The clinical evaluation of Vi-one chlorhexidine mouthwash on plaque-induced gingivitis: A double-blind randomized clinical trial

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Abstract

Background: Chlorhexidine (CHX) is the most effective antiseptic mouthwash to date. Essential oil such as thymol, have inhibitory and biocidal effects on a range of bacteria.

Objective: To determine the effect of mouthwash containing CHX and thymol on plaque induced gingivitis.

Methods: This double-blind randomized clinical trial study was performed on 60 patients with plaque induced gingivitis who were randomly divided into two groups: Group I (CHX/thymol mouthwash-Vi-one) and Group II (CHX mouthwash-Behsa). Patients in each group underwent scaling and root planning and polishing, then were educated about BASS-Method brushing, and were recommended Oral-B toothbrushes and Pooneh toothpaste. The two groups were asked to rinse their mouths for 60 seconds twice a day, once in the morning and once at night, after brushing their teeth. Plaque index, gingival index, bleeding index and stain index were evaluated at baseline and 14 days later in Ramfjord teeth. Data analysis was conducted using SPSS version 21. Independent-samples t-test and paired-samples t-test were used for data analysis.

Results: The results showed that plaque index and gingival index significantly reduced in two groups (p<0.001). However, group I was significantly more efficient than group II (p<0.001, p=0.021 respectively). Similar results were observed in terms of bleeding index with the difference that the two groups did not differ significantly from each other (p=0.879). Both groups significantly increased the stain index. No remarkable difference was also observed between the two groups (p=0.754).

Conclusion: Based on the results of this study, we can conclude that the CHX/thymol mouthwash can be offered to patients with dental plaque-induced gingivitis, because it appears to be more effective in controlling dental plaque and gingivitis.

Trial registration: The trial was registered at the Iranian Registry of Clinical Trials (http://www.irct.ir) with the Irc ID: IRCT201602231760N45.

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Keywords: Mouthwashes, Chlorhexidine, Gingivitis, Periodontal index, Thymol

1. Introduction
Dental plaque is a sticky and soft biofilm that is formed on the tooth surface (1,2). It begins to form on tooth surfaces within only a few minutes after brushing the teeth. If the dental plaque is not eliminated, it can result in dental caries and periodontal diseases such as gingivitis and periodontitis (1). Periodontal diseases affect the tooth-supporting tissues; gingivitis is the mildest form of periodontal disease, which generally develops due to inadequate

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oral hygiene. The most evident signs of gingivitis are gingival inflammation and bleeding (3). The principal risk factor for gingivitis is the bacterial plaque which forms on the gingiva and tooth surfaces (2-4). Plaque control is the key to prevention and the first step in the treatment of periodontal diseases (5, 6), which consists of mechanical and chemical methods. The chemical method is more common and cost-effective (5); however, since it relies on the individual’s capabilities, it is not always reliable. Therefore, chemical methods are necessary as adjunctive methods for proper control of plaque and gingivitis (4, 5). Currently, a large number of mouthwashes are available to this end, of which chlorhexidine (CHX) has been introduced as the most effective chemical agent to control plaque (2, 4-6). The high efficacy of CHX, a cationic bisbiguanide molecule, has been attributed to its high substantivity in the oral cavity and also its bactericidal and bacteriostatic activities (7). CHX does not cause systemic poisoning due to its low absorption from the digestive system (8). It is rapidly adsorbed, due to its positive charge, to surfaces with a negative charge such as intraoral mucous membranes, salivary pellicles on tooth surfaces, tongue, salivary proteins, biofilm components, including bacteria, extracellular polysaccharides and glycoproteins (1, 9, 10). After it is adsorbed to the cell membrane of bacteria, it damages the membrane, resulting in leakage of intracellular components. CHX adsorbed to various surfaces is gradually released into the oral cavity. Slow release of CHX results in prolongation of its antibacterial activity in the oral cavity for several hours, which depends on different factors such as dose, intervals of use of the mouthwash, temperature, presence of tooth or prostheses and the salivary pH (1). Use of CHX mouthwash results in some side effects such as brown discoloration of teeth, some restorative materials and the tongue (2, 4, 8), interference with taste perception and a dryness and burning sensation in the oral cavity (1). In relation to anti-plaque and anti-gingivitis properties, some studies have been carried out on mouthwashes containing essential oil (EO) (for example: thymol, menthol, eucalyptol and methyl salicylate) and the results have shown that these mouthwashes, too, are effective in the treatment of gingivitis (11-14). A mouthwash has been manufactured in Iran, called Vi-one (0.2% CHX, without alcohol) and the manufacturer claims that it has the highest efficacy in preventing formation of bacterial plaque and prevents gingival inflammation and bleeding, with minimum side effects (15). The manufacturer claims that this mouthwash exhibits antimicrobial activity since it contains active components such as thymol (0.020%), which is one of the most important ingredients of thyme, and promotes the effect of CHX on prevention of plaque formation; in addition, it has a pleasant taste and exhibits antioxidative properties (15). Furthermore, it helps tooth remineralization because it has xylitol (0.040%) in its composition, and decreases the side effects of 0.2% CHX (15). The chemical composition of mouthwashes affects their efficacy (16); therefore, the antibacterial effect of CHX can differ between different brands. The present study was undertaken to determine the efficacy and side effects of CHX mouthwash in combination with thymol since no study was found on the effects of such a mouthwash.

