

## Effectiveness of Essential Oils against Pathogenic Bacteria: A Laboratory Study

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**ABSTRACT:** A laboratory investigation was carried out to assess the efficiency of specific essential oils against various pathogenic microorganisms. The study included testing the efficacy of each essential oil (ginger oil, garlic oil, turmeric oil, clove oil, eucalyptus oil, rosemary oil, aloe oil, and frankincense oil) against pathogenic bacteria (*Escherichia coli* and *Staphylococcus aureus*) to reduce their growth and reproduction, as well as determining which essential oil is more effective than others. According to the findings, garlic oil has a more potent antibacterial impact against *Staphylococcus aureus* than *Escherichia coli*, and clove oil has a more substantial antimicrobial effect than garlic oil, particularly against *Staphylococcus aureus*. Rosemary oil has a great capacity to suppress both types of pathogens. Aloe is also effective in inhibiting both types of bacteria. Because of the chemicals that inhibit bacterial growth in those oils, frankincense, and eucalyptus oil have a better inhibitory potential for *Staphylococcus aureus* than *Escherichia coli*. There was no inhibitory ability for turmeric oil or ginger oil. The study indicated that essential oils can inhibit bacterial development.

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### 1. INTRODUCTION

Humans have a long history of fighting diseases and epidemics, as no nation, people, or civilization, regardless of their strength in the face of these diseases, has ever been able to overcome them. In human societies' diversity and differences, numerous plant species have been used for medicinal purposes for thousands of years. In the face of disease-causing microorganisms, humans could produce antibiotics due to scientific and technical advancements; it was only a matter of time before he discovered effective medications. In the decades after World War II, the introduction of antibiotics marked the beginning of a new age. In the industrialized world, many important causes of mortality have been eradicated (such as TB, pneumonia, and others). ( Cartwright & Armstrong, 2020; Lachiusa, 2021). Still, the antibiotics, which had practically 100% efficacy against serious infections, began to lose their potency. Infectious microorganisms have evolved to be resistant to synthetic medications over time. There is an ongoing competition between the development of novel antibiotics and the emergence of antibiotic resistance. Consequently, there is a growing interest with alternative treatments such as essential oils. (Karadağlıoğlu et al., 2019).

Scientific studies have demonstrated that plants have significant biological activities against pathogenic organisms such as fungi and bacteria due to chemical compounds, the essential oils of medicinal plants are the most effective (Najmi, Z.etal.,2023; E Patterson et al., 2019). The present study revealed several important aspects of essential oils. Essential oils are the primary compounds responsible for the distinctive odor of plants, and they are simple to separate and extract from their producing plant organs using various distillation and extraction techniques. Essential oils are extracted from plants or their components (roots, leaves, bark, etc.), with the capacity to evaporate, distinguishing them from fatty oils entirely. Under standard settings, it evaporates and leaves no evidence behind. It is also lip soluble despite the absence of fatty components (Burt 2004; Sartoratto, A et al., 2004) .

Plant extracts include the plant's active compounds, mainly metabolic byproducts that can be used as water or alcohol extracts, oils, powders, or any other delivery system (Chouhan, S.etal., 2017 ; Ghannadi, A et al., 2012). Essential oils are highly complicated molecules consisting of over 200 different chemical components. Minimal chemical and structural variations between them influence their interaction and absorption (Akthar, M et al., 2014). This study investigates the antimicrobial properties of certain

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essential oils against various harmful pathogens including *Escherichia coli* and *Staphylococcus aureus*. The study seeks to show the importance of using plant -essential oils in inhibiting pathogens. The essential oils used in this study are ginger oil, garlic oil, turmeric oil, clove oil, eucalyptus oil, rosemary oil, aloe oil, and frankincense oil.

## 2. MATERIALS & METHODS

### 2.1 Materials

- i. Essential oils: A different group of ready-made essential oils were used (ginger oil, garlic oil, turmeric oil, clove oil, eucalyptus oil, frankincense oil, rosemary oil, and aloe oil).
- ii. The culture medium: We use MH agar (Muller Hinton Agar) for microorganisms used in antibiotic sensitivity testing.
- iii. The diluent: is Acetone.
- iv. Tools and equipment: Petri dishes, syringes, test tubes, digital ruler, conical flask, burner, hood, sensitive balance, autoclave, incubator, cylinder, Swab, alcohol for sterilization, and glassware.

