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Chemical Composition and Antibacterial Activity of Essential oils of Some Algerian and Turkish Medicinal Plants

Bazı Cezayir ve Türk Şifalı Bitkilerinin Uçucu Yağlarının Kimyasal Bileşimi ve Antibakteriyel Aktivitesi

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Abstract

In the present time, drug resistance in microbes is a very serious problem. Essential oils and other extracts of plants have evoked interest as sources of natural products. They have been screened for their potential uses as alternative remedies for the treatment of many ailments. In the present study, the in vitro antibacterial activity of the essential oils from ten aromatic plants Thymus fontanesii, Thymus vulgari, pulegium, Eugenia caryophyllata, Mentha Origanum vulgare, Laurus nobilis, salvia, Geranium. Lavender and Cinnamomum aromaticum was investigated. The oils compositions were characterized gas chromatography/mass spectrophotometrical analyses. The antibacterial activity of these oils were tested against seven pathogenic bacteria: Pseudomonas aeruginosa **ATCC** Escherichia coli ATCC 25922, Staphylococcus oxaR ATCC 43300, Staphylococcus aureus oxaS ATCC 25923, Staphylococcu aureus ATCC 33862, Bacillus cereus ATCC 11778, Bacillus subtilis ATCC 6633; by using the agar incorporation method to determine the Minimum Inhibitory Concentration (MIC) of each oil. The results have shown a greater antibacterial effect of all essential oils samples against Gram positive than the Gram negative bacteria. Pseudomonas aeruginosa was the only

Özet

Günümüzde, mikroplardaki ilaç direnci çok ciddi bir sorundur. Uçucu yağlar ve diğer bitki özleri, doğal ürünler kaynağı olarak ilgi uyandırdı. Birçok rahatsızlığın tedavisi için alternatif ilaçlar olarak potansiyel kullanımları için taranmıştır. Bu çalışmada, on aromatik bitkilerden Thymus fontanesii, Thymus vulgari, Mentha pulegium, caryophyllata, Origanum Eugenia Laurus nobilis, salvia, Sardunya, Lavanta ve Cinnamomum aromaticum'dan elde edilen uçucu vağların vitro antibakterivel in aktivitesi araştırılmıştır. Yağ bileşimleri, kromatografisi spektrofotometrik / kütle analizleri ile karakterize edildi. Bu yağların antibakteriyel aktivitesi, yedi patojenik bakteriye karsı test edildi: Pseudomonas aeruginosa ATCC 27853. Escherichia coli **ATCC** 25922. Staphylococcus aureus oxaR ATCC 43300, Staphylococcus aureus oxaS ATCC 25923, Staphylococcu aureus ATCC 33862, Bacillus cereus ATCC 11778, Bacillus subtilis ATCC 6633; Her bir yağın Minimum İnhibitör Konsantrasyonunu (MIC) belirlemek için agar ekleme yöntemini kullanarak. Sonuçlar, Gram esansivel örneklerinin Gram negatif yağ bakterilere göre Gram pozitif karşısında daha antibakteriyel etkisi olduğunu büyük bir göstermiştir. Pseudomonas aeruginosa, Laurus nobilis, salvia, sardunya, lavanta ve Mentha

bacterium not susceptible to *Laurus nobilis*, salvia, geranium, lavender and Mentha pulegium oils. The highest and broadest activity was shown by *Cinnamomum aromaticum* oil. These results suggest that these essential oils may be alternative natural source medicine to prevent and treat many bacterial diseases.

Keywords: Essential oils, Chemical compositions, Medicinal plants, Antibacterial activity.

pulegium yağlarına karşı duyarlı olmayan tek bakteriydi. En yüksek ve en geniş aktivite Cinnamomum aromaticum yağı tarafından gösterilmiştir. Bu sonuçlar, bu esansiyel yağların birçok bakteri hastalığını önlemek ve tedavi etmek için alternatif doğal kaynak ilaç olabileceğini düşündürmektedir.

Anahtar kelimeler: Uçucu yağlar, Kimyasal bileşimler, Tıbbi bitkiler, Antibakteriyel aktivite

Abbreviations: MIC, Minimum Inhibitory Concentration; EOs, Essential oils

1. INTRODUCTION

Traditional systems of medicine continue to be widely practiced on many accounts. Population rise, inadequate supply of drugs, prohibitive cost of treatments, side effects of several allopathic drugs and development of resistance to currently used drugs for infectious diseases have led to increased emphasis on the use of plant materials as a source of medicines for a wide variety of human ailments (Joy, Thomas, Mathew & Skaria, 1998). Medicinal and aromatic plants constitute a major source of natural organic compounds. It has been estimated that 14 - 28% of higher plant species are used medicinally and that 74% of pharmacologically active plant derived components were discovered after following up on ethnomedicinal use of the plants (Ncube, Afolayan & Okoh, 2008). Essential oils (EOs) and extracts from a wide variety of plants have long been used for medicinal purposes. They possess multiple antimicrobial. antibacterial, antifungal, anticancer, antiviral and antioxidant properties. The antimicrobial activities of essential oils have been recognized for many years and recently have been extensively researched. More particularly, essential oils and their components such as Cinnamaldhyde, Carvacrol, Eugenol Camphor are known effective agents against a wide variety of microorganisms including pathogens bacteria (Mahmoodi, Roomiani, Afshin, Akhondzadeh, Kamali & Taheri, 2012) .Scientifically these oils have been proved highly potent antimicrobial agents in comparison to antibiotics. The main objective of this study was to evaluate the chemical composition and the antibacterial activity of essential oils of some medicinal plants in an attempt to contribute to the use of this EOs as an alternative product.

2. MATERIAL AND METHODS

Our study was carried out at the laboratory of research on local animal products of Ibn-Khaldoun University, Tiaret, Algeria during the period from March to June 2013. *Origanum vulgare, Laurus nobilis* and *salvia* essential oils were purchased from Turkey in 2007, geranium and lavender essential oils were purchased from Algeria, the medicinal plants (*Thymus fontanesii, Thymus vulgaris* and *Mentha pulegium*) were collected from the region of Tiaret located in western Algeria during the year 2012.

