Essential Oils and Antimicrobial Effects

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Abstract— Essential oils are hydrophobic, typically fragrant plant extracts. These oils are usually obtained by steam distillation and contain a number of terpene hydrocarbons. Essential oils can be obtained from any part of the plant. Since ancient ages, essential oils are used for upper respiratory tract infections, skin diseases, kidney stones, stomach and liver diseases, the treatment of parasitic diseases. Recently, the resistance of bacteria to antibiotics and the treatment of some infections led researchers to natural resources. Essential oils have bactericidal potential, namely their inhibition doses on microorganisms and their lethal doses are close. Therefore, it is promising especially in the elimination of bacteria with multiple antibiotic resistance.

There are many studies on the antimicrobial effects of essential oils derived from various plant species. However, the antimicrobial effects of some plant families have proven to be stronger, such as, Lamiaceae, Apiaceae, Lauraceae, Myrtaceae, Rutaceae, Poaceae. They are effective among the microorganisms as *S. aureus*, *E. coli*, *Klebsiella* spp, *Salmonella* spp., *P. aeruginosa*, *L. monocytogenes*, *C. jejuni*, *C. albicans*, *Aspergillus* spp, *Cryptococcus* spp and *Fusarium* spp. Studies on how essential oils kill organisms have revealed some ideas about their mechanisms. İt has been reported that Gram (-) bacteria are more durable than Gram (+) to essential oils. In general, essential oils act by disrupting the cell membrane integrity of microorganisms. It has also been reported that they deform the intracellular structures depending on the dose and duration.

Although their advantage, the use of essential oils brings some risks. Unconscious use of essential oils that in the drug class Over-the-counter (OTC) may result in overdose, intoxication or drug interactions, resulting in serious injury and organ failure. The aim of this study was to review the potentials and possible risks of essential oils for antimicrobial activity

Index Terms— Antimicrobial, Plant, Essential oil, Volatile oil.

1 INTRODUCTION

Essential oils (EO) are aromatic compounds produced by plants as secondary metabolites. They are a mixture of hydrocarbons, alcohol, aldehydes, esters, ethers, ketones, oxides, phenols and terpenes with their characteristic smell [1]. EO are liquid, volatile, clear and rarely colored, soluble in lipid and generally have a lower solubility in organic solvents than water. They can be obtained from different parts of plants such as, flowers, leaves, stems, seeds, fruits, roots or bark. EO are very complex natural mixtures that can contain approximately 20-60 components in quite different concentrations [2]. Various factors affect the yield and effect of EO. These factors are; plant growing area, the season of collection, used plant organs and essential oil used to obtain the method. Different methods are used to obtain EO. The oldest and most common method is the hydrodistillation method. The method of hydrodistillation is based on boiling of water and plant material in a glass flask connected to the refrigerant for 2-8 hours, condensing the oil molecules moving with the water vapor in the refrigerant and separating from the water. EO yield varies between plant species [3]. In the Steam Distillation method, steam applied to the fresh plant material placed in the glass container drains the oil droplets into the collection container and the oil is condensed and separated from the water.

Other methods are vacuum distillation, Solvent extraction, Supercritical liquid extraction, Microwave extraction, compressed solvent extraction, Solid-phase micro extraction, versatile extraction and mechanical method [4].

2 ESSENTIAL OIL COMPONENTS

EO are not real oils, since they do not contain lipids, they are highly complex compounds consisting of about 20-60 components in various concentrations. EO generally consist of hydrocarbons and oxygenated derivatives of hydrocarbons. These derivatives include alcohols, acids, esters, aldehydes, ketones, phenol and phenol ethers, quinones, lactones, furan derivatives, oxides, amines and sulfur compounds [5]. Most of the substances found in EO are of terpene origin. Terpenes are one of the most common groups of natural products. For example, other spices such as, citrus fruits and cinnamon are including of several terpenes. Limonene and citral, camphor, pinene, eugenol, anethol, thymol, geraniol have been the subject of many studies are well known terpenes. Chemically, terpenes are defined as a group of molecules whose structure is various but having a certain number of isoprene units. Terpenes are classified according to the number of isoprene in the basic molecular skeleton; Monoterpenes (C10),

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Sesquiterpenes (C15), Diterpenes (C20), Sesterpenes (C25), Triterpenes (C30), Carotenoids (40). Most terpenes are hydrocarbons; however, they may also be oxygen-containing compounds such as, alcohols, ketones or aldehydes. Monoterpenes and Sesquiterpenes are essential components of essential oils [6]. The researchers observed the effects of some essential oils by isolating the major components of essential oils in order to make drug formulations and to achieve the standard of treatment. For exapmle, Lamiaceae family is one of the families with the highest EO production and yield. However, members of the lamiaceae family are known for their antimicrobial activity. Thymol and carvacrol are plant-specific terpenes belonging to the lamiaceae family. Antimicrobial studies have reported that thymol and carvacrol affect both gram-positive and gram-negative bacteria. Electron microscopy images showed that both terpenes can disrupt the stability of the membrane structure. Thymol and carvacrol are also reported to prevent biofilm formation [7].