**Table 1: Number of used tools**

No.	Tools	Number
1	Petri dish	12
2	Swap	12
3	Syringes	9
4	Test tube	12
5	Digital ruler	1
6	Conical flask	1
7	Cylinder	1
8	glassware	9

## 3. METHODS

### 3.1 Preparing the bacterial suspension

Prepare a suspension of *E. coli* bacteria by adding 1-4 colonies to 5 milliliters of sterile saline solution using a sterile cotton swab. Ensure continuous mixing to achieve a concentration comparable to the standard turbidity constant solution. Repeat the same process with *S.aureus* bacteria. (All equipment utilized have undergone rigorous sterilization using alcohol).

### 3.2 Preparing the cultural medium

The culture medium (MH agar) was made in accordance with the manufacturer's instructions. A total of 240 milliliters of distilled water was used to prepare 12 Petri dishes together with 9.12 grams of MH agar, using the following equation.  $38g/1000ml = X/240ml$ , where  $X = 9.12g$ .The culture media was sterilized using an autoclave at a temperature of 121°C for a duration of 15 minutes.

### 3.3 Preparation of Essential oils

Commercial essential oils of rosemary (*Rosmarinus officinalis* L.), garlic oil, clove oil, turmeric oil, = ginger oil, aloe oil, frankincense oil, and eucalyptus (*Eucalyptus globulus* L.) in the form of pure native essential oils with 100% main compounds were obtained from Super markets. The choice of essential oils is due to their wide spectrum of antimicrobial activity and their presence in domestic high-quality cosmetics and disinfectants (Orchard & van Vuuren, 2017).

Stock solutions were generated by diluting essential oils using appropriate solvent acetone. The purpose of adding acetone to the oil is to dissolve the bonds of the oil and facilitate its penetration of the cell membrane. Therefore, 1 ml of clove oil and 2 ml of acetone were extracted and combined in a glassware with vigorous shaking. We repeat the previously mentioned procedures with the remaining oils, ensuring that all instruments are sterilized.

### 3.4 Determination of Antibacterial Activity

Assessing the antibacterial effectiveness of essential oils using a series of sequential steps: Take roughly 3 milliliters of the created essential oil and 3 milliliters of Acetone using a syringe. *E. coli* and *S. aureus* were cultivated using a diffusion technique on the culture media. Create five little holes, a single one placed centrally and the remaining four distributed along the edges of each plate. Place two drops of garlic oil, clove oil, turmeric oil, and ginger oil in the four holes of the dish. Then, add approximately two drops of Acetone in the central hole. Cover the dishes and transfer them to an incubator set at a temperature of 37 °C for a duration of 24 hours. Repeat the same procedure with the remaining oils (rosemary oil, aloe oil, frankincense oil, and eucalyptus oil). The data was obtained and analyzed using Microsoft Excel.

4. RESULTS AND DISCUSSION

Table (2) and (3) show the effect of many essential oils on E. coli species and S.aureus species; after 24 hours of placing the dishes in the incubator, we took them out and measured the inhibitory zone for each oil with the digital ruler, as shown in figure (1):

Table 2: Efficiency of Essential oils on bacterial species.

Oils Species	garlic oil	clove oil	turmeric oil	ginger oil	Control
E.coli 22	NO	1.9mm	NO	NO	NO
E.coli 31	NO	2.2mm	NO	NO	NO
E.coli 13	NO	6.5mm	NO	NO	NO
S.aureus 146	0.5mm	12.1mm	NO	NO	NO
S.aureus 19	2.5mm	11.7mm	NO	NO	NO
S.aureus 3	2.4mm	12.3mm	NO	NO	NO

Table 3: Efficiency of Essential oils on bacterial species.

