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REVIEW ARTICLE

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Efficacy of mouthwashes based on natural products in biofilm formation and gingival inflammation in childhood: systematic review

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ABSTRACT

Introduction: Mouthwashes are used as complements to oral hygiene in children, to improve the control of dental biofilm. Products derived from herbs and plants have gained prominence in the market. **Objective:** To analyze scientific evidence on the effectiveness of mouthwashes made from natural ingredients in controlling biofilm and gum inflammation in children, compared to chlorhexidine. **Methods:** A qualitative systematic review was carried out, with data research in June 2024, using electronic databases and gray literature. **Results:** Six randomized clinical trials were selected as they met the inclusion criteria. To assess the risk of bias in the chosen studies, the Cochrane Collaboration's RoB 2 tool for randomized studies and the GRADE method were used to determine the level of evidence and strength of recommendation. It was observed that mouthwashes containing natural ingredients such as *Aloe Vera L.*, *Azadirachta indica L.*, Triphala, and *Cymbopogon citratus L.* presented results comparable to chlorhexidine in reducing dental biofilm and gingival inflammation, according to periodontal indices. The quality of evidence is moderate to low. Future clinical trials should understand the limitations associated with the different compositions and mechanisms of action of natural products. Due to the heterogeneity of the studies, no meta-analysis was performed. **Conclusion:** Although mouthwashes made with plant extracts appear to be a complementary alternative in supporting the mechanical control of dental biofilm in children, the evidence derives from studies with moderate to low methodological quality. Registration in the PROSPERO database: CRD42022321139.

Keywords: mouthwashes; child; biological products; periodontal diseases; chlorhexidine.

INTRODUCTION

There is a small number of epidemiological studies that discuss the presence of periodontal diseases in childhood. According to the American Academy of Periodontics¹, they can be divided into gingival diseases/conditions and periodontitis²⁻⁴. The presence of bacteria supra and subgingival biofilm is considered an essential factor in the development of periodontal disease^{5,6}.

In childhood, such evolution is associated with factors such as manual dexterity, differences in biofilm composition, and tooth eruption chronology^{7,8}. In addition, chemical control may be indicated as an adjuvant therapy, such as the use of chlorhexidine (CHX)^{9,10}. This substance is considered a broad-spectrum antiseptic and the gold standard in the chemical control of bacterial plaque¹¹. However, its long-term use can cause adverse effects such as stains on the teeth and tongue, and taste disturbances and oral mucosal alterations¹² and a potential risk of bacterial resistance¹³.

On the other hand, the use of mouthwashes based on natural products such as Aloe Vera L.¹⁴, *Punica granatum L.*¹⁵ has a potential inhibitory effect against oral microorganisms, when compared to synthetic oral antimicrobials^{16,17}. Among the main advantages, easy access by the population, revival of traditional practices, and low cost stand out, being evaluated through clinical and laboratory tests¹⁸⁻²⁰. Natural products also have side effects, associated with the dose and frequency used, which encourages clinical research in the development of substances with lower toxicity and better biocompatibility²¹⁻²⁴.

Based on this, this study aimed to evaluate scientific evidence of the effectiveness of natural products mouthwashes used as an adjunct in the treatment of periodontal disease in childhood, compared to chlorhexidine, through a systematic review.

METHODS

To carry out this qualitative systematic review, the recommendations for Preferred Reporting Items for Systematic Reviews and Meta-Analyses Protocols (PRISMA-P)²⁵ were followed in preparing the protocol for this study, which was registered in the PROSPERO database under protocol CRD42022321139.

For the preparation and description of the study, the established question was "Are mouthwashes based on natural products as effective in the treatment of periodontal diseases in children as chlorhexidine?"

Eligibility criteria

Randomized clinical trials without restriction of date and language were selected based on question PICO²⁶: Population (P) = children between 6 and 12 years old with periodontal disease; Intervention (I) = mouthwash based on natural products; Comparison (C) = chlorhexidine-based mouthwash; Outcome (O) = efficacy in the treatment of periodontal disease based on reduction in periodontal parameters related to plaque or gingivitis/periodontitis as an outcome measure (Figure 1).

Other types of publication were excluded, which associated the diagnosis of periodontal disease with systemic alterations, special needs or orthodontic treatment, deleterious habits such as smoking, submitted to oral prophylaxis, non-surgical and/or surgical periodontal therapy in the last six months. Before the study, pregnant and lactating women with a history of antibiotic and/or anti-inflammatory use in the last three months before the study were excluded.

Araujo et al. Efficacy of mouthwashes based on natural products in biofilm formation and gingival inflammation in childhood: systematic review. ABCS Health Sci. [Epub ahead of print]; DOI: 10.7322/abcshs.2024219.2958

Information sources and search strategy

The literature search was carried out on June 10, 2024, and the electronic databases Scopus, Medline via PubMed, Web of Science, Latin American and Caribbean Literature in Health Sciences (Lilacs), and Medicines Traditional, Complementary and Integrative (MCTI), via the Virtual Health Library (VHL) and Cochrane Library database were used as a source of studies^{25,26}. The search strategy used a combination of controlled vocabulary and free terms as shown in Table 1, following the syntax rules specific to each database and Boolean operators (OR, AND), organized according to the PICO strategy.

