Saudi Journal of Biological Sciences 28 (2021) 6145–6152

Contents lists available at ScienceDirect

Saudi Journal of Biological Sciences

journal homepage: www.sciencedirect.com

Original article

Chromatographic profiles and antimicrobial activity of the essential oils obtained from some species and cultivars of the *Mentheae* tribe (*Lamiaceae*)

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ARTICLE INFO

Article history:
Received 19 January 2021
Revised 21 June 2021
Accepted 22 June 2021
Available online 26 June 2021

Keywords:
Monarda didyma
Salvia sclarea
Thymus genus
Cultivar
Essential oil
GC–MS
Antimicrobial effect

ABSTRACT

The present study was focused on the chemical composition and antimicrobial activity of the essential oils (EsO) obtained from five *Lamiaceae* representatives grown in the south of Ukraine. Among them are *Salvia sclarea* L., *Monarda didyma* (cultivar 'Cambridge Scarlet'), *Thymus pulegioides* (cultivar '2/6-07'), *Thymus vulgaris* (cultivar 'Jalos'), and *Thymus serpyllum* L. The component analysis of the EsO was carried out by gas chromatography method coupled with mass spectrometry (GC–MS). The antimicrobial properties of the EsO were determined using the agar diffusion test against widespread pathogenic bacterial strains (*Staphylococcus aureus, Escherichia coli, Enterococcus faecalis, Streptococcus pyogenes*) and opportunistic yeast *Candida albicans*. The EsO of *Thymus serpyllum* and *Thymus vulgaris* (cultivar 'Jalos') displayed noteworthy antibacterial properties against a wide spectrum of the microorganisms. These antimicrobial properties could be attributed to the high content of aromatic monoterpenoid thymol (52.56% and 47.33%, respectively). The EsO of *Salvia sclarea* with the dominance of linalyl acetate (45.51%) and linalool (38.98%) as well as *Thymus pulegioides* (cultivar '2/6-07') containing α-citral (27.10%) and β-citral (17.11%) demonstrated the strongest antimicrobial effects on typical and clinical strains of *Staphylococcus aureus* with the inhibition zones in the range of 24.0–31.0 mm. The *Salvia sclarea* EsO demonstrated the most significant effect against clinical strains of *Candida albicans*. In conclusion, the present study revealed the chemical composition of five *Lamiaceae* species and cultivars grown in the south of Ukraine and considerable antimicrobial activity of the tested EsO, especially against the typical and clinical strains of *Staphylococcus aureus* and *Candida albicans*. The obtained results could be perspective for applying in the pharmaceutical industry and for the conservation of food and cosmetic products.

1. Introduction

Natural substances derived from medicinal plants represent a rich source for the development of new herbal medicinal products. Plant-derived compounds could be appropriate alternatives to synthetic active substances for the treatment of many diseases (Moghtader, 2012). EsO are considered as an alternative to synthetic antimicrobial medicines, especially antibiotics (Sharifi-Rad et al., 2017; Tariq et al., 2019). The ability of EsO to inhibit the growth of undesirable microorganisms in cosmetic and food products could solve the problem of the substitution for synthetic

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Peer review under responsibility of King Saud University.

Production and hosting by Elsevier

https://doi.org/10.1016/j.sjbs.2021.06.068
1319-562X/

preservatives which can cause serious problems for human health (Liu et al., 2017; Long et al., 2019; Park et al., 2012).

Among the different groups of Angiosperms, *Lamiaceae* Martinov family is a large taxonomical unit of plants. This family is unofficially divided into two groups according to the phytochemical peculiarities: the first group mainly produces EsO, while the second one biosynthesize components of the polar fraction (di- and triterpenoids, iridoids and polyphenols) (Marchioni, 2020). Such genera as *Thymus* L., *Salvia* L., and *Monarda* L. belong to the *Mentheae* Dumort. tribe of the *Nepetoideae* Burnett subfamily of the *Lamiaceae* family. They are characterized by the distinctive aromatic smell due to the accumulation of EsO in epidermal glandular trichoms (Lange and Turner, 2013). The representatives of these genera are important medicinal plants widely used in traditional and complementary medicine. The *Thymus* species are native to temperate regions in Europe, North Africa and Asia where they are used mainly to cure bronchitis and oral cavity infections (Jarić et al., 2015; Mohammadi et al., 2020; Mostafa et al., 2018). Representatives of the *Salvia* genus are widely spread in temperate and subtropical regions on the different continents and are often used for treating the illnesses related to the gastrointestinal and respiratory systems (Cui et al., 2015; Hudz et al., 2019; Koshovyi et al., 2020). The *Monarda* species are native to North America where they are widely used in folk medicine for the treatment of flu, cold, skin wounds, etc. (Fraternale, et al., 2006; Mattarelli et al., 2017). Many species from the abovementioned genera are widely cultivated throughout the world. The plant raw materials of such species as *Thymus serpyllum* (*Serpylli* herba) and *Thymus vulgaris* (*Thymi* herba) as well as *Salviae sclareae* aetheroleum are included to the 10th Edition of the European Pharmacopoeia (2020). The appearance of the new cultivars and chemotypes encourage the studies of their chemical composition and biological activity (Myadelets et al., 2014, Hudz et al., 2019; Hudz et al., 2020).

The comparative analysis of EsO and antimicrobial activity of promising species from the above-mentioned genera is of considerable scientific interest to researchers in the field of botany, plant physiology, microbiology, pharmacognosy and pharmaceutical technology. A different number of original papers and reviews about the EsO composition and activities of the tested species were published up to 15 December 2020 (data retrieved from PubMed via searching the words 'latin name of plant' and 'essential oil'): '*Monarda didyma* essential oil' – 4 results, '*Thymus serpyllum* essential oil' – 31, '*Thymus pulegioides* essential oil' – 1, '*Thymus vulgaris* essential oil' – 779, and '*Salvia sclarea* essential oil' – 47. As can be seen from these data, the *Thymus vulgaris* EsO is the most tested by different researchers. However, the existence of many cultivars and chemotypes as well as the influence of climatic conditions on the component composition of the EsO of *Lamiaceae* plants (Raafat and Habib, 2018; Vaičiulytė et al., 2016) encourages to investigate their composition and antimicrobial potential.

Searching for new antimicrobial compounds from natural resources became a necessity because antibacterial resistance is nowadays a serious threat to public health (Kot et al., 2019; Mulyaningsih et al., 2011; Tariq et al., 2019). Historically, discovering antibiotics led to the big management of infectious diseases worldwide through decreasing morbidity and mortality but it caused the development of many drug-resistant pathogens (Tariq et al., 2019). Antimicrobial effects of EsO as the complex mixtures of volatile organic compounds are due to the synergistic effect of their components (Jarić et al., 2015). EsO possess huge advantages over purified components. Among them is a low resistance of pathogenic microorganisms (Tariq et al., 2019). The EsO which are characterized by dominating aromatic compounds such as thymol or carvacrol can destroy the cellular architecture of pathogenic bacteria, leading to the rupture of membrane and leakage of ions and even molecules (Tariq et al., 2019). Using EsO against phytopathogens is very perspective because of their potential to save food without hazards for human health and the environment (Adebayo et al., 2013). As antimicrobial activity of EsO are exerted by their constituents, it is necessary to know their composition to foresee a possible role in the inhibition of pathogenic bacterial and fungi growth.

