EFFECT OF ESSENTIAL OILS ON SOME PATHOGENS THAT CAUSE ECZEMA

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In this study, the antimicrobial activity of essential oils obtained from *Thymbra spicata* L., *Lavandula angustifolia* Mill. and *Myrtus communis* L. on the pathogens causing eczema *Staphylococcus aureus* (ATCC 29213), *Staphylococcus epidermidis* (ATCC 12228), *Escheria coli* (ATCC 25922), *Acinetobacter baumannii* (ATCC 43498), *Pseudomonas aeruginosa* (ATCC 27853) ve *Candida albicans* (ATCC 90028) were investigated. The MIC and MBC values of the essential oils used in the study against the pathogens causing eczema were determined. As a result of the results obtained, antimicrobial activity of plant essential oils used in the study on test microorganisms was determined. Among the essential oils, it was found that the most effective essential oil was thyme followed by the lavender.

Keywords: Essential oil, Eczema, GC-MS, Antimicrobial

INTRODUCTION

Turkey; It has a rich vegetation due to its geographical location, climate and wide area. It also contains many medicinal and aromatic plants (Faydaoğlu ve Sürücüoğlu, 2011). Medicinal and aromatic plants are used as a drug in traditional and modern medicine to prevent, cure or maintain health (Anonim, 2012). Essential oils are obtained from various parts of the plant such as flowers, buds, seeds, leaves, branches, wood, fruit and roots, and approximately 1/3 of nearly 300 plant families growing in nature contain essential oil (Anonim, 2013). Although the mechanism of action of essential oils varies according to their active ingredients, they have antimicrobial, carminative, coloretic, sedative, diuretic, antispasmodic effects (Maksimovic ve ark., 2005).

Thymbra spicata L. is widely grown in the Aegean, Mediterranean and Southeastern Anatolia Regions. In bush form, it grows up to 15-50 cm. Flowering stems are ascending or steep and sometimes very branched. *Lavandula angustifolia* Mill. is a perennial plant with an average of 50 cm in semi-shrub form, growing up to a maximum of 1 m, with grayish green leaves, blue colored and fragrant flowers (Ceylan, 1996). *Myrtus communis* L., can be generally short and rarely 1-3 m long, especially in coastal areas where the Mediterranean climate prevails (Oğur, 1994). Perennial, evergreen, bush-shaped flowers are white in color, fruits are multi-seeded, blackish purple in color (İlçim and Dığrak, 1998).

Bacteria and yeast cause some diseases in the skin. The most common of these diseases is eczema. It is a psychosomatic skin disease that occurs for various reasons and is seen with symptoms such as redness, swelling, vesicles and itching on the skin. Gram-positive bacteria such as *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Escheria coli*, *Acinetobacter baumanii*, *Pseudomonas aeruginosa* and *Candida albicans* pathogens cause eczema. While *S. aureus* is common in nature, *S. epidermidis* disease is found in many parts of the body, especially human skin and upper respiratory mucosa (Gülbandılar, 2009; Eryılmaz ve Gürpınar, 2017). While *P. aeruginosa* is found in nature, *A. baumanii* is found in hospital environment, *E. coli* are common bacteria found in intestines (Enoch et al., 2009; Torlak, 2011). *C. albicans*, a yeast type fungus, is found in various parts of the body (Acarkan, 2014).

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Antibiotics are widely used in the treatment of eczema. The progressive increase in side effects caused by the use of synthetic origin substances in the treatment of diseases, the resistance of organisms against synthetic substances, and the gradual restriction of synthetic drugs have led people to seek alternative solutions in the treatment of the disease.

In this study, the effects of *T. spicata* L., *L. angustifolia* Mill. and *M. communis* L. essential oils on some pathogens (*S. aureus*, *S. epidermidis*, *E. coli*, *A. baumanii*, *P. aeruginosa* and *C. albicans*) causing eczema were investigated in vitro.