Gray literature was also consulted by accessing the Open Gray websites (<https://opengrey.eu>), Brazilian Digital Library of Theses and Dissertations (BDTD) (<https://bdtd.ibict.br>), Clinical Trials (<https://clinicaltrials.gov>), Brazilian Registry of Clinical Trials (ReBEC) (<https://ensaiosclinicos.gov.br>), and Google Scholar, in the first 100 records. A manual search was also carried out in the reference lists of selected articles to find studies relevant to the subject under research.

Then, the Rayyan²⁷ application was used to save and organize all references and read titles and abstracts. After removing duplicates, CCNA and VFFF reviewers screened studies by reading titles and abstracts based on inclusion and exclusion criteria. Following the methodological rigor of the systematic review for the development of the study, a consensus meeting was held between reviewers to define conflicts. Subsequently, full texts of potentially relevant articles were retrieved and thoroughly reviewed to verify their eligibility. After the complete reading of the selected articles, the results were compared, and divergences were resolved by a third reviewer (GACGC). Cohen's Kappa coefficient (κ) was used to assess the agreement between the two reviewers in the selection of articles. The Kappa value was 0.75, indicating substantial agreement, and a 95% confidence interval.

Data collection and bias analysis

To ensure the reliability of article selection and data extraction, calibration was performed between reviewers by discussing the inclusion/exclusion criteria and applying the data extraction form. Ten articles were then randomly selected for initial calibration. The initial agreement assessed by the Kappa index was 0.75. After discussing the articles with disagreements, the agreement increased to 0.90. Disagreements were resolved by consensus among the reviewers.

Data extraction was performed by CCNA and VFFF reviewers independently. Data were compared, and disagreements were resolved with the third GACGC reviewer. In case of missing data or indeterminate information, attempts were made to contact the first/corresponding author of the articles included in the study.

Through a spreadsheet created in Microsoft Excel (Microsoft 365, version 2019) to standardize the data to be extracted, the following information was collected: first author, year, country, study design, age group, number of patients, natural product and its concentration used in the intervention group and control, with chlorhexidine, as well as dosage used, study duration, periodontal indices analyzed, main results, statistical tests and possible adverse events in the experimental and control groups.

Two reviewers, CCNA and VFFF, independently conducted a risk of bias assessment of the included studies using the Cochrane Collaboration's ROB 2 Risk of Bias 2²⁸ tool for randomized clinical trials by checking five Bias domains. Studies were judged for each domain and classified on their overall assessment as either some concerns or low/high risk of bias.

Level of evidence

In accordance with GRADE²⁹ (Grading of Recommendations, Assessment, Development and Evaluation), the level of evidence for narrative analysis was established. This method synthesizes the available evidence through five key steps that assess factors such as risk of bias, imprecision, inconsistency, Indirectness, and the likelihood of publication bias. The goal is to evaluate the quality of the evidence and the strength of the recommendations across the six studies included in the systematic review.

RESULTS

Selection of studies

A total of 6,762 citations were retrieved from searches across all electronic sources. After reviewing the titles and abstracts to filter the content, 2,707 studies were removed due to duplication, resulting in 4,055 articles remaining. After reading the titles and abstracts, 3206 were excluded for not being associated with the theme proposed by the study, 826 did not meet the inclusion criteria, hierarchically: literature/systematic review (85).

The result was 23 studies that included participants aged between 06 and 12 years. However, in 14 of them, the age of the study participants was included, but without stratification by age group determined in the study, and in three, there were no associated periodontal indices, which were subsequently excluded. Only six articles advanced to the qualitative synthesis and data extraction stage.

Table 2 summarizes the characteristics of the 06 qualified RCTs^{14,23,24,30-32} that evaluated the comparison of clinical effects between manipulated/commercial experimental formulations of natural product mouthwashes and the positive control group with formulations

Araujo et al. Efficacy of mouthwashes based on natural products in biofilm formation and gingival inflammation in childhood: systematic review. ABCS Health Sci. [Epub ahead of print]; DOI: 10.7322/abcshs.2024219.2958

of chlorhexidine, in a total of 1574 participants. Three studies were conducted in India^{23,30,31}, one study in Brazil²⁴, in Syria¹⁴ and one in Egypt³².

All included studies were carried out after the year 2011³⁰, two between the years 2014³¹ and 2015²⁴, and three after the year 2020^{14,23,32}, which may indicate greater popularity and scientific interest in mouthwashes based on natural products, biocompatibility, accessibility, and diversity of raw materials.

These studies involved mouthwashes based on several natural products of plant origin and concentrations: *Aloe vera L.* 100%¹⁴, *Cymbopogon citratus L.* 0.25%²³, *Punica granatum L.* 15%²⁴, *Azadirachta indica L.* 2%³² and Triphala 0.60%^{30,31}, which is the combination of the extract of three fruits from the trees *Emblica officinalis*, *Terminalia Chebula*, and *Terminalia Belerica*³⁰. The control groups tested in such studies used CHX, in concentrations between 0.1%^{14,24,30-32} to 0.2%²³. In both groups, mouthwashes were used as an adjuvant therapy to prophylaxis and oral hygiene instructions. As for the duration of the intervention, different intervals were observed with the tested products: 5 days¹⁴, 14 days²⁴, 15 days³¹, 21 days^{23,32}, and 270 days³⁰, according to Table 2.

Microorganism control was evaluated by counting total bacterial colonies of *Streptococcus mutans/Lactobacillus sp.*³⁰ and *Streptococcus spp.*²⁴, obtained from salivary collection, showing standard deviation values at baseline and final time for each product, with a reduction in bacterial growth in both studies.

