Antimicrobial Effect of *Polylophium involucratum* Essential Oil on Oral Bacteria

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**ABSTRACT**

**Background & Aim:** Due to the prevalence of oral bacteria, their complications and increased microbial resistance and also recognition of the antibacterial properties of *Polylophium involucratum*, in this study, the antimicrobial properties of this plant essential oil were tested in different dilutions on oral bacteria at the Faculty of Microbiology of the Islamic Azad University of Lahijan in 2015.

**Experimental:** In this study, the antibacterial properties of *Polylophium involucratum* essential oil at different concentrations of 1: 6, 1: 12, 1: 24, 1: 48 and 1: 96, were tested on oral bacteria including *Streptococcus mutans* and *Streptococcus sanguinis* with disk fusion method.

**Results:** According to results, the *Polylophium involucratum* essential oil had a greater antibacterial effect on the *Streptococcus sanguinis* than the *Streptococcus mutans*. The highest inhibition zone values were observed against the *Streptococcus sanguinis*, ranging from 18mm to 27mm. Also, the antibacterial properties of *polylophium involucratum* essential oil were higher than other chemical drugs.

**Recommended applications/ industries:** The results of this study indicated the antibacterial effect of the *Polylophium involucratum* essential oil on some pathogenic bacteria, especially *Streptococcus mutans* and *Streptococcus Sanguis*, which can be used as a herbal antibiotic with fewer complications than usual antibiotics for treatment of disease.

1. Introduction

*Polylophium involucratum* is a plant from the Umbelliferae strain that located in the Apiaceae order. The seed of this plant has a sharp smell, and given that it has not been studied for the compounds in the essential oil of this plant, Plant seeds contain 3.8% V/W essential oil. Four substances, called limonene,
various chemical structures (Zaman, 2012). Plants are a vital source of medicinal and therapeutic substances. These compounds detected from secondary metabolites of plants show various activities against various types of diseases such as oral and dental diseases, cancer, liver disorders, diabetes, arteriosclerosis and inflammatory diseases. Polylophium involucratum (Pall.) Boiss is a family of Apiaceae, native to the highlands of northwestern Iran (Emad, 1999). Oral and dental diseases associated with dental caries can lead to major health problems (Mobble et al., 2010). Oral and dental illnesses are associated with poor quality of life and associated with chronic diseases and systemic diseases (Mobble et al., 2010). Hence, the urgent need to find an alternative to antibiotics and chemicals is important for the treatment of oral and dental illness. Since ancient times, humans used medicinal herbs to treat common diseases, and some of these traditional drugs are still included as part of permanent treatment, many community diseases (Salehnia, 1999).

There is now interest in using natural products in improving health and fitness. Oral and dental diseases associated with dental caries are major health problems. Periodontal diseases are recognized as a major public health problem worldwide. The most common cause of adult teeth is loss of teeth. This periodontal disease is associated with bacterial infection. Therefore, it appears that antibacterial therapy is involved (Weitzel et al., 2014). Therefore, antibacterial therapy seems to be a good way to improve the inflammation of the tissue (Salehnia, 1999). Staphylococcus mutans is one of the bacteria that are often found in the mouth and on human teeth. Phenolic and thymolin compounds can be the most effective ingredient on oral bacteria, and another important ingredient, carvacrol, is well soluble in alcohol and organic solvents. These materials are mainly stored in young leaves during plant growth. The tail is also used to relieve a cough. The alcoholic extract of this plant is used for sterilization (Vargas, 2012). The science of identifying and using medicinal herbs is as old as human life. Until the 19th century, the use of natural resources, mainly of plants, has been the main means of treating diseases. But the rapid progress in chemistry and the lack of natural resources led to the replacement of herbal drugs with new chemical compounds. But nowadays scientists of many different disciplines have turned to herbal medicines. Then there are some hypotheses and questions about that for example: a) Polylophium involucratum essential oil with different concentrations has different effects on oral bacteria, b) These materials can reduce oral bacteria, c) Which one of the oral bacteria is most affected against Polylophium involucratum essential oil? Oral and dental diseases associated with dental caries can cause major health problems. Oral diseases are associated with a decrease in quality of life and are associated with chronic diseases and systemic diseases.

Therefore, the urgent need to find an alternative route is very important for the treatment of oral and analgesics rather than antibiotics and chemicals. Regarding the oral intake of this plant, the importance of family plants and the lack of study on its secondary metabolites, the decision was made to investigate the antimicrobial effect of the plant in terms of efficacy on oral bacteria (Altes and Moreno, 1994). There is a little research on the effect of polylophium involucratum essential oil on oral bacteria, which is tested in this study. If the results of this study are positive, this essential oil can be used in mouthwashes formulation and even herbal remedies that are very effective in treatment of oral and dental infections.

2. Materials and Methods

2.1. Extraction of essential oil by distillation

Essential oil was extracted using the Clevenger apparatus. 100 grams of the dried plant was distilled after a little grinding, and the distillation continued until to collection of essential oil. After that, the essential oil was stored in a special container in the refrigerator until analysis.
2.2. Microbial samples

In this study, the standard microbial strains, *Streptococcus mutans* PTCC1683 and *Streptococcus sanguinis* PTCC 1440, were purchased from the Center for Scientific and Industrial Research of Iran.

