Original Article

Comparative Study Of Synthetic Antibiotic Amoxicillin With Volatile Oil Extracted From Some Traditional Medicinal Plants On Bacteria Human Pathogen (Streptococcus pyogenes, Staphylococcus aureus, Pseudomonas aeruginosa, Escherichia coli)

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Abstract

The extensive utilization of antibiotics in clinical practice, agriculture, and veterinary settings contributes to the emergence of antibiotic resistance in infectious microorganisms. This escalating problem poses a significant challenge in effectively treating pathogenic microbes. The study assessed the comparative antibacterial effects of Amoxicillin, a modern drug, and various essential oils against both Gram-positive (S. pyogenes and S. Aureus) and Gram-negative (P. aeruginosa and E. coli) bacteria. Amoxicillin exhibited moderate inhibition, while essential oils, including Basil and Eucalyptus, showed variable effects. Basil oil displayed limited inhibition against S. pyogenes but strong inhibition (21 mm) against S. aureus. Eucalyptus oil demonstrated limited inhibition against S. pyogenes, strong inhibition (19 mm) against S. aureus, and moderate inhibition against P. aeruginosa. Cypress oil effectively inhibited S. aureus (20 mm) but was ineffective against other bacteria. Notably, Dill oil demonstrated robust inhibitory effects, especially against S. pyogenes (16 mm), S. aureus (18 mm), and P. aeruginosa (15 mm), with moderate inhibition against E. coli (13 mm). The study underscores the potential of essential oils, particularly Dill oil, as viable alternatives or supplements to traditional antibiotics, emphasizing their diverse antibacterial properties.

Keywords: Bactria, Amoxillin, Volatile oil, Medicinal plants, Antibacterial.

Introduction

Medicinal and aromatic plants constitute a large part of natural flora and are considered an important resource in various fields such as the pharmaceutical, flavor and fragrance, perfumery, and cosmetic industries **(Swamy & Sinniah, 2015).** At present, more than 80% of the global population depends on traditional plant-based medications for treating various human health problems **(Sudipt, et al., 2012; & Arumugam, et al., 2016).**

The proliferation of drug-resistant microbial pathogens poses a significant and critical challenge to the effective treatment of infectious diseases **(Owlia et al., 2009)**. Gram-positive and gram-negative bacteria are responsible for severe and life-threatening infections, particularly in immune compromised patients **(Lestari, 2004)**. Gram-positive bacteria like S. aureus play a predominant role in post-operative wound infections, toxic shock syndrome, and food poisoning, while gram-negative bacteria such as E. coli, naturally present in the human intestine, can lead to lower urinary tract infections, cholecystitis, or septicemia **(Jose & Reddy, 2010)**. Numerous studies have showed that the escalating resistance rates of S. aureus and E. coli to antibiotics **(Mubita et al., 2008; Arredondo & Cuevas, 2008; Chambers & Deleo, 2009)**.

Essential oils derived from Medicinal and Aromatic Plants possess aromatic qualities due to a diverse blend of chemical compounds belonging to different families, including terpenes, aldehydes, alcohols, esters, phenols, ethers, and ketones **(Degenhardt et al., 2009; & Akthar et al., 2014)**. These essential oils hold significant global market potential due to their distinctive flavor, fragrance attributes, and biological activities **(Swamy et al., 2015; & Swamy & Sinniah, 2016)**.

Utilized in aromatherapy and the treatment of various conditions such as cardiovascular disease, diabetes, Alzheimer's, and cancer, essential oils offer versatile therapeutic applications **(Ali et al., 2015)**. Researchers have long acknowledged the antimicrobial effects of essential oils and their chemical constituents **(Duschatzky et al., 2005; Lang & Buchbauer, 2012).** Additionally, studies have shown the synergistic effects of combining two or more essential oil components against a variety of human pathogens **(Koroch et al., 2007; & Nazzaro et al., 2013)**.

More recently, the prevalence of antimicrobial drug resistance has prompted researchers to discover novel antimicrobial lead molecules to treat various human pathogens (**Rudramurthy et al., 2016)**. Some of the presently available synthetic drugs fail to inhibit many pathogenic microbes. In addition, the use of synthetic chemicals for the control of pathogenic microorganisms is limited because of their carcinogenic effects, acute toxicity, and environmental hazard potential. In this regard, the exploitation of essential oils to control epidemic multidrug resistant pathogenic microorganisms can be useful to combat various infectious diseases **(Mulyaningsih et al., 2010)**.