Oils Species	Rosemary oil	aloe oil	frankincense oil	eucalyptus oil	Control
E.coli 22	3.7mm	NO	NO	NO	NO
E.coli 31	9.8mm	3.5mm	NO	NO	NO
E.coli 13	4.2mm	2.4mm	NO	NO	NO
S.aureus 146	5.7mm	5.8mm	3.9mm	3.1mm	NO
S.aureus 19	7.6mm	10.1mm	5.4mm	2.5mm	NO
S.aureus 3	4.7mm	4.2mm	2.3mm	5.8mm	NO

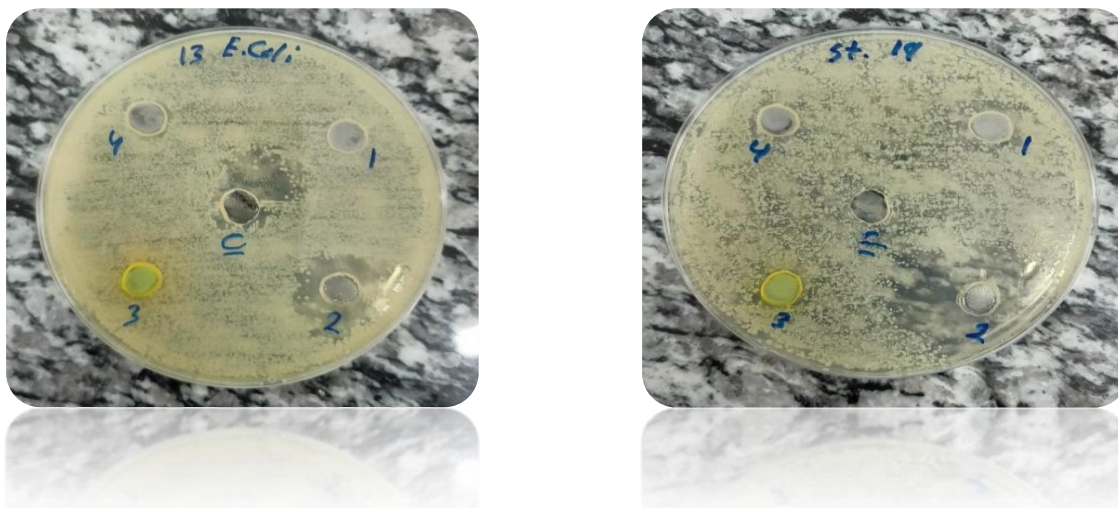


Figure 1 (a),(b): Inhibition zones of essential oils against bacterial species  
 Note: 1= = garlic oil, 2= = clove oil, 3= = turmeric oil and 4= = ginger oil.