MATERIALS AND METHODS

T. spicata, *L. angustifolia* and *M. communis* essential oils were used as material in the study. The antimicrobial activities of these essential oils against *S. aureus* (ATCC 29213), *S. epidermidis* (ATCC 12228), *E. coli* (ATCC 25922), *A. baumannii* (ATCC 43498), *P. aeruginosa* (ATCC 27853) and *C. albicans* (ATCC 90028). pathogens were investigated. Antimicrobial activity studies were conducted in Hatay Mustafa Kemal University. Activity studies of essential oils were carried out by tube dilution method. The group without essential oil was selected as the negative control, and gentamicin and fluconazole were used as the drug control. Dimethyl sulfoxide (DMSO) of 1% was used to dissolve essential oils in the medium. Mueller-Hinton Broth was used for bacteria in the tube dilution method for the production of microorganisms and Saboraud Dextroz Broth for *C. albicans*. It has been determined by controlled experiments that 1% concentration of DMSO is non-toxic on the growth of microorganisms.

Obtaining Essential Oils

The essential oils used in the study were obtained from the leaves of *T. spicata* and *M. communis* plants naturally found in the flora of Hatay. And *L. angustifolia* essential oil was collected from plants that were previously cultivated in Hatay Mustafa Kemal University in full blooming period and obtained by water distillation method from leaves and herbs of plants.

Determination of Essential Oil Components

The determination of essential oil components was carried out under the following conditions with the Thermo Scientific ISQ Single Quadrupole model GC device. TR-FAME MS model, 60 m length column was used. Helium (99.9%) was used as the carrier gas at a flow rate of 1 mL/min. Ionization 22 energy was set at 70 eV, mass range m/z 1.2-1200 amu. Scan Mode was used for data collection. The MS transfer line temperature is 250 °C, the MS ionization temperature is 220 °C, the injection port temperature is 220 °C, the column temperature is 50 °C at the beginning and has been increased to 220 °C with a temperature increase rate of 3 °C/min. The structure of each compound was identified with the Xcalibur program using mass spectra (Wiley 9).

Antimicrobial Activity Tests

Microorganism colonies taken with the loop were suspended in Phosphate Buffered Saline (PBS), which is a phosphate buffer solution. $1x10^8$ bacteria/ml compared to Mc Farland turbidity tube no 0.5; *C. albicans* dilution was prepared to be $1x10^5$ and these

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dilutions were used as inoculum. The determination of the antimicrobial activities of essential oils was evaluated in accordance with the National Committee for Clinical Laboratory Standards (NCCLS) criteria. Tested bacterial and fungal microbial strains were suspended in PBS (phosphate buffered water) to McFarland 0.5, and those containing bacterial strains Mueller-Hinton agar and Sabouraud Dextrose agar for *C. albicans* were inoculated on plates. In the study, dimethyl sulfoxide (DMSO) was used as a solvent to dissolve essential oils. Non-toxic concentration (1%) of the selected concentration of DMSO on microorganisms was used. The concentrations of the essential oils tested in the study (10.24, 5.12, 2.56, 1.28, 0.64, 0.32, 0.16, 0.08, 0.04 and 0.02 µg/ml) were used. DMSO was used as negative control. Amikacin, gentamicin, and nystatin were used as reference drugs for gram-positive anti-bacterial activity, gram-negative anti-bacterial activity, and antifungal activity, respectively. All microorganism plates were incubated at 37 °C and the results were evaluated after 24th hour of incubation for bacteria and after 48th hour of incubation for *C. albicans*. Essential oil concentrations that inhibit apparent growth were considered to be minimum inhibitory concentrations (MICs). In addition, minimal bactericidal and fungicidal concentrations were determined by seeding on Mueller Hinton agar and Saboraud Dexroz agar from the next dilutions with the final concentration without visible growth.

