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INFLUENCE OF DISTILLATION METHOD ON THE CHEMICAL COMPOSITION OF ESSENTIAL OILS ISOLATED FROM DIFFERENT PARTS OF SWEET BASIL (*OCIMUM BASILICUM* L.) CV 'CINNAMON'

WPŁYW METODY DESTYLACJI NA SKŁAD CHEMICZNY OLEJKÓW ETERYCZNYCH WYIZOLOWANYCH Z RÓŻNYCH CZĘŚCI BAZYLII POSPOLITEJ (*OCIMUM BASILICUM* L.) ODMIANY 'CINNAMON'

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Streszczenie. Skład chemiczny olejków eterycznych, wyizolowanych na drodze hydrodestylacji i destylacji z parą wodną z kwiatostanów i liści z łodygami bazylii pospolitej odmiany 'Cinnamon', badano metodą GC-MS. (E)-Cynamonian metylu, linalol i estragol stanowiły dominujące składniki wszystkich analizowanych olejków. Aczkolwiek, porównując obie metody destylacji, wykazano, że w przypadku olejków otrzymanych z kwiatostanów zawartość (E)-cynamonianu metylu, estragolu, eugenolu i (Z)-cynamonianu metylu była większa w olejku wyizolowanym na drodze hydrodestylacji, natomiast zawartość β-selinenu, β-eudesmolu i α-selinenu była większa w olejku otrzymanym na drodze destylacji z parą wodną. W przypadku olejków wyizolowanych z liści z łodygami więcej (E)-cynamonianu metylu i (Z)-cynamonianu metylu stwierdzono w olejku otrzymanym na drodze hydrodestylacji; zawartość linalolu, β-eudesmolu i estragolu była większa w olejku otrzymanym na drodze destylacji z parą wodną.

Key words: essential oil composition, hydrodistillation, linalool, methyl cinnamate, sweet basil 'Cinnamon', water-steam distillation.
Słowa kluczowe: bazylia pospolita 'Cinnamon', cynamonian metylu, destylacja z parą wodną, hydrodestylacja, linalol, skład olejku eterycznego.

INTRODUCTION

Basil (*Ocimum basilicum* L.) is an annual aromatic plant, native to the tropical and subtropical regions of Asia, Africa and South America (Telci et al. 2006). Since antiquity the herb has been used for chest infections, digestive problems, headaches, migraines, head-colds and culinary purposes (Davis 2005; Sellar 2005). Today, it is widely used for its therapeutic properties and for production of essential oil (Raghavan 2007).

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The leaves and flowering tops of basil are perceived as carminative, stomachic, antispasmodic, local anesthetic and anticonvulsant (Chalchat and Ozcan 2008; Hassanpouraghdam et al. 2010). The essential oil, extracted from leaves and flowers, possesses antifungal, insect-repelling and antiseptic activities (Reuveni et al. 1984; Dube et al. 1989). The oil of the plant is also used in food, cosmetic and aromatherapy industries, due to its pleasant aroma and antimicrobial activity (Viña and Murillo 2003; Suppakul et al. 2003; Zheljazkov et al. 2008).

Many cultivars of basil are in common use for both culinary application and essential oil extraction. They vary in size, color, aroma and have different value on the world market (Raghavan 2007). Some cultivars are rich in essential oils and could offer a new source of oil for industry (Švecova and Neugebauerova 2010).

Scented basils (lemon, anise, cinnamon) can be used fresh or dried in the place of regular sweet basil in sorbets, honeys, vinegars and baked goods. Dried leaves and flowers can be used in potpourris (Abduelrahman et al. 2009). Basil 'Cinnamon', also called Mexican spice basil, is commonly used in hot drinks, raw dishes, fruit salads, jellies and marinades for meats. Due to its distinctive cinnamon taste and aroma, the herb is also used as an alternative to ground, dried cinnamon (Gardner 1998; Ogden 1990).

Essential oil from basil flowers or whole plant is extracted by hydrodistillation or steam distillation. Depending on the harvested plant part, two grades of oil are obtained, i.e. flower oil and herb oil. Flower oil is more expensive and has a superior note (Bonnardeaux 1992).

The quality of the oil, its composition and aroma, depends on the variety, season, development stage and distillation technique.

The aim of this work was to investigate the influence of distillation method on the chemical composition of essential oils isolated from different parts of sweet basil (*Ocimum basilicum* L.) cv Cinnamon.