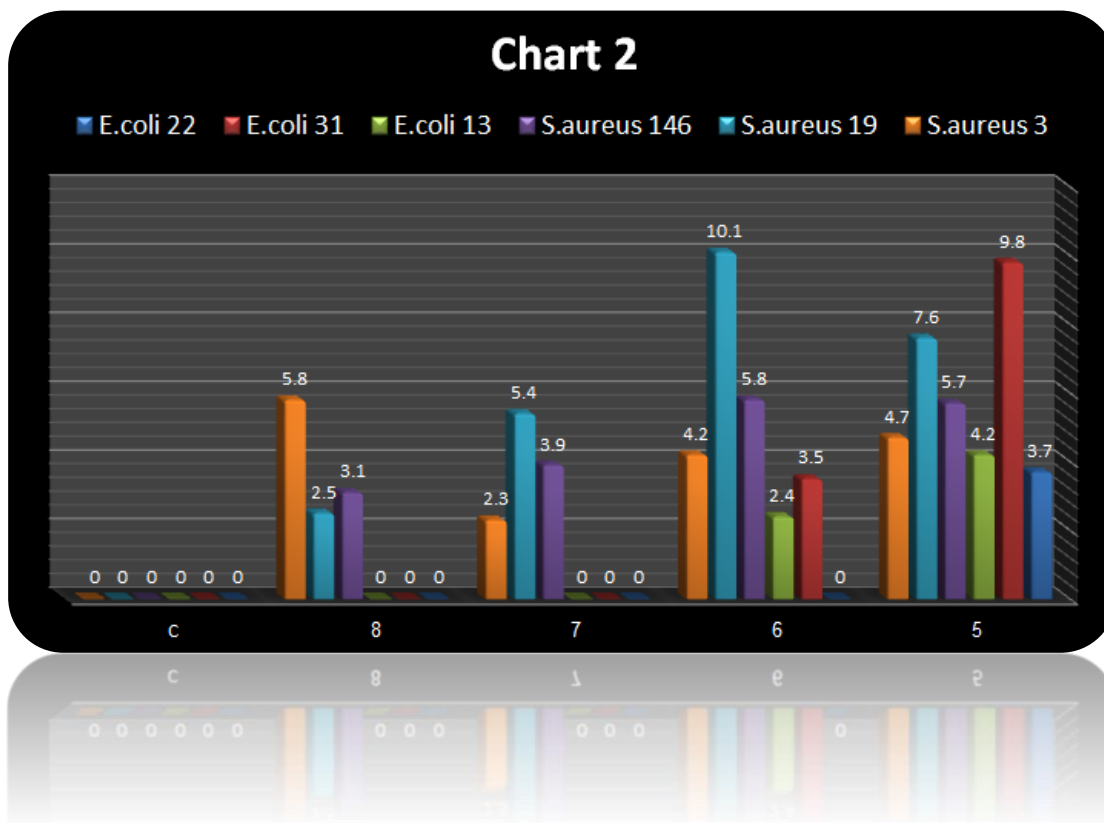


Figure 2: Efficiency of Essential oils on bacterial species.

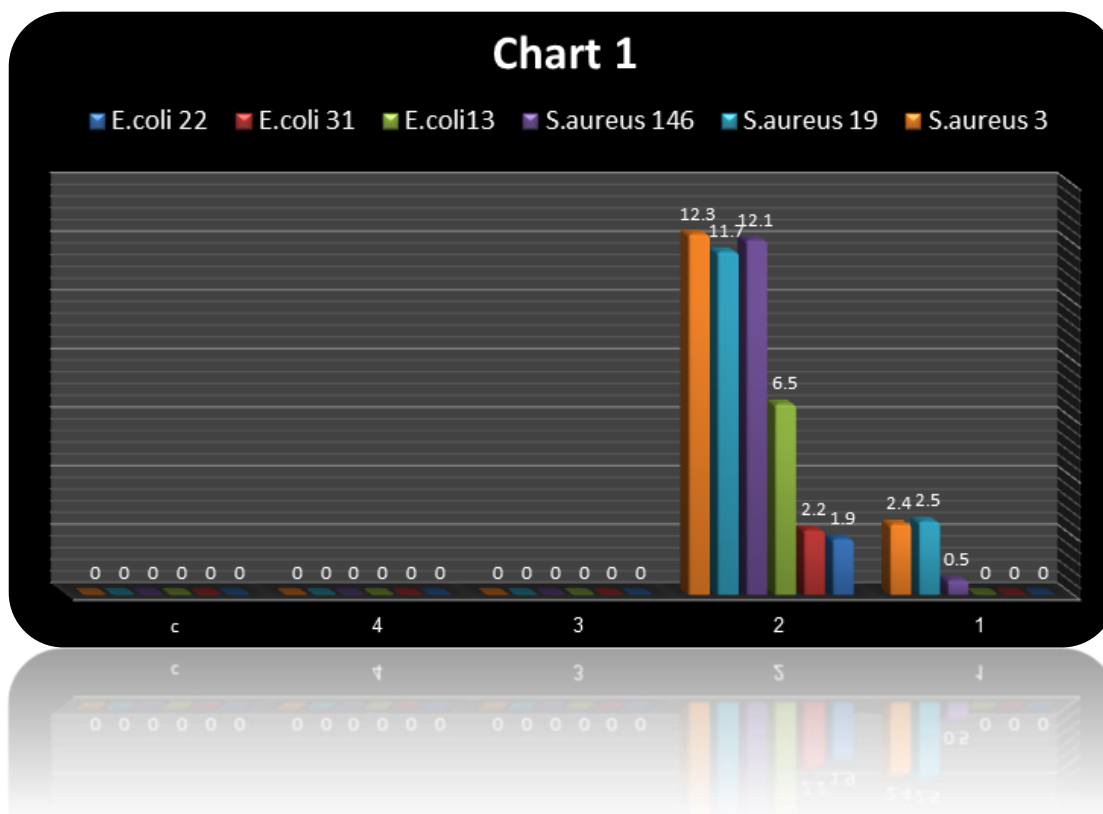


Figure 3: Efficiency of Essential oils on bacterial species.