The primary aim of this paper was to study the chemical composition of EsO obtained from five *Lamiaceae* species (*Salvia sclarea* L., *Monarda didyma* (cultivar 'Cambridge Scarlet'), *Thymus pulegioides* (cultivar 2/6–07), *Thymus vulgaris* (cultivar 'Jalos'), *Thymus serpyllum* L.) cultivated in Ukraine. The secondary aim was to reveal the antimicrobial potential the tested EsO against the typical and clinical strains of pathogenic microorganisms.

2. Materials and methods

2.1. Plant material

The aerial parts of *Monarda didyma* (cultivar 'Cambridge Scarlet') (CCS), *Salvia sclarea, Thymus pulegioides* (cultivar '2/6-07') (C2), *Thymus vulgaris* (cultivar 'Jalos') (CJ), and *Thymus serpyllum* were collected in the flowering stage in Kherson region (Ukraine) in 2016. The voucher specimens were deposited at the Herbarium of the Sector of Mobilization and Conservation of Plant Resources of the Rice Institute of the NAAS (Plodove, Kherson region, Ukraine) and the Department of Analytical Chemistry of Opole University (Poland).

2.2. Essential oil isolation procedure

The EsO were obtained via hydrodistillation for 4 h with a Clevenger-type apparatus according to European Pharmacopoeia (2020) using fresh raw materials. The oils were kept at 4 °C in sealed amber bottles before analysis by GC–MS and antimicrobial studies.

2.3. General experimental procedures of GC–MS

The Hewlett Packard HP 6890 series GC system chromatograph (Hewlett Packard, Waldbronn, Germany) was employed for this study. It was coupled with the Hewlett Packard 5973 mass selective detector (Hewlett Packard, Waldbronn, Germany). The chromatograph was equipped with the non-polar, high-temperature ZB-5HT (5% diphenyl- and 95% dimethylpolysiloxane) capillary column Phenomenex (Torrance, CA, USA) of length of 30 m with inner diameter of 0.32 mm and film thickness of 0.25 μm. The split ratio was 20:1 and 1 μL of a sample was introduced. Helium was a carrier gas, its flow rate was 2 mL/min. The analyses were performed at the temperature range of 40–280 °C and the heating rate was 10 °C/min; injector temperature was 250 °C (Hudz et al., 2020).

The volatile compounds of the tested EsO were identified by comparing the mass spectra data with spectrometer database of the NIST 11 Library and their retention index calculated against *n*-alkanes (C_9–C_{20}). Each chromatographic analysis was repeated three times. The average value of the relative composition of the EsO percentage was calculated from the peak areas.

2.4. Antimicrobial assay

Antimicrobial activity was determined using the agar diffusion test (Kryvtsova et al., 2019). The bacterium inocula in the physiological solution were adjusted to the equivalent of 0.5 McFarland standard. Then 100 μL of bacteria inoculates were evenly spread on the surface of Muller-Hinton agar (incubated at a temperature

of 37 ± 2 °C for 24 h). 100 μL of the yeasts inoculates were spread on SDA agar (incubated at 35 ± 2 °C for 48 h). 10 μL of the EsO were introduced into wells (6 mm in diameter). The diameters of the inhibition zones were measured in millimeters including the well diameter.

The following bacteria and yeasts from the American Type Culture Collection (ATTC) were used: *Candida albicans* ATCC 885–653; *Staphylococcus aureus* ATCC 25923; *Escherichia coli* ATCC 25922; *Enterococcus faecalis* ATCC 29212; *Streptococcus pyogenes* ATCC 19615; reference *Staphylococcus aureus CCM 3953* biofilm-forming strain. The clinical strains of bacteria and yeasts (*Staphylococcus aureus, Escherichia coli, Streptococcus pyogenes*, *Enterococcus faecalis*, *Candida albicans*) isolated from the oral cavities of patients suffering from inflammatory periodontium and pharynx. We identified the bacteria and fungi based on macromorphology, micromorphology, physiological and biochemical tests using ENTEROtest, STREPTO-test, STAPHYLO-test produced by Erba Lachema. The clinical strains with multiple resistances at least to two classes of antibiotics were chosen. The following antibiotics were used as positive controls: gentamicin (10 mcg/disk); ampicilin (10 mcg/disk), amoxicillin/clavulanic acid (20/10 mcg/disk); nystatin (100 UI) and itraconazol (30 mcg/disk) for *Candida* strains. Dimethyl sulfoxide (DMSO) was used as a negative control.

Each antimicrobial assay was performed at least three times. The obtained data were expressed as mean ± standard deviation (x ± SD) of three measurements. The Tukey's test was applied for comparisons of means; differences were considered significant at $p < 0.05$.

3. Results

3.1. GC–MS analysis of essential oil

GC–MS analysis of the tested EsO revealed their component compositions (Table 1).

As can be seen from the obtained results, thymol was the predominant component of the EsO isolated from the *Thymus vulgaris* CJ (47.33%), *Thymus serpyllum* (52.56%) and *Monarda didyma* CCS (47.54%) herbs. *o*-Cymene (14.34%) and γ-terpinene (11.57%) were the major components of *Thymus vulgaris* CJ besides thymol. Isothymol methylether (11.28%) was the important component of *Thymus serpyllum* in addition to thymol. α-Citral (27.10%) and β-citral (17.11%) were the major components of *Thymus pulegioides* C2 EsO.

Eucalyptol (17.82%) was the second predominant component of the EsO *of Monarda didyma* CCS besides thymol. The tested *Salvia sclarea* EsO was characterized by the dominance of linalyl acetate (45.51%) and linalool (38.98%).

It should be noted that thymol, *o*-cymene, linalool, caryophyllene, terpinen-4-ol, α-terpineol, and 1-octen-3-ol are the common compounds of the EsO of all the investigated species. Some of minor compounds can be regarded as the specific markers of the investigated species, for instance, aromadendrene (0.12%) was found only in *Thymus serpyllum* EsO, pinocarveol (0.11%) in *Thymus pulegioides* C2, whilst β-thujene (0.60%), 3-carene (0.10%) and many others were found only in the EsO of *Thymus vulgaris* CJ.

Table 1
Chemical composition of the EsO of five *Lamiaceae* species determined by GC–MS.

