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CLINICAL IMPACT OF SODIUM FLUORIDE, GUAVA LEAVES EXTRACT, AND POMEGRANATE PEEL EXTRACT MOUTHRINSES ON DENTAL PLAQUE IN CHILDREN: A RANDOMIZED CLINICAL STUDY

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Abstract

Background: The oral environment facilitates microbial growth and biofilm development, which plays a key role in dental plaque accumulation and the deterioration of gingival health. **Aim:** To evaluate and compare the effectiveness of three distinct mouthrinses—sodium fluoride, guava leaf extract, and pomegranate peel extract—on dental plaque accumulation and health of gingiva in children. **Methods:** a random clinical study involving 40 healthy children aged 8 to 12 years. Participants were randomly assigned to four groups (n = 10 per group): one control group using distilled water, and three intervention groups receiving sodium fluoride, guava leaf extract, or pomegranate peel extract mouthrinses. Gingival and plaque indices were measured at baseline (Day 0), and subsequently on Days 7 and 14. **Results:** showed a significant difference **between fluoride, guava, and pomegranate mouthrinse groups**) compared to the water group in gingival and plaque indices. **Conclusion** Mouthrinses containing sodium fluoride, guava leaf extract, and pomegranate peel extract significantly improved gingival health and reduced plaque accumulation over a 14-day period. These natural and conventional agents may serve as effective adjuncts in pediatric oral hygiene.

Keywords: Mouthrinse, Gingival Index, Pomegranate, Guava Leaf, Plaque Index, Sodium Fluoride.

INTRODUCTION

Dental plaque consists of multiple microbial biofilm environments on the exposed tooth surface composed of a certain thickness of densely packed bacteria embedded in an organic matrix of polymers of bacterial origin and saliva. It represents a major etiological agent linked to oral pathologies such as dental caries and periodontal disease. [1]. According to several recent studies; *S. mutans and Lactobacillus sp.* are types of cariogenic microorganisms that are embedded in oral biofilm and have received the most attention because of the potential impact of their acid production on the demineralization of tooth enamel [2, 3].

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Plaque control could easily be controlled with increased awareness of oral hygiene. Prevention of plaque can be achieved either by mechanically brushing oral tissues, flossing between teeth, or by using chemical agents such as dentifrices and topical mouthwashes to reduce bacterial plaque [4].

Fluoride mouth rinse is the most effective chemotherapeutic mouthwashes employed to prevent dental caries and the buildup of dental plaque. Fluoride possesses two distinct methods of action: firstly, it inhibits acid-induced demineralization of dental tissue by bacteria. Secondly, it enhances the remineralization of partially demineralized enamel during the early stages of dental caries by presence of calcium and phosphate ions in saliva. The .primary drawbacks of these chemotherapeutic drugs are their toxicity and the emergence of microbial resistance [5].

To overcome the obstacles of these chemical agents, the search for new and safe therapeutic agents continues. Guava leaf (*Psidium guajava*) extract (GLEs) have phytochemical bioactive substances such as tannins, flavonoids, vitamins (C&A), and alkaloids that have antibacterial and antioxidant effects and maintain the function of the immune system [6].

Additionally, natural phytochemicals are isolated from some fruits, like pomegranate peel. It is a natural fruit, mainly consumed because of its pleasant flavor antibacterial effects, antioxidant properties, and high nutritional values. Pomegranate peel extract (PPE) has polyphenolic antimicrobial activity [7].

The objective of the current research was clinically evaluate and compare the effect of fluoride, guava leaf extract, and pomegranate peel extract mouth rinses on gingival and plaque indices.

METHODS

Study Design

This randomized controlled clinical trial involved forty healthy and cooperative children, comprising both boys and girls, aged 8-12 years attending the outpatient dental clinic of the Pediatric and Preventive Dentistry and Dental Public Health Department at Suez Canal University.

The study aimed to evaluate and compare the effectiveness of three distinct mouth rinses -sodium fluoride, guava leaves aqueous extract and pomegranate aqueous extract- on reducing dental plaque in children. Extra-oral and intra-oral examinations were documented for all patients utilizing examination sheets.