As shown in Table 3, dental biofilm control was measured at baseline and at different study intervals, represented by the mean, median, and standard deviation of the following parameters: Quigley and Hein¹⁴, Silness and Loe^{23,24,30,31} plaque index. To evaluate gingival inflammation, bleeding indices on probing, Ainamo and Bay^{14,24,32} and gingival Loe and Silness^{14,23,30,31} were used on probing. Mahmoud et al.³² showed greater reductions in the

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bleeding rate after the use of mouthwashes based on *Azadirachta indica* L. 2%, while for Nobrega et al.²⁴ there was no efficacy of the mouthwash based on *Punica Granatum* L. compared to the control group. Four studies^{14,23,30,31} revealed no difference between herbal mouthwashes and CHX, used as an adjunct, to control inflammation and dental biofilm.

About side effects, only one study¹⁴ provided a questionnaire at the end of the period of application of each type of mouthwash for all children, through three questions about burning sensation in the oral cavity, taste alteration, and teeth pigmentation, with no report by any of the children who participated in the study. All articles included mentioned supervision of children by parents/guardians during and after using mouthwash.

Differences in diluents were observed when handling mouthwashes based on natural products. Two of them were performed with the use of hydroalcoholic solution²⁴ and ethyl alcohol²³. Other studies did not mention the diluent medium used. Due to the heterogeneity of results, as differences in heterogeneity of diluents, dosage, and natural product used, it was not possible to perform a meta-analysis.

Risk of bias assessment

Figure 2 shows the risk of bias analysis performed according to the ROB 2 tool²⁸. Three studies^{23,30,32} showed risk of bias in the selection of the reports' results in the "Other biases". The study by Alnouri et al.¹⁴ had "low risk of bias" in all other domains, being considered as a reference among the articles included in this systematic review. Three studies were considered at "high risk of bias": the study by Akula et al.²³, for presenting methodological problems in the process of randomization and outcome measurement, as well as the study by Mahmoud et al.³² and the study by Bajaj and Tandon³⁰ showed a moderate risk of bias due to the presence of confounding factors in the result analysis.

Assessment of the certainty of the level of evidence by the GRADE tool

As indicated in Table 4, the plaque index exhibited a low level of evidence across the five clinical trials that used it as an outcome. The certainty of the evidence was rated as low, with results being inconsistent, likely due to clinical heterogeneity or variations related to the interventions. In addition, when analyzing the bleeding index on probing, there is a serious inaccuracy due to the number of participants being less than 400. Regarding the gingival index, there is low certainty of evidence related to only one study¹⁴.

DISCUSSION

There is a growing scientific interest in the use of natural products in the health area due to their biocompatibility and lower toxicity¹⁸. Literature search^{33,34} previously carried out evaluated the effectiveness of such an approach in Dentistry. However, this systematic review provides a comprehensive analysis of scientific studies, based on the certainty of evidence, directed to the use of mouthwashes based on natural products in the treatment of periodontal diseases in children in the mixed dentition phase, between 06 and 12 years old.

In this age group, aspects such as histological and anatomical alterations of the periodontal tissue, such as increased probing depth, related to the exfoliation of deciduous teeth and eruption of permanent teeth, reduced manual dexterity for effective mechanical control of plaque³⁵ and the beginning of colonization of Gram-negative bacteria such as *Prevotella intermedia* and *Tannerella forsythia*^{8,36} suggest a greater susceptibility to periodontal diseases such as gingivitis³.

In this systematic review, six qualified randomized clinical studies were included^{14,23,24,30-32}, and found heterogeneity of natural products researched, based on plant

Araujo et al. Efficacy of mouthwashes based on natural products in biofilm formation and gingival inflammation in childhood: systematic review. ABCS Health Sci. [Epub ahead of print]; DOI: 10.7322/abcshs.2024219.2958

extracts, a reflection of the progressive market search for natural products that have antiplaque efficacy and antimicrobial action against oral microorganisms²¹. In all studies, the initial periodontal analysis was about gingivitis, without relating to the diagnosis of periodontitis. Possibly, this fact must be related to the lower prevalence of this pathology among the age group determined for the study, since gingivitis represents about 80% of periodontal diseases in children with mixed dentition^{3,4}.

The studies included participants between 8 and 12 years old, associating mouthwash as a supporting method in oral hygiene. In this age group, some peculiar aspects are relevant in the evolution of studies. All of them included guidance on supervising the use of mouthwash for parents and guardians, a fundamental step in maintaining the safety of using mouthwashes in children^{37,38}. It is worth mentioning that, according to the recommendations of the American Dental Association (ADA), the use of mouthwash should be indicated for children over 06 years of age due to the improvement in their physiological capacity to perform mouthwash safely.

Five qualified studies^{14,23,30-32} found that the natural products *Cymbopogon citratus L.*, *Aloe Vera L.*, *Triphala*, and *Azadirachta indica L.* had similar or better effects than CHX in reducing plaque and gingival indices when compared at baseline and the end of the study. Despite a significant reduction in such indices, these results do not indicate that herbal products can be replaced by CHX because, until now, there is not enough scientific evidence to demonstrate the superiority of natural products and their possible side effects when compared to CHX^{18,39,40}. For Martins et al.²², mouthwashes based on phenolic compounds are less effective when compared to synthetic antimicrobials in improving mean plaque index and reducing biofilm over time in children and adolescents.