Component	IR	Area [%] ± SD				
		Thymus vulgaris (cultivar 'Jalos')	*Thymus pulegioides* (cultivar '2/6-07')	*Thymus serpyllum*	*Salvia sclarea*	*Monarda didyma* (cultivar 'Cambridge Scarlet')
β-thujene	929	0.60 ± 0.02	–	–	–	–
α-pinene	935	0.32 ± 0.03	–	–	–	–
camphene	950	0.39 ± 0.01	–	–	–	–
β-phellandrene	975	0.09 ± 0.01	–	–	–	–
β-pinene	978	0.20 ± 0.01	–	–	–	–
1-octen-3-ol	981–982	0.68 ± 0.02	0.37 ± 0.02	2.46 ± 0.14	0.03 ± 0.00	1.99 ± 0.08
3-octanone	987–988	0.03 ± 0.00	0.10 ± 0.01	0.12 ± 0.04	–	0.17 ± 0.04
β-myrcene	991–992	1.37 ± 0.04	0.07 ± 0.01	0.43 ± 0.05	0.27 ± 0.02	
3-octanol	995	0.11 ± 0.00	0.16 ± 0.02	0.17 ± 0.03	–	0.87 ± 0.03
α-phellandrene	1003–1004	0.16 ± 0.01	–	0.09 ± 0.02	–	–
3-carene	1010	0.10 ± 0.00	–	–	–	–
terpinolene	1017	1.02 ± 0.02	–	0.58 ± 0.02	–	–
o-cymene	1025–1029	14.34 ± 0.21	0.40 ± 0.02	4.50 ± 0.27	0.08 ± 0.01	0.34 ± 0.01
β-terpinene	1030	–	–	0.22 ± 0.08	–	–
limonene	1030	–	–	–	0.13 ± 0.01	–
eucalyptol	1032–1033	2.65 ± 0.03	0.31 ± 0.02	0.44 ± 0.01	–	17.82 ± 0.40
trans-β-ocimene	1040–1041	–	–	0.06 ± 0.01	0.03 ± 0.00	–
β-ocimene	1050–1051	0.06 ± 0.01	–	0.06 ± 0.00	0.02 ± 0.00	–
γ-terpinene	1060–1063	11.57 ± 0.09	0.20 ± 0.00	3.15 ± 0.26	–	0.47 ± 0.00
cis-β-terpineol	1070	1.07 ± 0.17	0.07 ± 0.00	0.87 ± 0.04	–	2.03 ± 0.02
cis-linalol oxide	1074	–	0.52 ± 0.04	–	0.41 ± 0.02	–
1-nonen-3-ol	1082	–	–	0.12 ± 0.03	–	0.03 ± 0.01
α-terpinolene	1089	–	0.12 ± 0.00	0.09 ± 0.00	–	0.03 ± 0.00
trans-linalol oxide	1090	0.14 ± 0.02	0.51 ± 0.03	–	0.42 ± 0.02	–
trans-β-terpineol	1094–1097	0.20 ± 0.01	–	0.12 ± 0.02	–	0.61 ± 0.01
linalool	1100	2.85 ± 0.05	4.84 ± 0.21	1.20 ± 0.02	38.98 ± 0.60	0.42 ± 0.01
thujone	1114	0.08 ± 0.00	0.07 ± 0.01	–	–	–
trans-*p*-menth-2-en-1-ol	1124	0.06 ± 0.02	–	0.05 ± 0.02	–	0.09 ± 0.00
pinocarveol	1141	–	0.11 ± 0.01	–	–	–
2-bornanon	1152	–	0.44 ± 0.03	–	–	–
camphor	1153	1.19 ± 0.06	0.22 ± 0.01	–	–	0.08 ± 0.00
verbenol	1154	–	0.11 ± 0.01	–	–	–
p-menthone	1156	–	–	0.09 ± 0.01	–	–
cis-verbenol	1157	–	0.16 ± 0.02	–	–	–
endo-borneol	1169	2.06 ± 0.70	0.22 ± 0.01	0.13 ± 0.02	–	0.13 ± 0.01

(*continued on next page*)

Table 1 (*continued*)

Component	IR	Area [%] ± SD				
		Thymus vulgaris (cultivar 'Jalos')	*Thymus pulegioides* (cultivar '2/6-07')	*Thymus serpyllum*	*Salvia sclarea*	*Monarda didyma* (cultivar 'Cambridge Scarlet')
menthol	1176	–	–	0.06 ± 0.02	–	
terpinen-4-ol	1180	1.04 ± 0.05	0.92 ± 0.05	1.26 ± 0.03	0.10 ± 0.00	1.18 ± 0.02
α-terpineol	1192–1194	0.34 ± 0.02	0.56 ± 0.02	0.23 ± 0.01	5.85 ± 0.09	8.26 ± 0.21
trans-*p*-menth-8-en-2-one	1204	–	–	0.03 ± 0.01	–	–
nerol (*cis*-geraniol)	1233	–	2.93 ± 0.18	–	0.62 ± 0.04	–
isothymol methyl ether	1238	0.07 ± 0.01	–	11.28 ± 0.31	–	5.85 ± 0.10
thymol methyl ether	1246–1247	1.32 ± 0.06	1.16 ± 0.21	4.15 ± 0.01	–	1.06 ± 0.07
β-citral	1247	–	17.11 ± 0.22	–	–	–
thymoquinone	1258	–	–	0.15 ± 0.06	–	0.15 ± 0.03
linalyl acetate	1264–1265	–	–	0.08 ± 0.01	45.51 ± 0.45	–
geraniol	1261	–	0.12 ± 0.02	–	–	–
α-citral	1274	–	27.10 ± 0.17	–	0.12 ± 0.02	–
geraniol acetate	1288	–	0.98 ± 0.04	–	–	–
carvenone	1286	–	–	0.03 ± 0.02	–	–
bornyl acetate	1287	0.09 ± 0.01	–	–	–	0.05 ± 0.01
carvacrol	1290	–	–	0.13 ± 0.04	–	–
thymol	1299–1305	47.33 ± 0.02	2.48 ± 0.15	52.56 ± 0.28	0.45 ± 0.08	47.54 ± 0.16
p-thymol	1309–1310	4.67 ± 0.23	–	4.01 ± 0.49	–	4.82 ± 0.46
geranyl formate	1321	–	1.07 ± 0.15	–	–	–
thymol acetate	1357	–	–	0.16 ± 0.02	–	0.21 ± 0.01
nerol acetate	1366	–	1.23 ± 0.24		1.80 ± 0.05	–
α-copaene	1379	–	–	0.05 ± 0.01	–	–
α-cubebene	1379	–	–	–	0.30 ± 0.03	–
geranyl acetate	1385	–	6.24 ± 0.57	–	3.11 ± 0.02	–
β-bourbonene	1387–1388	–	–	0.11 ± 0.01	0.15 ± 0.01	0.21 ± 0.01
caryophyllene	1419–1424	2.23 ± 0.06	0.18 ± 0.00	3.13 ± 0.20	0.60 ± 0.01	1.08 ± 0.02
β-cubebene	1433	–	–	0.08 ± 0.01	–	0.09 ± 0.00
aromadendrene	1443	–	–	0.12 ± 0.01	–	–
humulene	1458	0.08 ± 0.00	–	0.18 ± 0.01	–	0.06 ± 0.00
alloaromadendrene	1465	–	0.19 ± 0.00	–	–	–
cis-β-farnesene	1465	–	–	0.15 ± 0.01	–	–
γ-muurolene	1480	0.03 ± 0.00	–	0.15 ± 0.05	–	0.21 ± 0.00
germacrene D	1484	0.04 ± 0.00	–	0.76 ± 0.06	–	2.05 ± 0.06
β-selinene	1495	–	–	0.04 ± 0.00	–	–
γ-elemene	1499	–	0.66 ± 0.05	0.32 ± 0.03	–	–
α-muurolene	1502	–	–	0.04 ± 0.00	–	–
β-bisabolene	1511	–	1.31 ± 0.08	4.94 ± 0.12	–	–
γ-cadinene	1517	0.03 ± 0.00	–	–	–	0.15 ± 0.00
δ-cadinene	1527	0.08 ± 0.00	–	0.34 ± 0.02	–	0.25 ± 0.01
spathulenol	1584	–	–	0.28 ± 0.05	–	
caryophyllene oxide	1589	–	6.19 ± 0.22	0.56 ± 0.05	–	0.14 ± 0.04

3.2. Antimicrobial assay of the essential oils

The results of the antimicrobial activities study of the EsO of five *Lamiaceae* species against typical and clinical strains of pathogenic microorganisms are presented in Table 2.