The parents or legal guardians of each kid provided informed permission regarding the nature of the therapy. Each one had comprehensive dental treatments as necessary until all selected children were free of caries.

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Inclusion and exclusion criteria

The children were selected according to inclusion criteria; Patients had fully erupted their first permanent molars and first premolar. They were advised not to take any therapeutic agents including antibiotics, steroids, or anti-inflammatory medications for two weeks during this trial. Children should not make any orthodontic appliance at the time of study. In addition, children should not use fluoride toothpaste any topical fluoride application, or chew xylitol gum at the time of study [8]. The exclusion criteria where children had crowding teeth. Children are mouth breathing. Children had allergies to dental products. Parents and children refuse to participate in the study. Patients refused to follow up.

Ethical approval and Clinical trial registration

The present trial was conducted at the Pediatric and Preventive Dentistry and Dental Public Health Department of the Faculty of Dentistry, Suez Canal University, after clearance from the Research Ethical Committee (REC) of the Faculty of Dentistry, Suez Canal University, according to rules from Helsinki, number (241/2019). Registration of Clinical Trials: The study is registered on ClinicalTrials.gov with ID: NCT06013735, launched on August 24, 2023.

Randomization and Allocation

This study is a randomized clinical control trial performed on (40) healthy children selected from a pool of 95 children aged (8-12) years who attend the outpatient clinic of Pediatric and Preventive Dentistry and Dental Public Health Department, Faculty of Dentistry, Suez Canal University. Selection and randomization were carried out by using random wheel selector into four groups in website https://wheelofnames.com/. Grouping and allocation based on the type of mouthwash chosen by the pointer when children rolled the wheel as follows:

- Water group: this group, serving as the control group, consists includes ten children rinsed with distilled water (control group)
- The sodium fluoride mouth rinse group (0.05%) [9] includes ten children rinsed with sodium fluoride aqueous solution (positive control)
- The Guava group (0.5%) [8] includes ten children rinsed with guava leaves aqueous extract mouth rinse
- Pomegranate group (0.5%) [10] Include ten children rinsed with pomegranate peel aqueous extract

Clinical Procedures:

Each child was examined extra and intra-oral, all detected carious teeth were restored and all participants took a new brush to encourage them. Also, gingival condition as (color, size, shape, and bleeding on probing), and a plaque was detected if it was heavy, medium, and light by necked eye according to Löe and Silness index, 1963 [11].

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In other hand, each child was evaluated at (0-baseline) for gingival index and plaque index and all participants were divided into four groups: Water group, Sodium Fluoride group, Guava group, and Pomegranate peel group according to treatment modalities.

Each child was instructed to brush their teeth with a water-wetted toothbrush, without toothpaste twice daily, using Modified Stillman's brushing technique for two weeks. Each child used mouth rinse according to different treatment modalities demonstrated for all treated children.

Each child was instructed to rinse with 15 ml of the solution for 1 minute according to different modalities, followed by expectoration of the residual mouth rinse. On follow-up 7 days and 14 days, all patients were asked not to eat or drink anything at the morning day of investigation only rinsing with mouth rinse in the dental clinic [12].