RESULTS AND DISCUSSION

Essential Oil Rates and Components of Plants Used in the Study

According to the results obtained, thyme essential oil ratio was 3.00%, lavender essential oil ratio was 2.90%, and the essential oil ratio obtained from myrtle leaves was 1.25%. When we examine the essential oil components of the thyme, the highest component was determined as Carvacrol with 55.30%, followed by o-Cymene with 13.51% and-Terpinene with 12.30%. When the components of the essential oils of the lavender were examined, it was found that the highest component was Linalool with 18.03%, followed by α-Bisabolol with 17.44% and Linalyl acetate with 8.76%. When we examine the main components of the essential oils of the myrtle, the essential oil components were determined as 33.80% Eucalyptol, 25.42% α-Pinene and 10.75% Linalool, respectively.

Effectiveness of Essential Oils on *S. aureus*

The effectiveness of thyme, lavender and myrtle essential oils on *S. aureus* bacteria was examined in terms of MIC values, it was determined that thyme essential oil inhibited bacterial growth at 0.02 µg/ml, lavender essential oil at 0.32 µg/ml, and myrtle essential oil at 0.64 µg/ml. When the effectiveness of thyme, lavender, myrtle essential oils on *S. aureus* bacteria was examined in terms of MBK values, it was determined that thyme essential oil showed a bactericidal effect at 0.04 µg/ml, lavender essential oil at 0.64 µg/ml, and myrtle essential oil at 1.28 µg/ml.

Effectiveness of Essential Oils on *S. epidermidis*

The effectiveness of thyme, lavender and myrtle essential oils on *S. epidermidis* was examined in terms of MIC values, 0.02 µg/ml of thyme essential oil, 0.16 of lavender essential oil. It has been determined that µg/ml, myrtle essential oil inhibits bacterial growth at 0.32 µg/ml. When the effectiveness of thyme, lavender and myrtle essential

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oils on *S. epidermidis* was examined in terms of MBC values, it was determined that thyme essential oil showed a bactericidal effect at 0.04 µg/ml, lavender essential oil at 0.32 µg/ml, and myrtle essential oil at 0.64 µg/ml.

Table 1. Efficacy concentrations of essential oils against *S. aureus*

Essential Oil Concentration (µl/ml)	*T. spicata*		*L. angustifolia*		*M. communis*	
	MIC	MBC	MIC	MBC	MIC	MBC
0.01	+	+	+	+	+	+
0.02	-	+	+	+	+	+
0.04	-	-	+	+	+	+
0.08	-	-	+	+	+	+
0.16	-	-	+	+	+	+
0.32	-	-	-	+	+	+
0.64	-	-	-	-	-	+
1.28	-	-	-	-	-	-

+: There is reproduction, -: There is no reproduction

Table 2. Efficacy concentrations of essential oils against *S. epidermidis*

Essential Oil Concentration (µl/ml)	*T. spicata*		*L. angustifolia*		*M. communis*	
	MIC	MBC	MIC	MBC	MIC	MBC
0.01	+	+	+	+	+	+
0.02	-	+	+	+	+	+
0.04	-	-	+	+	+	+
0.08	-	-	+	+	+	+
0.16	-	-	-	+	+	+
0.32	-	-	-	-	-	+
0,64	-	-	-	-	-	-

+: There is reproduction, -: There is no reproduction

Effectiveness of Essential Oils on *E. coli*

The effectiveness of thyme, lavender and myrtle essential oils on *E. coli* was examined in terms of MIC values, it was determined that thyme and lavender essential oils inhibit bacterial growth at 0.32 µg/ml and myrtle essential oil at 0.64 µg/ml. When the effectiveness of thyme, lavender and myrtle essential oils on *E. coli* was examined in terms of MBC values, it was determined that thyme and lavender essential oils showed a bactericidal effect at 0.64 µg/ml and myrtle essential oil at 1.28 µg/ml.

Effectiveness of Essential Oils on *A. baumannii*

The efficiency of thyme, lavender and myrtle essential oils on *A. baumannii* was examined in terms of MIC values, it was determined that thyme essential oil inhibits bacterial growth at 0.02 µg/ml, lavender essential oil at 0.32 µg/ml, and myrtle essential oil at 0.64 µg/ml. When the effectiveness of thyme, lavender and myrtle essential oils on *A. baumannii* was examined in terms of MBK values, it was determined that thyme essential oil had a bactericidal effect at 0.04 µg/ml, lavender essential oil at 0.64 µg/ml, and myrtle essential oil at 1.28 µg/ml.

