

Chemical Composition of the Essential Oils of Three *Ocimum basilicum* L. Cultivars from Serbia

Aleksandra S. ILIĆ^{1*}, Mališa P. ANTIĆ², Slavica C. JELAČIĆ²,
Tatjana M. ŠOLEVIĆ KNUDSEN³

¹Technical College of Applied Studies, Nemanjina 2, 12000 Požarevac, Serbia; a.ilic.vts@gmail.com (*corresponding author)

²University of Belgrade, Faculty of Agriculture, P.O. Box 14, 11080 Belgrade, Serbia; mantic@agrif.bg.ac.rs; jelacic@agrif.bg.ac.rs

³University of Belgrade, Institute of Chemistry, Technology and Metallurgy, Njegoševa 12, P.O. Box 473, 11000 Belgrade, Serbia; tsolevic@chem.bg.ac.rs

Abstract

Basil essential oil (BEO) contains a wide range of chemical compounds whose content may vary depending on chemotypes, environmental conditions, agronomic techniques and particularly the origin of the plant. In our present study, essential oils (EOs) were isolated by hydrodistillation method from dry herbs of three basil cultivars and analyzed by GC-FID and GC-MS. Two of the tested cultivars belong to sweet basil group (B₁ and B₂) while the third one was large leafed 'Genovese' basil (B₃). EO content in the dry herb was 0.65%, 0.41% and 0.62% respectively. The main classes of compounds of B₁EO and B₃EO were sesquiterpene hydrocarbons (38.39% and 37.95%), oxygenated monoterpenes (25.44% and 28.04%) and phenylpropanoids (17.43% and 15.71%). The main constituents of both EOs were monoterpene alcohol linalool (13.68% and 15.38%), phenoyl derivate eugenol (10.83% and 8.97%) and sesquiterpene hydrocarbon α -bergamotene (8.12% and 9.25%). In both EOs, epi-bicyclosesquiphellandrene was detected in considerable amount (7.03% and 8.07%). The most abundant compound classes in B₂EO were oxygenated monoterpenes (52.07%), sesquiterpene hydrocarbons (24.27%) and phenylpropanoids (10.95%). Linalool was the dominant compound (40.97%), followed by epi-bicyclosesquiphellandrene (8.70%) and methyl chavicol (7.92%). The results showed complex chemical composition of BEOs and pointed out the presence of biologically active compounds of importance for different branches of the pharmaceutical, chemical and food industry. Although there are differences in the chemical composition of the BEOs, the obtained results show that all of the tested cultivars are rich in compounds which are responsible for biological activities.

Keywords: basil, essential oil, GC-FID, GC-MS, hydrodistillation

Introduction

The family Lamiaceae is widely distributed over the world. It comprises over 5,000 medicinal and aromatic plant species whose EOs have multiple applications (Sakkas and Papadopoulou, 2017; Piras *et al.*, 2018). Basil (*Ocimum basilicum* L.) is a widely known member of Lamiaceae family. At present, this annual aromatic plant, native to Southeast Asia, is globally cultivated and has significant economic value (Varga *et al.*, 2017). Basil has been grown traditionally as a decorative, medicinal, seasoning and ritual herb (Jelačić *et al.*, 2011). Additionally, basil is mostly cultivated for its EO, which has broad pharmaceuticals and industrial uses (Shiwakoti *et al.*, 2017). BEO has been used in the food industry, especially in vegetables, meat and dairy products. It has been used as a flavouring as well as a

natural agent for increasing the shelf life of food products (Riveros *et al.*, 2015; Sharafati-Chaleshtori *et al.*, 2015; Piras *et al.*, 2018). It is also used in commercial fragrances, oral care products and aromatherapy (Labra *et al.*, 2004).