2.1. Essential Oils Extraction

The aerial parts (leaves and flowers, 30 g) of the plant Thymus fontanesii, Thymus vulgaris, Mentha pulegium, the flower buds of clove (Eugenia caryophyllata) and the bark of Cinnamomum aromaticum were dried at room temperature, hydrodistilled for 3 h using a Clevenger apparatus type (British Pharmacopoeia, 1998). The oils were dried over anhydrous sodium sulfate and stored in the dark at 2-4°C. The yield of the essential oils was 1.56% (v/w), 2.39%, 7.45%, 1.90%, and 1.46% for Thymus vulgaris, Thymus fontanesii, Eugenia caryophyllata, Mentha puligium and Cinnamomum aromaticum, respectively.

2.2. Gas Chromatography-Mass Spectrometry (GC-MS)

GC analysis was carried out using a Shimadzu 2010 Plus gas chromatograph coupled to a

Shimadzu QP2010 Ultra mass selective detector. The seperation was performed by means of a Restek Rxi-5MS capillary column, 60 m length, 0.25 mm i.d. and a 0.25 μm phase thickness. The split mode was used. The oven program was as follows: Initial temperature was 60 °C for 2 min, which was increased to 240 °C at 3 °C min⁻¹, 250 °C was maintained for 4 min. Helium (99.999%) was used as carrier gas with a constant flow-rate of 1 mL min⁻¹. Detection was carried out in electronic impact mode (EI); ionization voltage was fixed to 70 eV. Scan mode (40-450 m/z) was for mass acquisition. The volatile compounds were identified by comparison of their retention indices (relative to C7-C30 alkane standards), and matching mass spectral data with those held in FFNSC1.2 and W9N11 library of mass spectra and literature comparison (Bicchi et al., 2008). This part of the study has been performed in the department of chemistry, Faculty of Sciences, Karadeniz Technical University, Trabzon Turkey.

2.3. Evaluation of The Antibacterial Activity

2.3.1. Bacterial Strains and Inoculums Standardization

Reference strains of Pseudomonas aeruginosa ATCC 27853, Escherichia coli ATCC 25922, Staphylococcus aureus oxaR ATCC 43300, Staphylococcus aureus oxaS ATCC 25923, Staphylococcu aureus ATCC 33862, Bacillus cereus ATCC 11778 and Bacillus subtilis ATCC 6633 were kindly provided by the university hospital center of Mustapha Pasha, Algiers (Algeria). Prior to the experiment the strain was maintained by subculture in the specific media; the inoculum suspension was obtained by taking five colonies from 24 hours culture, then suspended in 5 ml of sterile saline (0.85% NaCl) and shacked for 15 seconds. The density was adjusted to the turbidity of a 0.5 McFarland Standard (equivalent to 1×10^8 cfu/mL).

2.3.2. Minimum Inhibitory Concentration Measurement (MIC)

The MIC values of essential oils have been determined by using an agar incorporation technique method. Essential oils were incorporated into Mueller-Hinton media with increasing concentrations. The mixture was shacked moderately and poured into plates, then

standard inoculums of 0.5 McFarland of each bacterial strains was inoculated and the plates were incubated at 37°C for 24 hours. The MIC was determined based on the lowest concentration of essential oils that inhibited the growth of the tested organisms.

3. RESULTS AND DISCUSSION

3.1. Results

The chemical composition of the tested EOs is presented in Table 1 and the antibacterial activity of the tested EOs is shown in Table 2.

3.2. Discussion

The antibacterial activity of the ten selected essential oils against seven bacterial species is summarized in Table 2. The results revealed that the selected essential oils showed antibacterial activity with varying magnitudes. Cinnamomum aromaticum bark had the highest antibacterial effect, particularly against resistant strains Pseudomonas aeruginosa ATCC 27853 and Staphylococcus aureus oxaS ATCC 25923 with MIC values of 1 and 0.67 µl/ml, respectively. Previous studies showed that essential oils of aromatic plants have antimicrobial activity against bacteria and fungi. Their antimicrobial activity is attributed to the presence of small terpenoid and phenolic compounds. components with phenolic structures, such as carvacrol and eugenol were highly active against the tested microorganisms. Members of this class be either bactericidal or known to bacteriostatic agents, depending on the used concentration (Pelczar, Chan, & Krieg, 1988). These compounds were strongly active despite their relatively low capacity to dissolve in water.

The chemical composition of essential oils depends on climatic, seasonal, and geographic conditions, harvest period and distillation technique. In addition, their antibacterial activity depends on the type, composition concentration of the essential oils, the type and concentration of the target microorganism, the composition of the substrate, the processing and the storage conditions (Karataş & Ertekin, 2010). The activity of the oils would be expected to relate to the respective composition of the volatile plant oils.

