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EO-Based Mouthwashes – Is There Something That Should Be Known?

SUMMARY

The possibility to utilize the antibacterial and antibiofilm potential of essential oils (EOs) in dentistry is recognized and best seen in the design of EO-based mouthwashes. The up-to-date results, obtained with the most commonly used formulation being consisted of thymol, eucalyptol, menthol, and methyl salicylate (Listerine), proved its efficacy against plaque and gingival inflammation. However, novel alcohol-free products remain to be further investigated. Commercially available mouthwash containing curcumin, clove oil, mentha oil, eucalyptol, thymol and tea tree oil, and the mouthwash based on ginger EO (α-zingiberene, β-bisabolene, β-sesquiphellandrene, curcumene), reduced plaque index and improved periodontal status of the patients. The plaque index was also reduced and overall healing was improved after the surgical removal of the third molar by mouthwash containing John wort oil. In addition, Cinnamomum zeylanicum EO-based mouthwash and mouthwash containing peppermint oil were efficient against stomatitis and xerostomia, respectively. In addition to beneficial effects achieved in the mouth, i.e. at the appropriate site of application, novel investigations pointed-out that EO-based mouthwashes could be recognized as efficient in the reduction of bacterial and viral aerosols. This has been shown for Listerine, and for several mouthwashes containing following active substances of plants' EO origin: α-farnesene, β-farnesene, farnesol, nerolidol, α-bulnesene, eremanthin, β-sesquiphellandrene, spiroether, cinnamylacetate, cinnamaldehyde, eugenol, menthol, and carvacrol. This finding could be of special interest, in order to be included in further clinical studies related to COVID -19 pandemic.

Key words: COVID-19, Essential Oil, Mouthwash, Periodontal Disease, Oral Hygiene, Plaque

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REVIEW PAPER (RP)

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Introduction

Essential oils (EOs) are plant derivates extracted from the plant's stems, leaves, flowers, bark, fruits, or roots. They are complex mixtures of many components, mainly characterized with several ones (2 or 3) presented in the highest abundance. The main constituents usually belong to terpenes, terpenoids, or phenylpropanoids, all having a considerable influence on EO's overall potentials. In addition to their bioactivity observed for each of them, it is worth to note that interactions of the active constituents within the EOs could improve and multiply the overall effectivity. The antibacterial, antibiofilm, antiviral, anti-inflammatory, and antioxidant potential, are just some of their various attributes, all being well-established in the literature[1-9] .

The intention to utilize some of the aforementioned EO activities in dentistry, such as antibacterial and especially antibiofilm, makes perfect sense, considering that bacteria embedded in biofilm polysaccharide matrix

cause the majority of dental diseases (caries, gingivitis, periodontitis, and stomatitis). For that reason, it is not surprising that EOs are implemented within toothpastes, mouthwashes, dental gels, but also in some dental materials[10,11]. Their well-established antibacterial and antibiofilm properties, together with the appropriate organoleptic features (i.e. pleasant, refreshing taste), make them perfect starting material for prosperous bioactive and consumers-acceptable formulations for oral hygiene maintenance.

The idea that mouthwashes should be used as an adjunct to daily oral hygiene routine is not the new one and it is based on their ability to chemically induce bacterial death and/or disruption of polysaccharide matrix within the specific microcomunity i.e. plaque (biofilm). The best confirmation that this idea is well founded, as well as that its best realization could be achieved by application of EOs and/or their active constituents, is the fact that the most commonly known mouthwash nowadays is certainly EO-based mouthwash Listerine. Taking this into account, the first section of this review will be dedicated to present up-to-date overview of the outcomes achieved with Listerine mouthwashes. Second part of this review will bring to the light other EO based mouthwashes and summarize their literature-presented potentials. Finally, having been introduced with the EO-based mouthwashes and their efficacy, the last part of the review will try to answer to the extremely topical issue: if there is a potential beneficial role of the EO-based mouthwashes in pandemic/epidemic of COVID-19 and similar communicable diseases transmissible by aerosols.