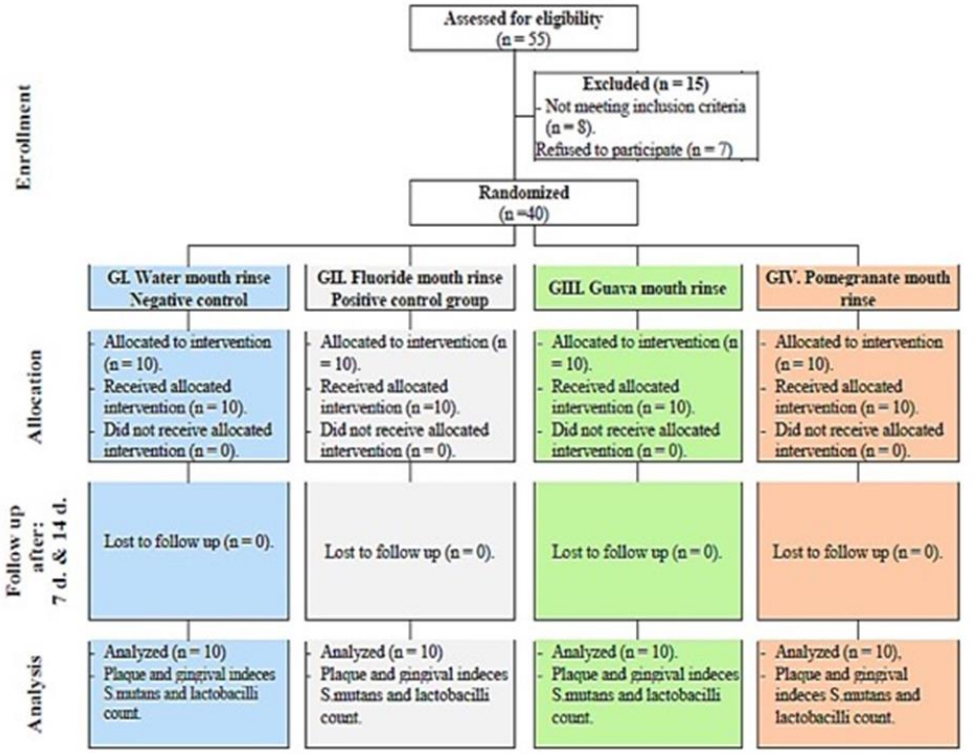

Figure 1: CONSORT diagram

Mouthwashes rinse preparation

The mouthwash rinse for sodium fluoride, guava leaves extract and pomegranate peel extract were prepared from freshly collected plant materials. All mouthwash was formulated at the Pharmaceutics Department, Faculty of Pharmacy, Mansoura University.

Sodium fluoride mouthrinse: The mouth rinse formulations were prepared by solubilizing sodium fluoride powder from Sigma Aldrich company in ethanol with constant stirring at moderate speed (100 rpm) in a 250 mL beaker. Then the other ingredients were added

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with constant stirring. The total weight was adjusted to 100 grams with distilled water [8]. The concentration *of* sodium fluoride is 0.05% for daily use [9].

Guava mothrinse was produced from guava leaves harvested from guava trees, thoroughly cleaned, air-dried for five days, pulverized, and stored in specialized bags. Guava extraction was conducted utilizing 1 kilogram of leaf powder and 2.5 L of 95% ethyl alcohol for 3 days at ambient temperature [13]. The concentration of the supernatant was carried out using a rotary evaporator set at 40 °C. The Rotavap maker states that solvent evaporation is facilitated by a high-efficiency glass condenser and collected in a bottle, significantly enhancing evaporation efficiency. The mouthwash formulations were created by dissolving guava extract in ethanol while maintaining steady stirring at a moderate speed of 100 rpm in a 250 mL beaker. Subsequently, the remaining components were incorporated with continuous agitation. The entire weight was calibrated to 100 grams using distilled water [14].

Pomegranate mouthrinse was formulated using mature pomegranate fruits (*Punica granatum*) obtained from the local market. The pericarps were hand-removed from pomegranates weighing 165 kg. Subsequently, the material underwent air drying for 10 days, was crushed into a powder, and then 1 kilogram of this powder was packaged in plastic lock bags [10]. One kilogram of pericarp was combined with 2.5 liters of 95% ethyl alcohol for 3 days at ambient temperature [13]. The concentration of the supernatant was carried out using a rotary evaporator set at 40 °C. The solvent evaporation is facilitated by a high-efficiency glass. Gummy extracts were maintained in the bakery at 4 ºC before use [12].

Mouthrinses formulations were created by solubilizing pomegranate peel extract in ethanol while maintaining steady stirring at a moderate speed of 100 rpm in a 250 mL beaker, followed by the addition of additional components with continuous stirring. The total weight was calibrated to 100 grams using distilled water [14].

METHODS OF CLINICAL EVALUATION

Gingival index

The gingival condition was evaluated by the naked eye (bleeding,redness, inflammation, and color change). Clinical evaluation by measuring gingival index scores according to Löe and Silness, 1963. It was used for the assessment of gingival condition changes. a blunt periodontal probe was passed along the gingival sulcus of selected teeth in the mesial, buccal, lingual, and distal sides for each upper (first right molar, right lateral incisor, and left first premolar) teeth and lower (left first molar, left lateral incisors and right first premolar) teeth, then waited for one minute to see the bleeding tendency.