2. Material and Methods

2.1. Trial design and participants

This study was a randomized clinical trial that was conducted from December 22, 2015 to June 22, 2016. This double-blind randomized clinical trial study was performed on 60 patients, who referred to the Department of Periodontology, Faculty of Dentistry, Babol University of Medical Science.

2.2. Selection criteria

The inclusion criteria for participation in this study were: patients who were able to cooperate with similar plaque index (Silness and Loe), having chronic moderate-severe plaque induced gingivitis, being the age of 25 to 50 years old. The exclusion criteria were as follows: patients with history of systemic disease that could influence periodontium, patients who smoked, patients who were under periodontal treatment during the past 6 months and patients who received antibiotics during the last month.

2.3. Interventions

Eligible patients were included in the present study after the study procedures were explained to them by the operator and after they signed informed consent forms. Patients in each group underwent scaling, root planning and polishing, then were educated about BASS-Method brushing and recommended the Oral-B toothbrush and Pooneh toothpaste. Patients were randomly given a bottle of the mouthwash that each bottle contained Vi-one 0.2% CHX or Behsa 0.2% CHX. The two groups were asked to rinse their mouths for 60 seconds twice a day, once in the morning and once at night, after brushing their teeth and not to rinse their mouths for two hours after using the respective mouthwashes and abstain from eating; they were expected to continue these procedures for 2 weeks.

2.4. Outcomes

The following clinical parameters were recorded at baseline and 2 weeks later in Ramfjord teeth: Plaque index (PI, Silness and Loe), Gingival index (GI, Silness and Loe), Bleeding index (BI, Barnette), and Stain index (SI, Green and Vermillion).
2.5. Sample size
The sample size was calculated to be 60 patients. This sample size was calculated based on the results of previous study (5) by assuming the test power of 80% and a confidence level of 95%.

2.6. Randomization and blinding
Randomization was done by one of the researchers who did not have a role in the treatment of the participants. The randomization sequence was computer-generated, with the randomization itself conducted through IBM® SPSS® Statistics version 21 (IBM® Corp., Armonk, NY, USA) (random number generation). After coding identical bottles containing the mouthwashes, each patient was randomly given a bottle of the mouthwash by an operator who was blinded to the type of the mouthwash in each bottle. Each bottle contained Vi-one 0.2% CHX or Behsa 0.2% CHX.

2.7. Statistical methods
Data analysis was conducted using SPSS version 21 software. We used independent-samples t-test and paired-samples t-test to analyze the primary and secondary outcomes. The statistical difference was significant at p<0.05. Also, before performing the statistical analyses, the normality of the variables' distribution was examined using the K-S test.

2.8. Research ethics
The proposal for this thesis research was presented to the Ethics Committee of Babol University of Medical Sciences (MUBABOL.REC.1394.188). Participation in this study was voluntary and patients could leave the study in each stage. Indeed, in this study, for ethical considerations, the participants were informed about the objective and nature of the study, and each participant provided her written consent in her native language (Persian) prior to the study. Also, we were committed to keeping all of the participants’ information confidential. Mouthwash that had been given to patients was standard, was approved by the Ministry of Health and is available in pharmacies across the country.

3. Results
The mean PI and GI decreased significantly after intervention in both groups, but the mean difference of the group that rinsed Vi-one was significantly higher than the Behsa group (p<0.001, p =0.021 respectively) (Table 1). According to the table of gingival index, before the study, all patients who used Vi-one mouthwash had moderate gingivitis and after the study, all of them had mild gingivitis. Before the study, 29 patients who used Behsa mouthwash had moderate gingivitis and one patient had severe gingivitis, and after the study, 27 patients had mild and three of them had moderate gingivitis. Mean values (standard deviations) of the percentage of bleeding for the Vi-one mouthwash initially and finally were 83.75 (28.70) and 19.44 (10.40), respectively, and for the Behsa mouthwash were 89.31 (26.90) and 25.97 (14.46), respectively. Analysis of this data show that the mean BI in the Vi-one group decreased more than that in the Behsa group, but there were no significant differences between the two groups (p=0.879). Mean values (standard deviations) of the percentage of staining for the Vi-one mouthwash initially and finally were 41.81 (18.71) and 107.50 (27.23), respectively, and for the Behsa mouthwash were 45.56 (21.99) and 109.58 (27.83), respectively. Analysis of this data show that the SI increased in both groups, with greater increase in the Vi-one group, but no statistically significant differences were observed between the two groups (p=0.754).