The content and chemical composition of the BEO has been the subject of many studies. The yield from different plant parts varies between 0.2-1.9% with the main components being linalool, methyl chavicol, eugenol and methyl cinnamate, as well as 1,8-cineole, methyl eugenol, geraniol, geranial, neral and α -bergamotene (Marotti *et al.*, 1996; Labra *et al.*, 2004; Sakkas and Papadopoulou, 2017). Based on the distribution and abundances of the main compounds in EO composition, Marotti *et al.* (1996) described three chemotypes: (1) linalool, (2) linalool / methyl chavicol and (3) linalool/eugenol chemotypes. In recent study which comprised 85 accessions of *O. basilicum*, based on seven major compounds, Varga *et al.* (2017)

proposed intraspecific characterization into five chemotypes: (A) linalool, (B) linalool/trans- α -bergamotene, (C) linalool/methyl chavicol, (D) linalool/trans-methyl cinnamate and (E) methyl chavicol. Chemotypes A and C can be considered to be European chemotypes. Chemotype D is a Tropical chemotype, while Chemotype E is described as a Reunion chemotype (Varga *et al.*, 2017). The composition of BEO is highly dependent on the chemotype, phenological stage of the plant as well as on other factors (growing and agroclimatic conditions, drying and distillation method) and thus also affects the biological activities (Marotti *et al.*, 1996; Klimánková *et al.*, 2008; Taie *et al.*, 2010; Wesolowska *et al.*, 2012; Shiwakoti *et al.*, 2017).

BEOs have been reported to possess numerous biological properties. A number of authors have mentioned antioxidant (Juliani and Simon, 2002; Božin *et al.*, 2006; Taie *et al.*, 2010; Beatović *et al.*, 2013; Shirazi *et al.*, 2014; Riveros *et al.*, 2015; Sharafati-Chaleshtori *et al.*, 2015; Elgndi *et al.*, 2017; Shiwakoti *et al.*, 2017; Stanojević *et al.*, 2017), antimicrobial (Božin *et al.*, 2006; Carović-Stanko *et al.*, 2010; Soković *et al.*, 2010; Beatović *et al.*, 2013; Stefan *et al.*, 2013; Joshi, 2014; Shirazi *et al.*, 2014; Sharafati-Chaleshtori *et al.*, 2015; Silva *et al.*, 2015; Chauhan *et al.*, 2017; Sakkas and Papadopoulou, 2017; Stanojević *et al.*, 2017; Piras *et al.*, 2018), anticancer (Taie *et al.*, 2010; Shirazi *et al.*, 2014; Elgndi *et al.*, 2017) and insect-repelling properties (Perumalsamy *et al.*, 2014) of BEO due to its phenolic and aromatic constituents.

The content and composition of EO are very important parameters for assessing the quality of basil and its application as raw material for different branches of pharmaceutical, food and chemical industries (Jelačić *et al.*, 2011). Therefore, the aim of the present study was to investigate the chemical constituents of the BEOs from three cultivars planted in the Republic of Serbia.

Materials and Methods

Plant material

Dried basil herb B₁ was obtained from “Bilje Borča” Belgrade while B₂ material was obtained from Institute for Medicinal Plant Research “Dr Josif Pančić” Belgrade in 2016. The materials were identified and representative herbarium specimens were deposited at “Bilje Borča” (serial No. L018-05-15) and the Institute for Medicinal Plant Research “Dr Josif Pančić” (serial No. 02740216). The B₃ material was ‘Genovese’ basil from the collection of the Institute for Crop Sciences of the Faculty of Agriculture in Belgrade and the Plant Gene Bank of Serbia where it is deposited under DB code (DB-01).

Isolation of the essential oils

The plant material was milled in a laboratory electric mill immediately before extraction and 20 g was subjected to hydrodistillation for 2 h using a Clevenger type apparatus according to the procedure IV of the Yugoslavian Pharmacopoeia (1984). The EOs were dried over anhydrous sodium sulphate and stored in sealed vials at 4 °C until further analysis. The yield of EO was calculated based on dry weight of plant material and expressed as % (mL EO/100g of dry plant material).

Essential oil analysis

Qualitative and quantitative analysis was carried out using GC-FID and GC-MS. For GC-MS analyses, an Agilent 7890N gas chromatograph with a HP5-MS capillary column (30 m length; 0.25 mm inner diameter; 0.25 μ m film) was used. The following temperature program was employed: 60 °C for 0 min; then 3 °C min⁻¹ to 280 °C and then held for 20 min. Helium was used as the carrier gas with the flow rate 1 mL min⁻¹. The GC was coupled to a Hewlett-Packard 5972 MSD operated at 70 eV and scanning masses in the 40-550 range. The peaks were identified by comparison of their retention indices (calculated relative to n-alkanes) to the literature data (Adams, 2007; Babushok *et al.*, 2011), and by comparison of their mass spectra to the mass spectra in the databases (NIST/EPA/NIH mass spectral library NIST2000, Wiley/NBS registry of mass spectral data, 7th ed., electronic versions).

