Antibacterial Effect of Essential Oils (Clove Oil, Castor Oil and Ginger Oil) Against Human Pathogenic Bacteria

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Abstract

Essential oils are volatile, natural, complex compounds which are produced as secondary metabolites by plants for their protection against various microorganisms as well as pests. A wide range of plants have been explored for their essential oils in the past few decades. The study was conducted to determine the antibacterial activity of essential oils against human pathogenic bacteria which were gram positive (Staphylococcus aureus and Streptococcus pyogenes) as well as gram negative (Escherichia coli, Klebsiella pneumoniae, Pseudomonas aeruginosa, and Shigella sonnei). Five ml of three different oils, i.e. clove oil, castor oil, and ginger oil, were taken in a test tube so that each oil had four different concentrations. Four concentrations of (0, 25, 50 and 75) μL of oils were mixed with 1000, 975, 950 and 925 μL of DMSO respectively to make it a volume of 1ml. It was observed that clove oil was effective against the entire gram positive as well as gram negative bacteria that were used. The inhibition zone was greatest in the case of clove oil at 75 μL against P. aeruginosa (23 mm) and the smallest zone of inhibition was shown by castor oil against K. pneumoniae (12 mm). Other oils were sensitive as well as resistant to the bacteria. Hence, it is found that different oils have shown inhibitory activity towards different pathogens to a variable extent. However, clove oil was inhibitory to all the bacteria in all concentrations.

Keywords: Antibacterial activity; essential oil; pathogenic bacteria

Introduction

Aromatherapy, which believes that essential oils and other aromatic molecules have therapeutic benefits, has sparked a surge in interest in essential oils in recent decades. They are frequently used in fragrances, cosmetics, soaps, cleaning goods, and other items, as well as in food and beverage flavoring (Alizadeh, 2013; Al-Qudah et al., 2014). As contemporary antibiotic treatments, higher plant products with evidence-based activities against fungus and harmful bacteria are playing an increasingly important role (Gundidza, 1993). Furthermore, many researchers are still interested in finding novel antibacterial natural compounds. Despite the fact that their antibacterial and antifungal effects have been demonstrated to be significantly less potent than commercially available synthetic medicines (Bidlack et al., 2000; Giamperi et al., 2002), plant oils are a source of very promising natural ingredients for the production of new antimicrobial drugs. Essential oils (EOs) are volatile liquids derived from herbs, spices, and various plants, and can include up to 50 distinct components in various ratios (Burt, 2004). They are volatile chemicals of...
terpenoid and non-terpenoid origins that are produced via separate biosynthetic pathways and have different main metabolic precursors (Bakkali et al., 2008).

Infectious illnesses caused by bacteria are still one of the major sources of morbidity and death in people and animals today. Bacteria are known to have the genetic capacity to acquire and spread resistance to therapeutic drugs (Nascimento et al., 2000). Multiple drugs resistance to currently available medicines is reducing their efficacy, resulting in severe failures in the treatment of infectious diseases (Hancock, 2005). Methicilin resistance in Staphylococci, penicillin resistance in Pneumococci, vancomycin resistance in Enterococci, and various gram-negative bacteria resistances are examples (Norrby et al., 2005). In light of microorganism resistance and the possible absence of new antimicrobial medicines, new chemicals capable of inhibiting bacteria's resistance mechanism must be developed, therefore aiding disease management, treatment, and eradication (Oluwatuyi et al., 2004). As a result, there is a strong desire to develop natural and efficient cures for such diseases through the use of essential oils. The ability of different essential oils to suppress certain infections varies depending on the kind of essential oil, the concentration utilized, and the pathogen against which the oil is employed (Friedman et al., 2004). This study aims to unravel the antibacterial effect of Clove oil [Syzygium aromaticum (L.) Merr. & Perry], Castor oil (Ricinus communis L.) and Ginger oil (Zingiber officinale Roscoe) available in the market against gram positive (Staphylococcus aureus and Streptococcus pyogenes) and gram negative (Escherichia coli, Pseudomonas aeruginosa, Klebsiella pneumoniae and Shigella sonnei). Therefore, the present study has tried to figure out the antimicrobial efficacy of essential oils available in the market of Dharan.