Listerine: up-to-date

Listerine was developed in 1879. by Dr. Joseph Lawrence and pharmacist Jordan Wheat Lambert. This formulation contained a highly specific mixture of essential oil constituent's i.e thymol, eucalyptol, menthol and methyl salicylate, and was initially intended as a surgical antiseptic. The formulation was designed and named after the work of Joseph Lister, the father of the modern antisepsis[12]. Although formulation proved its activity, it was introduced for many other things rather than what was formulated for, and it took more than 5 decades to establish its position as the highly effective mouth rinse in prevention of periodontal diseases and biofilm (i.e plaque) formation[12].

The Listerine activity could be assigned to the antibacterial and antibiofilm activity of its constituents. It is common knowledge that the antibacterial activity of the EOs' constituents is a result of bacterial membrane penetration and disruption, consequently causing leakage of the microelements and finally leading to bacterial cell death[2,3,13,14]. Although mechanisms of antibiofilm effect has not been studied extensively so far, available literature data indicate that natural substances (including the EOs and their constituents), could induce antibiofilm activity by multiple mechanisms: (i) by affecting different stages of biofilm formation, such as planktonic cell adhesion, formation of microcolonies and biofilm maturation, as well as (ii) by interfering with cellular characteristic and metabolic pathways being essential for biofilm formation, such as cell communication (quorum sensing), cell flagellation and motility, cell surface properties (i.e. hydrophobicity and roughness), production of adhesins and polysaccharide extracellular matrix[15]. If it can be understood that plaque, the main cause of periodontal disease, is nothing more than a bacterial micro-community within the extracellular matrix settled on gingiva, it is easy to link why the Listerine proved its efficiency within the field.

Although it is advised for almost all patients as a support to ordinary oral hygiene routine, it is probably the most important for those that cannot achieve ideal manually cleaning including elderly patients, patients with physical or mental limitations, and/or patients with lower levels of manual dexterity. Additionally, Listerine is also recognized as valuable for patients with specific dental conditions including malposed teeth, bridge-work, as well as the one in the orthodontic treatment procedure. In the after mentioned cases, proper oral hygiene may be impaired, leading to the presence of residual biofilm causing consequently disease onset (i.e. caries, gingivitis). Accordingly, EO-based mouthwashes are considered a valuable supplement to toothbrushes and flossing[1, 16-19].

At the end of 2019., Takenaka *et al*.[1] summarized the results of many systematic reviews and meta-analyses on the subject, and stressed that previous results clearly demonstrated the efficiency of EO mouthwash (Listerine) against dental biofilm and gingival inflammation. These results showed that Listerine provides statistically significant decrease of plaque and gingival indices. Interestingly, although a comprehensive analysis was provided, Listerine remained in the research interest focus. Contrary to previous outcome, a recent study by Hunsrisakhun *et al*.[20] did not demonstrate any additional benefits of using alcohol-free EO containing 0.05% fluoride mouth rinse (Listerine) when compared to clear 0.05% fluoride mouthwash. Moreover, another recent clinical study had not demonstrated a greater influence on periodontal pocket depth (PPD) reduction in patients with periodontitis after additional four- and six-month use of EO-based mouthwash (Listerine) following scaling and root planning (SRP) compared to only SPR[21].

Interestingly, the previously mentioned study conducted by Hunsrisakhun *et al*.[20] tested alcohol-free Listerine product. This information may be of interest, because it spreads a light over still controversial question about the alcohol role within Listerine products. Alcohol, within an EO-based mouth rinse, serves as an agent that dissolves and solubilizes EO constituents, responding for the product's long-lasting. Incorporation of alcohol in mouth rinse has limited its use in certain patient populations and/or conditions (i.e children, alcoholics,

those of certain religious beliefs, and patients with oral mucositis). In order to overcome these drawbacks, in 2012. Listerine non-alcohol products were launched. This additionally resulted in growing research interest on the subject, especially whether alcohol significantly supports the antiplaque potential of EO constituents within mouthwash or not. Studies are still scarce and Lynch *et al*.[22] found no difference between mouthwashes, while contrary, Spudidaro *et al.*[23] displayed that alcohol-based mouthwashes containing EOs were more efficient compared to alcohol-free ones. It could be expected that alcohol-containing EO-based Listerine will hold its position as reliable adjunct to conventional (routine) oral hygiene in nearest future, while non-alcohol version will undergo the further time testing. Accordingly, further investigations will be needed to justify and explain whether the alcohol vehicle enhances the antibiofilm properties of EOs within mouthwash or not[1].