For GC-FID analyses, an Agilent 4890A gas chromatograph with a HP5-MS capillary column (30 m length; 0.25 mm inner diameter; 0.25 μ m film) was used. The temperature program employed was the same as the one used for GC-MS analyses. Hydrogen was used as carrier gas (1 mL min⁻¹). The GC was coupled to a FID detector operating at 300 °C.

Statistical analysis

The analysis of variance (ANOVA) was carried out using a significance level of $p \leq 0.05$. The least significant difference (LSD) test, if necessary, was used to determine the significant differences between tested basil cultivars.

Results

The yield of BEOs

All cultivars analyzed in the present research yielded light yellow EOs. The contents of B₁EO, B₂EO and B₃EO were 0.65% (0.65 mL EO/100 g of dry plant material), 0.41% and 0.62% respectively. Comparing the obtained yields of BEO using LSD test, it can be concluded that there is a statistically significant difference between the observed cultivars in their content of the EOs.

Content of EO in basil was investigated by different authors. Jelačić *et al.* (2011) found that the yield of EO from aerial parts of 10 tested basil populations varied from 0.87% to 1.84%. Božin *et al.* (2006) and Elgndi *et al.* (2017) reported the yield of BEO from Serbia to be 0.37% and 0.67%, respectively. Beatović *et al.* (2015) reported the yield of EO from twelve *Ocimum* species varied from 0.65% to 1.90%. Literature data of Romanian basil cultivars show the EO level from 0.2% to 0.6% (Benedec *et al.*, 2009). The yield of obtained BEO in Republika Srpska (Bosnia and Herzegovina) was 0.4% (Stanojević *et al.*, 2017). A study by Wesolowska *et al.* (2012) showed that the content of EO in herbs of three tested basil cultivars ranged from 0.38% to 0.55%. Content of the EO of basil cultivars from Albania was ranged from 0.11% to 3.40%, while content of the EO obtained from aerial parts of 10 Italian basil cultivars was in the range from 0.3% to 0.8% (Marotti *et al.*, 1996; Cheliku *et al.*, 2015).

It can be concluded that the content of BEO

determined in our present research is in good agreement with the literature data.

Chemical composition of BEOs

The chemical composition of examined BEOs is presented in Table 1. Overall, 60 volatile constituents in the

three BEOs were identified: 38 in B₁E₀ (94.99% of the total oil), 42 in B₂E₀ (90.85% of the total oil) and 30 compounds in B₃E₀ (95.45% of the total oil). Statistical analysis of the results obtained by GC-MS analysis was conducted for all constituents with the content higher than 1% of the EO.

Table 1. Chemical composition of *Ocimum basilicum* L. essential oils from three tested cultivars

