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COMPARATIVE STUDY OF ANTIMICROBIAL PROPERTIES OF OREGANO, CLOVE AND NEEM OIL USING THE DISC DIFFUSION METHOD

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ABSTRACT

The threat of antimicrobial resistance (AMR) has emerged as one of the biggest global health challenges of our time. Without prompt action, AMR is likely to become the leading cause of death in the world by 2050. The crisis is further fueled by the overuse of antibiotics, which has caused the emergence of multidrug-resistant pathogens. Essential oils from plants have been receiving attention as alternatives to antimicrobial drugs due to their antimicrobial nature. This research paper set out to compare the antimicrobial activity of oregano oil, clove oil, and neem oil against the gram- negative pathogen Escherichia coli using the disc diffusion method. Oregano oil showed the highest antimicrobial activity with a zone of inhibition (ZOI) of 22 mm, followed by clove oil at 18 mm and neem oil at 13 mm. The better performance of oregano oil is due to its active components, carvacrol and thymol, which disrupt microbial cell membranes. Clove oil which is rich in eugenol, also showed significant activity, while neem oil, containing limonoids and flavonoids, showed comparatively lower efficacy. These studies establish the possibility that essential oils can be prospective antimicrobial agents derived from natural sources that can potentially bring a solution in managing AMR. Further applications into both human therapy and industrial utilization must, therefore, be pursued.

KEYWORDS: Antimicrobial resistance, essential oils, oregano oil, clove oil, neem oil, disc diffusion method, Escherichia coli.

INTRODUCTION

When a microorganism evolves to develop tolerance against the antimicrobial medication, the observed phenomenon is referred as antimicrobial resistance.[1] In recent years, antimicrobial resistance has become a worldwide threat to human health. It is reported that antimicrobial resistance alone, was responsible for more than a million deaths worldwide in 2019.[2,3] If immediate actions are not taken, it is estimated then by 2050 Antimicrobial resistance will be the leading cause of mortality among all other diseases in the world.[4]

The rapid and careless use of antibiotics in recent years have only added fuel to the fire by accelerating the antimicrobial resistance in pathogens.[5] Multidrug-resistant and extensively drug-resistant infections are becoming an increasing threat in many developing countries and without rapid action such problem will continue to become more troubling to future generations.[6] Essential oils as potential therapy for antimicrobial resistance is gaining transaction in recent years.[7,8] Essential oils (EOs) are complex volatile chemicals that are produced in different parts of plant like flowers, buds, leaves, stems, seeds, fruits, woods etc.[9] Essential oils have long history of usage in therapeutics.[10] The antimicrobial property of essential oil is due to their major and minor low-molecular weight terpenes, terpenoids, phenylpropenes and aliphatic components present in their chemical constitution.[11] Oregano, Clove and neem essential oil are among the most studied essential oils for antibacterial and antifungal activities due to potent bioactive compounds present in them.[12,13,14] Oregano oil (from *Origanum vulgare)* have been shown to disrupt microbial cell membranes due to carvacrol and thymol present in it.[15] Clove oil (From S*yzygium aromaticum)* has eugenol which is known for its broad-spectrum antimicrobial and antioxidant properties.[16] Neem oil (from *Azadirachta indica)* has complex mixture of limonoids and flavonoids which results in antimicrobial activity.[17] In present research study the antimicrobial property of oregano, clove and neem oil were determined and compared for antibacterial activity against E coli, a common human pathogen and representative organism for antimicrobial resistance in gram negative bacteria.[18]

MATERIAL AND METHODS

Essential oils: Oregano, neem and clove essential oil.

Microbial Strain: Common human pathogen Escherichia *coli were* selected for testing.

Disc Diffusion Method: Disc diffusion method was employed.[19]

1. Mueller-Hinton agar plates were prepared for determining the antimicrobial strength.

Ingredient	Amount
Beef Extract	2.0 g
Acid Hydrolysate of Casein	17.5 g
Starch	1.5 g
Agar	17.0 g
Distilled Water	1 liter
pH (adjusted to neutral at 25 °C)	-

2. The culture plates were inoculated with microbial suspension of E. coli using striking method.
3. Sterile paper discs of whatman filter paper (6 mm diameter) were saturated with essential oil and placed on the inoculated agar surface.
4. The plates were then incubated at 37°C for 24 hours.
5. The zones of inhibition (ZOI) around each essential oil were measured in millimeters (mm) with the help of a caliper and compared for assessment of antimicrobial activity.

RESULTS

The zones of inhibition produced by each essential oil were measured to assess the antimicrobial activity and the results were recorded.

Essential Oil	*Zone of inhibition against E. coli* (mm)
Oregano Oil	22
Clove Oil	18
Neem Oil	13

Oregano oil exhibited superior antimicrobial properties compared to clove and neem oils. Clove oil also showed significant activity but was less effective than oregano oil. Neem oil had the least effect among the three oils tested.

CONCLUSION

With the rising problem of antimicrobial resistance (AMR), the current research gives a hope in finding a possible substitute of natural origin such as essential oils against the microbial pathogens. Of the three plant-based oils studied, oregano oil stood out with a strong antimicrobial activity of 22 mm ZOI, killing the common pathogenic bacterium, Escherichia coli, by the action of carvacrol and thymol. Clove oil also exhibited significant values, due to its rich content of eugenol leading to 18 mm of ZOI, whereas neem oil with unique limonoid and flavonoid combinations had moderate efficacy of 13 mm ZOI.

These findings emphasize the need to look into plant-based solutions in the fight against AMR. Essential oils, such as oregano, clove, and neem, provide a natural and sustainable approach and a safer alternative to synthetic antibiotics, which are losing their effectiveness. While this study is focused on E. coli, the findings open up a whole avenue for further research in the broader applications of essential oils in human therapy, agriculture, and industrial settings. Harnessing the power of nature can help take meaningful steps toward addressing one of the most pressing health schallenges of our time. In conclusion, the essential oils are not just fragrant plant extracts; they are also the game-changers in the fight against antimicrobial resistance. Let's continue exploring, innovating, and integrating these natural remedies into our fight for a healthier future.

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