volume 3 Issue 2 (Page: 1-34), 2021 ISSN: 2582-0826

Figure 10 and 11: Microbiological powder (Brain heart infusion broth) for transport medium preparation

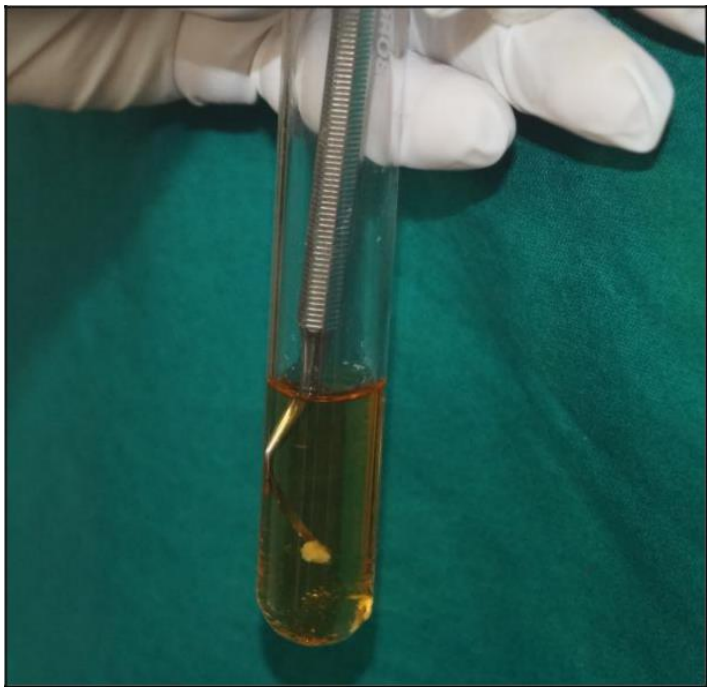


Figure 12: Samples transferring to BHI

Figure 13: Incubator- Samples incubated

Figure 14: Inoculating loop dipping in to the inoculums

International Journal of Drug Research and Dental Science
Volume 3 Issue 2 (Page: 1-34), 2021 ISSN: 2582-0826

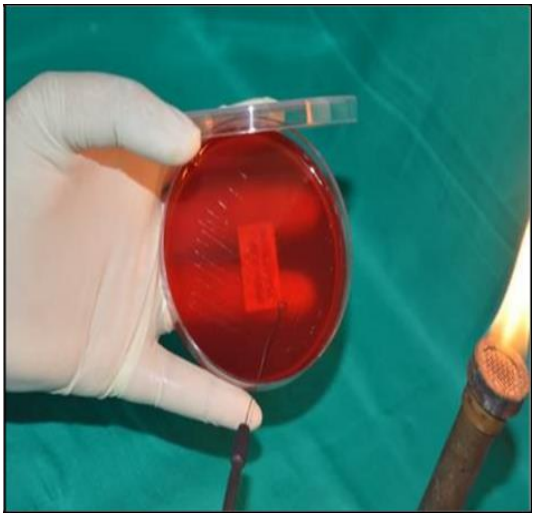


Figure 15- Streaking to different medias (Blood agar, Choclate agar and Mac Conkey agar plates)

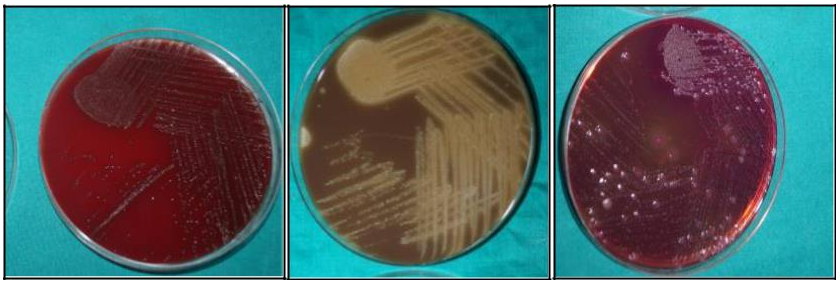
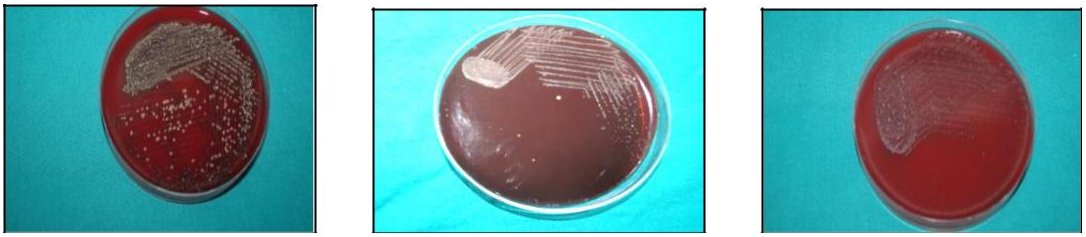


Figure 16- Bacterial colony formed in different medias (Blood agar, Choclate agar and Mac Conkey agar plates) after incubation

Microbiological Analysis

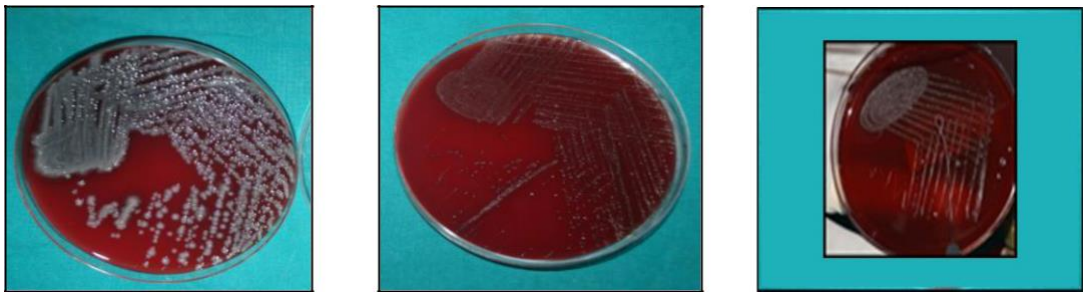
Baseline **14th Day** **21st day**

Chlorhexidine mouth wash (0.2%)



Green tea mouth wash

International Journal of Drug Research and Dental Science
Volume 3 Issue 2 (Page: 1-34), 2021 ISSN: 2582-0826



Essential oil mouth wash
Figure 17: Bacterial colonies formed associated with the usage of mouth washes

Containing Green tea, CHX (0.2%) and Essential oil in patients undergoing orthodontic treatment in baseline, 14th day and 21st day

Figure 18: Bacterial colony count counted by colony counter

International Journal of Drug Research and Dental Science
Volume 3 Issue 2 (Page: 1-34), 2021 ISSN: 2582-0826

Statistical Analysis

In this study, Data will be expressed in terms of mean and standard deviation.

Comparison between the groups will be done using ANOVA with post hoc test or Kruskal Wallis test.

Pair-wise comparisons between the time points within each group will be done using Paired t-test or Wilcoxen sign rank test.

P<0.05 will be considered significant.

Results

Table 1: shows a comparison of a gingival index between three groups on the first day, 14th day, and 21st day, we observed that there is a significant difference in mean GI on the first day and 14th day (p<0.05) whereas no significant difference after 21st day.