Partially erupted teeth were excluded. The gingival index scores varied from 0 to 3, where 0 indicates no inflammation, 1 denotes mild inflammation with slight color changes, minimal edema, and no bleeding upon probing; 2 signifies moderate inflammation characterized by moderate glazing, redness, and bleeding upon probing; and score 3

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reflects severe inflammation, characterized by intense redness, gingival enlargement (hypertrophy), and spontaneous bleeding [11]**.**

Gingival index of each tooth can be obtained by adding the values of each tooth (mesial, buccal, distal, and lingual) and dividing by the sum number of the sides of one tooth (4). The individual gingival index was determined by averaging the scores of all indexed teeth, calculated as the total gingival score divided by the number of teeth assessed. Inflammation severity was classified as follows: 0.1–1.0 = mild, 1.1–2.0 = moderate, and 2.1–3.0 = severe. [11].

Plaque Index

The plaque index was assessed according to the Löe and Silness index [11]. The selected teeth were dried with air and included the following: in the upper arch — the first right molar, right lateral incisor, and left first premolar; and in the lower arch — the left first molar, left lateral incisor, and right first premolar. These teeth were visually examined under adequate lighting using the naked eye and a periodontal probe.

Examination focused on the cervical (supra-gingival) third of each tooth surface — buccal, lingual, mesial, and distal. When plaque was not visible to the naked eye, a periodontal probe was used to detect its presence.

The plaque index was scored on a scale from 0 to 3 as follows: **Score 0:** No plaque; no soft deposits are detected on the tip of the probe, **score 1:** A thin film of plaque adheres to the tooth surface and is only visible on the tip of the probe when passed across the surface; not visible to the naked eye, and **Score 2:** Moderate accumulation of plaque within the gingival pocket, along the gingival margin, and on the adjacent tooth surface; visible to the naked eye. Finally, Score **3:** Heavy accumulation of soft deposits in the gingival pocket and/or on the gingival margin and tooth surface; clearly visible.

The plaque index for each participant was calculated by summing the scores for all examined surfaces and dividing by the number of teeth assessed (n = 6). The oral hygiene condition was then categorized as follows: **0:** Excellent, **0.1–0.9:** Good, **1.0–1.9:** Fair and **2.0–3.0:** Poor.

Statistical analyses

Data were collected, verified, cleaned, and organized into tables and figures using Microsoft Excel 2016. Outliers were identified and managed using IBM SPSS Statistics version 29.0 (IBM Corp., Armonk, NY, USA).

The normality of the data distribution was assessed using the Shapiro-Wilk and/or Kolmogorov-Smirnov tests at a significance level of 0.05 to determine whether parametric or non-parametric statistical tests were appropriate.

Descriptive statistics, including both graphical and numerical methods, were used to summarize the data. Results were expressed as mean ± standard deviation (SD). For inferential analysis, comparisons among the four experimental groups over multiple time points were conducted using two-way repeated measures analysis of variance (ANOVA).

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To evaluate intra-group changes over time, one-way repeated measures ANOVA was used, while inter-group differences at each time point were assessed using one-way ANOVA.

Where significant differences were identified, Duncan's Multiple Range Test (DMRT) was applied for post hoc comparisons among treatment groups, with the significance level set at $p < 0.05$.

RESULTS

Gingival tissue was clinically assessed at the **baseline (0-day)** using the Gingival Index and Plaque Index according to the method described by **Löe and Silness**. At baseline, the gingiva exhibited **severe inflammation**, characterized by pronounced redness, hypertrophy, and rounding of the free gingiva, along with visible food debris (Figures 1a, 2a, 3a, and 4a).

The clinical measurements, including the Gingival Index and Plaque Index, were evaluated at **7 days** and **14 days** for all treatment groups based on the different treatment modalities. The observed changes in gingival condition and plaque accumulation were documented at these time points are illustrated in Figures 1b, 1c, 2b, 2c, 3b, 3c, 4b, and 4c.