Traditional plants have been proved to be a novel source in the search of antimicrobial compounds **(Kaushik et al., 2010)**. Several types of essential oils have been studied to control a wide range of bacteria like Clove bud oil showed a strong antibacterial effect against Salmonella typhi, Staphylococcus aureus, and Pseudomonas aeruginosa **(Conner, 1993).** Likewise, carvacrol, eugenol, and thymol essential oils were inhibited E. coli **(Kim et al., 1995)**. **(Arora & Kaur, 1999)** studied the antimicrobial activity of water extract of clove and black pepper on E. coli. In another study, Ocimum basilicum essential oils were strongly antimicrobial against B. thermosphacta, E. coli, L. innocua, L. monocytogenes, P. putida, S. typhimurium, S. putrefaciens, M. flavus. **(Teixeira et al., 2013).**

Considering essential oils significant potential as natural sources of antimicrobial medications as an alternative to synthetic pharmaceuticals for the management of harmful microorganisms. This study aimed to investigate the antimicrobial effectiveness of Basil, Eucalypts, Monterey Cypress, and Dill plants essential oils against pathogenic bacteria S. pyogenes, S. aureus, P. aeruginosa, E. coli in vitro conditions. All of plants assayed in this study are commonly used as medicinal plants in different localities of Kurdistan, and other parts of the world.

Material and method

Plants collection

The fresh vegetative parts of the plant were collected in October 2023, as shown in **Table 1**, from fields in different regions of Duhok, Kurdistan, Iraq. The study was conducted at the Pharmacy Department of Duhok Polytechnic University, and the plants were identified by the authors

Essential oil extraction:

The volatile oil was obtained through hydrodistillation, following the method outlined by **(Afrudin et al., 2015)**. One hundred grams of the fresh vegetative parts were cut into small pieces, mixed with 500 ml of distilled water, and then subjected to extraction using a Clevenger apparatus for 3 hours. After boiling for 3 hours, an extract containing essential oil, water, and other plant compounds was obtained. The essential oil was separated from the extract using 15 ml of chloroform, divided into 3 stages, with 5 ml of chloroform used in each stage. Subsequently, the chloroform was evaporated at 70°C, following the procedure outlined by **(Harfouch et al., 2019)**

Antibacterial activity:

We studied the antibacterial activity of the essential oil against Gram-positive strains (S. pyogenes, S. aureus) and Gram-negative strains (P. aeruginosa and E. coli). These strains were obtained from the laboratory section of Vajeen Hospital in Duhok city and were maintained on nutrient agar at a temperature of 4°C.

Culture preparation:

The Gram-positive S.pyogenes strains were isolated from urine collected from urinary tract infections. S. aureus was isolated from swabs collected from a wide variety of infected superficial skin and mucous membrane areas. The Gram-negative P.aeruginosa strains were isolated from infection areas or wound swabs, while E. coli strains were isolated from stool-urine collected during intestinal infections (diarrhea). These strains were submitted to Vajeen Hospital and identified using standard bacteriological techniques. The cultures were maintained by subculture on Mueller-Hinton agar Petri dishes, which were then prepared using nutrient agar for the disc diffusion method.

Table 1 Medicinal plant tested for their antibacterial in the study.

Plants	Common name	Family	Part used	Origin	Traditional used
Ocimum basilicum L.	Basil	Lamiaceae	Leave, stem	Locally	Historically, basil has been used as a medicine to cure renal problems, warts, worms, constipation, headaches, coughs, and diarrhea. **(Simon et al., 1971-1980)**
Eucalypts tereticornis Sm.	Eucalypts	Myrtaceae	leaves	Locally	Traditional medicine uses the eucalyptus plant to treat diabetes, obesity, and insulin resistance. Insulin resistance is a result of chronic inflammation of adipose tissue. **(Ceballos et all., 2018)**
Cupressus macrocarpa Hortw.	Monterey cypress	Cupressaceae	leaves	Locally	Cypress leaves are used as a contraceptive and to cure laryngitis, diabetes, inflammation, toothaches, and stomach pain **(Selim et al., 2014).**
Anethum graveolens L	Dill	Apiaceae	Leave, stem	Locally	Dill plants have helpful impacts on the control and management of diabetes and cardiovascular problems **(Goodarzi et al., 2018).**

Disc diffusion Method

The antibacterial screening of the plant extracts against (Ocimum basilicum, Eucalypts tereticornis, Cupressus macrocarpa, Anethum graveolens) bacteria was carried out by determining the zone of inhibition using disc diffusion method after 24 h **(Sahoo et al., 2006).**

Statistical analysis

Agar disc diffusion assay was performed in triplicate under strict aseptic conditions to ensure consistency of all findings.

Result and discussion

Our study showed that results in Table 2 presents the comparative zone inhibition for synthetic antibiotic Amoxicillina a modern drug, and various essential oils against Gram-positive (S. pyogenes and S. Aureus) and Gram-negative (P. aeruginosa and E. coli) bacteria.