When the gingival index is compared between the three groups, Chlorhexidine (0.2%) group has the least gingival index followed by the green tea group and essential oil group on the first day, 14th day & 21st day.

When the gingival index was compared between the time points in each group, it was observed that there is a significant decrease from the first day to the 21st day also there is a significant difference in mean GI in each group. From the post hoc test, it was observed that there is a significant difference between all the time points (Graph 1).

		First day		14th day		21st day		
		Mean	**Std. Deviation**	**Mean**	**Std. Deviation**	**Mean**	**Std. Deviation**	**P-value**
Gingival Index	Essential Oil Group	1.95	0.042	1.36	0.222	1.1	0.176	<0.001
	Chlorhexidine Group (0.2%)	1.55	0.474	1.2	0.258	1.05	0.158	0.006
	Green Tea Group	1.73	0.435	1.22	0.249	1.1	0.211	<0.001
P-value		0.029		0.043		0.789		

Table 1: Comparision of Gingival Index Associated with the Usage of Mouth Washes Containing Green Tea, CHX (0.2%) and Essential Oil in Patients Undergoing Orthodontic Treatment

Table 2: shows the comparison of a papillary bleeding index between the three groups on the first day, 14th day, and 21st day. It was observed that there is a significant difference in mean PBI on 14th day and the 21st day. Whereas no difference on 1st day.

The papillary index was compared between time points in each group. It was observed that there is a significant difference in mean PBI in each group. From the post hoc test, it was observed that there is a significant difference between all the time points a decrease in the papillary bleeding score is observed from the first day to the 21st day.

International Journal of Drug Research and Dental Science
Volume 3 Issue 2 (Page: 1-34), 2021 ISSN: 2582-0826

When the papillary index is compared between the three groups, the Chlorhexidine group has the least papillary index followed by the green tea group and essential oil group on the first day, 14th day & 21st day (Graph 2).

		First day		14th day		21st day		
		Mean	**Std. Deviation**	**Mean**	**Std. Deviation**	**Mean**	**Std. Deviation**	**P-value**
Papillary Bleeding Index	Essential Oil Group	2.4	0.527	1.9	0.568	1.4	1.4	0.003
	Chlorhexidine Group (0.2%)	2.3	0.483	1.5	0.527	1	0	<0.001
	Green Tea Group	2.6	0.516	1.8	0.422	1.1	0.316	<0.001
P-value		0.418		0.046		0.001		

Table 2: Comparision of Papillary Bleeding Index Associated with the Usage of Mouth Washes Containing Green Tea, CHX (0.2%) and Essential Oil in Patients Undergoing Orthodontic Treatment

Table -3 shows the comparison of bacterial colony count associated with the usage of three mouthwashes on the first day, 14th day, and 21st day, it was observed that there is no significant difference in mean Bacterial count ($p>0.05$) on the first day, whereas there was a significant difference after 14th and 21st day. When bacterial colony count is compared between the time points in each group, it was observed a significant difference in the chlorhexidine group and green tea group. Whereas it was not observed in the essential oil group.

When the bacterial count is compared between the three groups, the Chlorhexidine group has the least bacterial colony count followed by the green tea group and the essential group on the first day, 14th day & 21st day.

When the bacterial count was compared between the time points in each group, it was observed that there is a significant decrease from the first day to the 21st day. (Graph 3).

		First day		14th day		21st day		
		Mean	**Std. Deviation**	**Mean**	**Std. Deviation**	**Mean**	**Std. Deviation**	**P-value**
Bacterial Colony Counts	Essential Oil Group	89200	8107.61	79500	7321.961	61700	8165.646	0.103
	Chlorhexidine Group (0.2%)	85100	11656.662	56200	9437.514	27300	4808.557	<0.001
	Green Tea Group	89500	5562.773	76000	6110.101	55200	7130.529	<0.001
P-value		0.469		<0.001		<0.001		

Table 3: Comparision of Bacterial Colony Count Associated with the Usage of Mouth Washes Containing Green Tea, CHX (0.2%) and Essential Oil in Patients Undergoing Orthodontic Treatment

Table 4: shows a comparison between the two groups on the first day using Tukey's post hoc tests. It was observed that, a significant difference in gingival index between essential oil and chlorhexidine group on the first day.

First Day	Group		P-value
Gingival Index	Essential Oil Group	Chlorhexidine Group (0.2%)	.023
	Chlorhexidine Group (0.2%)	Green Tea Group	.208
		Green Tea Group	.534

Table 4: Comparision between the Two Groups Using Tukey's Post HOC Tests on the First Day

Table 5: shows a comparison between the two groups on the 14th day using Tukey's post hoc tests. It was observed that a significant difference between the essential oil and chlorhexidine group in the papillary index, and bacterial colony count in the chlorhexidine group is significantly different from the essential oil and green tea group.

Dependent Variable	Group		P-value
Gingival Index	Essential Oil Group	Chlorhexidine Group (0.2%)	.061
		Green Tea Group	.088
	Chlorhexidine Group (0.2%)	Green Tea Group	.982
Papillary Bleeding Index	Essential Oil Group	Chlorhexidine Group (0.2%)	.036
		Green Tea Group	.398
	Chlorhexidine Group (0.2%)	Green Tea Group	.398
Bacterial Colony Counts	Essential Oil Group	Chlorhexidine Group (0.2%)	<0.001
		Green Tea Group	.096
	Chlorhexidine Group (0.2%)	Green Tea Group	<0.001

Table 5: Comparision between the Two Groups Using Tukey's Post HOC Tests on the 14th Day

Table 6: shows the comparison between the two groups on the 21st day using Tukey's post hoc tests. It was observed that the papillary index of the chlorhexidine group is significantly different from the essential oil and green tea group. The bacterial colony count in the chlorhexidine group, essential oil, and green tea group are significantly different.

21st day	Group		P-value
Papillary Bleeding Index	Essential Oil Group	Chlorhexidine Group (0.2%)	.002
		Green Tea Group	.010
	Chlorhexidine Group (0.2%)	Green Tea Group	.800
Bacterial Colony Counts	Essential Oil Group	Chlorhexidine Group (0.2%)	.000
		Green Tea Group	.000
	Chlorhexidine Group (0.2%)	Green Tea Group	.000

Table 6: Comparision between the Two Groups Using Tukey's Post HOC Tests on the 21st Day

Conclusion:

Table 7: shows a comparison of the CHX (0.2%) group using Tukey's post hoc tests. When the gingival index is compared between different time points in the chlorhexidine group, it was observed that there is no significant decrease from the first day to the 14th day and from the 14th day to the 21st day but a significant decrease is observed from the first to 21st day. When the PB index is compared between different time points in the chlorhexidine group, it was observed that there is a significant decrease from the first day to the 14th day and from the 14th day to the 21st day. When bacterial colony count is compared between different time points in the chlorhexidine group, it was observed that there is a significant decrease from the first day to the 14th day and from the 14th day to the 21st day.