3 MEDICAL USES OF ESSENTIAL OILS

The use of EO is much older than their nomenclature and stabilization. One of the advanced societies in medicine, ancient Egypt used plant extracts and essential oils to treat wounds, store food, and even embalming [4]. It is known that essential oil can be obtained from more than 3000 plant species, of which about 300 have commercial value. They have been used in food, agriculture, animal husbandry, cosmetics, aromatherapy for a long time. EO themselves or some components are of great interest in the field of pharmacology [8]. It is common practice to use plants for healing in various parts of the world. With the recognition of the symptoms and treatment mechanisms of diseases, new methods have been developed for the effective use of plant-based active substances. The use of essential oils for the treatment of cancer called the plague of the age has been the subject of research and some promising results have been obtained.

One of the most important problems encountered in the treatment of cancer is that chemotherapeutics that exhibit cytotoxic effects especially affect cells with high proliferative index. Therefore, the dose required for treatment is reduced [9]. Studies have shown that monoterpenes have a tumor suppressing effect. In addition, the chemoprevention effects of monoterpenes protect healthy cells from the harm of treatment (such as, preventing DNA interaction) [10]. Ramadan et al. Reported that Tea tree EO induces apoptosis in human malignant melanoma (A-375) and squamous cell carcinoma (HEp-2) cell lines [12]. In contrast, turmeric essential oil showed anti-carcinogenic effects by inhibiting the isoforms of cytochrome p450 (CYP1A1, CYP1A2, CYP2B1 / 2, CYP2A, CYP2B and CYP3A) enzymes that are involved in the activation of carcinogens [13].

In biological systems, the deterioration of the balance between free radicals and antioxidants with scavenging action against them is defined as oxidative stress. Oxidative stress plays important role in the pathogenesis of atherosclerosis, cardiovascular diseases, diabetes, aging and degenerative diseases. Reactive oxygen species are molecules with very high reactivity and are formed as a result of normal metabolism in the cell organelles, especially mitochondrium, or by the effect of environmental stimuli. Although it is still a subject of debate among researchers, it is now known that oxidative stress can lead to tissue and organ damage if not balanced. The importance of terpenes (the main component of aromatic species of Lamiaceae family) under the antioxidant effect of EO emphasized in experimental studies. Today, many people use EO as food supplements to protect against various diseases including cancer [14].

In addition to anticarcinogenic, antimutagenic and antioxidant effects, essential oils are very valuable in the treatment of wounds caused by various reasons. The skin is the largest organ that protects the human body from external factors [15]. The deterioration of skin integrity for any reason makes the organism vulnerable to pathogenic microorganisms. In particular, large and deep surface wounds disrupt the organism's homeostasis. Contamination of wounds with microorganisms can delay healing and affect the functioning of organs, causing malformation and death [16].

Ancient people have also used EO to treat wounds. Wound healing is a physiological process in which many different cells and molecules work together. The healing process takes place in the form of inflammation reaction, cell proliferation and synthesis and remodeling of the extracellular matrix components [17]. EO exhibit immunomodulatory effect, particularly by inducing apoptosis and cell proliferation, which can accelerate the wound healing process [18]. Infections are a major problem in wound healing and often delay treatment. Colonization and biofilm formation can trigger systemic reactions with delayed wound healing process. Microorganisms that cause widespread wound infection are shown in table 1 [19]. The antimicrobial effect of the therapeutics to be used in wound treatment is a feature desired by healthcare professionals. The resistance to antibiotic agents used today is a serious public health problem. EO of plant origin offer a wide potential of antimicrobial action with their various components. Antimicrobial essential oils and the microorganisms to which they are effective are shown in Table 2.