As can be seen from the results of antimicrobial activity studies, the EsO of all the analyzed species possess the marked antimicrobial properties against the typical and clinical strains of gram-positive (*Staphylococcus aureus, Streptococcus pyogenes, Enterococcus faecalis)* and gram-negative (*Escherichia coli)* pathogenic bacteria as well as yeast *Candida albicans*. It should be noted that the clinical isolates of microorganisms used in this study were insensitive to such antibiotics as ampicillin and gentamicin, and clinical strains of *Candida albicans* were insensitive to antifungal drugs nystatin and itraconazol.

The highest antimicrobial activity against both the typical and clinical strains of *S. aureus* was detected for the EsO of *Thymus vulgaris* CJ, *Thymus serpyllum* and *Salvia sclarea*. The other EsO showed slightly lower antimicrobial activity relative to these test cultures. The EsO of *Monarda didyma* CSS was very active against both the typical and clinical strains of *Staphylococcus aureus. Thymus vulgaris* CJ and *Thymus serpyllum* EsO significantly inhibited the *Escherichia coli* growth. *Thymus serpyllum* EsO was quite active against *Streptococcus pyogenes.* The highest anti-candidal effect was detected for the EsO of *Thymus vulgaris* CJ, *Thymus serpyllum* and *Salvia sclarea*. Generally, all the tested EsO are characterized by the presence of antifungal activity against *Candida albicans* as the growth inhibition zones were greater than 20.0 mm for most of them. Thus, *Thymus serpyllum* and *Thymus vulgaris* CJ EsO were found to possess the antimicrobial activity.

4. Discussion

It was found that thymol and its isomers (*p*-thymol, carvacrol) were the predominant compound of three tested EsO (*Thymus vulgaris* CJ, *Thymus serpyllum* and *Monarda didyma* CCS) as well as many other *Nepetoideae* species of the *Lamiaceae* family (Hudz et al., 2020; Moghtader, 2012; Nutrizio et al., 2020; Vaičiulytė, et al., 2016). Predominant compounds can be considered as biologically active components of EsO while the minor ones can discriminate species, subspecies, cultivars or chemotypes.

There is no doubt that it is very important to know the limit content of some bioactive compounds in EsO with undesired properties. The main components of the tested EsO do not possess any toxic properties like psychoactive neoclerodane diterpene salvinorin A of *Salvia divinorum* (Siebert, 2004). The content of the toxic component thujone in the *Salvia sclarea* EsO oil is regulated by the EU (maximum 0.2% in the EsO) (European Pharmacopoeia, 2020). As can be seen from the Table 1, thujone was presented in the tested EsO in the very small quantities: 0.08% in *Thymus vulgaris* CJ and 0.07% in *Thymus pulegioides* C2.

Table 2
Antimicrobial activities of the tested EsO against the typical and clinical strains of pathogenic microorganisms.

Test culture	*Thymus vulgaris* (cultivar 'Jalos'	*Thymus pulegioides* (cultivar '2/6-07')	*Thymus serpyllum*	*Salvia sclarea*	*Monarda didyma* (cultivar 'Cambridge Scarlet')	*AMP*	*AMO*	*GEN*	*NIS*	*ITR*
S. aureus ATCC 25923	30.0 ± 0.25^{a}	25.0 ± 0.25^{a}	31.0 ± 1.00^{a}	30.0 ± 0.75^{a}	30.0 ± 0.50^{a}	27.0 ± 1.0^{a}	28.0 ± 1.00^{a}	20.0 ± 0.75	0	0
S. aureus clinical strain	31.0 ± 0.20^{a}	24.0 ± 0.20^{a}	28.0 ± 0.50^{b}	30.0 ± 1.00^{a}	25.0 ± 0.25^{b}	0	0	0	0	0
E. faecalis ATCC 29212	25.0 ± 0.50^{b}	11.0 ± 0.55 ^{g}	30.0 ± 1.25^{a}	20.0 ± 1.00^{d}	10.0 ± 0.30 ^{g}	0	17.5 ± 0.50^{c}	18.0 ± 1.00	0	0
E. faecalis clinical strain	19.0 ± 1.00^{d}	10.0 ± 0.25 ^{g}	17.0 ± 0.30 ^{g}	17.0 ± 0.75^{e}	9.0 ± 0.25 ^{g}	0	8.00 ± 0.50^{e}	0	0	0
S. pyogenes ATCC 19615	12.0 ± 0.25^{e}	17.0 ± 0.50^{d}	21.0 ± 0.80^{e}	20.0 ± 0.75^{d}	18.0 ± 0.25^{e}	0	18.0 ± 0.25^{b}	0	0	0
S. pyogenes clinical strain	12.0 ± 0.35^{e}	17.0 ± 0.75^{d}	20.0 ± 0.50^{f}	18.0 ± 0.75^{e}	17.0 ± 0.80^{e}	0	12.0 ± 0.50^{d}	0	0	0
E. coli ATCC 25922	30.0 ± 0.75^{a}	15.0 ± 0.25^{e}	25.0 ± 0.20^{c}	19.0 ± 0.55^{e}	15.0 ± 1.00^{f}	0	17.0 ± 0.75^{c}	19.0 ± 1.00	0	0
E. coli clinical strain	30.0 ± 1.00^{a}	13.0 ± 0.20^{f}	21.0 ± 0.75^{e}	19.0 ± 0.25^{e}	14.0 ± 1.00^{f}	0	0	0	0	0
C. albicans ATCC 885–653	24.0 ± 0.20^{b}	19.5 ± 0.30^{c}	23.0 ± 1.20^{d}	23.0 ± 0.15^{c}	21.0 ± 0.50^{d}	0	0	0	20.0 ± 1.0^{a}	21.0 ± 0.75^{a}
C. albicans clinical strain	23.0 ± 0.35^{c}	21.0 ± 1.00^{b}	24.5 ± 0.50^{c}	25.0 ± 0.45^{b}	22.0 ± 0.55^{c}	0	0	0	0	0

Notes: the data were statistically significant as compared with the control ($P < 0.05$); letters indicate the inhibition zones significantly different one from other according to Tukey's test.
Positive controls: *AMP – ampicillin, AMO – amoxicillin/clavulanic acid, GEN – gentamicin* for bacteria; *NIS – nistatin, ITR – itraconazol* for *C. albicans;* «0»– no inhibition.

Such ecological factors as climate, altitude, pathogen attack to plants, cultivation systems, in combination with the genetic background can considerably modify EsO composition through interfering with a plant's secondary metabolism (Marchioni et al., 2020; Mohammadi et al., 2020). The season of harvesting had also the significant effect on the chemical composition of EsO (Verma et al., 2011).

Nutrizio et al. (2020) revealed that the EsO of Croatian *Thymus serpyllum* accumulated linalool (0.30–15.03%), thymol (0.48–9.43%) and carvacrol (0.47–15.20%). The major components of the *Thymus serpyllum* EsO grown in Western Himalaya were thymol (19.4–60.1%), γ-terpinene (0.3–13.8%) and *p*-cymene (3.5–10.4%). (E)-nerolidol, caryophyllene oxide, myrcene and borneol chemotypes of wild *Thymus serpyllum* were found in Estonia (Paaver et al., 2008). The contents of (E)-nerolidol, caryophyllene oxide, myrcene and borneol were in the range of 1.7–70.1%, 1.4–45.0%, 0–20.2% and 0–19%, respectively, while the contents of thymol and carvacrol were very low (0–4.0%, and mean 0.96 and 0.62%, respectively) (Paaver et al., 2008). Paaver et al. (2008) concluded that thymol and carvacrol are not the main components of the Estonian wild thyme EsO. Loziene et al. (1998) revealed five chemotypes of *Thymus serpyllum* on the base of hierarchical cluster analysis: linalool chemotype, geranial/geraniol/neral one, thymol one, carvacrol/-terpinene/*p*-cymene one and thymol/carvacrol/*p*-cymene/γ-terpinene. The geranial/geraniol/neral chemotype is the most frequently found, followed by the phenolic types. The linalool chemotype was recorded only one time. To whatever degree, we can state that the tested EsO of *Thymus serpyllum* cultivated in Ukraine is the thymol/isothymol methyl ether chemotype.