Table1. Chemical composition of essential oils

Sample Code	Name of Compound	Area (%)	RI
Eugenia	•	, ,	
caryophyllata			
1	α –Pinene	0.27	944
2	Benzaldehyde	0.97	971
3	Trimethylbenzene	0.45	1003
4	Eucalyptol	0.42	1041
5	Benzenepropanol	0.88	1170
6	Endo-Borneol	0.13	1175
7	Benzylidene malonaldehyde	0.37	1229
8	Benzylidene acetaldehyde	57.75 0.25	1279
9	Bornyl acetate	0.25	1294
10	Eugenol	2.34	1365
11 12	E-caryophyllene Alloaromadendrene	0.17 7.99	1434 1762
13	β –Cedrene	0.63	1702
14	Pentacosane	4.03	2506
15	10, 12-Tricosadiynoic acid methyl ester	23.33	2609
Thymus vulgaris	15, 12 Theosadiynoic acid methyl ester	43.33	2007
1 nymus vuigaris	α –Pinene	7.83	944
2	Benzene,1,2,3-trimethyl	6.82	976
3	Myrcene	2.63	995
4	Benzene,1,3,5-trimethyl	24.65	1002
5	α -Terpene	3.55	1025
6	p-Cymene	27.38	1032
7	γ –Terpinene	12.36	1066
8	Carvacrol	12.77	1303
Thymus			
fontanesii			
1	α –Thufene	0.34	945
2	Isocumane	2.91	962
3	Benzene,1,2,3-trimethyl	13.16	976
4	Benzene, 1, 3, 5-trimethyl	52.37	1002
5	γ –Terpinene	8.41	1066
6	Undecane	5.03	1100
7	Carvacrol	17.78	1303
Mentha pulegium			
1	<i>p</i> -Menthan-3-one	5.32	1163
2 3	<i>p</i> -Menth-4(8)-en-3-one	23.81	1249
3	Cinnamaldehyde	60.39	1278
4	<i>p</i> -Beritone	7.16	1352
5	Pentocosene	3.32	2500
Origanum			
vulgare	Dannar 1 2 2 War da 1	10.07	076
1	Benzene 1,2,3 trimethyl	12.27	976
2 3	Benzene 1,3,5 trimethyl	66.90 5.80	1002
3 4	Undecane	5.89	1100
4	Carvacrol	14.94	1303
Salvia			
Saivia 1	α –Pinene	3.50	944
2	Camphene	3.40	960
3	β –Pinene	3.48	988
4	Eucalyptol	30.10	1041
5	Linalool	3.57	1103
6	2-Bornanone	20.29	1156
7	L-Borneol	17.28	1176
8	Bornyl acetate	6.57	1294
9	E- Caryoplhyllene	5.52	1434
10	Carophyllene oxide	6.27	1600
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Table 1 continue			
Laurus nobilis			
1	β –Pinene	5,5365	988
2	Eucalyptol	48,5014	1041
3	2- Bornanone	14,7950	1156
4	Endo-Borneol	12,3463	1175
5	Cinnamaldehyde	15,4148	1278
6	Iso Bornyl acetate	3,4059	1294
Lavander			
1	α –Pinene	3.17	944
2	Camphene	7.01	960
3	Benzene 1,3,5 trimethyl	6.92	1002
4	D-Limonene	1.07	1036
5	Eucalyptol	25.47	1040
6	Fenchone	43.95	1097
7	2- Bornanone	12.40	1156
Geranium			
1	Benzene 1,3,5 trimethyl	13.86	1002
2	Menthone	19.00	1161
3	Isomenthone	20.20	1172
4	Citronellol	30.81	1229
_ 5	Citronellyl formate	16.13	1276
Cinnamomum			
aromaticum			
1	α -Pinene	41.35	944
2	Camphene	21.68	960
3	2-Ethytoluene	11.69	970
4	Mesitylene	15.58	1002
5	Eucalyptol	6.60	1040
6	Cinnamaldehyde	3.09	1278

Table 2. The antibacterial activity of ten selected essential oils against bacterial specie.

MIC Values of Essential Oils (μl/ml)							
Strains EOs	S.aureus ATCC 33862	S.aureus ATCC 43300	S.aureus ATCC 25923	B.cereus ATCC 11778	B. subtilis ATCC 6633	E.coli ATCC 25922	P.aeruginosa ATCC 27853
Laurus nobilis	3	3	2	1	1.6	5	_
Salvia	3	3	3	2	2	7	_
Origanum vulgare	3.44	5.26	3.44	2.56	3.44	3.44	2.04
Thymus fontanesii	3.44	5.26	3.44	5.26	3.44	5.26	4
Thymus vulgaris	2.56	2.56	2.56	2.56	2.04	5.26	3
Geranium	5.26	5.26	5.26	3.44	3.44	11.11	_
Lavender	3	2	11.11	11.11	11.11	3	_
Eugenia caryophyllata	3.44	3.44	3.44	3.44	3.44	5.26	2.2
Mentha pulegium	1	1	1.2	1	0.8	1.6	_
Cinnamomum aromaticum	0.2	0.2	0.67	0.25	0.2	0.33	1

(-): no inhibition

It could be stated that factors like functional groups, configuration and chemical structure play a role in the activity of the constituents comprising essential oils against microbes. In this aspect, constituents of essential oils cause an increase in the permeability of the cell membrane and thus leads to the leakage of the vital intracellular components of the bacteria outside the membrane.

This causes a disturbance in the equilibrium of inorganic ions and possible impairment of bacterial enzyme system and cell respiration (Abu-Darwish. Al-Ramamneh. E.A.M.. Kyslychenko, V., Sergeevna, K. & Uliana, 2012). Essential oils can coagulate the cytoplasm and damage lipids and proteins. Burt (2004) showed that the antibacterial effects of the whole essential oils are stronger than their major components when they are individually studied. Thus, attention should be paid to the important role of the minor components. This indicated that synergistic effects existed between the major and minor compounds of the essential oils when they are combined together than they are acting separately. It can be seen that the growth of tested bacteria responded differently to the essential oils and their components, which indicates that different components may have different modes of action or that the metabolism of some bacteria is able to better overcome the effect of the oil or adapt to it. Gram negative bacteria are in general more resistant than Gram positive one (Soković, Glamočlija, Marin, Brkić & Griensven, 2010).

Better effectiveness of essential oils against Gram-positive bacteria may be due to volatile action of essential oils and to the absence of lipopolysaccharide layer in Gram positive bacteria that might function as an effective barrier against any incoming bio-molecules (Delaquis, Stanich, Girard, & Mazza, 2002).

The lower susceptibility of Gram-negative bacteria to the essential oils may be explained by diffusion limitations of essential compounds through their external membrane caused by the presence of a hydrophilic barrier. Although this barrier is not totally impermeable, it hinders the transport of macromolecules and hydrophobic components (Pierozan et al., 2009).