1. Water Mouthrinse Group

(a) **(b)** **(c)**

Fig. (1): a) A photograph of a 10-year-old patient at baseline (0 days) showing severe gingival inflammation, characterized by redness, rounding of the free gingiva, and visible food debris in the water-treated group. b) A photograph of a patient 10 years old 7 days after oral hygiene measures, showing persistent inflammation and roundation of free gingiva in water treated group. c) A photograph of a patient 10 years old after 14 after oral hygiene measures, showing persistent inflammation and roundation of free gingiva and bleeding in water-treated groups

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2. Fluoride Mouth Rinse Group

(a) (b) (c)

Fig. (2): a) A photograph of a patient 9 years old in a 0-base line showing roundation and redness in free gingiva and food debris in a fluoride-treated group. b) A photograph of a patient 9 years old in 7days after oral hygiene measure showing decreased roundation of free gingiva and decreased inflammation of gingiva in fluoride-treated groups. c) A photograph of a patient 9 years old 14 days after oral hygiene measures showing more decreased roundation of free gingiva and more decreased inflammation of gingiva in the fluoride-treated group.

3. Guava Mouthrinse Group

(a) (b) (c)

Fig. (3): a) A photograph of a patient 10 years old in 0-base line, increased redness of free gingiva and increased inflammation of gingiva in guava treated group. b) A photograph of a patient 10 years old 7 days after oral hygiene measure, decreased redness of free gingiva and slightly decreased inflammation of gingiva and bleeding free gingiva in guava treated group. c) A photograph of a patient 10 years old 14 days after oral hygiene measure, decreased redness of free gingiva and more decreased inflammation of gingiva in guava treated group.

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4. Pomegranate Mouth Rinse Group

(a) **(b)** **(c)**

Fig. (4): a) A photograph of a patient 11 years old in a 0-base line showing roundation and redness in free gingiva and food debris in a fluoride-treated group. b) A photograph of a patient 11 years old in 7days after oral hygiene measure showing decreased roundation of free gingiva and decreased inflammation of gingiva in the fluoride-traup. c) A photograph of a patient 11 years old 14 days after oral hygiene measures showing more decreased roundation of free gingiva and more decreased inflammation of gingiva in the fluoride-treated group

Gingival index (GI) scores

Table 1 presents the Gingival Index (GI) scores as means with standard deviations. Repeated measures ANOVA analysis revealed that: In the Water group, there were no significant changes in gingival index scores across baseline, 7 days, and 14 days ($p > 0.05$). In contrast, the Fluoride, Guava, and Pomegranate groups showed highly significant differences in gingival index scores between the different time points ($p < 0.001$).

Table 1: The Gingival Index is presented as the mean and standard deviation for all treated groups

Group	Gingival Index			ANOVA RM
	Baseline	7 days	14 days	
Water	1.89±0.23 a	1.64±0.16 a	1.53±0.03 a	**0.449 ns**
Fluoride	1.85±0.12 a	0.98±0.13de	0.48±0.06 f	**<0.001*** **
Guava	1.88±0.22 a	1.40±0.14 b	1.10±0.09 cd	**<0.001*** **
Pomegranate	1.80±0.11 a	1.20±0.07 c	0.95±0.09 e	**<0.001*** **
ANOVA	**0.723 ns**	**<0.001*** **	**<0.001*** **	
Two-way Repeated Measure ANOVA				
Group	<0.001***			
Time	<0.001***			
Group x Time	<0.001***			

*, **, and *** indicate significance levels at $p < 0.05$, $p < 0.01$, and $p < 0.001$, respectively; “ns” denotes non-significant results with $p > 0.05$.

[a,b] Means followed by different letters *vertically* or *horizontally* are significantly different according to DMRTs at 0.05 level.

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Table 2 presents the percentage changes in the Gingival Index (GI) from baseline (Day 0) to Day 7 and Day 14. The Water group exhibited the smallest reduction in GI scores, with a decrease of -18.94%, which was not statistically significant according to the paired t-test ($p > 0.05$). Conversely, the Fluoride group showed the largest reduction, with a -74.38% decrease in GI scores from baseline to Day 14. This reduction was highly significant ($p < 0.001$), indicating marked improvement in gingival health.

