Effect of Chlorhexidine, Ozonated Olive Oil and Olive Oil Mouthwash on Oral Health Status of Patients with Gingivitis: A Randomised Controlled Trial

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Authors’ contributions
This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

Background: Effective plaque control is important to prevent gingival and periodontal diseases. In recent years, Olive oil and Ozonated olive oil are gaining paramount importance in dentistry because of its minimal side effects as compared to chemical agents.
Aim: To assess the effect of olive oil, ozonated olive oil, and chlorhexidine mouthwash on oral health status of patients with gingivitis.
Materials and Methods: The present double blinded, parallel designed, randomized clinical trial was carried out among 66 gingivitis patients in the Department of Periodontics, Saveetha Dental College and Hospitals, Chennai, India. Participants were assigned to three groups: 22 participants in each group [Group A (CHX mouthwash), Group B (OOO - Ozonated olive oil) and Group C (OO - olive oil)]. Complete ultrasonic scaling was done and subjects were asked not to use any oral hygiene aids and recalled after three days and the gingival index and OHI(S) were noted...
The development of a preventive regimen that targets the microbial risk factor is the most comprehensive and successful approach toward gingivitis. The gingivitis is curable and easy to revert back to healthy gingiva, but once it worsens to periodontitis, the grade depends on their recovery. Prevention is better than cure, hence the maintenance of oral health is the best way to prevent all these. Oral health is multifaceted and includes the ability to speak, smile, smell, taste, touch, chew, swallow and convey a range of emotions through facial expressions with confidence and without pain, discomfort and disease of the craniofacial complex. Oral hygiene can be defined as the maintenance of teeth and gums in a healthy condition. The most reliable methods of oral hygiene maintenance are mechanical methods of tooth cleaning using toothbrushes in adjuvant with chemotherapeutic agents [17]. Even though brushing removes the debris out of the oral cavity, there is a high chance of debris over interdental areas [18].

One of the traditional Indian remedies is oil pulling which was familiarized by Dr. F. Karach [26]. Oil pulling is an ancient ayurvedic therapy where a tablespoon of oil is used to gargle or rinse all over the mouth every morning [27]. This process should be done for 5 mins, but the ideal usage is 2 to 3 mins. The swishing of oil activates enzymes and draws toxins out of the blood. Their antioxidant effects damage the cell wall of microorganisms and destroy them [28]. The emulsification process which occurs due to agitation of oil in the mouth leads to the formation of a soapy layer, which can alter the adhesion of the bacteria on the tooth surface, remove superficial worn out squamous cells, and improve oral hygiene. Since it inhibits bacterial adhesion, it also prevents plaque coaggregation over the mouth [29].

At present, there are a number of indigenous natural medicinal products which deserve due

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**Keywords:** Chlorhexidine; oil pulling; olive oil; ozonation; innovative; mouthwash.

**1. INTRODUCTION**

The most common diseases of the mouth are gingivitis and periodontitis. Gingivitis can be defined as gum disease with inflammation of the gums, where the gums will appear red and puffy, and will usually bleed during tooth-brushing or dental examination [1]. Gingivitis is a mild condition of inflammation which is usually asymptomatic, hence they are unnoticed. The primary etiology of gingivitis is plaque, but there are several aggravating factors including habits like smoking, stress, genetic factors, systemic diseases and hormonal distress [2-10]. Untreated gingivitis will cause periodontitis, which manifests as increased pocket depth, recession, furcation involvement, mobility, bone loss [11-16].
recognition for their contribution to improving oral health. Olive oil (OO) has the following advantages over the standard and commercially available mouthwashes [30]. It causes no staining and no allergic reactions. It is available at the same cost of chlorhexidine mouthwash [31]. One of the real considerations is, olive oil is a substance we use over our kitchen daily, so there is no need of getting a separate one. Considering these benefits, oil pulling therapy with OO could be promoted as a measure for the prevention of oral disease [32].

Currently, ozone therapy is gaining popularity as a modern noninvasive method of treatment. It is a powerful oxidizing agent with a high antimicrobial power against oral pathogens [33]. Ozonation is a process of addition of superoxide o(-) ion to the normal oxygen molecule. This can be done only in the superoxide state and they are highly unstable. Their stability is purely because of the resonance of the molecule [34]. The olive is superheated and oxygen gas is superheated to achieve a renaissance phase and combined together to form an ozonated olive oil (OOO) [35]. Ozone (O3), when in contact with organic fluids, causes the formation of reactive oxygen molecules (O2) which influence the cellular metabolism, tissue repair, and antimicrobial effect [36]. In addition, ozone therapy can be systemically or locally applied, cost-effective and has restricted intolerance or contraindication with minimal side effects.

Our team has extensive knowledge and research experience that has translated into high quality publications [37-56]. Through extensive literature search, it was revealed that there is a lack of adequate studies testing the efficacy of freshly prepared olive oil and ozonated olive oil. Hence, the rationale of this study was to evaluate the effectiveness of OO and OOO over chlorhexidine mouthwash on oral health status of patients with gingivitis.