Compound	Content (%)			RI ^a	RI ^b
	B ₁ E ₀	B ₂ E ₀	B ₃ E ₀		
<i>α</i> -Pinene	1.07	0.07	1.95	939	943
Camphene	-	0.45	-	954	957
Benzaldehyde	-	0.02	-	961	963
<i>β</i> -Pinene	2.90	0.45	2.18	980	982
2-Carene	-	0.06	-	1001	1005
p-Cymene	-	0.02	-	1026	1024
<i>β</i> -Phellandrene	1.56	-	3.49	1030	1028
Eucalyptol (1,8-cineole)	5.98	2.12	6.20	1034	1033
<i>cis</i> - <i>β</i> -Ocimene	-	0.23	-	1039	1043
<i>trans</i> - <i>β</i> -Ocimene	-	0.02	-	1050	1051
<i>γ</i> -Terpinene	0.20	0.04	-	1060	1064
<i>trans</i> -Linalool oxide	-	0.99	-	1083	1085
Linalool	13.68	40.97	15.38	1099	1103
Camphor	1.15	0.53	1.63	1144	1147
Menthone	0.19	-	-	1154	1152
Isomenthone	-	0.16	-	1164	1161
Borneol	-	0.28	1.59	1165	1163
<i>β</i> -Terpineol	1.16	-	-	1166	1167
Menthol	-	0.15	-	1169	1170
<i>α</i> -Terpineol	0.91	0.48	-	1190	1189
Estragole (methyl chavicol)	4.13	7.92	5.26	1196	1194
Ethyl octanoate	0.40	-	0.61	1196	1199
Carvone	0.15	0.89	0.14	1242	1245
Geraniol	0.49	4.80	0.11	1258	1254
Geranial	-	0.13	-	1270	1268
Isobornyl acetate	1.73	-	2.91	1286	1289
Thymol	0.34	0.16	-	1302	1300
<i>trans</i> -Anethole	-	1.54	-	1301	1305
Myrtenyl acetate	-	-	0.08	1327	1325
1,5,5-Trimethyl-6-methylene- cyclohexene	0.13	-	-	1338	1335
<i>α</i> -Cubebene	0.75	0.08	2.61	1351	1350
Eugenol	10.83	0.62	8.97	1357	1352
Geraniol acetate	-	0.57	-	1386	1390
<i>β</i> -Cubebene	4.76	-	-	1388	1388
<i>β</i> -Elemene	4.01	2.63	4.47	1391	1390
Methyleugenol	2.13	0.69	1.48	1410	1406
<i>β</i> -Caryophyllene	-	0.22	-	1423	1426
<i>α</i> -Bergamotene	8.12	1.95	9.25	1433	1430
<i>β</i> -Copaene	1.25	-	3.92	1433	1435
<i>cis</i> - <i>β</i> -Farnesene	0.67	-	1.10	1444	1447
<i>α</i> -Humulene	1.40	0.78	1.36	1456	1460
<i>γ</i> -Gurjunene	-	-	1.05	1479	1478
D-Germacrene	-	3.42	-	1480	1483
Epi-bicyclosequiphellandrene	7.03	8.70	8.07	1488	1487
<i>β</i> -Guaiane	2.34	-	-	1489	1490
<i>α</i> -Selinene	-	0.72	-	1494	1492
Bicyclgermacrene	1.85	0.86	-	1495	1495
<i>α</i> -Muurolene	-	3.41	-	1497	1498
<i>γ</i> -Muurolene	3.71	-	0.54	1474	1476
Bulnesene	-	1.58	5.58	1505	1502
<i>cis</i> -Calamenene	2.50	-	-	1521	1520
<i>cis</i> -Nerolidol	0.52	-	-	1535	1538
Spathulenol	3.70	0.68	2.25	1577	1580
Caryophyllene oxide	0.37	-	-	1582	1586
<i>α</i> -Cadinol	1.41	0.87	-	1632	1637
<i>β</i> -Eudesmol	-	0.43	-	1648	1649
<i>α</i> -Bisabolol	-	-	2.10	1685	1681
Hexahydrofarnesyl acetone	0.29	0.13	0.48	1836	1833
Phytol	0.98	0.11	0.62	2122	2120
Phytol-acetate	0.20	-	0.07	2225	2229
Total identified (%)	94.99	90.85	95.45		1

- - compound not detected.

RI^a – Relative retention indices on a column with dimethylsilicone stationary phase with 5% phenyl groups reported in literature (Adams, 2007; Babushok *et al.*, 2011; NIST database, electronic version).

RI^b – Relative retention indices experimentally determined and calculated against n-alkanes (C8–C32) on the HP-5MS column.

Based on statistical analysis of the selected constituents, it can be concluded that there were no statistically significant differences between the examined BEOs.

The main classes of compounds of all the examined BEOs were sesquiterpene hydrocarbons (38.39%, 24.27% and 37.95%), oxygenated monoterpenes (25.44%, 52.07% and 28.04%) and phenylpropanoids (17.43%, 10.93% and 15.71%). Monoterpene hydrocarbons and oxygenated sesquiterpenes were present in low amounts in all the analyzed BEOs (< 10%). The relative abundances of the main classes of compounds are shown in Table 2.