Table 2: Change and Change (%) and Pearson's correlation presenting the change in the gingival index in water, fluoride, guava, and pomegranate groups with time 7 and 14 days

Group	7 days		14 days		Paired t-test	Correlation	
	Change	Change %	Change	Change %		R	p
Water	-0.255	**-13.48**	-0.358	**-18.94**	**0.207 ns**	-0.68	**0.224 ns**
Fluoride	-0.871	**-46.97**	-1.379	**-74.38**	**<0.001*** **	-0.973	**<0.001*** **
Guava	-0.475	**-25.33**	-0.775	**-41.33**	**<0.001*** **	-0.896	**<0.001*** **
Pomegranate	-0.604	**-33.49**	-0.858	**-47.58**	**<0.001*** **	-0.947	**<0.001*** **
ANOVA	**<0.001*** **	**<0.001*** **	**<0.001*** **	**<0.001*** **			

*, **, *** significant at *p*<0.05, <0.01, <0.001 respectively; ns, non-significant at p>0.05

Plaque Index Scores:

In Table (3), Plaque Index (PI) data are presented as means and standard deviations in Table 1. Repeated measures ANOVA showed a statistically significant difference in PI scores across baseline, 7 days, and 14 days in the Water group ($p < 0.05$). In contrast, the Sodium Fluoride, Guava, and Pomegranate groups exhibited highly significant differences in PI scores between the time points ($p < 0.001$).

Table (3): Plaque index presented as mean and standard deviation

Group	Plaque Index (Mean ± SD)			ANOVA RM
	Baseline	7 days	14 days	
Water	1.85±0.27 a	1.63±0.36 a	1.46±0.29 b	**0.032***
Fluoride	1.89±0.37 a	0.72±0.31 d	0.30±0.15 e	**<0.001*** **
Guava	1.82±0.26 a	1.48±0.30 b	1.22±0.18 c	**<0.001*** **
Pomegranate	1.85±0.28 a	1.22±0.16 c	0.90±0.07 d	**<0.001*** **
ANOVA	**0.926 ns**	**<0.001*** **	**<0.001*** **	
Two-way Repeated Measure ANOVA				
Corr. Model	<0.001***			
Group	<0.001***			
Time	<0.001***			
Group x Time	<0.001***			
*, **, and *** indicate significance levels at $p < 0.05$, $p < 0.01$, and $p < 0.001$, respectively; "ns" denotes non-significant results with $p > 0.05$.				
[a,b] Means followed by different letters vertically or horizontally are significantly different according to DMRTs.				

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Table (4): Change and Change % and Pearson's correlation presenting the change in Plaque index in water, fluoride, guava, and pomegranate groups with time (0, 7, and 14)

Group	7 days		14 days		Paired t-test	Correlation	
	Change	Change %	Change	Change %		r	p
Water	-0.211	**-12.16**	-0.387	**-20.92**	0.045*	-0.470	**0.008 ** **
Fluoride	-1.171	**-62.03**	-1.588	**-84.11**	<0.001***	-0.894	**<0.001*** **
Guava	-0.338	**-18.54**	-0.604	**-33.18**	<0.001***	-0.719	**<0.001*** **
Pomegranate	-0.633	**-34.23**	-0.950	**-51.35**	<0.001***	-0.890	**<0.001*** **
ANOVA	<0.001***	<0.001***	<0.001***	<0.001***			
*, **, and *** indicate significance levels at $p < 0.05$, $p < 0.01$, and $p < 0.001$, respectively; "ns" denotes non-significant results with $p > 0.05$.							

DISCUSSION

Herbal mouth rinses were investigated for their potential benefits in controlling plaque and inflammation. These rinses have the potential to act as adjunctive agents to conventional oral hygiene practices, especially in the prevention and control of gingivitis [15]. This aligns with the findings of John et al. [16], the American Dental Association (ADA) advises that mouthrinses should modulate the oral microbiome by selectively eliminating pathogenic bacteria while preserving the beneficial commensal microorganisms within the oral cavity. However, herbal mouthwashes may be less effective than chemical agents such as chlorhexidine (CHX) in achieving comparable antimicrobial efficacy [17].

In the current study, dehydration of leaves and peel to obtain extract occurred in fresh air not with heating to avoid evaporation essential oils and heat that may adversely affect on active substance in both.