2. MATERIALS AND METHODS

2.1 Study Population

The present double-blinded, parallel designed randomized clinical trial was carried out in the Department of Periodontics, Saveetha Dental College and Hospitals, Chennai, India. A total of 66 patients with gingivitis within the age group of 35-45 years were enrolled. The ethical clearance was obtained from the Institutional Ethical Committee (IHEC/SDC/UG- 1880/20/320) and a written informed consent was obtained from all the study participants.

2.2 Inclusion criteria

Participants within the age group of 35-45 years who were systemically healthy, presence of at least 20 teeth, probing depth of 1-3 mm, presence of bleeding on probing (BOP) in at least 30% of the sites were included in the study.

2.3 Exclusion criteria

Participants allergic to herbal extracts, smokers, pregnant or lactating mothers, participants under long term medications, systemically compromised patients were excluded from the study.

2.4 Test Solutions

Group 1: Chlorhexidine Mouthwash (Clohex Plus® mouthwash, Dr Reddy's Lab Ltd., Hyderabad, India).

Group 2: OOO (PurO3 olive oil, PurO3 Ltd, Arkansas, USA)

Group 3: OO (Cardia life extra virgin olive oil, kaleesuwari refinery Pvt Ltd., chennai, India).

2.5 Study Design

A pilot study was conducted using similar oils and mouthwashes to check the feasibility of the study. The prevalence of gingivitis was 80% in the pilot study. Considering the dropouts, the sample size was inflated by 20%, hence the sample size was 66 with 22 participants in each group [Group A (CHX mouthwash), Group B (OOO - Ozonated olive oil) and Group C (OO - olive oil)]. Participants were assigned to the groups by a person not involved in the study. All the subjects were provided with their assigned mouthrinses and were divided into Group 1, Group 2 and Group 3 randomly using a simple lottery method with 22 participants in each group. All the mouthrinses were dispensed in identical bottles thereby ensuring subject masking. The examiner and the participants were also blinded with regard to the mouthrinse allocated to them thereby ensuring a double-blinded study. Subjects were instructed to use 10 ml of mouthwash for 1 min twice daily after tooth brushing for a period of 1 month. Complete
ultrasonic scaling was done for all the participants and the subjects were provided with a standard tooth brush, standard tooth paste and they were advised to brush their teeth by following modified bass technique and recalled after three days and gingival index and OHIS was recorded (baseline). Then subjects were provided with respective mouthwashes for a period of 15 days. The gingival index was noted again after 1 month.

2.6 Clinical Parameters

• Oral Health Index(Simplified) (OHI-S)
• Loe and Silness gingival index (GI)

All the parameters were recorded at baseline (T1) and after 15 days (T2).

2.7 Statistical Analysis

The data was analyzed using Statistical Package for Social Sciences (SPSS Software, Version 23.0). Descriptive and inferential statistics were done for data summarization and presentation. One-way ANOVA was used to compare the mean values of OHI(S) and gingival index between the groups. Tukey's HSD post hoc test was done to find means that are significantly different from each other. Also, student's paired t-test was used to compare the mean values of OHI(S) and gingival index within the groups.

2.8 Preparation of Ozonated Olive Oil

Ozonated OO was prepared by passing ozone gas through commercially available OO (PurO3) using an ozone generator (Ozone Engineers). The output was titrated to 2 g/h for about 2 min to adjust the concentration of ozone to 0.01 ppm. Since half-life of ozone is only 20 min and it was freshly prepared every day just before the usage.

3. RESULTS

A total of 66 study participants were enrolled in this study and were divided into three groups each of 22 participants. Group A - CHX, Group B - OOO and Group C - OO.

One-way ANOVA showed there was no statistically significant difference between the baseline GI and OHI(S) values as compared to the three mouthwash groups (p=0.865), but there was a statistically significant difference (p=0.000) observed between the three mouthwash when compared after 15 days. The baseline TI values between the three groups were statistically not significant (p=0.865), whereas after 15 days, there was a statistically significant difference (p=0.000). (Table 1)

Tukey's HSD post hoc test was done to find means that are significantly different from each other. A statistically significant difference between Group 1 and Group 3 was observed in terms of post GI (p=0.000) and post OHI(S) (p=0.000) but statistically no significant difference was observed between Group 1 and Group 2 in terms of post GI (p=0.171) and post OHI(S) (p=0.393). (Table 2)

Student's paired t-test was done to compare the mean values of gingival index and OHI(S) within the groups. The mean difference between the baseline and post gingival index and baseline and the post OHI(S) was statistically significant in both Group 1 and Group 2 with the p value of 0.000. Whereas, no statistically significant difference was observed between the baseline and post GI (0.24) and baseline and post OHI(S) (0.27) in Group 3 (Table 3).