The study by Nóbrega et al.²⁴ concluded that 6.25% pomegranate peel-based mouthwash (*Punica Granatum L.*) was not effective in reducing dental biofilm and gingival inflammation,

Araujo et al. Efficacy of mouthwashes based on natural products in biofilm formation and gingival inflammation in childhood: systematic review. ABCS Health Sci. [Epub ahead of print]; DOI: 10.7322/abcshs.2024219.2958

despite its inhibitory effect on oral streptococci counts. However, randomized clinical studies^{41,42} demonstrate that *Punica granatum* extract inhibits the adhesion of microorganisms associated with bacterial plaque. This can be explained by the influence of factors such as preparation or extraction method, concentration of active compounds used in the study, on the anti-inflammatory and antimicrobial effect¹⁵.

Differences were observed in the dosage and intervention time of use of mouthwashes used in the intervention and control groups (Table 2). The absence of a standard dose in the prescription of natural products makes it difficult to compare with CHX⁴³. However, it is important to emphasize that establishing safe doses, degree of toxicity, and cost-effectiveness of natural products should be considered for a better understanding of their therapeutic indication in periodontal diseases^{21,39}.

In control groups, CHX was used at different concentrations. The lowest, found in the study by Bajaj and Tandon³⁰, CHX 0.1%, showed a similar effect to the use of *Triphala* 0.6% in the prevention and formation of plaque after 09 months. According to Haydari et al.¹² and James et al.⁴⁴, CHX 0.1%, twice a day, offers the same clinical benefits as a 0.2% solution, requiring longer use, in addition to producing fewer side effects such as pigmentation and reduction of bitter taste, favoring acceptance by children.

All studies used plant extracts with secondary metabolites such as flavonoids, tannins, terpenoids, phenolic compounds, alkaloids, myrcene, saponin, xanthones, anthocyanins, among others, found in several plants with antimicrobial activity^{45,46}. However, the mechanism of antiplaque actions and reduction of gingival inflammation of mouthwashes with natural products is still unclear due to their metabolic complexity⁴⁷.

In the analysis of the methods of preparing mouthwashes based on natural products, it was necessary to contact the authors for information, but there was no further clarification. Two

Araujo et al. Efficacy of mouthwashes based on natural products in biofilm formation and gingival inflammation in childhood: systematic review. *ABCS Health Sci.* [Epub ahead of print]; DOI: 10.7322/abcshs.2024219.2958

studies^{23,24}, during the preparation stage of natural products, used hydroalcoholic solutions. This fact can alter the result, since alcohol can modify the antimicrobial action^{34,48}. In addition, factors such as extraction methods, climate, and harvest time, among others, can change the concentration of active compounds, thus influencing the potential antimicrobial effect of natural extracts⁴⁹. Therefore, it is suggested that future studies use isolated metabolites and test the chemical composition of the product to identify the mechanism of action and possible interactions between the compounds present.

To evaluate the effectiveness of natural products in treating periodontal diseases in mixed dentition⁵⁰, the study utilized the gingival index (GI), bleeding on probing index (BOP), and plaque index (PI) both at the beginning and during the clinical follow-up phase. Except for Nóbrega et al.²⁴, the means and statistical analysis (p-value) of other studies showed a reduction in biofilm and gingival bleeding comparable to the use of chlorhexidine. In the systematic review by Chen et al.⁴⁰, mouthwashes based on natural products showed potential anti-inflammatory, antimicrobial, and antioxidant properties. Despite the improvement in the different evaluated parameters, in most trials, there was no statistically significant difference between the tested group and CHX, indicating that further studies are needed to verify the positive effects of plant extracts as adjuvant therapy in periodontal disease in childhood.

Adaptations of data measurement in the periodontal indices used during the mixed dentition and description of which teeth or faces were examined were not described^{23,24,30-32}. However, it is relevant to consider that exfoliation of deciduous teeth and eruption of permanent teeth affect gingival conditions in this age group^{18,50}. Pseudopockets can make it difficult to identify periodontal disease, and the indices used should be restricted to erupted permanent dental units, reducing the potential methodological bias^{51,52}.

Only one study¹⁴ evaluated the side effects of Aloe Vera-based mouthwash, through a questionnaire applied to children about burning sensation in the oral cavity, alteration in taste, and teeth pigmentation, with no reports. However, there is still no consensus on potential side effects, and a greater number of studies are still essential to evaluate the safety of herbal mouthwashes and characteristics such as toxicity and biocompatibility, contributing to the understanding of the mechanisms of action of natural products^{33,53}. CHX, despite the numerous side effects such as staining, desquamation of the intraoral mucosa⁵⁴, should still be considered the most indicated therapeutic approach in periodontal diseases in children.

The six clinical trials were submitted to an instrument for evaluating results (Figure 2) using the Cochrane Collaboration tool for randomized studies (RoB 2.0), which analyzes the data presented and methodological items that must be considered when evaluating an article. Three studies^{23,30,32} showed a high risk of bias. Additionally, two studies were classified as having some concerns^{24,31} and Alnouri et al.¹⁴, with low risk of bias. Biases were found to be associated with the lack of description of the reviewer's calibration in obtaining periodontal and plaque indices, information on product handling, and blinding of participants and researchers. Given these findings, it is essential to develop new studies with rigorous methodology to evaluate the proposed criteria.

Another aspect of fundamental importance when choosing a mouthwash refers to its substantivity, which is determined by its therapeutic efficacy and bactericidal and bacteriostatic effect over time. Studies prove that CHX has 12h substantivity, being highly resolving as a complementary approach in periodontal diseases in children^{44,55}. However, the substantivity of mouthrinses based on natural products is still unknown, and further studies are needed to assess their microbiological potential to determine their optimal dosage and duration of action.