Amoxicillin displayed moderate inhibitory effects, with values of (10 mm) against S. pyogenes, (12 mm) against S. aureus, (8 mm) against P. aeruginosa, and (9 mm) against E. coli. Among the essential oils, Basil and Eucalyptus exhibited variable inhibitory effects. Basil oil demonstrated limited inhibition against S. pyogenes (8 mm) but strong inhibition (21 mm) against S. aureus and (13 mm) against P. aeruginosa, while showing no inhibition against E. coli. Eucalyptus oil showed limited inhibition against S. pyogenes (8.5 mm), strong inhibition (19 mm) against S. aureus, and moderate inhibition (10 mm) against P. aeruginosa, while also not inhibiting E. coli. Cypress oil displayed strong inhibition (20 mm) against S. aureus but was ineffective against the other bacteria. Dill oil exhibited robust inhibitory effects, particularly against S. pyogenes (16 mm), S. aureus (18 mm), and P. aeruginosa (15 mm), with moderate inhibition against E. coli (13 mm). These findings underscore the potential of essential oils, especially Dill oil, as effective alternatives or supplements to traditional antibiotics, highlighting their diverse antibacterial properties.

Many chemical compounds found in aromatic and medicinal plants have antibacterial properties. These are produced in order to shield the plants from microbialpathogens. Essential oils' antibacterial qualities are mostly determined by their chemical composition and the concentration of their primary ingredients **(Nazzaro et al., 2013)**. These chemical substances are secreted in response to particular biotic and abiotic stressors through a sequence of molecular interactions **(Holley & Patel, 2005)**. Every substance may have a unique way of working against microorganisms. Generally, a sequence of biochemical events in the bacterial cell mediate the mechanism of antibacterial action, and these reactions depend on the kind of chemical ingredients found in the essential oil **(Burt, 2004; & Nazzaro et al., 2013)**. Furthermore, differing bacterial architecture, such as that of Gram-positive and Gram-negative bacteria, which have varied cell membrane compositions, also affects the antibacterial activity of essential oils **(Hammer & Carson, 2011; & Raut & Karuppayil, 2014).**

Numerous mechanisms have been proposed to explain the antibacterial action of essential oils. The main mechanism by which essential oils destabilise cells is by breaking down membrane integrity and increasing permeability. This causes a disruption in a variety of cellular processes, such as membrane-coupled energy production, membrane transport, and other metabolic regulatory activities. Essential oils may help a number of critical functions, including energy conversion, nutrition digestion, the production of structural macromolecules, and the secretion of growth regulators, by disrupting the cell membrane **(Oussalah et al., 2006).** Essential oils have the potential to impact both the cytoplasm and the cell's outer membrane **(Kim et al., 1995; & Nazzaro et al., 2013)**. Essential oils readily permeate bacterial cell membranes because of their lipophilic nature. It has been found that the essential oils of different medicinal and aromatic plants increase the permeability of bacterial cell membranes, causing cellular components to flow out and ions to be lost (**Saad et al., 2013; & Raut & Karuppayil, 2014)**. Additionally connected to lower membrane potentials, disrupted proton pumps, and ATP depletion are the antibacterial properties of essential oils. **(Turina et al., 2006).**

Table (2). Zone inhibition of synthetic antibiotic Amoxillin and essential oils used in the study (mm).

Plant volatile oil	**Gram- positive**		**Gram- negative**	
	Streptococcus pyogenes	**Staphylococcus Aureus**	**Pseudomonas aeruginosa**	**Escherichia coli**
Amoxillin	**10**	**12**	**8**	**9**
Ocimum basilicum	**8**	**21**	**13**	**No inhibition**
Eucalypts tereticornis	**8.5**	**19**	**10**	**No inhibition**
Cupressus macrocarpa	**No inhibition**	**20**	**7**	**No inhibition**
Anethum graveolens	**16**	**18**	**15**	**13**

Values of the well's diameter following a 24-hour incubation period against a Bactria strain exposed to volatile oil extracts in an agar disc diffusion assay, as well as the observed diameter zone of inhibition (mm).

This alteration in the cell organization may cause a cascade effect, resulting in other cell organelles being affected (**Carson et al., 2002)**. Likewise, **Cox et al., (1999 and 2000)** have shown that tea tree oil reduces S. aureus and E. coli growth by changing the permeability of cells, causing more intracellular K+ ions to leak out, and interfering with cell respiration. The cytoplasmic membrane and cell wall are penetrated by the essential oils, which may disturb the arrangement of molecules of polysaccharides, phospholipid bilayers, and different fatty acids **(Longbottom et al., 2004; & Raut & Karuppayil, 2014)**. All these events may be responsible for the coagulation of inner cellular components in the cytoplasm and break down of the bonds between the lipid and protein layers **(Burt, 2004).**