Dependent Variable	Day		P-value
Gingival Index	14th Day	21st Day	.563
		First Day	.058
	21st Day	First Day	.005
Papillary Bleeding Index	14th Day	21st Day	.030
		First Day	.001
	21st Day	First Day	<0.001
Bacterial Colony Counts	14th Day	21st Day	<0.001
		First Day	<0.001
	21st Day	First Day	First Day

Table 7: Comparision between Each Group Using Tukey's Post HOC Tests Post HOC Tests: Chlorhexidine Group (0.2%)

Table 8: shows a comparison of the green tea group using Tukey's post hoc tests. When the gingival index is compared between different time points in the green tea group, it was observed that there is a significant decrease from the first day to the 14th day and from the 14th day to the 21st day but no significant decrease is observed from the 14th day to 21st day. When the PB index is compared between different time points in the green tea group, it was observed that there is a significant decrease from the first day to the 14th day, first to 21st day, and from the 14th day to the 21st day. When bacterial colony count was compared between different time points in the Green tea group, it was observed that there is a significant decrease from the first day to the 14th day, first to 21st day, and from 14th day to 21st day.

International Journal of Drug Research and Dental Science
Volume 3 Issue 2 (Page: 1-34), 2021 ISSN: 2582-0826

Dependent Variable	Day		P-value
Gingival Index	14th Day	21st Day	.672
		First Day	.003
	21st Day	First Day	<0.001
Papillary Bleeding Index	14th Day	21st Day	.003
		First Day	.001
	21st Day	First Day	<0.001
Bacterial Colony Counts	14th Day	21st Day	<0.001
		First Day	<0.001
	21st Day	First Day	<0.001

Table 8: Post HOC Tests: Green Tea Group

Table 9: shows a comparison of the essential oil group using Tukey's post hoc test. When the gingival index is compared between different time points in an essential group, it was observed that there is a significant decrease from the first day to 14th day, 14th day to 21st day, and from first to 21st day. When the papillary bleeding index is compared between different time points in an essential group, it was observed that there is no significant decrease from the first day to the 14th day and from the 14th day to the 21st day but a significant decrease is observed from the first to 21st day.

Dependent Variable	Day		P-value
Gingival Index	14th Day	21st Day	<0.001
		First Day	<0.001
	21st Day	First Day	<0.001
Papillary Bleeding Index	14th Day	21st Day	.113
		First Day	.237
	21st Day	First Day	.002

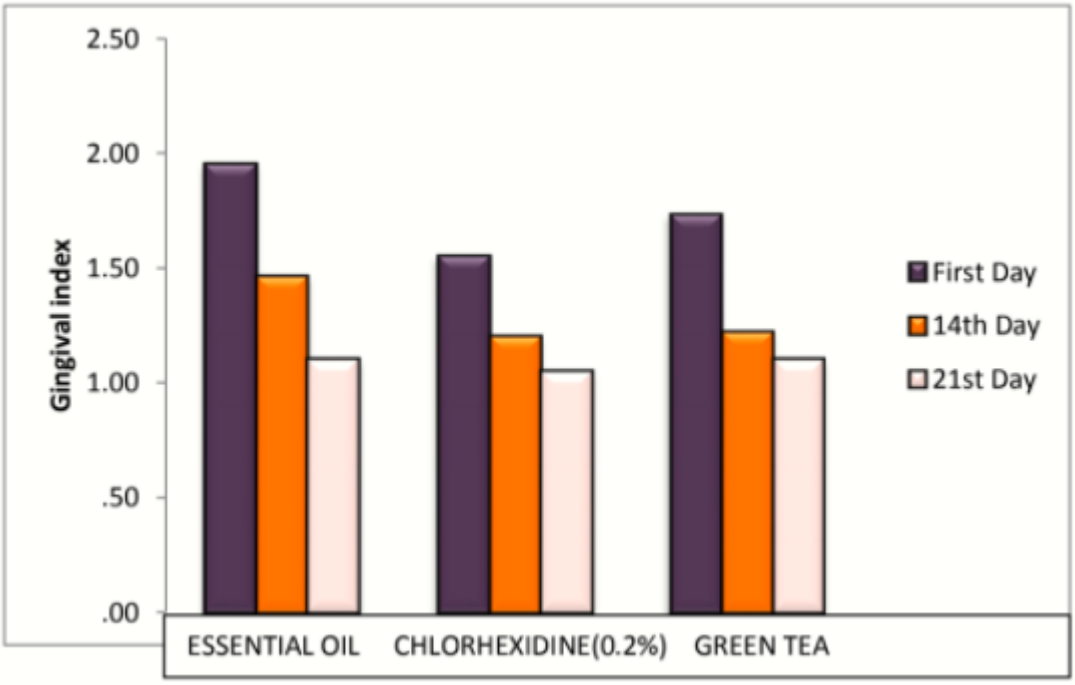
Table 9: Post HOC Tests Essential Oil Group

Table 10: shows the comparison of the percentage of decrease in bacterial count in three groups from baseline to 14th day and 21st day. when comparing the three groups chlorhexidine showed the highest percentage of reduction in bacterial colony count (33.96% reduction in colony count from baseline to 14th day and 51.42 % reduction from14th day to 21st day) followed by the green tea group (15.08% reduction in colony count from baseline to 14th day and 27.37 % reduction from 14th day to 21st day) and essential oil group (6.39% reduction in colony count from baseline to 14th day and 2, 16 % reduction from14th day to 21st day).

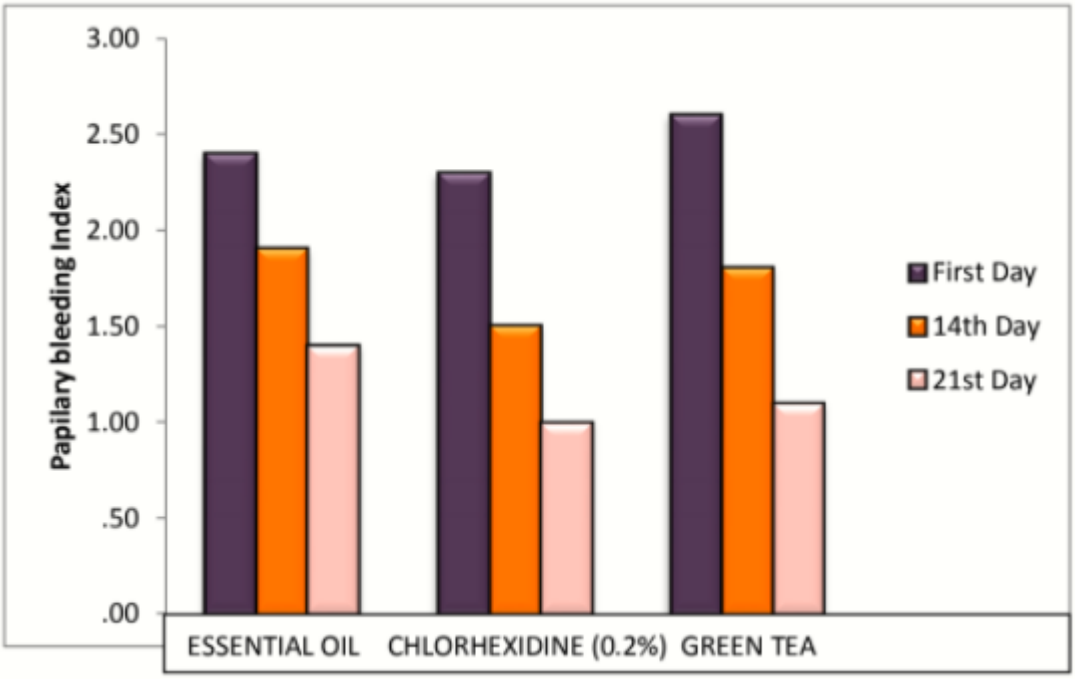
Undergoing Orthodontic Treatment on 14th day and 21st day from the Baseline		
Group	**14th day**	**21st day**
Essential Oil Group	6.39%	2.16%
Chlorhexidine Group	33.96%	51.42%
Green Tea Group	15.08%	27.37%