**Materials and Method**

**Preparation of Oils of Different Concentration**

Three essential oils (Clove oil, Castor oil, and Ginger oil) were obtained at a market in Dharan, Nepal. The branding of those essential oils, as well as the name of the business, are, however, kept out of the current study. The microbiology lab at the Central Campus of Technology in Dharan, Nepal, was used to investigate its antibacterial properties.

Five milliliters of essential oils were placed in a test tube to create sample oils in four different concentrations. To create

<table>
<thead>
<tr>
<th>Oils</th>
<th>0 µL</th>
<th>25 µL</th>
<th>50 µL</th>
<th>75 µL</th>
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</thead>
<tbody>
<tr>
<td>Clove oil</td>
<td>0 µL oil + 1000 µL DMSO</td>
<td>25 µL oil + 975 µL DMSO</td>
<td>50 µL oil + 950 µL DMSO</td>
<td>75 µL oil + 925 µL DMSO</td>
</tr>
<tr>
<td>Castor oil</td>
<td>0 µL oil + 1000 µL DMSO</td>
<td>25 µL oil + 975 µL DMSO</td>
<td>50 µL oil + 950 µL DMSO</td>
<td>75 µL oil + 925 µL DMSO</td>
</tr>
<tr>
<td>Ginger oil</td>
<td>0 µL oil + 1000 µL DMSO</td>
<td>25 µL oil + 975 µL DMSO</td>
<td>50 µL oil + 950 µL DMSO</td>
<td>75 µL oil + 925 µL DMSO</td>
</tr>
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Results
Oils have a hydrophobic nature, and hence the solvent DMSO is used to allow diffusion of oils from the wells so that the organisms present around the wells can be acted upon by the oils and inhibit their growth. The zone of inhibition (ZOI) was measured in millimeters (mm) in four different concentrations (0, 25, 50, and 75 µL). Hence, the antibacterial activity of the essential oils was determined.

Effects of Oils on Test Bacteria
Tests of different oils against the test bacteria were performed by the agar well diffusion method. It was observed that clove oil was effective against the entire gram positive as well as gram negative bacteria that were used (Fig. 1). Clove oil showed a highest inhibition zone against P. aeruginosa (23 mm), which was followed by E. coli (22 mm), S. pyogenes (18 mm) (Fig. 4) and S. aureus (17 mm). Clove oil was less effective against S. sonnie (16 mm) and K. pneumonia (16 mm) (Fig. 1). Overall, clove oil was found effective against all bacteria. However, castor oil was found effective against three bacteria and ineffective against the remaining three bacteria. P. aeruginosa was highly affected by castor oil with a ZOI of 15 mm (Fig. 4), followed by S. pyogenes (13 mm) and K. pneumonia (12 mm) in a 75 µl concentration (Fig. 2). Moreover, ginger oil was found to be effective against all the sample bacteria except P. aeruginosa. K. pneumonia was highly inhibited by ginger oil with an inhibition zone of 16 mm, which was followed by S. aureus (14 mm), S. sonnie (12.5 mm), E. coli (12 mm) and S. pyogenes (11 mm) (Fig. 4) in 75 µl concentration (Fig. 3).
Discussion