| Table 1. One-way ANOVA comparison of mean gingival index and OHI(S) between the study groups |
|-----------------------------------------------|-----------------|-------------------|-----------------|-----------------|-----------------|
| Variable                      | Group 1 (Chlorhexidine mouthwash) | Group 2 (OOO) | Group 3 (OO) | p value        |
| Pre GI                        | 2.681±1.037  | 3.409±1.543  | 3.5±1.144  | 0.865          |
| Post GI                       | 1.409±1.225  | 1.476±1.081  | 2.343±1.1154 | 0.000          |
| Pre OHI(S)                    | 4.54±1.33     | 4.8±1.012    | 4.36±1.000 | 0.47           |
| Post OHI(S)                   | 0.76±0.36     | 1.05±1.65    | 2.30±1.479 | 0.000          |
Table 2. Tukey’s HSD post Hoc pairwise comparison of mean gingival index and OHI(S)

<table>
<thead>
<tr>
<th>Groups</th>
<th>Post GI Mean difference</th>
<th>p value</th>
<th>Post OHI(S) Mean difference</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group 1 (CHX mouthwash)</td>
<td>-0.067</td>
<td>0.171</td>
<td>-0.29</td>
<td>0.338</td>
</tr>
<tr>
<td>Group 2 (OOO)</td>
<td>-0.934</td>
<td>0.000</td>
<td>-1.54</td>
<td>0.000</td>
</tr>
<tr>
<td>Group 3 (OO)</td>
<td>0.067</td>
<td>0.171</td>
<td>0.29</td>
<td>0.338</td>
</tr>
<tr>
<td>Group 2 (OOO)</td>
<td>-0.867</td>
<td>0.000</td>
<td>-1.25</td>
<td>0.000</td>
</tr>
</tbody>
</table>

Table 3. Mean comparison of gingival and OHI(S) within the study groups using student’s paired t test

<table>
<thead>
<tr>
<th>Groups</th>
<th>Mean GI p value</th>
<th>Mean OHI(S) p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group 1 (CHX mouthwash)</td>
<td>2.681±1.037 0.000</td>
<td>4.54±1.33 0.000</td>
</tr>
<tr>
<td>Group 2 (OOO)</td>
<td>3.409±1.543 0.000</td>
<td>4.8±1.012 0.000</td>
</tr>
<tr>
<td>Group 3 (OO)</td>
<td>3.5±1.144 0.24</td>
<td>4.36±1.000 0.27</td>
</tr>
</tbody>
</table>

4. DISCUSSION

The present study assesses the effect of olive oil, ozonated olive oil and chlorhexidine mouthwash on oral health status of patients with gingivitis.

The present study showed that the mean OHI(S) at baseline was statistically not significant between the three groups (p=0.47). However, the mean OHI(S) after 15 days was highly significant between CHX mouthwash and OO (p=0.000) and OOO and OO (p=0.000); showing that both chlorhexidine mouthwashes and OOO were equally effective in preventing plaque formation (p=0.338). Kamnath et al studied the clinical efficacy of aloe vera and tea tree oil and observed that both herbal and chlorhexidine mouthwashes showed significant reduction in OHI(S) scores after 1 month of usage [57]. Botelho et al studied the clinical efficacy of essential oil mouth rinse and observed that both essential oil mouth rinse and chlorhexidine mouthwashes showed significant reduction in OHI(S) after 1 month of usage [58].

Also, the present study revealed that the mean GI after 15 days was highly significant between CHX mouthwash and OO (p=0.000) and OOO and OO (p=0.000); showing that both chlorhexidine mouthwashes and OOO were equally effective in reducing inflammation (p=0.171). This might be due to the substantivity of CHX and OOO, which adhere to the tissues such as oral mucosa and teeth [59]. This helps to maintain a potent sustained release, which, in turn, reduces the bacterial count and prevents the accumulation of dental plaque and so the gingivitis [60]. Haas AN et al studied the clinical efficacy of essential oils containing mouthwashes and observed that they showed significant reduction in gingival index scores after 1 month of usage [61]. Richards et al studied the clinical efficacy of essential oil mouthwash and observed that both herbal and chlorhexidine mouthwashes showed significant reduction in gingival index scores after 1 month of usage [62]. Nardi GM et al studied the clinical efficacy of ozonated olive oil chlorhexidine mouthwashes, they showed significant reduction in gingival index scores after 15 days of usage [33]. Similar results were obtained in the studies of Claffey N et al [63] and Eid Alroudhan I et al [20]. Our findings are in accordance with the previous studies.

From the study results, it can be stated that the OOO had a promising antiplaque and antigingivitis property as similar as the chlorhexidine mouthwash. However, further long term follow-up studies are needed to substantiate the present finding and hence can be used as an adjunct to scaling and root planning in the management of gingival diseases.
5. CONCLUSION

The present study suggests that efficacy of OOO was equally effective in reducing plaque and gingivitis as compared to chlorhexidine mouthwash. Therefore, oil pulling using OOO can be a better alternative to chlorhexidine mouthwash.

DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

CONSENT AND ETHICAL APPROVAL

The ethical clearance was obtained from the Institutional Ethical Committee (IHEC/SDC/UG-1860/20/320 and a written informed consent was obtained from all the study participants.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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