Pseudomonas aeruginosa was the only bacterium not susceptible to Laurus nobilis, Salvia, geranium, Lavander and Mentha pulegium oils and was therefore excluded from further assays. Many researchers confirm that the structure of the cell wall plays role in the resistance of *P. aeruginosa* to essential oils and their components. An outer lipopolysacharide wall is reported to be present in Gram negative bacterium and acts to prevent the entrance of toxic agents.

It seems evident that there is a relationship between the high activity of the Thymus and Oregano type oils and the presence of phenol components, such as carvacrol. The high antimicrobial activity of these essential oils could be explained by their high percentage of phenol components. It seems likely, that carvacrol interferes with the activity of cell wall enzymes like chitin synthase/chitinase as well as α - and β glucanases of fungi (Gaunt, Higgins & Hughes, 2005). Thus carvacrol in thyme and Oregon essential oils is able to increase cell membrane permeability and allows accumulation of yocimene and terpinen-4-ol in cytoplasmic membranes to toxics levels (Di pasqua, De feo, Villiani & Mauriello, 2005).

Soković et al. (2010) studied the antibacterial activity of these oils and their main components and they found that carvacrol is the dominant component in *Origanum vulgare* oil. Thymol and pcymene are the major components in *Thymus vulgaris* oil. *O. vulgare* and *T. vulgaris* essential oils possess a higher antibacterial effect than streptomycin. Carvacrol showed the strongest antibacterial activity.

Data presented in a study done by Fani et Kohanteb (2017) revealed strong inhibitory activity of *Thymus vulgaris* oil on some oral pathogens, including *S. pyogenes*, *S. mutans*, *C. albicans*, *A. actinomycetemcomitans*, and *P. gingivalis*.

Another study done by Ozkalp, Sevgi, Ozcan and (2010) showed Oregano oil that possessed stronger antibacterial activity against Escherichia coli**RSKK** 340, Klebsiella pneumonia **RSKK** 06017, Pseudomonas aeruginosa RSKK 06021, Salmonella enteritidis RSKK 96046, Streptococcus pyogones RSKK 413/214, Bacillus cereus **RSKK** 1122, Staphylococcus aureus **RSKK** 96090 and methicilline-resistant Staphylococcus aureus (MRSA) compared with the antibiotic ampicillin. Chaudhry Masood, Saeed and Tariq (2007) found that the oregano oil exhibited significant

inhibitory activity against *Citrobacter*. spp, *Salmonella typhi* and *Escherichia coli*. It has long been acknowledged that oregano oil is among the most active against strains of *E. coli* and also presents antimicrobial activity against pathogenic microorganisms like *H. pylori* (Stamatis, Kyriazopoulos, Goleyou, Basayiannis, Skaltsas, & Skaltsa, 2003). Many studies reported that oregano oil was active against GNB viz., *Proteus vulgaris*, *Aeromonas hydrophila*, *Klebsiella pneumoniae* and *Escherichia coli* (Baydar, Osman, Ozkan, & Karadoan, 2004).

Stojković, glamočlija, Ana ćirić, Nikolić, Ristić, and Soković (2013) found that the essential oil of O. vulgare was rich in carvacrol (64.50%), while p-cymene and γ -terpinene were also dominant constituents with slightly lower percentage (10.90% and 10.80%). T. vulgaris essential oil was abundant with thymol (48.92%), followed by p-cymene (18.99%).

Tohidpour, Sattari, Omidbaigi, Yadegar, and Nazemi (2010) found in their study that *T. vulgaris* essential oil showed better inhibitory effects than *Eucalyptus globulus* essential oil against Methicillin-resistant *Staphylococcus aureus* (MRSA).GC analysis of *T. vulgaris* resulted in thymol as the oil major compound whereas GC/MS assay exhibited eucalyptol as the most abundant constitute of *Eucalyptus globulus*.

In our study, *Eugenia caryophyllata* essential oil possessed a strong antibacterial effect against all tested bacteria and on the resistant strain *Escherichia coli* ATCC 25922. The major compounds of this oil are Benzylidene acetaldehyde 57.75%, 10, 12-Tricosadiynoic acid methyl ester 23.33%, Alloaromadendrene 7.99%, Pentacosane 4.03%, and Eugenol 2.34%. The chemical composition of our essential oil is different from result of other studies.

Oulkheir et al. (2017) have shown that the essential oil of *Eugenia caryophyllata* has antibacterial activity against four Gram-negative bacteria and two Gram-positive bacteria.

Carneiro et al. (2017) studied the chemical composition and biological activities of *Eugenia klotzschiana* essential oil the result of this study showed that the major compounds were α -copaene (10.6 %) found in oil from leaves, β -bisabolene (17.4 %) in the essential oil from dry leaves and α -(E)-bergamotene (29.9 %) in oil from flowers. The result of the antibacterial

activity showed that only the anaerobic bacterium *P. nigrescens* and the aerobic bacterium *S. mutans* were significantly inhibited by all essential oils under study.

Alma, Ertas, Nitz, and kollmannsberger (2007) found in their study that the major compounds of the essential oil from clove cultivated in Mediterranean region of turkey were eugenol 87%, eugenyl acetate 8.01% and β caryophyllene 3.56%.

In a study done by Chaieb et al. (2007) they found that the major components present in the clove bund oil were eugenol (88.6%), eugenyl acetate (5.6%), β -caryophyllene (1.4%) and this oil has a strong antioxidant activity when compared with the synthetic antioxidant (tert-butylated hydroxytoluene). The tested oil has a significant effect against 53 pathogenic *Candida* species.

Sabahat and Perween (2008) found that clove essential oil showed strong antibacterial activity against one hundred Gram-negative bacilli belonging to 10 different species.