The predominant compound of *Thymus pulegioides* EsO from Serbia was linalyl acetate (40.0%) (Stojanović et al., 2014). The percentage of carvacrol and its precursors γ-terpinene and *p*-cymene in the EsO of Lithuanian *Thymus pulegioides* ranged between 16.88 and 29.29%, 20.60–24.43%, and 5.54–11.33% respectively, depending on the influence of meteorological conditions and growth stage (Vaičiulytė et al., 2016). Mockute and Bernotiene (1999) revealed that the main components of the citral-geraniol chemotype of *Thymus pulegioides* EsO collected in wild habitats of Lithuania were geraniol (14.9–30.8%), geranial (α-citral) (9.7–19.7%), β-caryophyllene (6.0–11.4%) and nerol (4.1–11.8%). The EsO of carvacrol chemotype of *Thymus pulegioides* EsO contained carvacrol (16.0–22.2%), β-bisabolene (11.1–20.2%), β-caryophyllene (11.1–19.1%), γ-terpinene (5.8–16.2%), *p*-cymene (5.5–10.4%), and thymol (3.3–9.8%) (Mockute and Bernotiene, 1999). The revealed components of this carvacrol chemotype of *Thymus pulegioides* are characteristic for the *Thymus* genus (Mockute and Bernotiene (1999)). Five chemotypes of *Thymus pulegioides* ssp *pulegioides* collected in Vilnius city according to their odour and chemical composition were revealed by Mockute and Bernotiene (2003). The first one was the citral/geraniol chemotype (lemon odour) contained 18.4–30.2% of cis- and *trans*-citrals (neral and geranial), 14.4–23.2% of geraniol, 9.4–13.1% of β-caryophyllene and 5.5–12.1% of nerol. The second chemotype was geranial ("sweet" odour) which contained 23.1– 25.5% of geraniol, 10.8–13.5% of β-caryophyllene, and 9.5–10.2% of β-bisabolene. The third chemotype was caryophyllene one with "spicy" odour and contained 18.0–19.7% of β-caryophyllene, 14.5–15.9% of β-bisabolene, 12.5–14.6% of germacrene D and 8.2–13.8% of (E)-nerolidol. The fourth γ-terpinene/*p*-cymene chemotype had "hydrocarbon" odour and composed of 15.5–17.2% γ-terpinene, 7.7–17.0% *p*-cymene, 4.6–13.5% carvacrol and 9.6–10.5% β-caryophyllene. The fifth chemotype was carvacrol one which possess the "phenolic" odour and contained carvacrol (24.4–33.3%), β-caryophyllene (9.5–14.9%) and β-bisabolene (10.5–12.9%) (Mockute and Bernotiene, 1999).

EsO of *Monarda* species can accumulate a high content of aromatic compounds (up to 90%) (Myadelets et al., 2014). The composition of EsO from *Monarda didyma* and *Monarda fistulosa* aerial parts grown in two Italian sites differed significantly, especially in thymol content (62% vs. 31%, respectively) (Mattarelli et al., 2017). Thymol (64.9%), methyl ether thymol (7.6%), and *p*-cymene (9.5%) were revealed among 37 identified compounds in the EsO of the herb of *Monarda didyma* cultivated in the Omsk Region (Russia) (Myadelets et al., 2014). It seems that Myadelets et al. (2014) were dealing with the thymol chemotype of *Monarda didyma* as well. The most abundant compounds of *Monarda didyma* oil grown in Italy were oxygenated monoterpenes, and thymol (19.4%), its methyl ether (19.9%) and linalool (17.1%) prevailed among them (Marchioni et al., 2020). Linalool was the main component of the specific chemotype of *Monarda didyma* grown in France (Carnat et al., 1991). It should be speculated that the tested EsO isolated from the Ukrainian *Monarda didyma* CCS belongs to the thymol-eucalyptol chemotype due to prevailing these two compounds (47.54% and 17.82%, respectively).

GC–MS analysis of EsO isolated from the Indian *Salvia sclarea* herb showed that monoterpenoids linalyl acetate (61.33%) and linalool (17.59%) were its major components (Kumar Singh et al., 2019). Raafat and Habib (2018) revealed the availability of five chemotypes of Lebanon *Salvia sclarea* due to their EsO composition which differed significantly in the contents of linalool (10.75–38.07%), α-terpineol (6.45–13.40%) and linalyl acetate (1.0–35.28%) as the most abundant compounds. Cai et al. (2006) determined the chemical composition of the EsO of four *Salvia sclarea* samples obtained from Huabao Fragrance & Flavor Co. Ltd (Shanghai, China). The growing places of them are therefore not known. These samples of EsO mainly contained: linalyl acetate (29.50–49.83%) and linalool (17.03–28.76%) (Cai et al., 2006). Hudaib et al. (2001) stated about the following composition of the tested samples of the EsO of *Salvia sclarea* growing in Italy and Russia: linalyl acetate (42.75–55.72%), linalool (9.01–11.97%), germacrene D (4.35–7.58%), and *β*-caryophyllene (3.00–4.24%). These authors conclude that EsO from infected materials of *Salvia sclarea* of Italian origin contained higher percentages of sesquiterpene hydrocarbons (e.g. germacrene D and *β*-caryophyllene), monoterpene alcohols (e.g. α-terpineol) and diterpenoids (mainly sclareol) than non-infected ones. The most abundant components of the Turkish *Salvia sclarea* EsO were caryophyllene oxide (24.1%), sclareol (11.5%), and spathulenol (11.4%) (Yuce et al., 2014). The main components of four EsO of the aerial parts of *Salvia sclarea* collected from eleven provinces in Iran were linalool (12.17–21.41%), linalool acetate (13.06–52.61%) and D-germacrene (0–17.69%) (Rajabi et al., 2014).

It could be speculated that the major compounds of the investigated EsO impart their specific odour and biological properties, while minor components define the chemosystematic significance. Thymol (5-methyl-2-(propan-2-yl)phenol) is a monoterpene phenol, which is used in traditional medicine due to various pharmacological properties, including antibacterial, antifungal, antioxidant, and antispasmodic (Nagoor Meeran et al., 2017). Thymol is as a component of herbal medicinal preparations, cosmetic products, including mouthwashes, and as a food flavoring (Memar et al., 2017).

A lot of studies have revealed the antibacterial and antifungal properties of thymol and its isomer carvacrol (2-methyl-5-(propan-2-yl)phenol), especially against antibiotic-resistant pathogenic bacteria and *Candida albicans* (Memar et al., 2017). Thymol and carvacrol are active ingredients of the EsO of many *Lamiaceae* species with promising antimicrobial as well as antioxidant effects (Memar et al., 2017; Shanaida et al., 2018). They can inhibit the growth of both gram-positive and gram-negative microorganisms and have the antibiofilm effects (Sharifi-Rad et al., 2017). Thymol and carvacrol, the major compounds of *Thymus vulgaris* and *Origanum vulgare* EsO collected in Serbia, showed the highest antimicrobial activity against food-borne pathogens such as *Pseudomonas aeruginosa, Salmonella enteritidis,* and *Staphylococcus aureus* (Soković et al., 2010). Rattanachaikunsopon and Phumkhachorn (2010) revealed that aromatic monoterpenoid *p*-cymene enhanced the inhibitory effects of carvacrol in the studies against *Vibrio cholerae* and other foodborne pathogens.