Table 10: Percentage Decrease in Bacterial Count with the Usage of Mouth Washes Containing Green Tea, Chlorhexidine (0.2%) and Essential Oil in Patients

International Journal of Drug Research and Dental Science
Volume 3 Issue 2 (Page: 1-34), 2021 ISSN: 2582-0826

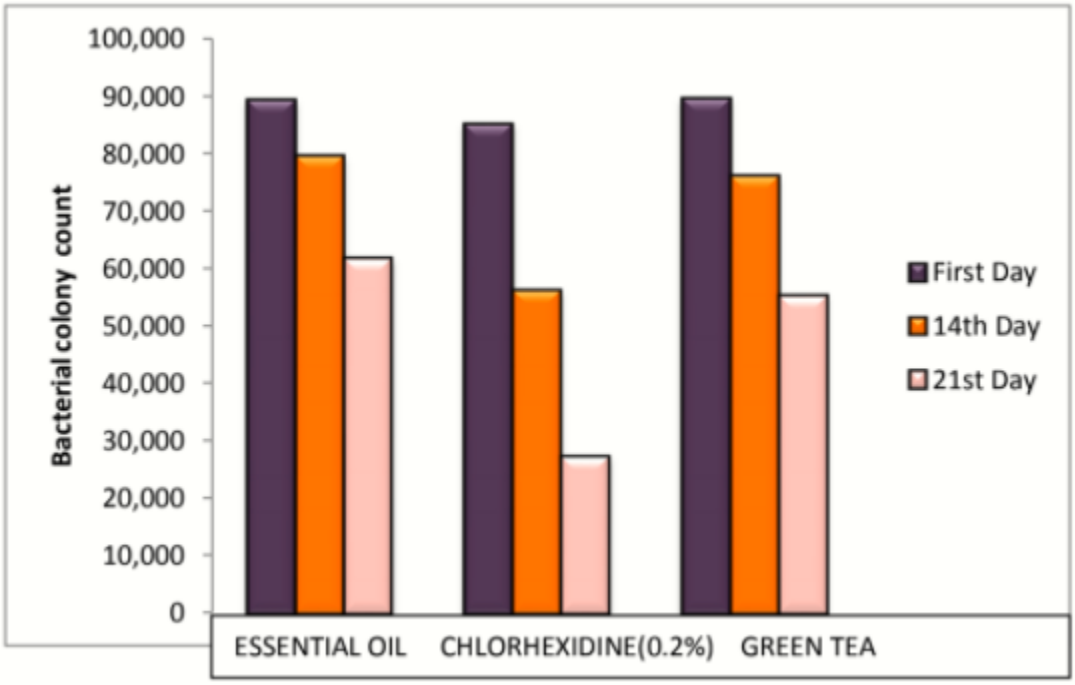


Graph 1: Comparision of Gingival Index Associated with the Usage of Mouth Washes Containing Green Tea, CHX (0.2%) and Essential Oil in Patients Undergoing Orthodontic Treatment



Graph 2: Comparision of Papillary Bleeding Index Associated with the Usage of Mouth Washes Containing Green Tea, CHX (0.2%) and Essential Oil in Patients Undergoing Orthodontic Treatment

International Journal of Drug Research and Dental Science
Volume 3 Issue 2 (Page: 1-34), 2021 ISSN: 2582-0826



Graph 3: Comparision of Bacterial Colony Count Associated with the Usage of Mouth Washes Containing Green Tea, CHX (0.2%) and Essential Oil in Patients Undergoing Orthodontic Treatment

Discussion: Dental plaque is a biofilm that actually adheres over the tooth surface is the common cause of periodontal diseases. Malocclusion is one of the most common dental disorders and is capable of increasing the risk of periodontal diseases.[65] Maintaining oral hygiene is not a major problem while using removable appliances because they can be withdrawn from the oral cavity. However fixed orthodontic appliances provide additional surfaces for the collection of food debris and plaque resulting in increased bacterial colonization and subsequently, accentuate their luxuriant growth in supragingival and subgingival region.[66]

Patients derive both functional and aesthetic benefits from orthodontic treatment. It was evident that patients wearing orthodontic appliances have a problem in maintaining good oral hygiene. Inadequate oral home care among orthodontic patients may make them more prone to develop gingivitis during orthodontic treatment. Therefore educating and motivating these patients, to maintain their oral health and providing recommendations for oral home care aid to improve their compliance, remains the cornerstone for achieving optimal oral hygiene results.s

Chemical plaque control agents act as effective adjuncts to mechanical plaque control in preventing plaque formation and gingival inflammation in patients undergoing orthodontic treatment. The idea of employing a chemical agent which acts in an identical manner to a toothbrush and removes bacteria from the tooth surface in an attractive proportion. The chemical agent contained in the mouth rinse would be expected to reach all tooth surfaces and thereby be totally effective.[66]

Chlorhexidine is an antimicrobial agent.[67] It is commonly available in the forms of mouth rinses, gels, varnish, toothpaste, spray, and sugar-free chewing gums. It acts on the inner cytoplasmic membrane, hence it is a membrane-active type of substance. It is dicationic at PH levels above 3.5. It prevents plaque accumulation, hence it is an antiplaque and antigingivitis agent and reduces the adherence of P. gingivalis to epithelial cells.

It can be bacteriostatic or bactericidal depending on the dose. It acts against a wide array of bacteria, including gram-negative and gram-positive bacteria dermatophytes and lipolytic viruses.

Chlorhexidine is a cationic bigunaide, which is bacteriostatic in low and bactericidal in higher concentration. Chlorhexidine gluconate at a concentration of 0.2% is regarded as the gold standard in the reduction of plaque formation and as a local bacterial compound.[68]

The mode of action of chlorhexidine is mainly binding to microbial cell membranes and damaging the surface structure leading to an osmotic imbalance and prescription of cytoplasm causing cell death. The chlorhexidine may inhibit the early adhesion of plaque bacteria on tooth surfaces.[69]

Essential oil mouthwash has proven to be effective at controlling inflammation and supragingival biofilms. They have the ability to alter the cell surface of specific microorganisms and eliminate their enzymatic activity. They can also inhibit the endotoxins of gram-negative pathogens. In vitro and in vivo studies have shown how essential oil mouthwashes can penetrate the dental biofilm and have bactericidal effect.[70]

Tea (infusion of dried leaves of camellia Sinensis) is the most popular and widely consumed beverage in the world today. Its polyphenolic component has been reported to possess antioxidant, antimicrobial, antimutagenic, antidiabetic, hypocholesterolemic, anti-inflammatory, and cancer-preventive properties.[71]

During recent years, the health benefits of green tea have been extensively researched and in recent studies, a correlation between taking green tea and reducing periodontal diseases is discovered. Green tea has a higher concentration of polyphenols, antioxidants, and thus more therapeutic benefits). Catechin suppresses periodontal inflammation, thereby providing better health and gingival health.[72]

Many of the biological properties of green tea has been ascribed to the catechin fraction, which constitutes up to 30%of the dry leaf weight. These potent antioxidants comprise free catechin such as (+) catechin, (+) Gallo catechin, (-) epicatechin, and (-) epigallocatechin and galloyl catechin such as (-) epicatechin gallate (Ecg), (-) gallocatechin gallate. Green tea also contains carotenoids, tocopherols, ascorbic acid, minerals such as Cr, Mn, Se, or Zn and certain phytochemical components.[73]

When the gingival index was compared on the baseline, 14th, and 21st day between the three groups, it was observed that the chlorhexidine group showed a decrease in the gingival index followed by green tea mouthwash and essential oil mouthwash.