Other EO-based mouthwashes and their possible utilization

It was expected that success in plaque reduction of EO-based formulation Listerine will lead to novel research, sumerized in Table 1. In line with this, a significant reduction in plaque index (PI) scores and clinical parameters improvement (Table 1) in patients with periodontitis and rheumatoid arthritis were observed when the experimental mouthwash (consisting of curcumin, clove oil, mentha oil, eucalyptol, thymol, and tea tree oil) was tested. In this non-surgical periodontal therapy, SRP alone led to decrease of clinical attachment level (CAL) and periodontal pocket depth (PPD), however, the greater improvement of these parameters were achieved when SRP was employed with additional EO -based mouthwash[24].

The study conducted by Faria *et al.*[25] investigated the efficacy of *Zingiber officinale* EO mouthwash on patients with fixed orthodontic appliances. The authors presented that the *Zingiber officinale* EO mouthwash had high initial bactericidal effect and proved some reduction of PI and bleeding index after the 7 days of use. Interestingly, their result showed that placebo (flavored distilled water) also had impact on PI. Since the intergroup analysis was not provided in this study, it couldn't be certainly asserted additional benefits of EO within mouthwash.

In another study, St. John's wort oil in olive oil based mouthwash reduced efficiently PI after the surgical procedure of wisdom tooth[26]. The data of this study confirms that the use of EOs showed similar results as the positive control (chlorhexidine gluconate mouthwash) regarding postoperative complications such as swelling, trizmus, pain, infection and wound healing. Therefore, as an important part of oral hygiene maintenance, additional usage of EO- based mouthwashes after wisdom tooth surgical removal was suggested.

Table 1. EO-based mouthwashes and their clinical relevance

Mouthwash and its composition	EO-Constituents	Study group	Reduction of the	Benefit in clinical status	Reference
Commercially available mouthwash	curcumin, clove oil, mentha oil, eucalyptol, thymol and tea tree oil	patients with chronic periodontitis and rheumatoid arthritis	plaque index	regarding clinical attachment level and periodontal pocket depth	24
***Zingiber officinale* EO mouthwash** Sodium lauryl sulfate, sorbitol polysorbate 80, *Z. officinale* essential oil, saccharin, distilled water	α-zingiberene β-bisabolene β-sesquiphellandrene ar-curcumene, germacrene,	patients with fixed orthodontic appliances	plaque index , bleeding index	n.p.*	25
St. John's wort EO mouthwash *Hypericum perforatum* oil in *Olea europaea* oil	β-caryophyllene and 2-methyl-octane α-pinene [27]	after the surgical removal of third molar	plaque index	regarding swelling, mouth opening, pain, , infectious complications (i.e., alveolar osteitis), and periodontal healing	26
***Cinnamomum zeylanicum* EO mouthwash** Carboxymethylcellulose, propylene glycol, tween 80, *C. zeylanicum* EO, brown dye, purified water	eugenol, caryophyllene, benzyl benzoate and linalool	oral candidiasis within denture stomatitis	n.d.	reduction of signs and symptoms of the oral candidiasis	28
Pepermint oil mouthwash peppermint oil, propolis, xylitol, ethyl alcohol, and water	menthol, menthone, menthofuran, 1,8-cineole, and menthyl acetate[29]	patients with xerostomia	gingival index, and mean salivary rate (SR) score reduction	dentinal hypersensitivity reduction and reliving dry mouth symptoms, as well as less plaque acumulation	30

*n.d.- the information has not been determined within the study

Interestingly, some of the researchers expanded the idea of the utilization of EO-based mouthwashes irrigants or sprays within other fields[7,8,31,32]. For example Araujo *et al*.[28] investigated the mouthwash and spray containing EO extracted from *C. zeylanicum* leaves against oral candidiasis within denture stomatitis. The results proved efficiency of the formulations in terms of reduction of oral candidiasis signs and symptoms as well as greater reduction of *Candida* spp. isolates from oral mucosa and dentures. Another intraoral spray efficiency (consisting of peppermint oil, propolis, xylitol, ethyl alcohol, and water) was investigated by Balyan *et al.*[30]. The oral spray proved its efficiency in terms of gingival index (GI) reduction, mean salivary rate (SR) score, and dentinal hypersensitivity. Moreover, plaque regrowth reduction was also demonstrated, making altogether quite promising its use to relieve symptoms of xerostomia.