Gram- positive and gram- Negative bacteria are susceptible to the potent antibiotic properties of Dill essential oil, which may be attributed to plants that contain 4% essential oil. Major compounds found in dill essential oil include limonene (33%), carvone (30-60%), and α-phellandrene (20.61%), as well as dihydrocarvone, diterpene, pinene, cineole, myrcene, paramyrcene, dillapiole, isomyristicin, myristicin, myristin, apiol, furanocoumarin, and 5-(4”-hydroxy3”methyl-2”-butenyloxy) -6,7-furocoumarin, falcarindiol, oxypeucedanin, and oxypeucedanin hydrate **(Saiman, 2004).** The specific composition of plant volatile oils, the structural arrangement of those components and their functional groups, and any potential synergistic interactions between those components are all potential factors that could influence the oils' efficacy **(Osman et al., 2009; & Dhama et al., 2014)**. This result agree with the previous study **(Mahdi, 2016)** reported the Dill essential oil significantly reduced the growth of test bacteria: E. faecalis (15-25mm), P. mirabilis (20-25mm), P. aeruginosa (17-27mm), K. pneumonia (15-35mm), E. coli (20-52mm), and the widest inhibition zone diameters was showed on S. aureus (70-72mm).

The effect antibacterial Basil essential oil on inhibition growth bacteria these result may be due to depending on the variations in chemotype, leaf, and flower color, aroma, and origin of the plant, Basil essential oils exhibit a wide and varying array of chemical compounds **(Da-Silva et al. 2003).** The chief constituents include chavicol methyl ether (or estragole), linalool, and eugenol **(Omidbaigi et al. 2003; Hussain et al. 2008).** Studies in the literature suggest that linalool, a monoterpene, is the main ingredient responsible for antibacterial activity **(Ravid et al. 1997).** However, **Matasyoh et al., 2009; Begnami et al., 2010; Lawal et al., 2014; & Beatovic et al., 2015)** studied on five species of basil plant include (O. basilicum, O. gratissimum, O. kilimandscharicum, O. lamiifolium, O. suave) reduced the growth or Inhibited microorganisms C. albicans, C. tropicalis, C. glabrata, P. notatum, R. stolonifer, M. mucedo, A. ochraceus, A. versicolor, A. niger, A. fumigates, T. viride, P. funiculosum.

The extract from the leaf of the Eucalyptus plant exhibited varying degrees of antibacterial activity against two clinical isolates. Based on the above experiment, it can be concluded that the extract has significant growth-inhibiting effects on both Gram-positive and Gram-negative bacteria. This could be explained by the presence of components such as 1,8-cineole, citronellal, citronellol, citronellyl acetate, p-cymene, eucamalol, limonene, linalool, pinene, terpinene, terpinol, alloocimene, and aromadendrene **(Nezhad et al., 2009)**.These results are almost similar to those shown by other works on the antimicrobial activity of essential oil of Eucalypts leaves as well as those of similar species **(Fit et al., 209; Ait-Ouazzou et al., 2011; & Elaissi et al., 2011).** These result agreed with previous study, **Bachir & Benali (2012)** on E. globulus plants the results obtained showed that essential oil of the leaves has antimicrobial activity against gram negative bacteria E. coli as well as gram positive bacteria S. aureus.

Cupressus macrocarpa oil displayed strong inhibition against S. Aureus but was ineffective against the other bacteria. According **Salem et al., (2018)** reported that the volatile oil from C. macrocarpa inhibited both S. aureus and B. cereus, with minimum inhibition concentrations of 0.31 and 0.12 mg ml^{-1}, respectively, and the bactericidal effect was at 0.2 and 0.41 mg ml^{-1}, respectively. The results varied, as previously mentioned, due to variations in the essential oil's chemical composition and its bioactive components. Moreover, this result may be due to the component of C. macrocarpa essential oil showed an antibacterial effect, e.g., α-phellandrene (**Zhang et al., 2017),** camphor and limonene **(Han et al., 2019)**, γ-terpinene **(Giweli et al., 2012).** Terpenoids are biologically active compounds with higher effects on the resistance to pathogens **(Sharma et al., 2019). Zhang et al., (2018)** reported that terpinen-4-ol showed antibacterial activity against S. aureus and S. agalactiae. According to **Huang et al., (2021)**, three isomeric terpineols that were isolated from Cinnamomum longepaniculatum leaf oil—terpinen-4-ol, α-terpineol, and δterpineol—have the potential to be employed as antibacterial chemicals since they cause bacterial cell death by rupturing their cell wall and membrane. Monoterpenoids disrupt the physiological and biochemical mechanisms that allow bacteria to grow and proliferate**. (Pandey et al., 2016).** However, **El-Ghorab et al., (2007)** reported that the volatile oil of Egyptian C. macrocarpa strongly inhibited the growth of S. aureus, P. aeruginosa, E. coli, A. niger and C. albicans, and concluded that, this may be attributed to the presence of neral, geraniol, eugenol dihydro, carvacrol acetate and phenol (2, 6-dimethoxy). Moreover, **Manimaran et al., (2007)** reported the potent antimicrobial activity of the volatile oil of the Indian C. macrocarpa and concluded that, such activity mainly due to the volatile oil components i.e., caryophyllene, α terpineol etc. In addition, a comparative antimicrobial study was done on the volatile oils content of Cupressus glauca, C. funebris, C. lawsonia, C. macrocarpa and C. sempervirens. The results showed that C. macrocarpa possessed the potent antibacterial and antifungal activities at concentration of 100 mcg ml^{-1} while against C. albicans was 50 mcg mLl^{-1}**(Manivannan et al., 2005)**.