As for the level of evidence of the studies analyzed using GRADE²⁹, it was considered low to moderate (Table 4). Parameters consider a sample of less than 400 participants as a qualifier for imprecision. To adequately observe the proposed intervention, larger sample sizes are important, eliminating the hypothesis of a probable placebo effect, based on the need to perform a sample calculation. In addition, different means of intervention were used in the studies, and it was not possible to define whether the mouthwash based on a natural product is an effective adjuvant method in the evolution of periodontal disease during the mixed dentition.

In the analysis of the results, the heterogeneity of data such as sample size, difference between natural products studied, evaluation time of the intervention and control groups, did not allow performing a quantitative analysis through meta-analysis, being considered a limitation of this study. However, this fact does not change the validity of the results found.

Based on the results of this systematic review, there is no consensus on the indication of herbal mouthwashes for the treatment of periodontal diseases in children as a substitute for the use of chlorhexidine. There is a need for studies with adequate methodological rigor regarding the composition, concentration and mechanism of action of the active compound formulations, in addition to the evaluation of the cost-benefit and long-term efficacy of natural products to prove the efficacy of the use of natural dental products in children, to base the clinical decision to prescribe such products with quality and efficacy on scientific evidence.

Conclusion

It can be concluded that, despite the use of mouthwashes based on plant extracts, combined with the restriction of adverse effects and biocompatibility, these are presented as an auxiliary alternative in the mechanical control of bacterial plaque, in the age group of 6 to 12 years, the evidence from the studies is of low methodological quality and in relation to

Araujo et al. Efficacy of mouthwashes based on natural products in biofilm formation and gingival inflammation in childhood: systematic review. *ABCS Health Sci.* [Epub ahead of print]; DOI: 10.7322/abcshs.2024219.2958

periodontal parameters, such as GI and PI, and the certainty of the evidence is between low and moderate. Therefore, additional research is needed to establish efficacy, adequate dosage, and potential side effects at different concentrations before herbal mouthwashes can be recommended as a reliable alternative to traditional mouthwashes for children.

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Araujo et al. Efficacy of mouthwashes based on natural products in biofilm formation and gingival inflammation in childhood: systematic review. ABCS Health Sci. [Epub ahead of print]; DOI: 10.7322/abcshs.2024219.2958

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Table 1: Search strategy used in the electronic database.