Burst and Reinders (2003) indicated that clove oil was found effective against non-toxigenic strains of *E. coli* O157:H7. Similarly, in another study done by Lopez, Sanchez, Batlle, and Nerin, (2005) clove oil was found active against foodborne Gram-positive bacteria and Gramnegative bacteria

Recently Boukraâ, Alzahrani, Abdellah, Bakhotmah, and Hammoudi (2013) found that Eugenia caryophyllata essential oil has an antibacterial effect against **Pseudomonas** aeruginosa ATCC 27853. In a similar study Alzahrani, Boukraâ, Abdellah, Bakhotmah, and Hammoudi, (2014) found that the essential oil of Eugenia caryophyllata was the most effective against Aspergillus flavus and Aspergillus niger then the other tested oils: Thymus vulgaris, Thymus fontanisii, Origanum vulgaris Geranium.

In our study we found that the major compounds of *Mentha pulegium* essential oil are Cinnamaldehyde 60.39%, *p*-Menth-4(8)-en-3-on 23.81%, and p-Beritone 7.16% and this oil has a strong antibacterial effect against the tested strains except *Pseudomonas aeruginosa*.

Daniel, Daniel, Dellacassa, Davies, and Salvador (2002) studied the composition of *Mentha pulegium* essential oil collected from the

Uruguay, and they reported that the composition of this oil is characterized by a high content of pulegone (73.4%) and isomenthone (12.9%). Agnihotri et al. (2005) found that pulegone (65.9-83.1%), menthone (8.3-8.7%), isomenthone (3.8-4.0%), are the major compounds of *Mentha pulegium* from India.

Derwich, Benziane and Boukir (2010) studied the chemical composition and the antibacterial activity of the essential oils of *Mentha pulegium* from Morocco. The results obtained indicated that the major component was piperitone (35.56%), other components present in appreciable contents were: piperitenone (21.18%), alpha-terpineol (10.89%), pulegone (6.452%), pipéritone oxide (4.02%), menthol (3.28%), menthone (3.09%), neomenthol (2.80%), and these oils were found to be active against *Staphylococcus intermedius*, *Staphylococcus aureus*, *Micrococcus luteus*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Klebsielle pneumoniae* and *Proteus mirabilis*.

The result of our study showed that the essential oil of Cinnamomum aromaticum was the most active against the tested bacteria with major compounds α Pinene, Camphene and. Mesitylene. Prabuseenivasan, Manickkam, and Savarimuthu (2006) evaluated the antibacterial activity of 21 plant essential oils against six bacterial species four gram-negative bacteria (Escherichia coli, Klebsiella pneumoniae, Pseudomonas aeruginosa, Proteus vulgaris) and two gram positive bacteria (Bacillus subtilis and Staphylococcus aureus). They found that the essential oil of cinnamon was the most effective as an antibacterial agent. The antibacterial activity has been attributed to the presence of some active constituents in the oil. GC-MS study revealed cinnamaldehyde to be the major constituent of cinnamon oil.

The results of this work were found to be consistent with the work done by Bowels, Sackitey, and Williams (1995) showed that the essential oil of cinnamon inhibit the growth of *Staphylococcus aureus*. These findings are also quite similar with the results of Chao, Young, and Oberg, 2000) reporting that cinnamon bark oil fully inhibited the growth of some gram positive and gram-negative bacteria, fungi and yeasts. It has been proposed that cinnamaldehyde inhibit production of an essential enzyme by the bacteria

and/or cause damage to the cell wall of the bacteria.

In our study the major compound of Laurus nobilis essential oil is eucalyptol (1.8-cineole). This composition showed a similar pattern to those published from other geographical regions: 1.8-cineole was reported as the major component in the essential oil from Turkey, China, and Tunisia. The major components of this oil, 1.8cineole, has been known to exhibit antimicrobial activity against the bacterial strains E. coli, Pseudomonas aeruginosa, Salmonella typhi, Staphylococcus Staphylococcus aureus. (Sivropoulou, intermedius, Bacillus subtilis Nikolaou, Papanikolaou, Kokkini, Lanaras, & Arsenakis, 1997).

Derwich, Benziane and Bouki (2009) found that the major component of *Laurus nobilis* essential oil was 1,8-cineole (52.43%), and this oil has antibacterial effect against three bacterial strains: *Staphylococcus aureus*, *Staphylococcus intermedius* and *Klebsiella pneumonia*.

Moghtaderet and Farahmand (2013) found that the major components of laurus nobilis essential oil were 1,8-cineole (25.7%), sabinene (8.7%) and α -pinene (5.25%) and this oil has a strong antibacterial effect against 9 bacteria strains such as three Gram positive bacteria: Staphylococcus Staphylococcus epidermidis aureus, Streptococcus faecalis and six Gram negative bacteria: Pseudomonas aeroginosa, Shigella flexneri, Klebsiella pneuomoniae, Salmonella typhi, Serratia marcescens and Escherichia coli. The Effect of the essential oil of L. nobilis on tested bacteria was more than that of tetracycline antibiotic.

The result of our sturdy has shown that Salvia essential oil has antibacterial effect against all the tested bacteria except *Pseudomonas aeruginosa*. The major compounds of this oil were eucalyptol (1.8-cineole) Bornanone, Borneol, and Caryophyllen oxide.

Morteza, Sonboli, Ebrahimi and Hashemi (2007) demonstrated that *Salvia chloroleuca* oil exhibited moderate to high antimicrobial activity, especially for *Bacillus subtilis, Staphylococcus epidermidis* and *S. aureus.* Moreover, the study showed the antimicrobial activity of five major components of the oil, including 1, 8-cineole, α -pinene, β -pinene and β -caryophyllene.

Pitarokili, Tzakou, Loukis and Harvala (2003) had already demonstrated the antifungal activity of the *Salvia fruticosa* essential oil and the main components that are 1, 8-cineole and camphor. In this study, 1, 8- cineole was the most abundant component (16.9-48.3% of total oil) in all analyzed oils.

Khalil and Li (2011) in their study found that the essential oils of *Salvia officinalis* collected at two different altitudes in Syrian coastline were analyzed by gas chromatography. Plant's development stage and the ecological factors had impact on the qualitative composition of *Salvia officinalis* essential oil. The major compounds of these oils were 1, 8-cineol, camphor, borneol, α -pinene, β -pinene, camphene, β -myrcene and caryophyllene. The results presented showed that the essential oil of *Salvia officinalis* inhibited the growth of Gram-positive bacteria (*S. aureus* and *Streptococcus* group D) and gram-negative bacteria (*E. coli*, *S. typhi* and *P. aeruoginosa*).