Löe and Silness and Gingival and plaque index were employed in this study to assess gingival conditions, including color, contour, and bleeding on probing [14]. This index is a widely accepted and validated clinical method for evaluating gingival inflammation [18]. In other hand, The Plaque Index was also used to assess the severity and distribution of dental plaque accumulation among the different treatment groups [19].

Forty children, aged 8 to 12 years, were selected for participation in this study. This age group was chosen based on several factors: at this stage, the first permanent molars and premolars have erupted, which are essential for the evaluation using the Silness and Löe Plaque and Gingival Index. Additionally, children in this age range are typically well-educated and capable of performing effective oral hygiene measures, ensuring a reasonable degree of patient cooperation. This selection also mitigates concerns related to the swallowing of mouthwash, as well as the risk of fluoride toxicity in younger children under the age of 6. These criteria are in line with those of Tehrani et al. (20), who included children aged 8 to 12 years in their study. In contrast, Elkarkhy et al. (21) and Elsawy et al. (22) included children aged 6 to 12 years in their respective studies. Randomization was performed by using a random wheel selector into four groups on the website https://wheelofnames.com/ and the eligible children were randomly assigned to four

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groups in equal proportions (1:1:1:1) allocation ratio to avoid selection bias of enrolled subjects.

In this study, children were excluded if they had taken antibiotics, steroids, or anti-inflammatory medications within two weeks prior to or during the study period to minimize any potential effects on bacterial counts. This approach aligns with the findings. of Qiu et al. (23) demonstrated that antibiotics can influence the growth of cariogenic bacteria and the composition of dental plaque biofilm. Additionally, children with orthodontic appliances were excluded, as such devices may contribute plaque accumulation and elevate the levels of cariogenic microorganisms in the oral biofilm. This is supported by the study of Manphibool et al. (24).

Additionally, participants were instructed not to chew xylitol gum during the study period, as xylitol has been shown to reduce dental biofilm formation. This is in agreement with Söderling et al., (25) who found that habitual xylitol gum chewing showed plaque-reducing effects. Also, The children were instructed to avoid using fluoride toothpaste or any topical fluoride applications from the start of the study and throughout its duration to clearly assess the effect of the sodium fluoride (NaF) mouth rinse on gingival health. Also, Children had inflammation and bleeding on probing to see the effect of mouthwash in the following periods 7 and 14 days. In the current study, dehydration of leaves and peel to obtain extract occurred in fresh air, not with heating to avoid evaporation of essential oils and heat may affect active substances in both.

In this study, each participant rinsed with 15 mL of the designated solution for 1 minute, ensuring that the remaining mouthwash was expectorated and not swallowed. Rinsing was performed twice daily, once in the morning and once evening. Following tooth brushing, rinsing was performed for a duration of study.

These instructions were consistent with those of Sabri et al. (2021) [26]. Additionally, all participants were advised to brush their teeth prior to rinsing, in accordance with the American Dental Association (ADA) guidelines (2022) [27]. Patil et al. (28) emphasized that tooth brushing alone primarily cleans the buccal and lingual surfaces, but is ineffective in cleaning interproximal and subgingival plaque, especially around misaligned teeth or during the mixed dentition phase. Therefore, the use of flowable mouth rinses may help target these difficult-to-reach areas in children.

In this study, the brushing technique employed was a modified version of the Stillman's technique, which utilizes a vibratory zigzag motion with bristles angled at 45° to the long axis of the tooth. This technique is known to effectively remove dental plaque and promote gingival health. According to Patil et al. (2019), the modified Stillman's technique is one of the most effective methods for plaque control in children [29]. In contrast, Ganesh et al. (2020) reported that the horizontal scrub technique is often preferred for younger children, as they may have difficulty applying more complex brushing techniques [30]. To ensure uniform effectiveness in plaque removal, all participants were provided with new toothbrushes, both to standardize the brushing procedure and encourage compliance.

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In the current study, plaque detection was performed using a sterile supra-gingival sickle scaler, as outlined by Lu et al. (2022) [31]. This method contrasts with that of Zhang et al. (2022), who collected plaque samples using sterile cotton swabs [32].