Data Base	Search Strategy
SCOPUS	(((TITLE-ABS-KEY (periodontal AND diseases) OR TITLE-ABS-KEY (gingivitis) OR TITLE-ABS-KEY (chronic AND periodontitis) OR TITLE-ABS-KEY (periodontitis) OR TITLE-ABS-KEY (child) OR TITLE-ABS-KEY (child*) OR TITLE-ABS-KEY (dental AND plaque) OR TITLE-ABS-KEY (tooth AND plaque) OR TITLE-ABS-KEY (biofilms) OR TITLE-ABS-KEY (biofilm))) AND ((TITLE-ABS-KEY (phytotherapy) OR TITLE-ABS-KEY (phyto*) OR TITLE-ABS-KEY (herbal AND medicine) OR TITLE-ABS-KEY (biological AND products) OR TITLE-ABS-KEY (natural AND product*) OR TITLE-ABS-KEY (plants, AND medicinal) OR TITLE-ABS-KEY (plant*) OR TITLE-ABS-KEY (plant AND extracts) OR TITLE-ABS-KEY (plant AND oils) OR TITLE-ABS-KEY (essencial AND oil*) OR TITLE-ABS-KEY (plant AND preparations) OR TITLE-ABS-KEY (mouthwashes) OR TITLE-ABS-KEY (herbal AND mouthwashes) OR TITLE-ABS-KEY (rinse) OR TITLE-ABS-KEY (mouthwash*) OR TITLE-ABS-KEY (chlorhexidine))) AND ((TITLE-ABS-KEY (dental AND plaque AND index) OR TITLE-ABS-KEY (periodontal AND index)))
PubMed	((((((((((((((((((((((((((((((Periodontal diseases[MeSH Terms]) OR (Periodontal diseases[Title/Abstract])) OR (Gingivitis[MeSH Terms])) OR (Gingivitis[Title/Abstract])) OR (Chronic Periodontitis[MeSH Terms])) OR (Chronic Periodontitis[Title/Abstract])) OR (Periodontitis[MeSH Terms])) OR (Periodontitis[Title/Abstract])) OR (Child[MeSH Terms])) OR (Child[Title/Abstract])) OR (Child*[Title/Abstract])) OR (Dental plaque[MeSH Terms])) OR (Dental plaque[Title/Abstract])) OR (Tooth plaque[Title/Abstract])) OR (Biofilms[MeSH Terms])) OR (Biofilms[Title/Abstract])) OR (Biofilm[Title/Abstract])) AND (((((((((((((((((((((((((((Phytotherapy[MeSH Terms]) OR (Phytotherapy[Title/Abstract])) OR (Phyto*[Title/Abstract])) OR (Herbal Medicine[MeSH Terms])) OR (Herbal Medicine[Title/Abstract])) OR (Herbal[Title/Abstract])) OR (Herb*[Title/Abstract])) OR (Biological Products[MeSH Terms])) OR (Biological Products[Title/Abstract])) OR (Natural Product*[Title/Abstract])) OR (Plants, medicinal[MeSH Terms])) OR (Plants, medicinal[Title/Abstract])) OR (Plant*[Title/Abstract])) OR (Plant extracts[MeSH Terms])) OR (Plant extracts[Title/Abstract])) OR (Plant oils[MeSH Terms])) OR (Plant oils[Title/Abstract])) OR (Essencial oil*[Title/Abstract])) OR (Plant preparations[MeSH Terms])) OR (Plant preparations[Title/Abstract])) OR (Mouthwashes[MeSH Terms])) OR (Mouthwashes[Title/Abstract])) OR (Herbal mouthwashes[Title/Abstract])) OR (Rinse[Title/Abstract])) OR (Mouthwash*[Title/Abstract])) OR (Chlorhexidine[MeSH Terms])) OR (Chlorhexidine[Title/Abstract])) AND (((Dental plaque index[MeSH Terms]) OR (Dental plaque index[Title/Abstract])) OR (Periodontal Index[MeSH Terms])) OR (Periodontal Index[Title/Abstract]))"
Web of Science	#1 Periodontal diseases (Topic) or Gingivitis (Topic) or Chronic Periodontitis (Topic) or Periodontitis (Topic) or Child (Topic) or Child* (Topic) or dental plaque (Topic) or Tooth Plaque (Topic) or Biofilms (Topic) or Biofilm (Topic) #2 Phytotherapy (Topic) or Phyto*(Topic) or Herbal Medicine (Topic) or Biological Products (Topic) or Natural Product*(Topic) or Plants, medicinal (Topic) or Plant* (Topic) or Plant extracts (Topic) or Plant oils (Topic) or essencial oil* (Topic) or Plant preparations (Topic) or Mouthwashes (Topic) or herbal mouthwashes (Topic) or Rinse (Topic) or Mouthwash* (Topic) or Chlorhexidine (Topic) #3 Dental plaque index (Topic) or Periodontal Index (Topic) #1AND #2 AND #3
Lilacs/BVS	((Periodontal diseases OR Enfermedades Periodontales OR Doenças periodontais OR Gingivitis OR Gingivitis OR Gengivite OR Chronic Periodontitis OR Periodontitis Crônica OR periodontite crônica OR Periodontitis OR Periodontitis OR periodontite OR Child OR Niño OR Criança OR Child\$ OR Dental plaque OR Tooth plaque OR Placa dental OR placa dentária OR Biofilms OR Biopelículas OR biofilmes OR Biolfim) AND (Phytotherapy OR Fitoterapia OR Fitoterapia OR Phyto\$ OR Herbal Medicine OR Medicina de hierbas OR medicina herbária OR Herbal OR Herb\$ OR Biological Products OR Productos Biológicos OR Produtos Biológicos OR Natural Product\$ OR Plants, Medicinal OR Plantas Medicinales OR plantas medicinais OR Plant\$ OR Plant extracts OR Extractos Vegetales OR Extratos Vegetais OR Plant Oils OR Aceites Vegetales OR Óleos Vegetais OR Essencial oil\$ OR Plant Preparations OR Preparaciones de Plantas OR Preparações de Plantas OR Mouthwashes OR Antisépticos Bucales OR Antissépticos Bucais OR Herbal mouthwashes OR Rinse OR Mouthwash\$ OR Chlorhexidine OR Clorhexidina OR Clorexidina) AND (Dental plaque index OR Índice de Placa Dental OR Índice de Placa Dentária OR Periodontal Index OR Índice Periodontal OR Índice Periodontal))
MCTI/BVS	((periodontal diseases) OR (enfermedades periodontales) OR (doenças periodontais) OR (gingivitis) OR (gingivitis) OR (gengivite) OR (chronic periodontitis) OR (periodontitis crônica) OR (periodontite crônica) OR (periodontitis) OR (periodontitis) OR (periodontite) OR (child) OR (niño) OR (criança) OR (dental plaque) OR (placa dental) OR (placa dentária) OR (tooth plaque) OR (biofilms) OR (biopelícula)s OR (biofilmes)) AND ((phytotherapy) OR (fitoterapia) OR (fitoterapi)a OR (herbal medicine) OR (medicina de hierbas) OR (medicina herbária) OR (plants, medicinal) OR (plantas medicinales) OR (plantas medicinais) OR (plant extracts) OR (extractos vegetales) OR (extratos vegetais) or (plant oils) OR (aceites vegetales) OR (óleos vegetais) OR (plant preparations) OR (preparaciones de plantas) OR (preparações de plantas) OR (mouthwashes) OR (antisépticos bucales) OR (antissépticos bucais) OR (chlorhexidine) OR (clorhexidina) OR (clorexidina)) AND ((dental plaque index) OR (índice de placa dental) OR (índice de placa dentária) OR (periodontal index) OR (índice periodontal) OR (índice periodontal))
COCHRANE Library	Periodontal diseases OR Gingivitis OR Chronic Periodontitis OR Periodontitis OR Child OR Child* OR Dental plaque OR Tooh Plaque OR Biofilms OR Biofilm AND Phytotherapy OR Phyto* OR Herbal Medicine OR Biological Products OR Natural Product* OR Plants, medicinal OR Plant* OR Plant extracts OR Plant oils OR Essencial oil* OR Plant preparations OR Mouthwashes OR Herbal mouthwashes OR Rinse OR Mouthwash* OR Chlorhexidine AND Dental plaque index OR Periodontal Index.

Table 2: Summary of characteristics of included studies.