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Figures (2) and (3) show that garlic oil was more effective against *S. aureus* bacteria than *E. coli* bacteria, and the presence of allicin in garlic oil is responsible for this antibacterial action. ( Amal Rajab, 2020). It has been observed that clove oil is more efficient than garlic oil, particularly against *S. aureus* bacteria. Clove oil's high concentration of eugenol is responsible for its antibacterial properties ( Najmi, Z. et al., 2023). The diverse chemical compositions of essential oils, which contain phenolic chemicals and terpenes, are responsible for their observed antibacterial effect. The efficacy of cloves is consistent with previous studies by Burt (2004) emphasizing their strong antibacterial qualities. Turmeric, ginger, control oils, and acetone do not have inhibitory action.

The findings also indicated that rosemary oil exhibits a greater ability to suppress bacteria compared to other oils. This is attributed to the presence of carnosic acid and rosmarinic acid, which are effective against both types of bacteria (El-Shenawy, M. et al., 2015; Najmi, Z. et al., 2023). Subsequently, aloe oil, known for its numerous advantages, follows next. Furthermore, it has a strong bacteria-inhibiting capacity because it contains antibacterial compounds such as polyphenols (Celikel, N., and Kavas, G., 2008). Frankincense oil and eucalyptus oil have greater antibacterial action, suggesting their potential as natural antibacterial agents against *S. aureus* bacteria compared to *E. coli* bacteria due to the presence of antibacterial compounds. Acetone lacks the ability to inhibit the growth of pathogens. Studies conducted by Kumar Swamy, M. et al. (2016), Sandy Duba et al. (2018), Man, A. et al. (2019), and Najmi, Z. et al. (2023) supported the findings.

### CONCLUSION

The essential oils studied demonstrated different levels of effectiveness against foodborne bacteria. These oils can be utilized to restrict the growth of pathogens and prolong the rotting of many types of food. Additional research on the relationships between the antibacterial properties of the studied oils and food additives offers valuable information on their prospective applications in food and beverage industries. Furthermore, Nano essential oils could be utilized for eliminating bacterial biofilm. Essential oils found in many therapeutic plants exhibit antibacterial properties. Hence, it is proposed that the utilization of essential oil extracts derived from medicinal plants could serve as a viable substitute for antibacterial agents, thereby making a substantial contribution to the identification of new medicinal products.