When the papillary bleeding index was compared between the three groups on baseline, 14th day and 21st day it was observed that chlorhexidine shows the least papillary bleeding index followed by green tea mouth wash and essential oil mouthwash.

A comparison of the total CFU of various mouthwashes in this study revealed that 0.2% CHX mouthwash was the most effective in reducing the total microbial colony count (33.96% reduction in total bacterial colony count from baseline to 14th day and 51.42% reduction in total bacterial colony count from 14th day to 21st day), followed by green tea mouth wash (15.8% reduction in total bacterial colony count from baseline to 14th day and 27.37% reduction in total bacterial colony count from 14th day to 21st day) and essential oil mouth wash (6.39% reduction in total bacterial colony count from baseline to 14th day and 2.6% reduction in total bacterial colony count from 14th day to 21st day).

A similar study was done by Sriparna et al in 2015, evaluated the effect of green tea, Listerine and chlorhexidine mouthwashes in gingivitis patients and the results showed that the use of green tea is an effective antiplaque agent that is comparable to CHX mouthwash and can be used as an adjunct to regular mechanical plaque control practices and professional scaling in gingivitis patients.[38]

The results obtained may be due to variation in the composition of mouthwashes. The reduction in gingival index, papillary bleeding index, and microbial colony forming units from baseline to 21st day may be attributed to proper brushing that the use of mouthwashes in conjugation with oral hygiene procedures during orthodontic treatment plays an important role in maintaining oral hygiene in patients.

In the present study, subjects under green tea show clinical and microbiological effects which are comparable with the gold standard mouthwash chlorhexidine. One of the reasons for the reduction in gingival index, papillary bleeding index, and the total bacterial colony count would be attributed to levels of catechin, tannins, and astringent present in green tea. These findings are consistent with the other studies which have reported similar results with usage of green tea mouthwash.

The results of this study were comparable with a study conducted by Jenabian et al in 2012, where they compared the effects of green tea mouthwash with another placebo group using saliva, along with the routine mechanical plaque control methods, and they concluded that a significant improvement was observed in all periodontal and gingival indexes after the usage of green tea mouthwash.[30] When comparing the clinical and microbiological changes, microbiological changes are more evident in this study, maybe because most of the gingival and periodontal changes are caused by the anerobic microorganisms which are present in the subgingival regions. Changes in the aerobic microorganisms by collecting the supragingival plaque samples were done in this study this is one of the reasons for the variations in the clinical and microbiological changes after the usage of the three mouthwashes.

A study done by Osawak et al in 1991, showed that green tea catechin has been shown to be bactericidal against Porphyromonas gingivalis and Prevotella intermedia in vitro.[74] Another study done by Demecule M et al in 2000 showed that green tea catechin contains galloyil radicles that possess the ability to inhibit both eukaryotic and prokaryotic cell-derived collagenase, an enzyme that plays an important role in the disruption of collagen component in the gingival tissues of patients with periodontal diseases.[75] Okamoto M et al in 2004 showed the inhibiting effect of green tea catechin on cysteine proteinases. Catechin derivatives have been reported to inhibit certain proteinases of P. gingivalis and may reduce periodontal break down.[16]

The present study was carried out to find the clinical and microbiological changes after the usage of mouthwashes containing 0.2% CHX, green tea, and essential oils in patients undergoing orthodontic treatments and the results showed a significant improvement in clinical changes and reduction in the bacterial colony count after the usage of these three mouth washes in patients undergoing orthodontic treatment.

Conclusion: The practice of alternative medicine with the use of plant and plant products has evolved widely in the form of mouth rinses and toothpaste. They have been shown to possess beneficial effects in the control of plaque and gingival inflammation. moreover, the drug resistance associated with the misuse of chemotherapeutic agents can considerably be reduced with the use of plant extracts. Considering the fact that the chemical formulations of commercially available mouth rinses are chemically based, and have side effects which restrict their use.

Effective use of mouthwashes as supplements for tooth brushing has proved to be beneficial in oral hygiene and maintenance for patients who are undergoing orthodontic treatment. All three mouthwashes in this study were found to be effective by a reduction in total bacterial colony count during orthodontic treatment.

India has a rich source of herbal plant products with medicinal values, green tea can be used as an adjuvant to oral hygiene maintenance with a goal in the prevention as well as the prevalence of periodontal diseases due to its antibacterial and antioxidant properties. Green tea extracts may have numerous effects on periodontal pathogens and periodontal tissues. Greater concentration of catechins better the oral health benefits. Its application as local drug delivery systems like strips, chips, and fibers before the treatment of periodontal disease or in combination with regenerative materials to improve periodontal regeneration should be focused.

Within the limitations of the study, it was concluded that all the three mouthwashes in this study are equally effective by showing significant clinical and microbiological changes after the usage in patients undergoing orthodontic therapy. Chlorhexidine mouthwashes showed better clinical and microbiological changes after the usage followed by green tea mouthwashes and essential oil mouthwashes. Green tea mouthwash can be recommended as a safe anti-inflammatory, antimicrobial mouthwash to control gingival inflammation and to maintain good oral hygiene during orthodontic treatment.