Study	Country	Study design	AGE Group	Sample	Natural Product	Control Group	Duration of the Intervention	posology (amount/duration/frequency)
Bajaj e Tandon, 2011 ³⁰	Índia	RCT* double blind	08 to 12	1309	Triphala 0.60% (manipulated)	CHX** 0.10% (commercial)	270 days	10 ml; 02 min; 01x/day
Bhattacharjee <i>et al.</i> 2015 ³¹	Índia	RCT* double blind	08 to 12	57	Triphala 0.60% (manipulated)	CHX** 0.12% (manipulated)	14 days	NI ml; 30 seg; 02x/day
Nóbrega <i>et al.</i> 2015 ²⁴	Brazil	RCT* double blind	09 to 12	35	<i>Punica granatum</i> L. 6.25% (manipulated)	CHX** 0.12% (manipulated)	14 days	10 ml; 01 min; 02x/day
Alnouri <i>et al.</i> 2020 ¹⁴	Syria	RCT* Crossed, triple blind	08 to 12	17	<i>Aloe Vera</i> L. 100% (manipulated)	CHX** 0.12% (manipulated)	05 days	10 ml; 01 min; 01x/day
Akula <i>et al.</i> 2021 ²³	Índia	RCT*	9 to 12	60	<i>Cymbopogon citratus</i> L.0.25% (manipulated)	CHX 0.2% (commercial)	21 days	10 ml; 01 min; 02x/day
Mahmoud <i>et al.</i> 2022 ³²	Egypt	RCT*	08 to 10	96	<i>Azadirachta indica</i> L. 2% (manipulated)	CHX** 0.12% (manipulated)	21 days	05 ml; NI min; 02x/day

Table 3: Results found.

Study	Plaque Index Baseline/Final (intervention group/CHX)	Periodontal Parameters Baseline/Final (intervention group/CHX)	Statistical Test	Statistical Analysis (Time/intergroup)	Results
Bajaj e Tandod, 2011 ³⁰	*1T0=0,84 T1(90d)=0,74 T2(180d)=0,58 T3(270d)=0,49 T0= 0,76 T1(90d)=0,70 T2(180d)=0,72 T3(270d)=0,61	*3T0=0,59T1(90d)=0,53 T2(180d)=0,47 T3(270d)=0,40 T0=0,54 T1(90d)=0,50 T2(180d)=0,54 T3(270d) =0,46	Paired T Test	*1T0 /T3 p=0,826 *3T0/T3 p=0,048	Triphala 0.6% had an effect. Similar to CHX 0.1% and biofilm formation after 270 days
Bhattacharjee <i>et al.</i> 2015 ³¹	*1T0=1,11 T1 (14d) =0,76 T0=1,17 T1(14d) =0,73	*3T0=0,93 T1 (14d) =0,53 T0=0,72 T1(14d) =0,42	ANOVA Mann-Whitney U test	*1p=0,826 *3p=0,048	The efficacy of Triphala is comparable to CHX in reducing biofilm
Nóbrega <i>et al.</i> 2015 ²⁴	*1T0=0,74 T1(07d)=0,63 T2(14d)=0,69 T0=0,77 T1(7d)=0,24 T2(14d)=0,27	*4T0=8,07 T1(07d)=6,65 T2(14 dias)=5,71 T0=5,11 T1(7d)=6,38 T2(14d) =6,01	ANOVA Students' T-test	*1p<0,001 *4 p=0.815	P. granatum mouthwash was not effective in controlling dental biofilm
Alnouri <i>et al.</i> 2020 ¹⁴	*2T0=23,5 T1(03 d)=23,04 T2(05d)=16,5 T0=31,06 T1(14d)=33,32 T2(21d)=23,91	*3T0=19,4 T1(03 d)=24,50 T2(05d)=17,88 T0=35,59 T1(14d)=24,50 T2(21d)=21,76 *4T0=0,15 T1(03d)=0,87 T2(05d)=0,07 T0=0,82 T1(14d)=0,87 T2(21d)=0,19	Kruskal-Walli test Shapiro-Wilk test Mann-WhitneyUtest	*2T2 p=0,090 *3T2 P=0,377	Efficacy of Aloe Vera mouthwash in improving plaque and gingival indices in children.
Akula <i>et al.</i> 2021 ²³	*1T0= 1,23 T1(14d)= 1,05 T2(21d) =0,92 T0=1,26 T1(14d)=1,12 T2(21d)=1,06	*3T0=1,34 T1(14d)=1,11 T2(21d)=0,98 T0=1,38 T1(14d)=1,23 T2(21d)=1,11	ANOVA	*1T1 p=0,47; T2 p=0,7; T3 p=0,07 *3T1 p=0,58; T2 p=0,02 T3 p=0,001	Lemongrass action is comparable to CHX in reducing gingivitis and as an antiplaque agent.
Mahmoud <i>et al.</i> 2022 ³²	NI	*4T0=2,38 T1 (21 d) =1,89 T0=1,98 T1(21 d) =1,73	Mann-Whitney U test Students' T-test	*4p=0,010	Greater effectiveness of Neem extract in controlling bleeding

*1- Loe and Silness plaque index; *2 - Quigley and Hein plaque index; *3- Loe and Silness gingival index; *4 - Ainamo and Ray bleeding index; NI= does not inform; d= days

Source: selected studies

Table 4: GRADE evidence summaries (Grading of Recommendations, Assessment, Development and Evaluation).