TABLE 1
COMMON MICROORGANISMS CAUSING WOUND INFECTION

Microorganism	Incidence	Microorganism	Incidence
S. aureus	Chronic wounds	*Proteus sp.*	
S. epidermidis	Acute wounds	*Klebsiella sp.*	
Streptococcus pyogenes	Chronic wounds	*Propionibacterium acnes*	Acute wounds
P. aeruginosa		*Acinetobacter baumannii*	Chronic wounds
Stenotrophomonas maltophilia		*Candida ssp.*	Chronic wounds
E. coli	Chronic wounds	*Aspergillus ssp*	Colonize

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TABLE 2
ESSENTIAL OILS AND AFFECTED MICROORGANISMS

Common name	Plant	Microorganism	References
Tea tree oil	*Melaleuca alternifolia*	*MSSA* *MRSA* *Escherichia coli* *Pseudomonas aeruginosa* *Acinetobacter baumannii* *Aspergillus niger*	Oliva et al 2018 [20].
Thyme	*Thymus vulgaris*	*Escherichia coli* *Staphylococcus aureus* *Pseudomonas putida* *Candida albicans* *Campylobacter jejuni*	Imelouane et al 2009 [21].
Clove oil	*Syzygium aromaticum*	*Escherichia coli* *Staphylococcus aureus* *Bacillus subtilis* *Klebsiella pneumonia* *Pseudomonas aeruginosa*	Prabuseenivasan et al 2006 [22].
Betel oil	*Piper betle*	*Acinetobacter* *Klebsiella pneumonia* *Proteus vulgaris* *Enterecoccus fecalis* *Staphylococcus aureus* *Staphylococcus epidermidis*	Aumeeruddy-Elalfi et al 2015 [23].
Oregano oil	*Origanum vulgare*	*Escherichia coli* *Klebsiella pneumoniae* *Listeria monocytogenes* *Proteus mirabilis* *Proteus vulgaris* *Pseudomonas aeruginosa* *Candida albicans* *Streptococcus pyogenes* *Streptococcus mutans* *Salmonella enteritidis* *Bacillus cereus* *Staphylococcus aureus* *Streptococcus pneumoniae* *Micrococcus luteus* *Aspergillus ochraceus*	Özkalp et al 2010 [24]. Kulaksız et al 2018 [25].

TABLE 2
ESSENTİAL OİLS AND AFFECTED MİCROORGANİSMS
(CONT.)

Lavender oil	*Lavandula officinalis*	*Escherichia coli* *Staphylococcus aureus*	Martucci et al 2015 [26].
Mentha oil	*Mentha pulegium*	*Staphylococcus aureus* *Staphylococcus epidermidis* *Bacillus cereus* *Listeria monocytogenes* *Salmonella typhimurium* *Vibrio cholera* *Aspergillus niger* *Candida albicans*	Mahboubi and Haghi 2008 [27].
Curcumin oil	*Curcuma longa*	*Escherichia coli* *Staphylococcus aureus* *Streptococcus mutans* *Mycobacterium smegmatis* *Enterococcus faecalis* *Staphylococcus epidermidis* *Candida albicans* *Aspergillus niger* *Aspergillus flavus*	Mishra et al 2018 [28].
Rosemary oil	*Rosmarinus officinalis*	*Staphylococcus aureus* *Bacillus cereus* *Bacillus subtilis* *Bacillus pumilis* *Pseudomonas aeruginosa* *Salmonella poona* *Escherichia coli*	Hussain et al 2010 [29].
Juniper oil	*Juniper galbuli* *Juniperus communis*	*Haemophilus influenza* *Klebsiella pneumoniae* *Salmonella enterica* *Clostridium perfringens* *Enterococcus faecalis* *Staphylococcus aureus* *Candida albicans*	Zheljazkov et al 2017 [30].

4 Antmicrobial Mechanism of Essential Oils

Gram-negative bacteria are more resistant to EO than Gram-positive bacteria. The cell wall of gram-positive bacteria consists of a layer of peptidoglycan to which other molecules such as, teichoic acid and proteins bind. The cell wall structure of the gram-positive bacterium allows for the easy entry of hydrophobic molecules into the cells. Phenolic compounds found in EO generally show antimicrobial activity against Gram-positive bacteria. The effects depend on the amount of compound present; at low concentrations, they can interfere with enzymes involved in energy production, at high concentrations, they may denature of proteins. The cell wall of gram-negative bacteria is thinner and there is an outer membrane of lipoproteins outside this peptidoglycan structure. One of the reasons why gram negatives are more resistant is this different cell cover, and increased efflux pump expression is an important factor in the resistance of gram-negative bacteria [31].