The main constituent of all BEOs was monoterpene alcohol linalool (13.68%, 40.97% and 15.38%). In considerable amounts in B₁EО and B₃EО the following constituents were detected: eugenol (10.83% and 8.97%), α -bergamotene (8.12% and 9.25%), epi-bicyclosquiphellandrene (7.03% and 8.07%), eucalyptol (5.98% and 6.20%) and methyl chavicol (4.13% and 5.26%). Except from linalool, compounds with higher abundance in B₂EО were epi-bicyclosquiphellandrene (8.70%), methyl chavicol (7.92%), geraniol (4.80%), D-germacrene (3.42%) and α -muurolene (3.41%).

As B₁EО and B₃EО showed similarity in chemical composition, the major difference between these BEOs and B₂EО was in the content of eugenol. B₂EО had the lowest content of this phenol derivate (0.62%) in which the two other cultivars were rich (10.83% and 8.97%). Politeo *et al.* (2007) found that the antioxidant activity of BEO highly depends on the content of this compound. Accordingly, it can be expected that EOs from these two basil cultivars will be more effective antioxidants compared to B₂EО. Furthermore, B₁EО and B₃EО had higher concentration of eucalyptol (5.98% and 6.20%) relative to B₂EО (2.12%). Monoterpene alcohol geraniol was present in considerable amount in B₂EО (4.80%) while in two other EOs it was in trace. D-germacrene and α -muurolene were sesquiterpene hydrocarbons identified only in B₂EО while β -cubebene was identified in significant amount in B₁EО and it was absent in EOs of other investigated cultivars.

The differences in the chemical composition of the EOs analyzed can be explained as a consequence of differences in growing and agroclimatic conditions.

Many studies reported linalool as the main compound of investigated BEOs (Marotti *et al.*, 1996; Juliani and Simon, 2002; Wesolowska *et al.*, 2012; Cheliku *et al.*, 2015; Riveros *et al.*, 2015; Silva *et al.*, 2015; Elgndi *et al.*, 2017). In study of Beatović *et al.* (2015), which included twelve basil cultivars, the oxygenated monoterpenes were predominant in eight cultivars with linalool as the main constituent. Shirazi *et al.* (2014) and Sharafati-Chaleshtori *et al.* (2015) reported methyl chavicol as the main compound of BEO while eugenol was detected as compound with highest abundance by Piras *et al.* (2018). Presence of the sesquiterpene hydrocarbon epi-bicyclosquiphellandrene as a compound with higher abundance is comparable with investigation of Benedec *et al.* (2009). The chemical composition of BEO 'Genovese' has been well described in the literature and the composition and abundances of major components in our study is in accordance with earlier studies (Marotti *et al.*, 1996; Juliani and Simon, 2002; Labra *et al.*, 2004; Carović-Stanko *et al.*, 2010; Beatović *et al.*, 2013; Stefan *et al.*, 2013).

It can be concluded that the compositions of the BEOs investigated in our present research are in good agreement with the literature data.

Table 2. The relative abundances of the main classes of compounds in tested BEOs

Classes of compounds	Content (%)		
	B ₁ EО	B ₂ EО	B ₃ EО
Monoterpene hydrocarbons	5.73	1.34	7.62
Oxygenated monoterpenes	25.44	52.07	28.04
Phenylpropanoids	17.43	10.93	15.71
Sesquiterpene hydrocarbons	38.39	24.27	37.95
Oxygenated sesquiterpenes	6.00	1.98	4.35
Diterpenes	1.18	0.11	0.69
Other	0.82	0.15	1.09

Conclusions

In this study, the chemical composition of the EOs from three basil cultivars from Serbia was investigated. The contents of BEOs were in good agreement with the literature data. The statistical analysis demonstrated a statistically significant difference between the observed cultivars in their contents of the EOs.

It was concluded that the compositions of the BEOs investigated were in good agreement with the literature data. Some differences in abundances and distribution of their constituents were observed. However, the statistical analysis of the selected constituents did not reveal any statistically significant differences between the examined BEOs. The differences in the chemical composition of the EOs analyzed were explained as a consequence of differences in growing and agroclimatic conditions. Although there were differences between the tested BEOs, the obtained results showed that all of them were rich in compounds which are responsible for biological activities. Further studies of examined cultivars are required to determine their biological activities and applicability as food additives.