Certainty assessment							Certainty
Outcomes	No. of participants (studies)	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations	
Plaque index (IP)	5 RCTs	very serious	No serious	No serious	No serious	none	⊕⊕○○ Low ^{a b}
Bleeding Index on Probing (ISS)	3 RCTs	serious	No serious	No serious	serious	none	⊕⊕⊕○ Moderate ^{c d}
Gingival index (GI)	4RCTs	serious	No serious	No serious	No serious	none	⊕⊕○○ Low ^{a b c}

* The risk in the intervention group (and its 95% confidence interval) is based on the assumed risk of the comparator group and the relative effect of the intervention (and its 95% CI).

The. Risk of bias related to blinding of participants and investigators, and lack of outcome for all randomized participants.

B. It does not inform the manipulation of natural products.

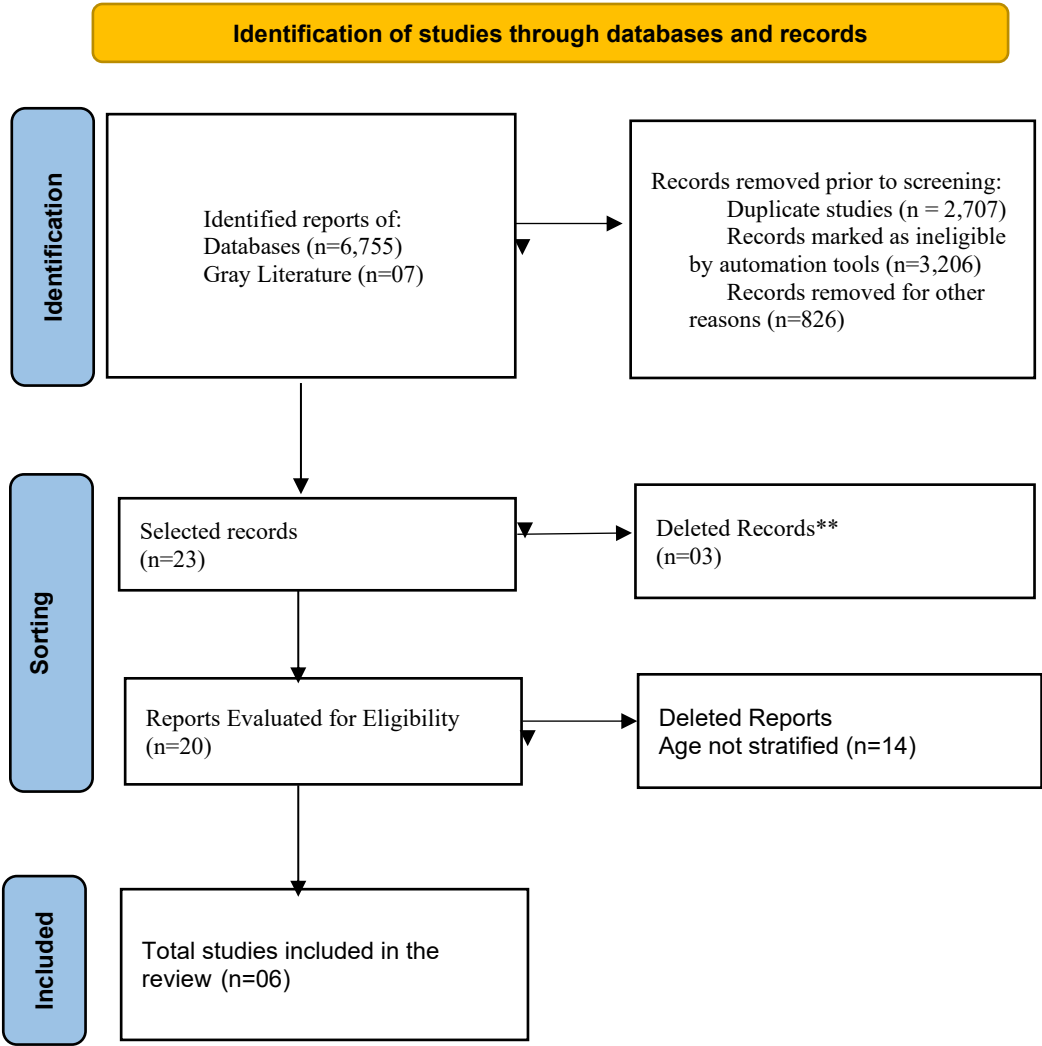
w. There is a very serious imprecision: the number of participants is less than 400.

d. lack of blinding of researchers

It is. Clinical heterogeneity: differences associated with interventions or outcomes.

RCT randomized clinical trial; NI - does not inform, Source: selected studies.

Figure 1: PRISMA flowchart of database search results



The PRISMA 2020 statement: an updated guideline for reporting systematic reviews

Figure 2: Risk of bias of included studies, according to the tool recommended by the Cochrane Collaboration's (RoB2)

	Randomization process	Deviations from intended	Missing results data	Results assessment	Incomplete result	Other biases
	D1	D2	D3	D4	D5	Overall
Akula <i>et al.</i> 2021 ²³	-	+	+	-	!	-
Alnouri <i>et al.</i> 2020 ¹⁴	+	+	+	+	+	+
Bajaj e Tandon 2011 ³⁰	+	+	-	+	!	-
Bhattacharjee <i>et al.</i> 2015 ³¹	+	+	+	+	!	!
Mahmoud <i>et al.</i> 2022 ³²	+	+	+	+	+	-
Nóbrega <i>et al.</i> 2015 ²⁴	+	+	+	+	!	!

Legend:

- Low risk of bias
- Some concerns
- High risk of bias