2.3. Disk saturation with essential oil and sensitivity test

Paper discs were used in this study. This method is one of the most useful laboratory methods for determining the susceptibility of microorganisms to antimicrobial agents. At first, the concentrations of 1:6, 1:12, 1:24, 1:48 and 1:96 of essential oil were prepared. After preparing the desired discs in sterile environments, they were washed with the desired essence at different concentrations. Then sterilized pins dried on a muller hinton agar culture medium that was inoculated with McFarland's standard concentration of the bacterium. The antimicrobial effect was measured on the basis of the non-growth halo on a millimeter scale. Erythromycin, Azithromycin, Penicillin, Tetracycline, Streptomycin and Ciprofloxacin were used for comparison under the same conditions as in the essential oil experiment.

3. Results and Discussion

According to results shown in Table 1, the *Polypodium involucratum* essential oil had a greater antibacterial effect on the *Streptococcus sanguinis* than the *Streptococcus mutans*. The highest inhibition zone values were observed against the medically important pathogens *Streptococcus sanguinis*, ranging from 18mm to 27mm. Also, the results indicated that the antibacterial properties of *polypodium involucratum* essential oil are higher than other chemical drugs and mouthwashes (Table 2). In many studies on the mechanism of action it was reported that the cell wall and cell membrane are affected and their permeability changes and release the intracellular contents, which can interfere with membrane function, such as electron transfer, Enzyme activity or absorption of nutrients.

In addition to the widespread use of medicinal plants in traditional medicine and industrial and edible uses, or as a seasoning, flavoring, and even preservative, today special attention is paid to these herbs and their derivatives for therapeutic use and therapeutic supplements in diseases (Czerepanov, 2007).

<table>
<thead>
<tr>
<th>Antibiotics</th>
<th>Disk content (µg)</th>
<th>The diameter of the non-growth halo(mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ciprofloxacin</td>
<td>30</td>
<td>24mm≥</td>
</tr>
<tr>
<td></td>
<td></td>
<td>27mm≤</td>
</tr>
<tr>
<td>Penicillin</td>
<td>10</td>
<td>24mm≥</td>
</tr>
<tr>
<td></td>
<td></td>
<td>17mm≤</td>
</tr>
<tr>
<td>Streptomycin</td>
<td>30</td>
<td>24mm≥</td>
</tr>
<tr>
<td></td>
<td></td>
<td>15mm≤</td>
</tr>
<tr>
<td>Tetracycline</td>
<td>100</td>
<td>22mm≥</td>
</tr>
<tr>
<td></td>
<td></td>
<td>18mm≤</td>
</tr>
<tr>
<td>Azithromycin</td>
<td>30</td>
<td>18mm≥</td>
</tr>
<tr>
<td></td>
<td></td>
<td>18mm≤</td>
</tr>
<tr>
<td>Erythromycin</td>
<td>10</td>
<td>21mm≥</td>
</tr>
<tr>
<td></td>
<td></td>
<td>15mm≤</td>
</tr>
</tbody>
</table>

Table 1. The mean diameter of the growth halo of the strains examined against the *Polypodium involucratum* essential oil.

Table 2. The diameter of the non-growth halo for *Streptococcus mutans* and *Streptococcus sanguinis*. There have been some reports on the antimicrobial effects of the herb extract and essential oils. Martos et al. (2008) studied the antifungal effect of lemon, orange and grapefruit oil on a number of fungi, including *Aspergillus niger*, and the results showed that the essential oils of these citrus fruits also have antifungal properties. Motamedifar et al. (2016) et al. examined the antimicrobial effect of ethanolic extract of *Peganum Harmala* seed on *Streptococcus mutans*, and compared it with 2% chlorhexidine. In another study, it was reported that lemon oil can have an antimicrobial effect better than another citrus (Weitzel et al., 2014). Studies by Danielle et al. (1989), Aderinkon et al. (1999) and Diamond and Al Zaid (2004) showed that chewing a wood called toothbrush...
wood could be effective in oral hygiene (Weitzel et al., 2014).

In summary, the results of this study indicate the antibacterial effect of the essential oil of this plant on some pathogenic bacteria, especially Streptococcus mutans and Streptococcus Sanguis, which can be used as a herbal antibiotic with fewer complications than usual antibiotics for treatment of the disease. Also, work on other extractive materials and methods, such as methanol and ethyl acetate, may have different functions. Clinical confirmation and pharmacological standardization are necessary before it is presented as an antibacterial agent.

4. Conclusion

The results of the current study, which was carried out in laboratory conditions, showed that some dilutions of Polylophium involucratum essential oil have antibacterial properties against common bacteria in dental caries. It was also found that Streptococcus sanguinis, the most important bacteria in the process of decay, are more susceptible to this essential oil of this plant. Also, the results indicated that the antibacterial properties of polylophium involucratum are higher than other chemical drugs and mouthwashes. Considering the beneficial properties of Polylophium involucratum herb and the limitations on the use of antibiotics and antimicrobial agents, it is suggested that further investigations should be made on plant compounds as an antiseptic. Also, Polylophium involucratum can be used as an alternative to antibacterial agents in the treatment of clinical infections or oral mucous membranes due to its lower toxicity. It also has synergistic effects with several antibiotics such as gentamicin and can increase the effect of these drugs and make it possible to use as a supplement to the drugs.

5. References


Salehnia, A. 1990. Antimicrobial effects of essential oil and green extract. Summary of the articles of the fourth seminar of medicinal plants of Iran. Tehran University of Medical Sciences, Tehran, Iran.