Table 1. Mean and mean difference of PI and GI of two groups

| Variable  | PI (Mean±SD) | GI (Mean±SD) |
|-----------|--------------|--------------|
|           | Before       | After        | Difference   | Before       | After        | Difference   |
| Type of mouthwash |              |              |              |              |              |              |
| Vi-one    | 1.65 ±0.062  | 0.515±0.143  | 1.143±0.161  | 1.533±0.209  | 0.608±0.184  | 0.925±0.225  |
| Behsa     | 1.65 ±0.047  | 0.820±0.122  | 0.830±0.120  | 1.504±0.244  | 0.738±0.198  | 0.765±0.292  |

4. Discussion
The present study was carried out to clinically evaluate the efficacy of Vi-one CHX mouthwash, as an adjunct in the treatment of dental plaque-induced gingivitis. In this study, the mean difference of PI and GI in the group that rinsed Vi-one was significantly higher than the Behsa group, which is not similar to the result of previous studies. Papaioannou et al. carried out a study on new 0.2% CHX without alcohol and conventional alcohol-containing 0.2% CHX, and reported that mean values of PI increased similarly for both solutions; however, these differences between initial and final values were statistically significant only for CHX without alcohol mouthwash. Similarly, the mean values for the GI showed small increases, but with no statistically significant differences between them (17).
studies might be attributed and Parikh.

In the study by Todkar et al., CHX mouthwash without alcohol exhibited better clinical

result in decreasing dental (CHX/thymol/alcohol resulted in decreasing dental (19). Based on the results of the present study and those of the studies above, it can be

alcohol (control group), with no significant differences between the two groups. The results of a

alcohol in 4 weeks). These two

term (<4 weeks) and long term (>4 weeks). These two

mouthwash (18). Charles et al. who reported that 0.12% CHX mouthwash resulted in significantly more calculus and stains compared to EO mouthwash and the control group (11). Neely reported, based on their study on the antimicrobial activity of thymol against oral pathogens, reported that thymol was effective in inhibiting the growth of oral pathogens; therefore, it can be useful for the preservation of oral hygiene (24). Didry et al. reported that thymol can be used for the treatment of oral infections alone or in combination with eugenol or carvacrol (25). Takeuchi et al. evaluated the effect of CHX/thymol varnish and fluoride varnish on the formation of dental biofilm in vitro and reported that CHX/thymol varnish inhibited biofilm formation but fluoride varnish did not exhibit such capability (26). Yucel-Lidberg et al. reported that in patients with chronic gingival inflammation, local use of CHX/thymol varnish was useful (27). Filoche et al. reported that the amount of chlorhexidine necessary to achieve an equivalent growth inhibition against Lactobacillus plantarum and Streptococcus mutans was several times lower when it was combined with EO compared to CHX alone; therefore, they concluded that EO might have a role in the development of new anti-caries treatments (28). In the present study, in both groups gingival bleeding decreased and tooth staining increased, with no significant differences between the two groups. The results of a study by Papaioannou et al. (17) in relation to staining, were consistent with those of the present study. The results of the present study were consistent with those of studies by Jose et al. on 0.2% CHX mouthwash with and without alcohol (1), by Leyes Borrajo et al. on 0.12% CHX, 0.05% sodium fluoride, 11% ethanol and the same mouthwash without alcohol (6) in relation to B I Charles et al. who reported that 0.12% CHX mouthwash resulted in significantly more calculus and stains compared to EO mouthwash and the control group (11). Neely reported, based on a review study, that formation of calculus and stain is higher with the use of CHX mouthwash compared to the use of an EO mouthwash (19). Based on the results of the present study and those of the studies above, it can be concluded that the presence of thymol in addition to CHX in Vi-one mouthwash does not possibly have a synergistic effect on BI and SI.
5. Conclusions
In brief, results of this study showed that Vi-one mouthwash is effective in reducing the Plaque Index and Gingival Index. Based on this result, we conclude that Vi-one mouthwash can be offered to patients with dental plaque-induced gingivitis, but we think that further studies are necessary to support this conclusion with stronger evidence.

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Trial Registration:
The trial was registered at the Iranian Registry of Clinical Trials (http://www.irct.ir) with the Irct ID: IRCT201602231760N45.

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Conflict of Interest:
There is no conflict of interest to be declared.

Authors' contributions:
All authors contributed to this project and article equally. All authors read and approved the final manuscript.

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