CONCLUSION

In conclusion, while synthetic antibiotic Amoxicillin exhibited moderate inhibitory effects, essential oils from Basil, Eucalyptus, and Monterey cypress plants showed that variable effects with notable inhibition against bactria. Whereas, Dill oil demonstrated robust inhibitory effects, especially against S. pyogenes, S.aureus, and P. aeruginosa, with moderate inhibition against E. coli. These findings underscore the potential of essential oils, particularly Dill oil, as effective alternatives or supplements to traditional antibiotics, emphasizing their diverse antibacterial properties. Further research is necessary to validate these results and explore the underlying mechanisms of the observed antibacterial effects.

REFERENCE

Ait-Ouazzou, A.; Lorán, S.; Bakkali, M., Laglaoui, A.; Rota, C., Herrera, A. & Conchello, P. (2011). Chemical composition and antimicrobial activity of essential oils of Thymus algeriensis, Eucalyptus globulus and Rosmarinus officinalis from Morocco. Journal of the Science of Food and Agriculture, 91(14): 2643-2651.

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: 1-7.

Ali, B.; Al-Wabel, N. A.; Shams, S.; Ahamad, A.; Khan, S. A. & Anwar, F. (2015). Essential oils used in aromatherapy: A systemic review. Asian Pacific Journal of Tropical Biomedicine, 5(8): 601-611.

Arora, D. S. & Kaur, J. (1999). Antimicrobial activity of spices. International journal of antimicrobial agents, 12(3): 257-262.

Arredondo-García, J. L. & Amábile-Cuevas, C. F. (2008). High resistance prevalence towards ampicillin, co-trimoxazole and ciprofloxacin, among uropathogenic Escherichia coli isolates in Mexico City. The Journal of Infection in Developing Countries, 2(05): 350-353.

Arumugam, G.; Swamy, M. K.; & Sinniah, U. R. (2016). Plectranthus amboinicus (Lour.) Spreng: botanical, phytochemical, pharmacological and nutritional significance. Molecules, 21(4):2-26.

Bachir, R. G. & Benali, M. (2012). Antibacterial activity of the essential oils from the leaves of Eucalyptus globulus against Escherichia coli and Staphylococcus aureus. Asian Pacific journal of tropical biomedicine, 2(9): 739-742.

Beatovic, D.; Krstic-Milosevic, D.; Trifunovic, S.; Siljegovic, J.; Glamoclija, J.; Ristic, M. & Jelacic, S. (2015). Chemical composition, antioxidant and antimicrobial activities of the essential oils of twelve Ocimum basilicum L. cultivars grown in Serbia. Records of Natural Products, 9(1), 62-75.

Begnami, A. F.; Duarte, M. C. T.; Furletti, V. & Rehder, V. L. G. (2010). Antimicrobial potential of Coriandrum sativum L. against different Candida species in vitro. Food chemistry, 118(1), 74-77.

Burt, S. (2004). Essential oils: their antibacterial properties and potential applications in foods—a review. International journal of food microbiology, 94(3): 223-253.

Carson, C. F.; Mee, B. J. & Riley, T. V. (2002). Mechanism of action of Melaleuca alternifolia (tea tree) oil on Staphylococcus aureus determined by time-kill, lysis, leakage, and salt tolerance assays and electron microscopy. Antimicrobial agents and chemotherapy, 46(6): 1914-1920.

Ceballos, S.; Guillén, A.; Muñoz, D. L.; Castaño, A.; Echeverri, L. F.; Acín, S. & Balcázar, N. (2018). Immunometabolic regulation by triterpenes of Eucalyptus tereticornis in adipose tissue cell line models. Phytomedicine, 50:109-117.

Chambers, H. F. & Deleo, F. R. (2009). Waves of resistance: Staphylococcus aureus in the antibiotic era. Nat Rev Microbiol. 7(9):629-641.

Conner, D. E. (1993). Naturally occuring compounds. Antimicrobials in foods,. . In: Davidison PM, Branen AL, editors. Antimicrobials in Foods. New York, NY, USA: Marcel Dekker, 441-468.

Cox, Gustafson, Mann, Markham, Liew, Hartland. & Wyllie. (1998). Tea tree oil causes K+ leakage and inhibits respiration in Escherichia coli. Letters in applied microbiology, 26(5): 355-358.