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References

- Adams RP (2007). Identification of essential oil components by gas chromatography/mass spectrometry. Allured Publishing Corporation, 4th ed. USA pp 69-351.
- Babushok VI, Linstrom PJ, Zenkevich IG (2011). Retention indices for frequently reported compounds of plant essential oils. Journal of Physical and Chemical Reference Data 40:1-47.
- Beatović DV, Jelačić SC, Oparnica CDJ, Krstić-Milošević DB, Glamočlija JM, Ristić MS, Šiljegović JD (2013). Chemical composition, antioxidative and antimicrobial activity of essential oil of *Ocimum sanctum* L. Hemijska Industrija 67(3):427-435.
- Beatović DV, Krstić-Milošević DB, Trifunović SS, Šiljegović JD, Glamočlija

- JM, Ristić MS, Jelačić SC (2015). Chemical composition, antioxidant and antimicrobial activities of the essential oils of twelve *Ocimum basilicum* L. cultivars grown in Serbia. *Records of Natural Products* 9(1):62-75.
- Benedec D, Oniga I, Oprean P, Tamas M (2009). Chemical composition of the essential oils of *Ocimum basilicum* L. cultivated in Romania. *Farmacia* 57(5):625-629.
- Božin B, Mímica-Dukić N, Simin N, Anckov G (2006). Characterisation of the volatile composition of essential oils of some *Lamiaceae* spices and the antimicrobial and antioxidant activities of the entire oils. *Journal of Agricultural and Food Chemistry* 54(5):1882-1828.
- Carović-Stanko K, Orlić S, Politeo O, Strikić F, Kolak I, Miloš M, Satović Z (2010). Composition and antibacterial activities of essential oils of seven *Ocimum* taxa. *Food Chemistry* 119:196-201.
- Cheliku N, Cvetkovikj KI, Stefkov G, Karapandzova M, Bardhi N, Qjazimi B, Kulevanova S (2015). Essential oil composition of five basil cultivars (*Ocimum basilicum*) from Albania. *Macedonian Pharmaceutical Bulletin* 61(2):11-18.
- Chouhan S, Sharma K, Guleria S (2017). Antimicrobial activity of some essential oils- Present status and future perspectives. *Medicines* 4:58-79.
- Elgndi MA, Filip S, Pavlič B, Vladić J, Stanojković T, Žižak Ž, Zeković Z (2017). Antioxidative and cytotoxic activity of essential oils and extracts of *Satureja montana* L., *Coriandrum sativum* L. and *Ocimum basilicum* L. obtained by supercritical fluid extraction. *Journal of Supercritical Fluids* 128:128-137.
- Jelačić SC, Beatović DV, Prodanović SA, Tasić SR, Moravčević DjZ, Vujošević AM, Vučković SM (2011). Chemical composition of the essential oil of basil (*Ocimum basilicum* L. Lamiaceae). *Hemijiska Industrija* 65(4):465-471.
- Joshi RK (2014). Chemical composition and antimicrobial activity of the essential oil of *Ocimum basilicum* L. (sweet basil) from Western Ghats of North West Karnataka, India. *Ancient Science of Life* 33(3):151-156.
- Juliani HR, Simon JE (2002). Antioxidant activity of basil. In: Janick J, Whipkey A (Eds). *Trends in new crops and new uses*. ASHS Press, Alexandria, VA, pp 575-579.
- Klimánková E, Holadová K, Hajslová J, Čajka T, Poustka J, Koudela M (2008). Aroma profiles of five basil (*Ocimum basilicum* L.) cultivars grown under conventional and organic conditions. *Food Chemistry* 107:464-472.
- Labra M, Miele M, Ledda B, Grassi F, Mazzei M, Sala F (2004). Morphological characterisation, essential oil composition and DNA genotyping of *Ocimum basilicum* L. cultivars. *Plant Science* 167:725-731.
- Marotti M, Piccaglia R, Giovanelli E (1996). Differences in essential oil composition of basil (*Ocimum basilicum* L.) Italian cultivars related to morphological characteristics. *Journal of Agricultural and Food Chemistry* 44(12):3926-3929.