Many essential oils have antibacterial action, as evidenced by numerous studies conducted in recent years. Because essential oils are highly permeable through the cell wall and cell membrane due to their lipophilic properties, they are predicted to be widely used as antibacterial and antifungal agents (Burt, 2004). The high concentration of terpenes, benzene derivatives, oxygenated compounds, and phenolic components such as thymol and carvacol, which are known to have significant antibacterial characteristics, might be linked to the oil’s antimicrobial capabilities (Burt, 2004). The interaction of essential oil components with polysaccharides, fatty acids, and phospholipids causes membrane breakdown, cellular content leakage, proton pump interference, and cell death (Edris, 2007; Oussalah et al., 2006). The results of multiple researches are difficult to compare, owing to the various test techniques, bacterial strains, and antibiotic sample sources employed. The essential oil of herbs and spices can have a wide range of compositions depending on the geographical location, variety, age of the plant, drying technique, and oil extraction method (Jerkovic et al., 2001). After the samples were put in corresponding wells created on MHA that had been swabbed with human pathogenic test bacteria, a zone of inhibition was detected in the research.

Due to the presence of a peptidoglycan layer beyond the outer membrane, essential oils are often more active against gram-positive bacteria (Sokovic et al., 2005). Gram negative bacteria, on the other hand, showed better sensitivity in this research than gram positive bacteria. This might be owing to the fact that the study used a smaller amount of gram-positive bacteria than gram negative bacteria for testing. The results indicated that clove oil was efficient at all concentrations in suppressing all of the test bacteria. It’s possible that eugenol and phenolic compounds are the major components responsible for clove oil’s antibacterial properties (Dorman and Deans, 2000). Hydrocarbon monoterpene has the lowest antibacterial action, but oxygenated compounds, particularly phenol-type compounds like thymol and carvacol, have a greater...
potential (Dorman and Deans, 2000). According to Knobloch et al. (1986), oxygenated monoterpenes have significant antibacterial action, especially on entire cells, whereas hydrocarbon derivatives have reduced antimicrobial activity due to their poor water solubility, which restricts their diffusion through the medium. As a result, the interaction of eugenol and phenolic compounds with bacteria's cell membrane phospholipids, which affects their permeability, might be the major stumbling block to their high antibacterial activity. They also maintain the ability to denature proteins (Chaieb et al., 2007).

Castor oil was not found effective against three pathogens (Shigella sonnei, Staphylococcus aureus and Escherichia coli) and showed inhibition zone against rest of three pathogens. Zarai et al. (2012) have reported the effect of castor oil against S. aureus, P. aeruginosa, K. pneumoniae and E. coli to be 24 mm, 8.2 mm, 6.2 mm and 4.2 mm respectively, which was self-extracted in laboratory. The antibacterial effects of this essential oil may be attributed to the comparatively high content of -pinene (16.88%), which is thought to actively impede microbe development (Dorman and Deans, 2000). However, P. aeruginosa was tolerant to ginger oil in our study, compared to Zarai et al. (2012). This might be due to the different strain of P. aeruginosa, as we did not differentiate the P. aeruginosa strains. Zarai et al. (2012) applied this oil against P. aeruginosa strains. Zarai et al. (2012) also reported the inhibitory effect against various infections. Clove and Ginger oils might be a promising source of alternative antimicrobial agents in the near future, and could play a key role in the identification of novel medicines for the treatment of many human diseases. As a result, essential oils are also the main source of the strong antibacterial action.

Conclusion
Different oils have been discovered to have varying degrees of inhibitory effect against various infections. Clove and ginger oils were shown to be highly efficient against the infections tested in humans. As a result, the oils that have demonstrated inhibitory effects are extremely important in the treatment of many human diseases. As a result, essential oils might be a promising source of alternative antimicrobial agents in the near future, and could play a key role in the identification of novel medicines for the treatment of a wide spectrum of pathogenic bacteria.

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Authors’ Contributions
Sraddha Dulal, Shiv Nandan Sah, and Sujan Chaudhary designed the manuscript. Laboratory work and data collection was carried out by Sraddha Dulal. Shiv Nandan Sah supervised the whole research. Manuscript preparation was done by Sujan Chaudhary and Chiranjibi Dangi. Final draft was critically revised and approved by all the authors.

Conflict of Interest
Authors declare to have no any conflict of interest with the present study.

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