Other study done by Karatas and Ertekin (2010) evaluated the antibacterial and antifungal activities of essential oils of four *Salvia* species. The results indicated that all essential oils exhibited antibacterial activity against *Bacillus subtilis, Escherichia coli* and *Staphylococcus aureus*. In addition, essential oils of *Salvia multicaulis* exhibited antibacterial activity against *Pseudomonas aeruginosa*. Essential oils of *Salvia syriaca* exhibited antibacterial and antifungal activities against all tested micro-organisms (*E. coli, Pseudomonas aeruginosa, S. aureus, Streptococcus pyogenes, B. subtilis* and *Candida albicans*).

In a study done by Ebani et al. (2018) Essential oils (EOs) from Salvia dolomitica and Salvia somalensis, were analyzed for composition and tested against bacterial and fungal pathogens isolated environmental from clinical and specimens. Both EOs showed similar percentages total monoterpenes and sesquiterpene hydrocarbons. The main constituents were 1,8cineole and b-caryophyllene in S. dolomitica and bornyl acetate and camphor in S. somalensis. The selected EOs have no relevant antifungal or compared antibacterial activities if conventional drugs.

In another study done by Çolak et al. (2018) the major compounds identified in essential oil of *S. staminea* were linally acetate (87.55%) and

linalool (22.05%) and this oil showed good antimicrobial activity against Gram-positive bacteria.

In our finding lavender essential oil has been active against all tested bacterial strains except aeruginosa Pseudomonas and compounds of this oil were fenchone 43.95%, eucalyptol (1.8-cineole) 25.47% and 2 Bornanone 12.40%. The results of a study done by Hui, He, Huan, Li, and AiGuo (2010) demonstrated that the essential oil of lavender consisted of 1,5-Dimethyl-1-vinyl-4-hexenyl but rated as the most abundant component, followed by 1,3,7-Octatriene, 3,7-dimethyl-, Eucalyptol , and Camphor . Lavender essential oil displays the strongest antioxidant activity against lipid peroxidation in a linoleic acid model system and good antibacterial activity against four rhinitisrelated bacteria including Staphylococcus aureus, Micrococcus ascoformans, Proteus vulgaris and Escherichia coli.

In a study done by Hossain, Heo, De Silva, Wimalasena, Pathirana, and Heo (2017) the essential oil of lavender (EOL) was examined for its antibacterial activity against thirty-eight strains of turtle-borne pathogenic bacteria belonging to seven species; *Aeromonas hydrophila*, *A. caviae*, *A. dhakensis*, *Citrobacter freundii*, *Proteus mirabilis*, *Salmonella enterica* and *Pseudomonas aeruginosa*. The results revealed that EOL was active against all tested turtle-borne pathogenic bacteria except *P. aeruginosa*.

Essential oil of geranium has an antibacterial effect against all tested bacteria except *Pseudomonas aeruginosa*. The major compounds of this oil are citronellol, isomenthone, menthone and citronellyl formate.

Bigos, Małgorzata, Danuta, and Sienkiewicz (2012) found in their study that the geranium oil obtained from Pelargonium graveolens showed a very strong antibacterial activity against the standard S. aureus strain (ATCC 43300) and seventy clinical S. aureus strains also obtained from the clinical materials and the chemical analysis of this oil showed that citronellol 26.7%, geraniol 13.4% representing the compounds of essential oil of geranium. The main constituents responsible for biological activity are citronellol, geraniol, linalool, isomenthone, nerol and citronellyl formate.

Renda, Celik, Korkmaz, Karagol, and Yayli (2016) evaluated the antimicrobial activity and analyses of the essential oils of six geranium l. species. The result of this study showed that the essential oils obtained by hydrodistillation were rich in benzene acetaldehyde (30%, 25.7%) (*G. asphodeloides, G. sanguineum*), caryophyllene (34.3%, 11.3%) (*G. psilostemon* and *G. robertianum*), hexadecanoic acid (36.2%, 15.1%) (*G. purpureum* and *G. pyrenaicum*).

The result of the antimicrobial activity of these oils against 10 microorganisms showed that these oils had antibacterial and antifungal activities against *staphylococcus aureus*, *bacillus cereus*, *mycobacterium smegmatis*, *candida albicans* and *saccharomyces cerevisiae*.

4. CONCLUSION

Development of resistance to chemotherapeutic agents shown by the microorganisms appears to be a continuous process since the time antibiotics were discovered. So every antibiotic has certain life span regarding its efficacy. Scientists have realized an immense potential in natural products serve as alternate source of combating infections in human beings which may also be of lower cost and lesser toxicity and side effects. The results of this study showed that the studied essential oils have an antibacterial effect against bacterial tested strains this effect was greater against Gram positive than the Gram-negative bacteria. These results demonstrated that essential oils of medicinal plants represent an economic source of natural mixtures of antibacterial compounds that can be as effective as modern medicine to combat pathogenic microorganisms and safe alternative to treat infectious diseases.

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REFERENCES

Abu-Darwish, M.S., Al-Ramamneh, E.A.M., Kyslychenko, V., Sergeevna, K. & Uliana, V. (2012). The antimicrobial activity of essential oils and extracts of some medicinal plants grown in

Ash-shoubak region – South of Jordan. *Pakistan Journal of Pharmaceutical Sciences*, 25, 239-246.

Agnihotri, V. K., Agarwal, S.G., Dhar, P.L., Thappa, R.K., Kapahi, B., Saxena, R.K. & Qazi, G.N. (2005). Essential oil composition of *Mentha pulegium* L. growing wild in the northwestern Himalayas India. *Flavour and Fragrance Journal*, 20, 607-610.