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Table 3. Efficacy concentrations of essential oils against *E. coli*

Essential Oil Concentration (µl/ml)	*T. spicata*		*L. angustifolia*		*M. communis*	
	MIC	MBC	MIC	MBC	MIC	MBC
0.01	+	+	+	+	+	+
0.02	+	+	+	+	+	+
0.04	+	+	+	+	+	+
0.08	+	+	+	+	+	+
0.16	+	+	+	+	+	+
0.32	-	+	-	+	+	+
0.64	-	-	-	-	-	+
1.28	-	-	-	-	-	-

+: There is reproduction, -: There is no reproduction

Table 4. Efficacy concentrations of essential oils against *A. baumannii*

Essential Oil Concentration (µl/ml)	*T. spicata*		*L. angustifolia*		*M. communis*	
	MIC	MBC	MIC	MBC	MIC	MBC
0.01	+	+	+	+	+	+
0.02	+	+	+	+	+	+
0.04	+	+	+	+	+	+
0.08	+	+	+	+	+	+
0.16	+	+	+	+	+	+
0.32	-	+	-	+	+	+
0.64	-	-	-	-	-	+
1.28	-	-	-	-	-	-

+: There is reproduction, -: There is no reproduction

Effectiveness of Essential Oils on *P. aeruginosa*

The effectiveness of thyme, lavender and myrtle essential oils on *P. aeruginosa* was examined in terms of MIC values; It was determined that thyme essential oil inhibits bacterial growth at 0.32 µg/ml, lavender essential oil at 10.24 µg/ml, and myrtle essential oil at 5.12 µg/ml. When thyme, lavender and myrtle communis essential oils were examined in terms of MBC values on *P. aeruginosa*, it was determined that thyme essential oil had a bactericidal effect at 0.64 µg/ml, lavender essential oil at 20.48 µg/ml, and myrtle essential oil at 10.24 µg/ml.

Effectiveness of Essential Oils on *C. albicans*

The effectiveness of thyme, lavender and myrtle essential oils on *C. albicans* was examined in terms of MIC values, it was determined that thyme essential oil inhibits fungus growth at 0.04 µg/ml, lavender essential oil at 2.56 µg/ml and myrtle essential oil at 0.16 µg/ml. When thyme, lavender and myrtle essential oils were examined in terms of MFC values on *C. albicans*, it was determined that thyme essential oil had a fungicidal effect at 0.04 µg/ml, lavender essential oil at 5.12 µg/ml, and myrtle essential oil at 0.32 µg/ml.

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Table 5. Efficacy concentrations of essential oils against *P. aeruginosa*

Essential Oil Concentration (µl/ml)	*T. spicata*		*L. angustifolia*		*M. communis*	
	MIC	MBC	MIC	MBC	MIC	MBC
0.16	+	+	+	+	+	+
0.32	+	+	+	+	+	+
0.64	+	+	+	+	+	+
1.28	+	+	+	+	+	+
2.56	+	+	+	+	+	+
5.12	-	+	+	+	-	+
10.24	-	-	-	+	-	-
20.48	-	-	-	-	-	-

+: There is reproduction, -: There is no reproduction

Table 6. Efficacy concentrations of essential oils against *C. albicans*

Essential Oil Concentration (µl/ml)	*T. spicata*		*L. angustifolia*		*M. communis*	
	MIC	MFC	MIC	MFC	MIC	MFC
0.01	+	+	+	+	+	+
0.02	+	+	+	+	+	+
0.04	-	-	+	+	+	+
0.08	-	-	+	+	+	+
0.16	-	-	+	+	-	+
0.32	-	-	+	+	-	-
0.64	-	-	+	+	-	-
1.28	-	-	+	+	-	-
2.56	-	-	-	+	-	-
5.12	-	-	-	-	-	-

+: There is reproduction, -: There is no reproduction

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