Linalool (3,7-Dimethyl-1,6-octadien-3-ol) is a monoterpene alcohol presented in many *Lamiaceae* species (Herman et al., 2016; Hudz et al., 2020). The addition of linalool increased the efficacy of *Thymus vulgaris* EsO against *Pseudomonas aeruginosa* (Herman et al., 2016). Linalool and α-terpineol demonstrated strong antimicrobial effect against the cariogenic and periodontopathic bacteria in low concentrations (below 0.4 mg/mL) (Park et al., 2012).

Citral (3,7-dimethylocta-2,6-dienal) has a specific lemon odour and antimicrobial activity. It significantly reduced the levels of such cytokines as IL-1β, IL-6, TNF-α and such oxidative factors as malondialdehyde and hydroxyl radicals in methicillin-resistant *Staphylococcus aureus*-infected mice and diminish their lung inflammatory infiltrates that indicate antioxidant, antimicrobial and anti-inflammation activities of citral (Long et al., 2019). The combinations of citral and linalool in nanoemulsion with sesame oil possessed the antibacterial effects against *Listeria monocytogenes* as a known food-borne pathogen (Prakash and Vadivel, 2020).

Mulyaningsih et al. (2011) found that eucalyptol, or 1,8-cineole (1,3,3-Trimethyl-2-oxabicyclo[2.2.2]octane), which was the second predominant component of the tested *Monarda didyma* CCS EsO, was very active against multidrug-resistant gram-negative bacteria such as *Escherichia coli, Pseudomonas aeruginosa*, *Klebsiella pneumoniae,* and *Acinetobacter* (Mulyaningsih et al., 2011). Sharifi-Rad et al. (2017) concluded that antimicrobial activity of EsO correlated mostly to the presence of molecules with aromatic structure or aldehydes, alcochols, ethers such as thymol, carvacrol, linalool, 1,8-cineole, etc.

Among investigated 72 *Lamiaceae* species, the EsO of *Thymus* genera were the most active against pathogenic microorganisms due to prevailing aromatic compounds (Karpiński, 2020). Among 79 EsO screened for antibiofilm ability and reducing virulence of the uropathogenic strains of *Escherichia coli,* thymol-rich *Thymus vulgaris* oil and carvacrol-rich *Origanum vulgare* oil as well as their major constituents, carvacrol and thymol, inhibited this pathogenic microorganism significantly (Lee et al., 2017). EsO of the Ethiopian *Thymus serrulatus* and *Thymus schimperi* containing thymol and carvacrol as the major components demonstrated the significantly high inhibition zones against tooth decay bacteria *Streptococcus mutans* and *Lactobacillus* (Damtie et al., 2020). Thymol (55.35%) and *p*-cymene (11.2%) were the major components of the EsO hydrodistillated from the Turkish *Thymus vulgaris*. This EsO demonstrated high antimicrobial activity. The highest inhibition zones were against *Bacillus cereus* (NRRL B3711*), Staphylococcus epidermidis* (ATCC 12228) and *Staphylococcus aureus (*ATCC 9144) (Gedikoğlu et al., 2019). The EsO of the Polish *Thymus vulgaris* which contained thymol (61.9%), *p*-cymene (10.0%), and γ-terpinene (10.0%) as predominant compounds demonstrated the strong bactericidal activity against multidrug-resistant strain of *Staphylococcus aureus (*Kot et al., 2019). The EsO of the Indian *Thymus vulgaris* and its major active compound thymol are characterized by the great anti-candidal potential alone and in combination with antifungal medicines (Jafri et al., 2020). Antimicrobial properties of *Thymus vulgaris* EsO were found against several clinical isolates of opportunistic infections (Kryvtsova et al., 2019).

The EsO of Monarda didyma (cultivar 'Scorpio') grown in Poland was rich in linalool, *p*-cymene, thymol and thymol methyl ether, and showed the prominent antimicrobial effect against *Candida albicans*, *Bacillus cereus* and *Pseudomonas fluorescens* (Wróblewska

et al., 2019). The EsO of *Monarda punctata* from China contained 75.2% of thymol and exhibited the significant antibacterial effect against various microbial infection of respiratory tract, i.e. the drug resistant strains of *Staphylococcus aureus* (Li et al., 2014). Fraternale et al. (2006) found that EsO obtained from the Italian *Monarda didyma* contained thymol (up to 57.3%), γ-terpinene (up to 14.3%) and *p*-cymene (up to 10.5%). Studying the antifungal activity of this EsO against four phytopathogenic fungi demonstrated that *Rhizoctonia solani* and *Botrytis cinerea* were the most sensitive (Fraternale et al., 2006). Adebayo et al. (2013) revealed that the EsO of three *Monarda* species (*M. fistulosa*, *M. didyma*, and *M. didyma* var. *80-1A*) exhibited the inhibitory effects on mold *Botrytis cinerea*. It should be mentioned that the main components of the *M. didyma* EsO were thymol (41.17%), γ-terpinene (15.88%), and carvacrol (15.20%). Var. 80-1A of *M. didyma* was characterized by the domination of linalool (55.38%) and geraniol (20.71%), and *M. fistulosa* contained mainly geraniol (61.83%) and geranyl formate (16.61%) (Adebayo et al., 2013). Gram-positive cocci *Staphylococcus aureus* and yeast *Candida albicans* were very sensitive to the EsO obtained from the *Monarda fistulosa* cultivated in Ukraine due to the prevailing aromatic compounds such as thymol (42.01%) and *p*-cymene (15.45%) (Shanayda and Pokryshko, 2015).

It was revealed that the *Salvia sclarea* EsO and antibiotic oxacillin have the synergistic effect against methicillin-resistant *Staphylococcus epidermidis*. This EsO contained such main components as linalyl acetate (38.67%), linalool (20.42%) and germacrene (5.31%) (Chovanová et al., 2016). The antimicrobial activity of the *Salvia sclarea* EsO was related to damaging the bacterial cell membrane, leading to the release of ATP or even DNA (Cui et al., 2015). The moderate microbiostatic inhibitory activity was evaluated for the Italian *Salvia sclarea* EsO against *Candida albicans, Staphylococcus aureus*, *Staphylococcus epidermidis* and *Esherichia coli*. The inhibition effect increased progressively with increasing of contact time (*Peana et al., 1999*).

5. Conclusion

The analyzed EsO of five Ukrainian *Lamiaceae* representatives could be characterized as such chemotypes: 'thymol-*o*-cymene-γ-terpinene' of *Thymus vulgaris* CJ, 'thymol-isothymol methyl ether' of *Thymus serpyllum, '*α-citral-β-citral' of *Thymus pulegioides* C2, 'linalyl acetate-linalool' of *Salvia sclarea,* and 'thymol-eucalyptol' of *Monarda didyma* CCS. Among the tested EsO, *Thymus serpyllum* and *Thymus vulgaris* CJ followed by *Salvia sclarea* demonstrated the widest spectrum of action and highest antimicrobial activity against the most of tested typical and clinical strains of widespread pathogenic microorganisms. The found antimicrobial effects could be assigned to the chemical composition of the tested EsO. The sensitivity of the tested microorganisms is related to specific compounds of the EsO or synergistic effects of different compounds. Overall, all the chemotypes of the tested *Lamiaceae* species are highly promising herbal substances for the development of safe and effective herbal products in the form of oral liquid or sprays for the complementary treatment of the diseases of the oral cavity caused by drug-resistant pathogenic bacteria (*Staphylococcus aureus, Escherichia coli, Enterococcus faecalis, Streptococcus pyogenes*) and opportunistic yeast *Candida albicans.* These EsO can be also natural preservatives for cosmetic and food products.