References

1. Preshaw PM, Seymour RA, Heasman PA. Current concepts in periodontal pathogenesis. Dent Update. 2004; 31(10):570-2, 574-8. https://doi.org/10.12968/denu.2004.31.10.570
2. Malhotra R, Grover V, Kapoor A, Saxena D. Comparison of the effectiveness of a commercially available herbal mouthrinse with chlorhexidine gluconate at the clinical and patient level. J Indian Soc Periodontol. 2011; 15(4):349-52. https://dx.doi.org/10.4103%2F0972-124X.92567
3. Davies TM, Shaw WC, Worthington HV, Addy M, Dummer P, Kingdon A. The effect of orthodontic treatment on plaque and gingivitis. Am J Orthod Dentofacial Orthop. 1991; 99(2):155-61. https://doi.org/10.1016/0889-5406(91)70118-g
4. Paolantonio M, Di girolamo G, Pedrazzoli V, et al. Occurrence of Actinobacillus actinomycetemcomitans in patients wearing orthodontic appliances. A cross-sectional study. J Clin Periodontol. 1996; 23(2):112-8. https://doi.org/10.1111/j.1600-051x.1996.tb00543.x
5. Mitchell L. Decalcification during orthodontic treatment with fixed appliances-- an overview. Br J Orthod. 1992; 19(3):199-205. https://doi.org/10.1179/bjo.19.3.199
6. Gunsolley JC. Clinical efficacy of antimicrobial mouthrinses. J Dent. 2010; 38 Suppl 1:S6-10. https://doi.org/10.1016/s0300-5712(10)70004-x
7. Mandel ID. Chemotherapeutic agents for controlling plaque and gingivitis. J Clin Periodontol. 1988; 15(8):488-98. https://doi.org/10.1111/j.1600-051x.1988.tb01020.x
8. Jones CG. Chlorhexidine: is it still the gold standard? Periodontol 2000. 1997; 15:55-62. https://doi.org/10.1111/j.1600-0757.1997.tb00105.x
9. Santos A. Evidence-based control of plaque and gingivitis. J Clin Periodontol. 2003; 30 Suppl 5:13-6. https://doi.org/10.1034/j.1600-051x.30.s5.5.x
10. Kaur H, Jain S, Kaur A. Comparative evaluation of the antiplaque effectiveness of green tea catechin mouthwash with chlorhexidine gluconate. J Indian Soc Periodontol. 2014; 18(2):178-82. https://dx.doi.org/10.4103%2F0972-124X.131320
11. Peter William Taylor, J M Hamilton-miller, Paul D Stapleton. Antimicrobial properties of green tea catechins. Food Sci Technol Bull. 2005; 2: 71–81. https://doi.org/10.1616/1476-2137.14184

12. Kushiyama M, Shimazaki Y, Murakami M, Yamashita Y. Relationship between intake of green tea and periodontal disease. J Periodontol. 2009; 80(3):372-7. https://doi.org/10.1902/jop.2009.080510
13. Overholser CD, Meiller TF, Depaola LG, Minah GE, Niehaus C. Comparative effects of 2 chemotherapeutic mouthrinses on the development of supragingival dental plaque and gingivitis. J Clin Periodontol. 1990; 17(8):575-9. https://pubmed.ncbi.nlm.nih.gov/2212088/
14. Anderson GB, Bowden J, Morrison EC, Caffesse RG. Clinical effects of chlorhexidine mouthwashes on patients undergoing orthodontic treatment. Am J Orthod Dentofacial Orthop. 1997; 111(6):606-12. https://doi.org/10.1016/s0889-5406(97)70312-3
15. Hirasawa M, Takada K, Makimura M, Otake S. Improvement of periodontal status by green tea catechin using a local delivery system: a clinical pilot study. J Periodont Res. 2002; 37(6):433-8. https://doi.org/10.1034/j.1600-0765.2002.01640.x
16. Okamoto M, Sugimoto A, Leung KP, Nakayama K, Kamaguchi A, Maeda N. Inhibitory effect of green tea catechins on cysteine proteinases in Porphyromonas gingivalis. Oral Microbiol Immunol. 2004; 19(2):118-20. https://doi.org/10.1046/j.0902-0055.2003.00112.x
17. Tirang Reza Neyestani, Niloufar Khalaji, Aazam Gharav. Black and green teas may have selective synergistic or antagonistic effects on certain antibiotics against Streptococcus pyogenes in vitro .J Nutr Environ Med.16, 2007 - Issue 3-4. 258-266 https://doi.org/10.1080/13590840701703934
18. Tufekci E, Casagrande ZA, Lindauer SJ, Fowler CE, Williams KT. Effectiveness of an essential oil mouthrinse in improving oral health in orthodontic patients. Angle Orthod. 2008; 78(2):294-8. https://doi.org/10.2319/040607-174.1
19. Yaegaki K, Tanaka T, Sato T, et al. Hop polyphenols suppress production of water-insoluble glucan by Streptococcus mutans and dental plaque growth in vivo. J Clin Dent. 2008; 19(2):74-8. https://pubmed.ncbi.nlm.nih.gov/18763691/
20. Hassani AS, Amirmozafari N, Ordouzadeh N, Hamdi K, Nazari R, Ghaemi A. Volatile components of Camellia sinensis inhibit growth and biofilm formation of oral streptococci in vitro. Pak J Biol Sci. 2008; 11(10):1336-41. https://doi.org/10.3923/pjbs.2008.1336.1341
21. Pandis N, Papaioannou W, Kontou E, Nakou M, Makou M, Eliades T. Salivary Streptococcus mutans levels in patients with conventional and self-ligating brackets. Eur J Orthod. 2010; 32(1):94-9. https://doi.org/10.1093/ejo/cjp033
22. Awadalla HI, Ragab MH, Bassuoni MW, Fayed MT, Abbas MO. A pilot study of the role of green tea use on oral health. Int J Dent Hyg. 2011; 9(2):110-6. https://doi.org/10.1111/j.1601-5037.2009.00440.x
23. Faria RL, Cardoso LM, Akisue G, et al. Antimicrobial activity of Calendula officinalis, Camellia sinensis and chlorhexidine against the adherence of microorganisms to sutures after extraction of unerupted third molars. J Appl Oral Sci. 2011; 19(5):476-82. https://doi.org/10.1590/s1678-77572011000500007
24. Moghbel A, Farjzadeh A, Aghel N, Agheli H, Raisi N. Evaluation of the Effect of Green Tea Extract on Mouth Bacterial Activity in the Presence of Propylene Glycol. Jundishapur J Nat Pharm Prod.2012; 7(2):56-60. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3941855/
25. Hegde RJ, Kamath S. Comparison of the and colony count changes in saliva following chlorhexidine (0.12%) mouth rinse, combination mouth rinse, and green tea extract (0.5%) mouth rinse in children. J Indian Soc Pedod Prev Dent. 2017; 35(2):150-155. https://doi.org/10.4103/jisppd.jisppd_13_17
26. Kudva P, Tabasum ST, Shekhawat NK. Effect of green tea catechin, a local drug delivery system as an adjunct to scaling and root planing in chronic periodontitis patients: A clinicomicrobiological study. J Indian Soc Periodontol. 2011; 15(1):39-45. https://doi.org/10.4103/0972-124x.82269