Alma, M., Ertas, M., Nitz, S. & kollmannsberger, H. (2007) Chimical composition and content of essential oil from the bud of cultivated Turkish clove (*Syzygium aromaticum L*). *Bioresources*, 2, 265-269.

Alzahrani, H.A., Boukraâ, L., Abdellah, F., Bakhotmah, B. & Hammoudi, S.M. (2014) *In vitro* synergistic action of honey and essential oils against two Species of *Aspergillus*. *British Journal of Pharmaceutical Research*, 4, 93-100.

Baydar, H., Osman, S., Ozkan, G. & Karadoan, T. (2004). Antibacterial activity and composition of essential oils from Origanum, Thymbra and Satureja species with commercial importance in Turkey. *Food Control*, 15, 169-172.

Bicchi, C., Liberto, E., Matteodo, M., Sgorbini, B., Mondello, L., Zellner, B.A. Rubiolo, P. (2008).Quantitative analysis of essential oils: a complex task. *Flavour and fragrance Journal*, 23, 382-391.

Bigos, M., Małgorzata, W., Danuta, K & Sienkiewicz, M. (2012). Antimicrobial activity of geranium oil against clinical strains of *Staphylococcus aureus*. *Molecules*, 17, 10276-10291.

Boukraâ, L., Alzahrani, H.A., Abdellah, F., Bakhotmah, B. & Hammoudi, S.M. (2013). Synergistic Effect of Monofloral Honeys and Essential Oils against *Pseudomonas aeruginosa*. *British Microbiology Research Journal*, 3,564-573.

Bowels, B.L., Sackitey, S. K. & Williams, A. C (1995). Inhibitory effects of flavour compounds on *Staphylococcus aureus* WRRCB 124. *Journal of Food Safety*, 15, 337-347.

Burst, S.A. & Reinders, R.D. (2003). Antibacterial activity of selected plant essential oils against *Escherichia coli* O157:H7. *Letters in Applied Microbiology*, 36, 162-167.

- Burt, S. (2004). Essential oils: their antibacterial properties and potential applications in foods-a review, *International Journal of Food Microbiology*, 94, 223–253.
- Carneiro, N., Alves, C. C.F., Alves, J., Egea, M. B., Martins, C H.G., Silva, T. S L C.& Miranda. M .L.D. (2017). Chemical composition, antioxidant and antibacterial activities of essential oils from leaves and flowers of *Eugenia klotzschiana* Berg (Myrtaceae). *Anais da Academia Brasileira de Ciências*, 3,1907-1915.
- Chaieb, K., Zmantar, T., Ksouri, R., Hajlaoui, H., Mahdouani, K., Abdelly, C. & Bakhrouf, A. (2007). Antioxidant properties of the essential oil of Eugenia caryophyllata and its antifungal activity against a large number of clinical Candida species. Mycoses, 50, 403–406.
- Chao, S.C., Young, D.G. & Oberg, C.J.(2000). Screening for inhibitory activity of essential oils on selected bacteria fungi and viruses. *Journal of Essential Oil Research*, 12, 639–649.
- Chaudhry Masood, N. A., Saeed, S. & Tariq, P. (2007). Antibacterial effects of oregano (*Origanum vulgare*) against Gram negative bacilli. *Pakistan Journal* of *Botany*, 39, 609-613.
- Çolak, N. U., Yıldırım, S., Bozdeveci, A., Yayli, N, . Çoşkunçelebi, K., Fandakl, S. & Yaşar, A. (2018). Essential Oil Composition, Antimicrobial and Antioxidant Activities of *Salvia staminea*. *Records of natural products*, 12, 86-94.
- Daniel, L., Daniel, P.E., Dellacassa, P., Davies, R. V. & Salvador., C. (2002). Essential oils of *Mentha pulegium* and *Mentha rotundifolia* From Uruguay. *Brazilian Archives of Biology and Technology*, 45, 519-524.
- Delaquis, P.J., Stanich, K., Girard, B. & Mazza, G. (2002). Antimicrobial activity of individual and mixed fractions of dill, cilantro, coriander and eucalyptus essential oils. *International Journal of Food Microbiology*, 74, 101-109.
- Derwich, E., Benziane, Z. & Boukir, A. (2010). GC/MS Analysis and Antibacterial Activity of the Essential Oil of *Mentha Pulegium* Grown in Morocco. *Research Journal of Agriculture and Biological Sciences*, 6, 191-198.
- Derwich, E., Benziane, Z. & Bouki, A. (2009). Chemical Composition and Antibacterial Activity of Leaves Essential Oil of *Laurus nobilis*

- from Morocco. Australian Journal of Basic and Applied Sciences, 3, 3818-3824.
- Di pasqua, R., De feo, V., Villiani, F. & Mauriello, G. (2005). *In vitro* antimicrobial activity of essential oils from mediterranean. Apiaceae, Verbenaceae and lamiaceae against foodorne pathogens and spoilage bacteria. *Annals of Microbiology*. 55, 139-143.
- Ebani, V.V., Nardoni, S., Bertelloni, F., Giovanelli, S., Ruffoni, B., C. D'Ascenzi, Mancianti, F. (2018). Activity of *Salvia dolomitica* and *Salvia somalensis* Essential Oils against Bacteria, Molds and Yeasts. *Molecules*, 23, 396.
- Fani, M.M. & Kohanteb, J. (2017). *In Vitro* Antimicrobial Activity Of *Thymus Vulgaris* Essential Oil Against Major Oral Pathogens. *Journal Of Evidence-Based Complementary & Alternative Medicine*. 22, 660-666.
- Gaunt, L.F., Higgins, S.C. & Hughes, J.F. (2005). Interaction of air ions and bactericidal vapours to control micro-organisms. *Applied Microbiology*, 99, 1324-1329.
- Hossain, S., Heo, H., De Silva, B.C.J., Wimalasena, S.H.M.P, Pathirana, H.N.K.S. & Heo, G.J. (2017). Antibacterial activity of essential oil from lavender (*Lavandula angustifolia*) against pet turtle-borne pathogenic bacteria. *Laboratory Animal Research*, 33, 195–201.
- Hui, L., He, L., Huan, L., Li, X. & AiGuo, Z. (2010). Chemical composition of lavender essential oil and its antioxidant activity and inhibition against rhinitis related bacteria. *African Journal of Microbiology Research*, 4, 309-313.
- Joy, P. P., Thomas, J., Mathew, S. & Skaria, B. P.(1998). Medicinal plants. *Kerala agricultural university Aromatic and Medicinal Plants Research Station Odakkali*, Asamannoor P.O., Ernakulam District, Kerala, India.
- Karataş, H. &Ertekin,S.(2010). Antimicrobial activities of the essential oils of four *Salvia* species from Turkey. *Journal of Medicinal Plants Research*, 4, 1238-1240.
- Khalil, R. & Zheng-Guo, L. (2011). Antimicrobial activity of essential oil of Salvia