Cox, S. D.; Mann, C. M.; Markham, J. L.; Bell, H. C.; Gustafson, J. E.; Warmington, J. R. & Wyllie, S. G. (2000). The mode of antimicrobial action of the essential oil of Melaleuca alternifolia (tea tree oil). Journal of applied microbiology, 88(1): 170-175.

Da-Silva, F.; Santos, R. H. S.; Diniz, E. R.; Barbosa, L. C. A.; Casali, V. W .D. & DeLima, R. R. (2003). Content and composition of basil essential oil at two different hours in the day and two seasons. Braz. J. Med. Plants, 6(1): 33-38.

Degenhardt, J.; Köllner, T. G. & Gershenzon, J. (2009). Monoterpene and sesquiterpene synthases and the origin of terpene skeletal diversity in plants. Phytochemistry, 70(15-16): 1621-1637.

Dhama, K.; Tiwari, R.; Chakraborty, S.; Saminathan, M.; Kumar, A.; Karthik, K. & Rahal, A. (2014). Evidence based antibacterial potentials of medicinal plants and herbs countering bacterial pathogens especially in the era of emerging drug resistance: an integrated update. International Journal of Pharmacology, 10(1): 1-43.

Duschatzky, C. B.; Possetto, M. L.; Talarico, L. B.; García, C. C.; Michis, F.; Almeida, N. V. & Damonte, E. B. (2005). Evaluation of chemical and antiviral properties of essential oils from South American plants. Antiviral Chemistry and Chemotherapy, 16(4): 247-251.

Elaissi, A.; Salah, K. H.; Mabrouk, S., Larbi, K. M.; Chemli, R. & Harzallah-Skhiri, F. (2011). Antibacterial activity and chemical composition of 20 Eucalyptus species' essential oils. Food Chemistry, 129(4): 1427-1434.

El-Ghorab, A. H.; El-Massry, K. F. & Shaaban, H. A. (2007). Effect of drying on the chemical composition of the Egyptian Cupressus macrocarpa (Hartw. ex Gordon) essential oils and their biological characteristics. Journal of essential oil bearing plants, 10(5): 399-411.

Fit, N. I.; Rapuntean, G.; Rapuntean, S.; Chirila, F. & Nadas, G. C. (2009). Antibacterial effect of essential vegetal extracts on Staphylococcus aureus compared to antibiotics. Notulae Botanicae Horti Agrobotanici Cluj-Napoca, 37(2): 117-123.

Giweli, A.; Džamić, A. M.; Soković, M.; Ristić, M. S. & Marin, P. D. (2012). Antimicrobial and antioxidant activities of essential oils of Satureja thymbra growing wild in Libya. Molecules, 17(5), 4836-4850.

Goodarzi, M. T.; Khodadadi, I.; Tavilani, H. & Abbasi Oshaghi, E. (2016). The role of Anethum graveolens L. (Dill) in the management of diabetes. Journal of tropical medicine. 1-11.

Hammer, K. A. & Carson, C. F. (2011). Antibacterial and antifungal activities of essential oils. Lipids and essential oils as antimicrobial agents, H. Thormar, Ed., pp. 255–306, John Wiley & Sons, London, UK.

Han, Y.; Sun, Z. & Chen, W. (2019). Antimicrobial susceptibility and antibacterial mechanism of limonene against Listeria monocytogenes. Molecules, 25(1); 2-15.

Harfouch, R. M.; Darwish, M.; Al-Asadi, W.; Mohammad, A. F.; Gharib, N. M. & Haroun, M. (2019). Antibacterial activity of essential oils of Rosmarinus officinalis, Salvia officinalis and Anthemis nobilis widespread in the Syrian coast. Research Journal of Pharmacy and Technology, 12(7): 3410-3412.

Holley, R. A. & Patel, D. (2005). Improvement in shelf-life and safety of perishable foods by plant essential oils and smoke antimicrobials. Food microbiology, 22(4): 273-292.

Huang, J.; Yang, L.; Zou, Y.; Luo, S.; Wang, X.; Liang, Y. & Wei, Q. (2021). Antibacterial activity and mechanism of three isomeric terpineols of Cinnamomum longepaniculatum leaf oil. Folia Microbiologica, 66: 59-67.

Jose, B. & Reddy, L. J. (2010). Evaluation of antibacterial activity of the leaf and flower essential oils of Gliricidia sepium from south India. International Journal of Applied Pharmaceutics, 2(2): 20-22.

Kaushik, P., Goyal, P., Chauhan, A. & Chauhan, G. (2010). In vitro evaluation of antibacterial potential of dry fruit extracts of Elettaria cardamomum Maton (Chhoti Elaichi). Iranian journal of pharmaceutical research: IJPR, 9(3): 287-292.

Kim, J.; Marshall, M. R. & Wei, C. I. (1995). Antibacterial activity of some essential oil components against five foodborne pathogens. Journal of agricultural and food chemistry, 43(11): 2839-2845.