### REFERENCES

1. AKTHAR, M. S., DEGAGA, B., & AZAM, T. (2014). ANTIMICROBIAL ACTIVITY OF ESSENTIAL OILS EXTRACTED FROM MEDICINAL PLANTS AGAINST THE PATHOGENIC MICROORGANISMS: A REVIEW. J. ISSUES ISSN, 2350, 1588.
2. AMAL RAJAB. 2020. ESSENTIAL OILS AND FIGHT ANTIBIOTICS. AGRIDAY WEBSITE.
3. BURT, S. (2004). ESSENTIAL OILS: THEIR ANTIBACTERIAL PROPERTIES AND POTENTIAL APPLICATIONS IN FOODS—A REVIEW. INTERNATIONAL JOURNAL OF FOOD MICROBIOLOGY, 94(3), 223-253.
4. CARTWRIGHT, A. C., & ARMSTRONG, N. A. (2020). A HISTORY OF THE MEDICINES WE TAKE: FROM ANCIENT TIMES TO PRESENT DAY. PEN AND SWORD HISTORY.
5. CELIKEL, N., & KAVAS, G. (2008). ANTIMICROBIAL PROPERTIES OF SOME ESSENTIAL OILS AGAINST SOME PATHOGENIC MICROORGANISMS. CZECH JOURNAL OF FOOD SCIENCES, 26(3), 174. □
6. CHOUHAN, S., SHARMA, K., & GULERIA, S. (2017). ANTIMICROBIAL ACTIVITY OF SOME ESSENTIAL OILS—PRESENT STATUS AND FUTURE PERSPECTIVES. MEDICINES, 4(3), 58. □
7. E PATTERSON, J., MCELMEEL, L., & P WIEDERHOLD, N. (2019). IN VITRO ACTIVITY OF ESSENTIAL OILS AGAINST GRAM-POSITIVE AND GRAM-NEGATIVE CLINICAL ISOLATES, INCLUDING CARBAPENEM-RESISTANT ENTEROBACTERIACEAE. NCBI.NLM.NIH.GOV
8. EL-SHENAWY, M. A., BAGHDADI, H. H., & EL-HOSSEINY, L. S. (2015). ANTIBACTERIAL ACTIVITY OF PLANTS ESSENTIAL OILS AGAINST SOME EPIDEMIOLOGICALLY RELEVANT FOODBORNE PATHOGENS. THE OPEN PUBLIC HEALTH JOURNAL, 8(1). □
9. GHANNADI, A., BAGHERINEJAD, M. R., ABEDI, D., JALALI, M., ABSALAN, B., & SADEGHI, N. (2012). ANTIBACTERIAL ACTIVITY AND COMPOSITION OF ESSENTIAL OILS FROM PELARGONIUM GRAVEOLENS L'HER AND VITEX AGNUS-CASTUS L. NCBI.NLM.NIH.GOV.
10. KARADAĞLIOĞLU, Ö. İ., ULUSOY, N., BAŞER, K. H. C., HANOĞLU, A., & ŞIK, İ. (2019). ANTIBACTERIAL ACTIVITIES OF HERBAL TOOTHPASTES COMBINED WITH ESSENTIAL OILS AGAINST STREPTOCOCCUS MUTANS. PATHOGENS, 8(1), 20.
11. KUMARA SWAMY, M., SAYEED AKHTAR, M., & RANI SINNIHAH, U. (2016). ANTIMICROBIAL PROPERTIES OF PLANT ESSENTIAL OILS AGAINST HUMAN PATHOGENS AND THEIR MODE OF ACTION: AN UPDATED REVIEW. NCBI.NLM.NIH.GOV
12. LACHIUSA, D. (2021). ADVENTURES IN HISTORIC CURES: AMUSING HOME REMEDIES, ANECDOTES, AND DEFINITIONS. [HTML]
13. MAN, A., SANTACROCE, L., JACOB, R., MARE, A., & MAN, L. (2019). ANTIMICROBIAL ACTIVITY OF SIX ESSENTIAL OILS AGAINST A GROUP OF HUMAN PATHOGENS: A COMPARATIVE STUDY. NCBI.NLM.NIH.GOV

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14. NAJMI, Z., CALOGERO SCALIA, A., DE GIGLIO, E., COMETA, S., COCHIS, A., COLASANTO, A., LOCATELLI, M., DANIEL COISSON, J., IRITI, M., VALLONE, L., & RIMONDINI, L. (2023). SCREENING OF DIFFERENT ESSENTIAL OILS BASED ON THEIR PHYSICOCHEMICAL AND MICROBIOLOGICAL PROPERTIES TO PRESERVE RED FRUITS AND IMPROVE THEIR SHELF LIFE. NCBI.NLM.NIH.GOV
15. SANDY DUBA; AHMED SHAMS EL-DIN SHAABAN; NAWRAS ELEPER; YAHYA QAMARI BIOACTIVITY OF ESSENTIAL OIL FROM LAURUS NOBILIS L. LEAVES AGAINST SOME HUMAN PATHOGENIC BACTERIA.(2018). ALEPPO UNIVERSITY RESEARCH JOURNAL.
16. SARTORATTO, A., MACHADO, A. L. M., DELARMELINA, C., FIGUEIRA, G. M., DUARTE, M. C. T., & REHDER, V. L. G. (2004). COMPOSITION AND ANTIMICROBIAL ACTIVITY OF ESSENTIAL OILS FROM AROMATIC PLANTS USED IN BRAZIL. BRAZILIAN JOURNAL OF MICROBIOLOGY, 35(4), 275-280.