Although the above-mentioned studies were interested to explore if EO-based mouthwashes may be used to treat specific clinical conditions, one interesting profound systematic review investigated the possibility of the EO-based mouth rinses to reduce the number of microorganisms in the dental aerosol[33]. Based on a fact that some dental procedures result in dissemination of microorganisms in the aerosol, it was consider important to explore if pre-procedural mouthrinsing can decrease total CFU. The study confirmed that that pre-procedural mouth rinses significantly reduce the number of microorganisms in the dental aerosol. Encouraging results are in line with the idea that pre-procedural utilization of EO-based mouthwashes may be of benefit in the reduction of pathogens in aerosols, especially in the COVID-19 pandemic era.

Is there a place for EO mouthwashes in COVID-19 pandemic?

In December 2019, in Wuhan (China) the coronavirus infectious disease (COVID-19) was detected. After three months WHO declared outbreak of a pandemic and recognized it as a public health emergency of international concern[34-35]. The term COVID-19 includes respiratory conditions that can vary from the common cold to severe pneumonia (accompanied with septic shock, and multi-organ failure), while pathogenic causative agent is denoted as severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). SARS-CoV-2 is primarily transmitted directly through respiratory droplets. However, indirect transmission is also possible trough hand-mediated transfer of the virus from contaminated objects or surfaces[35]. Dental offices are places with high increased risk of virus transmission either directly or indirectly. Firstly, dental procedures including activation of slow and high-speed handpieces, ultrasonic scalers, and air-water syringes produce aerosols which mostly affect doctors and assistants working in close proximity to patients[35]. Moreover, the contaminated dental aerosol may settle on surfaces in the dental office and may result in the contamination of other patients via indirect contact [35].

Therefore, the pre-procedural mouth rinses usage has been proposed, aiming to reduce viral load in saliva and oropharyngeal tissues of the patients and consequently to reduce viral load in the dental aerosol[35]. In the study conducted by Reis *et al.*[35], authors claim that EO based mouthwashes possess efficiency against many viruses similar to the inducer of the COVID-19, even against SARS-CoV-1. The explanation was in the fact that EO/EO constituents are capable to prevent the penetration of virus by rupture of viral envelope and consequent dispute of the membranes fusion. Furthermore, they also suggested that EO/ EO constituents can affect viral replication. Review conducted by Mendosa *et al*.[36] presented that the EO constituents (eucalyptol, menthol, methyl salicylate, and thymol) combined with ethanol (Listerine product) achieved some effect (being qualified as moderate to high) against SARS-CoV-2 in *in vitro* study. Carrouel *et al*.[37] also listed the concrete EO constituents as powerful against SARS-CoV-2 such as α-farnesene, β-farnesene, farnesol, nerolidol, α-bulnesene, eremanthin, β-sesquiphellandrene, spiroether, cinnamylacetate, eremanthin, cinnamaldehyde, eugenol, menthol, and carvacrol. Concerning limited number of the clinical studies with EO- based mouthwashes and COVID-19 virus[38], and being in accordance with the study of Reis *et al*.[35], the pre-procedural mouth rinse containing EO may potentially reduce SARS-CoV-2 load in saliva. Nevertheless, the future clinical trials are required.

Conclusions

Clinical studies showed that EO- based mouthwashes have a recognized role as a reliable adjunct in preventing and controlling periodontal disease. Concerning their application within different fields in dentistry as well as in the COVID pandemic, studies are still in infancy, however, the first results are encouraging, showing their beneficial activity in the healing after the surgical removal of wisdom teeth, but also in xerostomia and stomatitis. Moreover, EO-based mouthwashes demonstrated their significant role in bacterial/viral load reduction within aerosols in dental offices. Taking all into account, EO-mouthwashes could be the candidates for future studies which will explore their potential more thoroughly, and expand their utilization fields.

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