Funding

Co-author Nataliia Hudz thanks the International Visegrad Fund for the scholarship for the research related to the development of sprays on the base of natural products performed in University of Opole (Poland).

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

References

Adebayo, O., Bélanger, A., Khanizadeh, S., 2013. Variable inhibitory activities of essential oils of three *Monarda* species on the growth of *Botrytis cinerea*. Can. J. Plant Sci. 93, 987–995.
Cai, J., Lin, P., Zhu, X., Su, Q., 2006. Comparative analysis of clary sage (*S. sclarea* L.) oil volatiles by GC-FTIR and GC-MS. Food Chem. 99, 401–407.
Carnat, A.P., Lamaison, J.L., Remery, A., 1991. Composition of leaf and flower essential oil from *Monarda didyma* L. cultivated in France. Flavour Fragrance J. 6 (1), 79–80.
Chovanová, R., Mikulášová, M., Vaverková, Š., 2016. Modulation of mecA gene expression by essential oil from *Salvia sclarea* and synergism with oxacillin in methicillin resistant *Staphylococcus epidermidis* carrying different types of staphylococcal chromosomal cassette mec. Int. J. Microbiol. 6475837
Cui, H., Zhang, X., Zhou, H., Zhao, C., Lin, L., 2015. Antimicrobial activity and mechanisms of *Salvia sclarea* essential oil. Bot. Stud. 56 (1), 16.
Damtie, D., Mekonnen, Y., 2020. Antibacterial activity of essential oils from Ethiopian thyme (*Thymus serrulatus* and *Thymus schimperi*) against tooth decay bacteria. PLoS ONE 15 (10).
European Pharmacopoeia, 2020. 10th ed. Council of Europe: Strasbourg, France.
Fraternale, D., Giamperi, L., Bucchini, A., Ricci, D., Epifano, F., Burini, G., Curini, M., 2006. Chemical composition, antifungal and *in vitro* antioxidant properties of *Monarda didyma* L. essential oil. J. Ess. Oil Res. 18, 581–585.
Gedikoğlu, A., Sökmen, M., Çivit, A., 2019. Evaluation of *Thymus vulgaris* and *Thymbra spicata* essential oils and plant extracts for chemical composition, antioxidant, and antimicrobial properties. Food Sci. Nutr. 7 (5), 1704–1714.
Herman, A., Tambor, K., Herman, A., 2016. Linalool affects the antimicrobial efficacy of essential oils. Curr. Microbiol. 72 (2), 165–172.
Hudz, N., Makowicz, E., Shanaida, M., Białoń, M., Jasicka-Misiak, I., Yezerska, O., Svydenko, L., Wieczorek, P.P., 2020. Phytochemical evaluation of tinctures and essential oil obtained from *Satureja montana* herb. Molecules 25 (20), 4763.
Hudz, N., Yezerska, O., Shanaida, M., Horčinová Sedláčková, V., Wieczorek, P.P., 2019. Application of the Folin-Ciocalteu method to the evaluation of *Salvia sclarea* extracts. Pharmacia. 66 (4), 209–215.
Jafri, H., Ahmad, I., 2020. *Thymus vulgaris* essential oil and thymol inhibit biofilms and interact synergistically with antifungal drugs against drug resistant strains of *Candida albicans* and *Candida tropicalis*. J Med Mycol. 30, (1) 100911.
Jarić, S., Mitrović, M., Pavlović, P., 2015. Review of ethnobotanical, phytochemical, and pharmacological study of *Thymus serpyllum* L. Evid. Based Complement. Alternat Med. 101978
Karpiński, T.M., 2020. Essential oils of Lamiaceae family plants as antifungals. Biomolecules. 10 (1), 103.
Koshovyi, O., Raal, A., Kovaleva, A., Myha, M., Ilina, T., Borodina, N., Komissarenko, A., 2020. The phytochemical and chemotaxonomic study of *Salvia* spp. growing in Ukraine. J. Appl. Biol. Biotech. 8 (3), 29–36.
Kot, B., Wierzchowska, K., Piechota, M., Czerniewicz, P., Chrzanowski, G., 2019. Antimicrobial activity of five essential oils from Lamiaceae against multidrug-resistant *Staphylococcus aureus*. Nat. Prod. Res. 33 (24), 3587–3591.
Kryvtsova, M.V., Salamon, I., Koscova, J., Bucko, D., Spivak, M., 2019. Antimicrobial, antibiofilm and biochemichal properties of *Thymus vulgaris* essential oil against clinical isolates of opportunistic infections. Biosyst. Div. 27 (3), 270–275.
Kumar Singh, V., Das, S., Kumar Dwivedy, A., Kumar Chaudhari, A., Upadhyay, N., Dubey, N.K., 2019. Assessment of chemically characterized *Salvia sclarea* L. essential oil and its combination with linalyl acetate as novel plant based antifungal, antiaflatoxigenic and antioxidant agent against herbal drugs contamination and probable mode of action. Nat. Prod. Res. 1–6.
Lange, B.M., Turner, G.W., 2013. Terpenoid biosynthesis in trichomes - current status and future opportunities. Plant Biotechnol. J. 11, 2–22.
Lee, J.H., Kim, Y.G., Lee, J., 2017. Carvacrol-rich oregano oil and thymol-rich thyme red oil inhibit biofilm formation and the virulence of uropathogenic *Escherichia coli*. J. Appl. Microbiol. 123 (6), 1420–1428.
Li, H., Yang, T., Li, F.-Y., Yao, Y., Sun, Z.-M., 2014. Antibacterial activity and mechanism of action of *Monarda punctata* essential oil and its main components against common bacterial pathogens in respiratory tract. Int. J. Clin. Exp. Pathol. 7 (11), 7389–7398.
Liu, Q., Meng, X., Li, Y., Zhao, C.-N., Tang, G.-Y., Li, H.-B., 2017. Antibacterial and antifungal activities of spices. Int. J. Mol. Sci. 18, 1283.
Long, N., Tang, H., Sun, F., Lin, L., Dai, M., 2019. Effect and mechanism of citral against methicillin-resistant *Staphylococcus aureus in vivo*. J. Sci. Food Agric. 99 (9), 4423–4429.
Loziene, K., Vaiciūniene, J., Venskutonis, P.R., 1998. Chemical composition of the essential oil of creeping thyme (*Thymus serpyllum* s.l.) growing wild in Lithuania. Planta Med. 64 (8), 772–773.
Marchioni, I., Najar, B., Ruffoni, B., Copetta, A., Pistelli, L., Pistelli, L., 2020. Bioactive compounds and aroma profile of some Lamiaceae edible flowers. Plants. 9 (6), 691.