Koroch, A. R.; Rodolfo Juliani, H. & Zygadlo, J. A. (2007). "Bioactivity of essential oils and their components," in Flavours and Fragrances Chemistry, Bioprocessing and Sustainability, R. G. Berger, Ed., pp. 87–115, Springer, Berlin, Germany.

Lang, G. & Buchbauer, G. (2012). A review on recent research results (2008–2010) on essential oils as antimicrobials and antifungals. A review. Flavour and Fragrance Journal, 27(1): 13-39.

Lawal, O. A.; Ogunwande, I. A.; Omikorede, O. E.; Owolabi, M. S.; Olorunsola, F. F.; Sanni, A. A. & Opoku, A. R. (2014). Chemical composition and antimicrobial activity of essential oil of Ocimum kilimandscharicum (R. Br.) Guerke: a new chemotype. American Journal of Essential Oils and Natural Products, 2(1), 41-46.

Lestari, E. S. (2004). Antimicrobial resistance among Staphylococcus aureus and Escherichia coli isolates in the Indonesian population inside and outside hospitals.14th European Congress of Clinical Microbiology and Infectious Diseases. Prague / Czech Republic.

Longbottom, C. J.; Carson, C. F.; Hammer, K. A.; Mee, B. J. & Riley, T. V. (2004). Tolerance of Pseudomonas aeruginosa to Melaleuca alternifolia (tea tree) oil is associated with the outer membrane and energy-dependent cellular processes. Journal of Antimicrobial Chemotherapy, 54(2): 386-392.

Mahdi, N. M. (2016). Antibacterial Effect of Dill Seed Oil Anethum graveolens Anethum graveolens. Kufa journal for nursing sciences, 6(2): 1-8.

Manimaran, S.; Themozhil, S.; Nanjan, M. J. & Suresh, B. (2007). Chemical composition and antimicrobial activity of cone volatile oil of Cupressus macrocarpa Hartwig from Nilgiris, India. Natural Product Sciences, 13(4): 279-282.

Manivannan, R.; Kumar, M. S.; Jawahar, N.; Ganesh, E. S. & Jubie, S. (2005). A comparative antimicrobial study on the essential oil of the leaves of various species of cupressus. Ancient Science of Life, 24(3), 131-133.

Matasyoh, J. C.; Maiyo, Z. C.; Ngure, R. M. & Chepkorir, R. (2009). Chemical composition and antimicrobial activity of the essential oil of Coriandrum sativum. Food Chemistry, 113(2), 526-529.

Mubita, C.; Syakalima, M.; Chisenga, C.; Munyeme, M.; Bwalya, M. & Chifumpa, G. (2008). Antibiograms of faecal Escherichia coli and Enterococci species isolated from pastoralist cattle in the interface areas of the Kafue basin in Zambia. Veterinarski Arhiv; 78(2): 179-185.

Mulyaningsih, S.; Sporer, F.; Zimmermann, S.; Reichling, J. & Wink, M. (2010). Synergistic properties of the terpenoids aromadendrene and 1, 8-cineole from the essential oil of Eucalyptus globulus against antibiotic-susceptible and antibiotic-resistant pathogens. Phytomedicine, 17(13), 1061-1066.

Nazzaro, F.; Fratianni, F.; De Martino, L.; Coppola, R. & De Feo, V. (2013). Effect of essential oils on pathogenic bacteria. Pharmaceuticals, 6(12): 1451-1474.

Nezhad, F. M.; Zeigham, H.; Mota, A.; Sattari, M. & Yadegar, A. (2009). Antibacterial activity of Eucalyptus extracts on methicillin resistance Staphylococcus aureus. Research Journal of Biological Sciences, 4(8): 905-908.

Osman, Y. A. H.; Yaseen, E. M. & Farag, M. M. (2009). Antimicrobial effect of some essential oils mixtures. Journal of Applied Sciences Research, (September, 1265-1276.

Oussalah, M.; Caillet, S. & Lacroix, M. (2006). Mechanism of action of Spanish oregano, Chinese cinnamon, and savory essential oils against cell membranes and walls of Escherichia coli O157: H7 and Listeria monocytogenes. Journal of food protection, 69(5): 1046-1055.

Owlia, P., Saderi, H., & Rasooli, I. (2012). Antimicrobial characteristics of some herbal oils on Pseudomonas aeruginosa.

Owlia, P.; Saderi, H.; Rasooli, I. & Sefidkon, F. (2009). Antimicrobial characteristics of some herbal Oils on Pseudomonas aeruginosa with special reference to their chemical compositions. Indian J Pharmacol Res., 8 (2): 107-114.

Pandey, A. K.; Kumar, P.; Singh, P.; Tripathi, N. N. & Bajpai, V. K. (2017). Essential oils: Sources of antimicrobials and food preservatives. Frontiers in microbiology, 7: 1-13.