- officinalis L. collected in Syria. *African Journal of Biotechnology*. 10, 8397-8402.
- Lopez, P., Sanchez, C., Batlle, R. & Nerin, C. (2005). Solid- and Vapor-phase antimicrobial activities of six essential oils: susceptibility of selected food borne bacterial and fungal strains. *Journal of Agricultural and Food Chemistry*. 53, 6939-6946.
- Mahmoodi, A., Roomiani, L., Afshin, M. S. Akhondzadeh, B. Kamali, A. &Taheri, S. (2012). Chemical Composition and Antibacterial Activity of Essential Oils and Extracts from *Rosmarinus officinalis*, *Zataria multiflora*, *Anethum graveolens* and *Eucalyptus globules*. *Global Veterinaria*, 9, 73-79.
- Moghtader, M. & Farahmand, A. (2013). Evaluation of the antibacterial effects of essential oil from the leaves of *Laurus nobilis* L. in Kerman Province. *Journal of Microbiology and Antimicrobials*, 5, 13-17.
- Morteza, Y., Sonboli, A., Ebrahimi, S.N. & Hashemi, S.H. (2007). Antimicrobial activity of essential oil and major constituents of *Salvia chloroleuca*. *Zeitschrift für Naturforschung*, 63, 337–340.
- Ncube, N.S., Afolayan, A.J. & Okoh, A. I. (2008). Assessment techniques of antimicrobial properties of natural compounds of plant origin: current methods and future trends. *African Journal of Biotechnology*. 7, 1797-1806.
- Oulkheir, S., Aghrouch, M., El Mourabit, F., Dalha, F., Graich, H., Amouch, F. & Chadli, S. (2017). Antibacterial Activity of Essential Oils Extracts from Cinnamon, Thyme, Clove and Geranium Against a Gram Negative and Gram Positive Pathogenic Bacteria. *Journal of Diseases and Medicinal Plants*, 31-5.
- Özkalp, B., Sevgi, F., Özcan, M. & Özcan, M. M. (2010). The antibacterial activity of essential oil of oregan (*Origanum vulgare L.*). *Journal of Food Agriculture and Environment*, 8, 272 274.
- Pelczar, M.J., Chan, E.C.S. & Krieg, N.R. (1988). Control of microorganisms, the control of microorganisms by physical agents. *In Microbiology*, 469–509.
- Pierozan, M. K., Pauletti, G.F., Rota, L. Santos, A. C. A. d. A., Lerin, M., Di luccio, .A. J Vladimiroliveira, J. (2009). Chemical

- characterization and antimicrobial activity of essential oils of salvia L. species. *Ciência e Tecnologia de Alimentos Campinas*, 29, 764-770.
- Pitarokili, D., Tzakou, O., Loukis, A. & Harvala, C. (2003). Volatile metabolites from Salvia fruticosa as antifungal agents in soilborne pathogens. *Journal of Agricultural and Food Chemistry*, 51, 3294-3301.
- Prabuseenivasan, S., Manickkam, J. & Savarimuthu, I. (2006). *In vitro* antibacterial activity of some plant essential oils *.BMC Complementary and Alternative Medicine*, 6, 39.
- Renda, G., Celik, G., Korkmaz, B., Karagol, S.A. & Yayli, N. (2016). Antimicrobial Activity and Analyses of Six *Geranium* L. Species with Headspace SPME and Hydrodistillation. *Journal of essential oil bearing plants*, 19, (8).
- Sabahat, S. & Perween. T. (2008). *In vitro* antibacterial activity of clove against gram negative bacteria, *Pakistan Journal of Botany*, 40, 2157-2160.
- Sivropoulou, A., Nikolaou, C., Papanikolaou, E., Kokkini, S., Lanaras, T. & Arsenakis, M. (1997). Antimicrobial, Cytotoxic and Antiviral Activities of Salvia fruticosa Essential Oil. *Journal of Agricultural and Food Chemistry*, 45,197-201.
- Soković, M., Glamočlija, J., Marin, P.D., Brkić, D & Griensven, L.J.L.D.V. (2010). Antibacterial effects of the essential oils of commonly consumed medicinal herbs using an *in vitro* model. *Molecules*, 15, 7532-7546.
- Stamatis, G., Kyriazopoulos, P., Goleyou, S., Basayiannis, A., Skaltsas, S. & Skaltsa, H. (2003). *In vitro* anti-Helicobacter pylori activity of Greek herbal medicines. *Journal of Ethnopharmacology*, 88, 175-179.
- Stojković, D., glamočlija, J. Ana ćirić, M., Nikolić, M., Ristić, Š.J. & Soković, M. (2013) investigation on antibacterial synergism of *Origanum vulgare* and *Thymus vulgaris* essential oils. *Archives of Biological Sciences*, 65, 639-643.
- Tohidpour, A., Sattari, M., Omidbaigi, R., Yadegar, A. & Nazemi, J. (2010). Antibacterial effect of essential oils from two medicinal plants against Methicillin-resistant *Staphylococcus aureus* (MRSA), *Phytomedicine*, 17, 142-145.