Mattarelli, P., Epifano, F., Minardi, P., Di Vito, M., Modesto, M., Barbanti, L., Bellardi, M.G., 2017. Chemical composition and antimicrobial activity of essential oils from aerial parts of *Monarda didyma* and *Monarda fistulosa* cultivated in Italy. J. Ess. Oil Bear. Plants. 20 (1), 76–86.

Memar, M.Y., Raei, P., Alizadeh, N., Aghdam, M.A., Kafil, H.S., 2017. Carvacrol and thymol: strong antimicrobial agents against resistant isolates. Rev. Med. Microbiol. 28 (2), 63–68.

Mockute, D., Bernotiene, G., 1999. The main citral-geraniol and carvacrol chemotypes of the essential oil of *Thymus pulegioides* L. growing wild in Vilnius district (Lithuania). J. Agric. Food Chem. 47 (9), 3787–3790.

Mockute, D., Bernotiene, G., 2003. Five chemotypes of the essential oils of *Thymus pulegioides* L. growing wild in Lithuania. J. Essential Oil Bearing Plants. 6 (3), 139–147.

Moghtader, M., 2012. Antifungal effects of the essential oil from *Thymus vulgaris* L. and comparison with synthetic thymol on *Aspergillus niger*. J. Yeast Fungal Res. 3, 83–88.

Mohammadi, S., Tabrizi, L., Shokrpour, M., Hadian, J., Schulz, H., Riewe, D., 2020. Morphological and phytochemical screening of some *Thymus* ecotypes (*Thymus* spp.) native to Iran in order to select elite genotypes. J. Appl. Bot. Food Qual. 93, 186–196.

Mostafa, A.A., Al-Askar, A.A., Almaary, K.S., Dawoud, T.M., Sholkamy, E.N., Bakri, M.M., 2018. Antimicrobial activity of some plant extracts against bacterial strains causing food poisoning diseases. Saudi J. Biol. Sci. 25 (2), 361–366.

Mulyaningsih, S., Sporer, F., Reichling, J., Wink, M., 2011. Antibacterial activity of essential oils from *Eucalyptus* and of selected components against multidrug-resistant bacterial pathogens. Pharm. Biol. 49 (9), 893–899.

Myadelets, M.A., Domrachev, D.V., Kriklivaya, A.N., Vysochina, G.I., 2014. Dependence of the composition of essential oil of *Monarda didyma* L. (Lamiaceae) from the age and nature of plant materials. Chem. Plant Raw Mater. 1, 215–219 (in Russian).

Nagoor Meeran, M.F., Javed, H., Al Taee, H., Azimullah, S., Ojha, S.K., 2017. Pharmacological properties and molecular mechanisms of thymol: prospects for its therapeutic potential and pharmaceutical development. Front. Pharmacol. 8, 380.

Nutrizio, M., Pataro, G., Carullo, D., Carpentieri, S., Mazza, L., Ferrari, G., Chemat, F., Banović, M., Režek Jambrak, A., 2020. High voltage electrical discharges as an alternative extraction process of phenolic and volatile compounds from wild thyme (*Thymus serpyllum* L.): in silico and experimental approaches for solubility assessment. Molecules 25 (18), 4131.

Paaver, U., Orav, A., Arak, E., Mäeorg, U., Raal, A., 2008. Phytochemical analysis of the essential oil of *Thymus serpyllum* L. growing wild in Estonia. Nat. Prod. Res. 22 (2), 108–115.

Park, S.N., Lim, Y.K., Freire, M.O., Cho, E., Jin, D., Kook, J.K., 2012. Antimicrobial effect of linalool and α-terpineol against periodontopathic and cariogenic bacteria. Anaerobe. 18 (3), 369–372.

Peana, A.T., Moretti, M.D., Juliano, C., 1999. Chemical composition and antimicrobial action of the essential oils of *Salvia desoleana* and *S. sclarea*. Planta Med. 65 (8), 752–754.

Prakash, A., Vadivel, V., 2020. Citral and linalool nanoemulsions: impact of synergism and ripening inhibitors on the stability and antibacterial activity against *Listeria monocytogenes*. J. Food Sci. Technol. 57 (4), 1495–1504.

Raafat, K., Habib, J., 2018. Phytochemical compositions and antidiabetic potentials of *Salvia sclarea* L. essential oils. J. Oleo Sci. 67 (8), 1015–1025.

Rajabi, Z., Ebrahimi, M., Farajpour, M., Mirza, M., Ramshini, H., 2014. Compositions and yield variation of essential oils among and within nine *Salvia* species from various areas of Iran. Ind. Crop. Prod. 61, 233–239.

Rattanachaikunsopon, P., Phumkhachorn, P., 2010. Assessment of factors influencing antimicrobial activity of carvacrol and cymene against *Vibrio cholerae* in food. J. Biosci. Bioeng. 110, 614–619.

Shanaida, M., Hudz, N., Korzeniowska, K., Wieczorek, P., 2018. Antioxidant activity of essential oils obtained from aerial part of some *Lamiaceae* species. Int. J. Green Pharmacy. 12 (3), 200–204.

Shanayda, M.I., Pokryshko, O.V., 2015. Antimicrobial activity of essential oils of plants belonging to *Lamiaceae* Juss. family. Ann. Mechnikov Inst. 4, 66–69.

Sharifi-Rad, J., Sureda, A., Tenore, G.C., Daglia, M., Sharifi-Rad, M., Valussi, M., et al., 2017. Biological activities of essential oils: from plant chemoecology to traditional healing systems. Molecules 22, 70.

Siebert, D.J., 2004. Localization of salvinorin A and related compounds in glandular trichomes of the psychoactive sage, *Salvia divinorum*. Ann. Bot. 93, 763–771.

Soković, M., Glamočlija, J., Marin, P.D., Brkić, D., van Griensven, L.J., 2010. Antibacterial effects of the essential oils of commonly consumed medicinal herbs using an *in vitro* model. Molecules 15 (11), 7532–7546.

Stojanović, G., Jovanović, O., Petrović, G., Mitić, V., Jovanović, V.S., Jovanović, S., 2014. Composition of headspace volatiles and essential oils of three *Thymus* species. Nat. Prod. Commun. 9 (11), 1609–1612.

Tariq, S., Wani, S., Rasool, W., Shafi, K., Bhat, M.A., Prabhakar, A., Shalla, A.H., Rather, M.A., 2019. A comprehensive review of the antibacterial, antifungal and antiviral potential of essential oils and their chemical constituents against drug-resistant microbial pathogens. Microb. Pathog. 134, 103580.

Vaičiulytė, V., Butkienė, R., Ložienė, K., 2016. Effects of meteorological conditions and plant growth stage on the accumulation of carvacrol and its precursors in *Thymus pulegioides*. Phytochemistry 128, 20–26.

Verma, R.S., Verma, R.K., Chauhan, A., Yadav, A.K., 2011. Seasonal variation in essential oil content and composition of thyme, *Thymus serpyllum* L. cultivated in Uttarakhand Hills. Indian J. Pharm. Sci. 73 (2), 233–235.

Wróblewska, K., Szumny, A., Żarowska, B., Kromer, K., Dębicz, R., Fabian, S., 2019. Impact of mulching on growth essential oil composition and its biological activity in *Monarda didyma* L. Ind. Crop. Prod. 129, 299–308.

Yuce, E., Yildirim, N., Yildirim, N.C., Paksoy, M.Y., Bagci, E., 2014. Essential oil composition, antioxidant and antifungal activities of *Salvia sclarea* L. from Munzur Valley in Tunceli, Turkey. Cell. Mol. Biol. 60 (2), 1–5.