Raut, J. S. & Karuppayil, S. M. (2014). A status review on the medicinal properties of essential oils. Industrial crops and products, 62: 250-264.

Ravid, U.; Putievsky, E.; Katzir, I. & Lewinsohn, E. (1997). Enantiomeric composition of linalool in the essential oils of Ocimum species and in commercial basil oils. Flavour Fragr. J., 12: 293-296.

Rudramurthy, G. R.; Swamy, M. K.; Sinniah, U. R. & Ghasemzadeh, A. (2016). Nanoparticles: alternatives against drug-resistant pathogenic microbes. Molecules, 21(7), 836: 2-30.

Saad, N. Y.; Muller, C. D. & Lobstein, A. (2013). Major bioactivities and mechanism of action of essential oils and their components. Flavour and Fragrance Journal, 28(5): 269-279.

Safrudin, I.; Maimulyanti, A. & Prihadi, A. R. (2015). Effect of crushing of clove bud (Syzygium aromaticum) and distillation rate on main constituents of the essential oil. American Journal of Essential Oils and Natural Products, 2(3): 12-15.

Sahoo, S.; Kar, D. M.; Mohapatra, S.; Rout, S. P. & Dash, S. K. (2006). Antibacterial Activity of Hybanthus enneaspermus Against Selected Urinary Tract Pathogens. Indian journal of pharmaceutical sciences, 68(5): 653-655.

Saiman, L. (2004). Microbiology of early CF lung disease. Paediatric respiratory reviews, 5: S367-S369.

Salem, M. Z.; Elansary, H. O.; Ali, H. M.; El-Settawy, A. A.; Elshikh, M. S.; Abdel-Salam, E. M. & Skalicka-Woźniak, K. (2018). Bioactivity of essential oils extracted from Cupressus macrocarpa branchlets and Corymbia citriodora leaves grown in Egypt. BMC Complementary and Alternative Medicine, 18: 1-7.

Selim, S. A.; Adam, M. E.; Hassan, S. M. & Albalawi, A. R. (2014). Chemical composition, antimicrobial and antibiofilm activity of the essential oil and methanol extract of the Mediterranean cypress (Cupressus sempervirens L.). BMC complementary and alternative medicine, 14: 1-8.

Sharma, M.; Koul, A.; Sharma, D.; Kaul, S.; Swamy, M. K. & Dhar, M. K. (2019). Metabolic engineering strategies for enhancing the production of bio-active compounds from medicinal plants. Natural Bio-active Compounds: Biotechnology, Bioengineering, and Molecular Approaches, 3: 287-316.

Simon, J. E.; Chadwick, A. F.; Craker, L. E. Herbs: an indexed bibliography, (1971-1980). The scientific literature on selected herbs, and aromatic and medicinal plants of the temperate zone. Archon Books: Hamden, CT, 1984.

Sudipta, K. M.; Lokesh, P.; Rashmi, W.; Vijay, R. & Ssn, K. (2012). Phytochemical screening and in vitro antimicrobial activity of Bougainvillea spectabilis flower extracts. International journal of phytomedicine, 4(3): 375-379.

Swamy, M. K. & Sinniah, U. R. (2015). A comprehensive review on the phytochemical constituents and pharmacological activities of Pogostemon cablin Benth.: an aromatic medicinal plant of industrial importance. Molecules, 20(5): 8521-8547.

Swamy, M. K. & Sinniah, U. R. (2016). Patchouli (Pogostemon cablin Benth.): botany, agrotechnology and biotechnological aspects. Industrial Crops and products, 87:161-176.

Swamy, M. K.; Mohanty, S. K.; Sinniah, U. R. & Maniyam, A. (2015). Evaluation of patchouli (Pogostemon cablin Benth.) cultivars for growth, yield and quality parameters. Journal of essential oil bearing plants, 18(4): 826-832.

Teixeira, B.; Marques, A.; Ramos, C.; Neng, N. R.; Nogueira, J. M.; Saraiva, J. A. & Nunes, M. L. (2013). Chemical composition and antibacterial and antioxidant properties of commercial essential oils. Industrial crops and products, 43, 587-595.

Turina, A. D. V.; Nolan, M. V.; Zygadlo, J. A. & Perillo, M. A. (2006). Natural terpenes: self-assembly and membrane partitioning. Biophysical chemistry, 122(2): 101-113.

Zhang, J. H.; Sun, H. L.; Chen, S. Y.; Zeng, L. I. & Wang, T. T. (2017). Anti-fungal activity, mechanism studies on α-Phellandrene and Nonanal against Penicillium cyclopium. Botanical studies, 58: 1-9.

Zhang, Y.; Feng, R.; Li, L.; Zhou, X.; Li, Z.; Jia, R. & Yin, Z. (2018). The antibacterial mechanism of terpinen-4-ol against Streptococcus agalactiae. Current microbiology, 75: 1214-1220.