27. Deshpande N, Deshpande A, Mafoud S. Evaluation of intake of green tea on gingival and periodontal status: An experimental study. J Interdiscip Dentistry. May-Aug 2012. Vol-2, Issue-2. https://doi.org/10.4103/2229-5194.100603
28. Xu X, Zhou XD, Wu CD. Tea catechin epigallocatechin gallate inhibits Streptococcus mutans biofilm formation by suppressing gtf genes. Arch Oral Biol. 2012; 57(6):678-83. https://doi.org/10.1016/j.archoralbio.2011.10.021
29. Eckley B, Thomas J, Crour R, Nga P. Periodontal and microbiological status of patients undergoing orthodontic therapy. Hong Kong Dent J 2012; 9:11-20. https://dentistry.hsc.wvu.edu/media/1239/periodontal-and-microbiological-status-of-patients-undergoing-orthodontic-therapy.pdf
30. Jenabian N, Moghadamnia AA, Karami E, Mir a PB. The effect of Camellia Sinensis (green tea) mouthwash on plaque-induced gingivitis: a single-blinded randomized controlled clinical trial. Daru. 2012; 20(1):39. https://doi.org/10.1186/2008-2231-20-39
31. Balappanavar AY, Sardana V, Singh M. Comparison of the effectiveness of 0.5% tea, 2% neem and 0.2% chlorhexidine mouthwashes on oral health: a randomized control trial. Indian J Dent Res. 2013; 24(1):26-34. https://doi.org/10.4103/0970-9290.114933
32. Morozumi T, Kubota T, Abe D, Shimizu T, Nohno K, Yoshie H. Microbiological effect of essential oils in combination with subgingival ultrasonic instrumentation and mouth rinsing in chronic periodontitis patients. Int J Dent. 2013; 2013:146479. https://dx.doi.org/10.1155%2F2013%2F146479
33. Rassameemasmaung S, Phusudsawang P, Sangalungkarn V. Effect of green tea mouthwash on oral malodor. ISRN Prev Med. 2013; 2013:975148. https://dx.doi.org/10.5402%2F2013%2F975148
34. Araghizadeh A, Kohanteb J, Fani MM. Inhibitory activity of green tea (Camellia sinensis) extract on some clinically isolated cariogenic and periodontopathic bacteria. Med Princ Pract. 2013; 22(4):368-72. https://doi.org/10.1159/000348299
35. Sakanaka S, Aizawa M, Kim M, Yamamoto T. Inhibitory effects of green tea polyphenols on growth and cellular adherence of an oral bacterium, Porphyromonas gingivalis. Biosci Biotechnol Biochem. 1996; 60(5):745-9. https://doi.org/10.1271/bbb.60.745
36. Santos IR, Moreira AC, Costa MG, Castellucci e barbosa Md. Effect of 0.12% chlorhexidine in reducing microorganisms found in aerosol used for dental prophylaxis of patients submitted to fixed orthodontic treatment. Dental Press J Orthod. 2014; 19(3):95-101. https://dx.doi.org/10.1590%2F2176-9451.19.3.095-101.oar
37. Radafshar G, Ghotbizadeh M, Saadat F, Mirfarhadi N. Effects of green tea (Camellia sinensis) mouthwash containing 1% tannin on dental plaque and chronic gingivitis: a double-blinded, randomized, controlled trial. J Investig Clin Dent. 2017; 8(1). https://doi.org/10.1111/jicd.12184
38. Biswas S, K R, S S , B M S. Comparative evaluation of the effect of Green tea, Listerine and Chlorhexidine mouth washes in gingivitis patients: A randomized controlled trial. Sch. J. Dent. Sci., 2015; 2(1):104-112.
39. Priya BM, Anitha V, Shanmugam M, Ashwath B, Sylva SD, Vigneshwari SK. Efficacy of chlorhexidine and green tea mouthwashes in the management of dental plaque-induced gingivitis: A comparative clinical study. Contemp Clin Dent. 2015; 6(4):505-9. https://doi.org/10.4103/0976-237x.169845
40. Quintas V, Prada-lópez I, Donos N, Suárez-quintanilla D, Tomás I. Antiplaque effect of essential oils and 0.2% chlorhexidine on an in situ model of oral biofilm growth: a randomised clinical trial. PLoS ONE. 2015; 10(2):e0117177. https://doi.org/10.1371/journal.pone.0117177
41. Haerian-ardakani A, Rezaei M, Talebi-ardakani M, et al. Comparison of Antimicrobial Effects of Three Different Mouthwashes. Iran J Public Health. 2015; 44(7):997-1003. https://doi.org/10.1371/journal.pone.0117177

42. Dehghani M, Abtahi M, Sadeghian H, Shafaee H, Tanbakuchi B. Comparative evaluation of the effect of Green tea, Listerine and Chlorhexidine mouth washes in gingivitis patients: A randomized controlled trial. Sch. J. Dent. Sci., 2015; 2(1):104-112. https://dx.doi.org/10.4103%2F0976-237X.169845
43. Hambire CU, Jawade R, Patil A, Wani VR, Kulkarni AA, Nehete PB. Comparing the antiplaque efficacy of 0.5% Camellia sinensis extract, 0.05% sodium fluoride, and 0.2% chlorhexidine gluconate mouthwash in children. J Int Soc Prev Community Dent. 2015; 5(3):218-26. https://doi.org/10.4103/2231-0762.158016
44. Sargolzaie N, Rajabi O, Ehteshamfar A. Comparative evaluation of Green Tea-Aloe Vera mouthwash and chlorhexidine 0.2% on gingival indices (A randomized clinical trial). JDMT, Volume 5, Number 1, March 2016. https://jdmt.mums.ac.ir/article_6249_1da86e6abfc9d5d5ecad2ba96a698df3.pdf
45. Sarin S, Marya C, Nagpal R, Oberoi SS, Rekhi A. Preliminary Clinical Evidence of the Antiplaque, Antigingivitis Efficacy of a Mouthwash Containing 2% Green Tea - A Randomised Clinical Trial. Oral Health Prev Dent. 2015; 13(3):197-20346. https://doi.org/10.3290/j.ohpd.a33447
46. Lakade LS, Shah P, Shirol D. Comparison of antimicrobial efficacy of chlorhexidine and combination mouth rinse in reducing the Mutans streptococcus count in plaque. J Indian Soc Pedod Prev Dent. 2014; 32(2):91-6. https://doi.org/10.4103/0970-4388.130780
47. Jain I, Jain P. Comparative evaluation of antimicrobial efficacy of three different formulations of mouth rinses with multi-herbal mouth rinse. Oct-Dec 2016 . Vol 34. Issue 4. 315-323; https://doi.org/10.4103/0970-4388.191409
48. Thomas A, Thakur S, Mhambrey S. Comparison of the antimicrobial efficacy of chlorhexidine, sodium fluoride, fluoride with essential oils, alum, green tea, and garlic with lime mouth rinses on cariogenic microbes. J Int Soc Prev Community Dent. 2015; 5(4):302-8. https://doi.org/10.4103/2231-0762.161759
49. Arab S, Nouhzadeh malekshah S, Abouei mehrizi E, Ebrahimi khanghah A, Naseh R, Imani MM. Effect of Fixed Orthodontic Treatment on Salivary Flow, pH and Microbial Count. J Dent (Tehran). 2016; 13(1):18-22. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4983561/
50. Kim K, Jung WS, Cho S, Ahn SJ. Changes in salivary periodontal pathogens after orthodontic treatment: An in vivo prospective study. Angle Orthod. 2016; 86(6):998-1003. https://doi.org/10.2319/070615-450.1
51. Abdulbaqi HR, Himratul-aznita WH, Baharuddin NA. Evaluation of Salvadora persica L. and green tea anti-plaque effect: a randomized controlled crossover clinical trial. BMC Complement Altern Med. 2016; 16(1):493. https://doi.org/10.1186/s12906-016-1487-0
52. Arunachalam LT, Sudhakar U, Vasanth J, Khumukchum S, Selvam VV. Comparison of anti-plaque and anti-gingivitis effect of curcumin and chlorhexidine mouth rinse in the treatment of gingivitis: A clinical and biochemical study. J Indian Soc Periodontol 2017; 21:478-83. https://www.jisponline.com/text.asp?2017/21/6/478/223960
53. Raju R, Divya A, Rajendran G, John J R. Analogous assay between green tea mouthwash, listerine mouthwash and chlorhexidine mouthwash in plaque reduction, on orthodontic patients: a randomized cross-over study. Int J Community Med Public Health. 2017 May; 4(5):1429-1435. https://dx.doi.org/10.18203/2394-6040.ijcmph20171751
54. Sharma S, Bhuyan L, Ramachandra S, Sharma S, Dash KC, Dhull KS. Effects of green tea on periodontal health: A prospective clinical study. J Int Oral Health 2017; 9:39-44. https://dx.doi.org/10.4103/jioh.jioh_37_17
55. Kulkarni P, Singh DK, Jalaluddin M, Mandal A. Comparative Evaluation of Antiplaque Efficacy between Essential Oils with Alcohol-Based and Chlorhexidine with Nonalcohol-Based Mouthrinses. J Int Soc Prev Community Dent. 2017; 7(Suppl 1):S36-S41. https://dx.doi.org/10.4103/jispcd.JISPCD_131_17