- Perumalsamy H, Kim JY, Kim JR, Hwang KN, Ahn YJ (2014). Toxicity of basil oil constituents and related compounds and the efficacy of spray formulations to *Dermatophagoides farinae* (Acari: Pyroglyphidae). *Journal of Medical Entomology* 51(3):650-657.
- Piras A, Gonçalves MJ, Alves J, Falconieri D, Porcedda S, Maxia A, Salgueiro L (2018). *Ocimum tenuiflorum* L. and *Ocimum basilicum* L., two spices of Lamiaceae family with bioactive essential oils. *Industrial Crops and Products* 113:89-97.
- Politeo O, Jukić M, Miloš M (2007). Chemical composition and antioxidant capacity of free volatile aglycones from basil (*Ocimum basilicum* L.) compared with its essential oil. *Food Chemistry* 101:379-385.
- Riveros CG, Nepote V, Grossa NR (2015). Thyme and basil essential oils included in edible coating as a natural preserving method of oilseed kernels. *Journal of the Science of Food and Agriculture* 96(1):183-191.
- Sakkas H, Papadopoulou C (2017). Antimicrobial activity of basil, oregano and thyme essential oil. *Journal of Microbiology and Biotechnology* 27(3):429-438.
- Sharafari-Chaleshtori R, Rokni N, Rafieian-Kapaei M, Drees F, Salehi E (2015). Antioxidant and antibacterial activity of basil (*Ocimum basilicum* L.) essential oil in beef burger. *Journal of Agricultural Science and Technology* 17(3):817-826.
- Shirazi MT, Gholami H, Kavosi G, Rawshan V, Tafsiy A (2014). Chemical composition, antioxidant, antimicrobial and cytotoxic activities of *Tagetes minuta* and *Ocimum basilicum* essential oils. *Food Science and Nutrition* 2(2):146-155.
- Shiwakoti S, Saleh O, Poudyal S, Barka A, Qian Y, Zheljzkov VD (2017). Yield, composition and antioxidant capacity of the essential oil of sweet basil and holy basil as influenced by distillation method. *Chemistry and Biodiversity* 14(4):e1600417.
- Silva VA, Sousa JP, Guerra FQS, Pessôa HLF, Frietas AFR, Alves LBN, Lima EO (2015). Antibacterial activity of *Ocimum basilicum* essential oil and linalool on bacterial isolates of clinical importance. *International Journal of Pharmacognosy and Phytochemical Research* 7:1066-1071.
- Soković M, Glamočlija J, Marin PD, Brkić D, van Griensven LJLD (2010). Antibacterial effects of the essential oils of commonly consumed medicinal herbs using an *in vitro* model. *Molecules* 15:7532-7546.
- Stanojević LjP, Marjanović-Balaban ŽR, Kalaba VD, Stanojević JS, Cvetković DJ, Cakić MD (2017). Chemical composition, antioxidant and antimicrobial activity of basil (*Ocimum basilicum* L.) essential oil. *Journal of Essential Oil Bearing Plants* 20(6):1557-1569.
- Stefan M, Zamfirache MM, Padurariu C, Trută E, Gostin I (2013). The composition and antibacterial activity of essential oils in three *Ocimum* species growing in Romania. *Central European Journal of Biology* 8(6):600-608.
- Taie HAA, Salama ZAER, Radwan S (2010). Potential activity of basil plants as a source of antioxidants and anticancer agents as affected by organic and bio-organic fertilization. *Notulae Botanicae Horti Agrobotanici Cluj-Napoca* 38(1):119-127.
- Varga F, Carović-Stanko K, Ristić M, Grdiša M, Liber Z, Šatović Z (2017). Morphological and biochemical intraspecific characterization of *Ocimum basilicum* L. *Industrial Crops and Products* 109:611-618.
- Wesolowska A, Kosecka D, Jadcak D (2012). Essential oil composition of three sweet basil (*Ocimum basilicum*) cultivars. *Herba Polonica* 58(2):5-16
- Yugoslavian Pharmacopoeia (1984). *Yugoslavian Pharmacopoeia*, 4th ed. National Institute of Health Protection. Belgrade pp 126-127.