In this present study, the 0.05% sodium fluoride mouthwash treatment group demonstrated a highly significant reduction in the gingival index at 7 days (−46.97%) and at 14 days (−74.38%) compared to baseline values. This reduction may be attributed to the fluoride's potential to reduce gum inflammation by promoting tissue health around the teeth, thus supporting the healing of irritated gum tissues associated with gingivitis. These results align with earlier research conducted by Yaseen et al. (2020), Seyam et al. (2021), and Ren et al. (2023), who also reported significant decreases in the gingival index and bleeding tendency following the use of NaF mouth rinse [33–34].

In this study, the 0.5% pomegranate peel mouth rinse showed a significant decrease in the gingival index within study duration. This may be attributed to the antioxidant properties of pomegranate peel, which likely contributed to the reduction of gingival inflammation.

In the 0.5% GLE (Guava Leaf Extract) mouthwash group, a highly significant decrease in the gingival index was observed after 7 days (−25.33%) and 14 days (−41.33%) compared to baseline values. This reduction may be attributed to the anti-inflammatory properties of guava leaves. Guava leaves contain high levels of vitamin C, which is recognized for aiding in wound healing. Vitamin C plays a crucial role in collagen synthesis, which is essential for the regeneration of gum tissues, thereby accelerating recovery from gingivitis [35].

Moreover, the plaque index results in this study showed a substantial reduction in the NaF group after 7 days (- 62.03%), as well as a decrease in plaque index at 14 days (-84.11%) with teeth brushing compared to the 0-base line in the NaF group. These findings are consistent Wiraja et al. (2007) reported that daily use of a concentration of 0.05 % sodium fluoride mouthwash reduces plaque more effectively than 0.2% chlorhexidine with tooth brushing in fixed orthodontic patients and mouth rinses are just an additional means of plaque reduction [36]. This is may be fluoride has antibacterial action.

In the 0.5% pomegranate-treated group, a significant reduction in plaque index was observed, with a percentage reduction (−34.23%) at 7 days and (−51.35%) at 14 days, in conjunction with tooth brushing, compared to baseline values. This reduction may be attributed to the antioxidant and antimicrobial properties of pomegranate peel. These findings are consistent with an in vivo study by Menezes et al. (2006), which demonstrated that pomegranate peel extract (PPE) mouth rinses exhibit antibacterial activity against selected microorganisms in dental plaque, suggesting their potential as an alternative treatment for dental plaque bacteria [37].

The plaque index in the guava leaf extract mouthwash group showed a significant reduction, with a decrease of −18.54% at 7 days and −33.18% at 14 days, in conjunction with tooth brushing, compared to baseline values. This decrease could be due to the antimicrobial effects of guava leaf extract, which contains flavonoids, tannins, and

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eugenol—compounds known for their strong antibacterial activity, thereby supporting oral hygiene [35].

Limitation:

The study faced several limitations. First, recruiting participants who met the inclusion criteria proved challenging. Additionally, the preparation of the extracts required an extended amount of time, which added to the overall complexity of the study. Furthermore, encouraging children to adhere to the oral hygiene protocol was difficult, as motivating them to consistently follow the prescribed oral hygiene measures was a challenge.

Recommendations

The results of this study suggest the need for additional clinical trials involving larger populations and longer follow-up durations to thoroughly assess the effectiveness and safety of long-term use of pomegranate peel and guava leaf extract mouthrinses in managing dental plaque and maintaining gingival health in children.

CONCLUSION

In conclusion, sodium fluoride demonstrates superior anti-inflammatory and antibacterial effects. Herbal mouthwashes, such as those containing guava leaf and pomegranate extracts, have shown improvements in gingival health and plaque control in children. These herbal alternatives are safe and effective, making them a viable option for replacing chemical mouthwashes in pediatric oral care.

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Authors' contributions

Sh.H.M.Sh participated in clinical procedures, collecting samples, study's design, data gathering, register in clinical trials.gov and paper writing. M.S.M.S.F. helped statistical analysis and review the paper. A.M.A. helped with data collection and review the manuscript. The. Final manuscript reviewed and accepted by author Sh.M.O helped in methodology, share in statistical analysis writing and review the manuscript.

Data availability

The datasets generated and analysed during the current study are available from the corresponding author on reasonable request.

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