56. Melinda N. Leonarto,* Eddy H. Habar. The impact of mouth-rinsing using chlorhexidine gluconate 0.2% to the amount of plaque-causing bacteria colonies in fixed orthodontic users. J Dentomaxillofac Sci. August 2017, Volume 2, Number 2: 91-94. http://dx.doi.org/10.15562/jdmfs.v2i2.524
57. Radafshar G, Ghotbizadeh M, Saadat F, Mirfarhadi N. Effects of green tea (Camellia sinensis) mouthwash containing 1% tannin on dental plaque and chronic gingivitis: a double-blinded, randomized, controlled trial. J Investig Clin Dent. 2017; 8(1). https://doi.org/10.1111/jicd.12184
58. Yadav S, Kumar S, Srivastava P, Gupta KK, Gupta J, Khan YS. Comparison of efficacy of three different mouthwashes in reducing aerosol contamination produced by ultrasonic scaler: A pilot study. Indian J Dent Sci 2018; 10:6-10. https://doi.org/10.4103/IJDS.IJDS_103_17
59. Barroso H, Ramalhete R, Domingues A, Maci S. Inhibitory activity of a green and black tea blend on . J Oral Microbiol. 2018; 10(1):1481322. https://dx.doi.org/10.1080%2F20002297.2018.1481322
60. Karim et al. Tolerability of a green tea-based mouth rinse: A pilot study. Saudi J Oral Sci (2019)31,457-462. https://doi.org/10.1016/j.sdentj.2019.04.004
61. Moness Ali et al. Antibacterial efficacy of green tea in ECC. Tanta Dental Journal 16:6–11. https://www.tmj.eg.net/article.asp?issn=1687-8574;year=2019;volume=16;issue=1;spage=6;epage=11;aulast=Moness
62. Bernabe E and Flores C. Estimating arch length discrepancy through Little's Irregularity Index for epidemiological use. Eur. J. Orthod. 28 (2006) 269–273. https://doi.org/10.1093/ejo/cji112
63. Löe H. The Gingival Index, the Plaque Index and the Retention Index Systems. J Periodontol. 1967; 38(6): Suppl: 610-6. https://doi.org/10.1902/jop.1967.38.6.610
64. Fatah et al. Prevalence of Dental Plaque, Gingival Bleeding and Dental Calculus among Patients Attended Periodontal Department of School of Dentistry at University of Sulaimani. (IOSR-JDMS). .Volume 14, Issue 9 Ver. IV (Sep. 2015), PP 82-85.
65. Aghili H, Jafari nadoushan AA, Herandi V. Antimicrobial effect of zataria multiflora extract in comparison with chlorhexidine mouthwash on experimentally contaminated orthodontic elastomeric ligatures. J Dent (Tehran). 2015; 12(1):1-10. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4436321/
66. Isotupa KP, Gunn S, Chen CY, Lopatin D, Mäkinen KK. Effect of polyol gums on dental plaque in orthodontic patients. Am J Orthod Dentofacial Orthop. 1995; 107(5):497-504. https://doi.org/10.1016/s0889-5406(95)70117-6
67. Balagopal S, Arjun Kumar P. Chlorhexidine: The gold standard antiplaque agent. JPSR· December 2013.5(12):270-274.
68. Abouassi T, Hannig C, Mahncke K, et al. Does human saliva decrease the antimicrobial activity of chlorhexidine against oral bacteria?. BMC Res Notes. 2014; 7:711. https://doi.org/10.1186/1756-0500-7-711
69. Erry M. Arief et al: The effect of chlorhexidine and triclosan on undisturbed plaque formation for 72 hours duration. Dentofasial, Vol.9, No.1, April 2010:1-6. https://pdfs.semanticscholar.org/dea4/b452041dc3ed900a6e7a8183491bbf0bbd8f.pdf
70. Fine DH, Furgang D, Lieb R, Korik I, Vincent JW, Barnett ML. Effects of sublethal exposure to an antiseptic mouthrinse on representative plaque bacteria. J Clin Periodontol. 1996; 23(5):444-51. https://doi.org/10.1111/j.1600-051x.1996.tb00572.x
71. Miller J M. Antimicrobial Properties of Tea (Camellia sinensis L.). ANTIMICROB. AGENTS CHEMOTHER. Nov. 1995, p. 2375–2377. https://dx.doi.org/10.1128%2Faac.39.11.2375
72. Soukoulis S, Hirrsch R. The effects of a tea tree oil-containing gel on plaque and chronic gingivitis. Aust Dent J. July 2004 .49(2):78-83. https://doi.org/10.1111/j.1834-7819.2004.tb00054.x

73. Taylor P et al. Antimicrobial properties of green tea catechins. Food Sci Technol Bull. 2005; 2:71-81. https://doi.org/10.1616/1476-2137.14184
74. Osawa K, Matsumoto T, Yasuda H, Kato T, Naito Y, Okuda K. The inhibitory effect of plant extracts on the collagenolytic activity and cytotoxicity of human gingival fibroblasts by Porphyromonas gingivalis crude enzyme. Bull Tokyo Dent Coll. 1991; 32(1):1-7. https://pubmed.ncbi.nlm.nih.gov/1668071/
75. Demeule M, Brossard M, Pagé M, Gingras D, Béliveau R. Matrix metalloproteinase inhibition by green tea catechins. Biochim Biophys Acta. 2000; 1478(1):51-60. https://doi.org/10.1016/s0167-4838(00)00009-1

How to cite this Article: **Krishna Priya. B, Anil Melath, Anjali K, Mohammed Feroz T. P, Subair. K, Nanditha Chandran;** *"Comparative Evaluation of Clinical Changes and Microbial Flora Associated with Usage of Mouth Washes Containing Green Tea, Chlorhexidine (0.2%) and Essential Oils in Patients Undergoing Orthodontic Therapy";* Int. J. Drug Res. Dental Sci., 2021; 3(2): 1-34,
doi: https://doi.org/10.36437/ijdrd.2021.3.2.A
Source of Support: Nil, **Conflict of Interest:** Nil.
Received: 17-2-2021 **Revised:** 22-4-2021 **Accepted:** 30-4-2021