The mechanism of action of essential oils varies according to the components they contain. One of the most common antimicrobial mechanisms attributed to EO is the toxic effects on membrane structure and membrane function. EO first disrupt the structure of the cell membrane and then cell homeostasis and drag it to death. Another mechanism is inhibition of the electron transport system. Cell membrane is vital for bacteria. Essential oils don't only act on the membrane but are the first target membrane. After reaching the cell, EO can inhibit cell division by acting on proteins. Furthermore, their interactions with DNA and RNA have various mechanisms of action such as, inhibition of membrane regeneration and inhibition of the synthesis of enzymes necessary for life. EO components such as, eugenol, carvacrol, and cinnamaldehyde can inhibit membrane-bound ATPase activity. They also cause inhibition of other enzymes and impair metabolism. Glucose metabolism was impaired in bacteria cells exposed to carvacrol and glucose accumulation was determined in the cell [32]. EO can disrupt the quorum sensing between bacteria and may even affect bacterial virulence due to gene expression by acting on DNA [33].

The treatment of infections caused by antibiotic resistant bacteria is becoming increasingly difficult. The use of EO alone or in combination with antibiotics is promising to overcome antibiotic resistance. Data indicate that *Acinetobacter baumannii*, a gram-negative bacterium, develops resistance to almost all commercial antimicrobials (cephalosporins, penicillins, carbapenems, aminoglycosides, fluoroquinolones, sulbactam). However, when Coriandrum sativum, Aniba rosaeodora, Eucalyptus camaldulensis, Aniba rosaeodora, Pelargonium graveolens and Myrtus communis combined with antibiotics of essential oils obtained from plants containing rich in linalool showed bacteriostatic / bacteriocidal effect. Changes in the cell membrane of EO are important. EO, when used in combination with protein synthesis inhibitors such as, gentamicin, facilitate the entry of the antibiotic into the cell. Concomitant use of EO and antibiotics lowers MIC levels, contributing to reducing unwanted drug effects and toxicity [34].

Escherichia coli is a common infectious agent with multiple drug resistance. *E. coli*, a gram-negative bacterium, is naturally resistant to penicillin G. It is also resistant to broad-spectrum β-lactamases (due to β-lactamase production), carbapenems (due to carbapenemase production) and quinolones. The EO of certain species of the Lamiaceae family (such as, Rosmarinus officinalis, Hyptis folium, Thymus riatarum, Origanum vulgare) have killed multiple drug-resistant *E. coli* strains when used in combination with antibiotics (such as, chloramphenicol, tetracycline, Streptomycin) [35]. Nevertheless, many experimental studies have reported that the major components of EO such as, carvacrol, thymol, linalool alone or in combination with antibiotics are lower than those of original EO. This is explained by the synergistic effect of the complex components of EO [36].

5 Toxicity of Essential Oils

The fact that EO are natural does not make them completely reliable. EO are thought to be toxic if misused or often taken at high doses [37]. Oral administration of EO rather than topical applications is considered to be more risky by physicians. Specific toxicity is roughly divided into two types: acute and chronic. Acute toxicity is the most risky poisoning. Symptoms of oral poisoning with EO include nausea, vomiting, ataxia, confusion, convulsions, and coma [38]. The toxic dosage range is different for each EO. Reported cases of intoxication after oral intake are usually associated with high doses and children. Nevertheless, to date, death has occurred in very few cases of intoxication [39].

Most of the available information on toxicity is based on animal experiments. There are two ways to measure EO toxicity: oral and dermal. In this way LD50 and lethal dose are determined. This finding gives an idea about the toxic dose of EO. In addition, these findings should be supported by invivo and in vitro toxicology tests. It has been reported that the application of EO in pure or high concentrations on the skin surface may cause irritation and severe tissue necrosis [40,]. Liver and kidney failure, respiratory depression, cardiac diseases and seizures are among the reported risks due to the increase in dosage and duration of oral use. In its 2016 report, the American Association of Poison Control Centers reported that essential oil poisoning increased, people of all ages were affected, but children should be paid special attention [41]. Controlling the therapeutic doses of EO and selling them in pharmacies just like medicines will largely prevent poisoning.

6 Conclusions

Although essential oils have been used since ancient times, they are not known enough, their mechanisms have not been fully elucidated and the clinic has not been able to find the place they deserve. EO are a natural source of valuable active compounds that can be used in the treatment and prophylaxis of diseases. Determining and standardizing the effects of this natural resource by in vivo and in vitro studies will contribute to the solution of the antimicrobial problem that the world needs today.

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