MATERIAL AND METHODS

Plant. In 2010–2011, the field experiment was conducted at the Horticultural Experimental Station in Dołuje near Szczecin. The basil (*Ocimum basilicum* L.) cv Cinnamon seeds used in the experiment came from commercial seed company World Sementi Polish Distribution in Szczecin. Experiment was established in a randomized block design, in four replications. In both years of the experiment the seeds were sown directly into the open field on 23 May in rows 40 cm apart. The area of one plot was 1.44 m^2 (1.2 x 1.2 m^2), and of the whole experiment – 30 m^2. For 1.44 m^2 of the field, 0.86 g of seeds was used.

Mineral fertilization was quantified according to the results of the chemical analysis of the soil samples. Annually, 63 kg N · ha^{-1} and 30 kg K_2O · ha^{-1} were used for plant fertilization. During vegetation manual weeding and soil loosening were performed.

The basil herb was harvested at the beginning of flowering (5 August 2010 and 9 August 2011). The plants were cut at 10–15 cm above the ground surface and dried at room temperature in a shady and well ventilated place. The inflorescences and leaves with stems were separated manually after drying.

Isolation of the essential oils. For the isolation of essential oil by hydrodistillation (HD), Dean-Stark apparatus was used (Rahmat et al. 2006; Tajidin et al. 2012; Nur Ain et al. 2013). Thirty grams of dried inflorescences and leaves with stems (separately) along with 500 ml distilled water were subjected to hydrodistillation for 4 hours. The distillate was saturated with sodium chloride and methylene chloride was added. Then, the methylene chloride layer and water layer were separated by funnel. After dehydrated by anhydrous sodium sulphate, the solvent was removed at 40ºC in a rotary evaporator. The yellow oils obtained this way were weighed and stored in hermetically sealed dark glass flask in a refrigerator until GC-MS analysis.

Water-steam distillation (WSD) was carried out by passing steam into a 1-liter round-bottomed flask containing the dried plant material (30 g) and 500 ml of distilled water for 90 minutes (Charles and Simon 1990). The inflorescences and leaves with stems were distilled separately. The condensate (water and oil) was extracted three times with methylene chloride to completely extract the essential oil. Anhydrous sodium sulphate was added to methylene chloride to remove moisture. Methylene chloride was then removed by rotary evaporation at 40ºC. The yellow oils obtained were weighed and stored in hermetically sealed dark glass flask in a refrigerator until GC-MS analysis.

The content of the oils was calculated based on dry weight of plant material and expressed as % (w/w) in Table 1.

GC-MS analysis of essential oils. The qualitative GC-MS analysis of the oils was performed using an HP 6890 gas chromatograph coupled with HP 5973 Network Mass Selective Detector. The separation was achieved using an HP-5MS fused silica capillary column 30 meters length, 0.25 mm in diameter and with 0.25 μm thick stationary phase film (5%-phenyl-methylpolysiloxane).

The GC oven temperature was maintained at 40ºC for 5 minutes, then increased to 60ºC at a rate of 30ºC per minute, next to 230ºC at a rate of 6ºC per minute (kept constant for 10 minutes), and then increased to a final temperature of 280ºC at a rate of 30ºC per minute. The oven was held at this temperature for 30 minutes. The flow rate of helium (carrier gas) through the column was kept at 1.0 ml · min^{-1}. Samples of 2 μl (as solutions in methylene chloride) were injected with a split ratio of 5 : 1. The injector and the transfer line temperature was 280^0C, the ion source temperature was 230ºC , the solvent delay was 4 minutes. Mass spectra were taken at 70eV. Mass range was from 40 to 550 m/z. The total running time for a sample was 76 minutes.

The components of the essential oils were identified by comparison of their mass spectra with those stored in NIST 2002 and Wiley NBS75K.L mass spectral libraries or with authentic compounds (camphor, carvacrol, isoeugenol, (E)-methyl cinnamate) available in our laboratory and confirmed by comparison of their retention indices, either with those of authentic compounds or with data published in the literature (Sajjadi 2006; Adams 2007; Hussain et al. 2008; Dzida 2010).

The retention indices (RI) were calculated for all volatile constituents using a homologous series of n-alkanes (C_8–C_{20}) under the same chromatographic conditions which were used for the analysis of the essential oils.

The relative percentage amounts of the essential oil constituents were evaluated from the total peak area (TIC) using apparatus software.

Statistical analysis. Several results of the study (Table 1, 4, 5) were subjected to the analysis of variance which was performed with AWAR programme, made by Department of Applied Informatics, Institute of Soil Science and Plant Cultivation in Puławy. The means of two years were separated by the Tukey's test at p = 0.05. The statistical analysis of the results given in Table 4 and 5 was conducted for these components of the essential oil which concentration was higher than 0.50%.

RESULTS

Hydrodistillation and water-steam distillation of inflorescences and leaves with stems of basil 'Cinnamon' provided a yellow liquids with characteristic cinnamon-like smell (Table 1).

Table 1. Essential oil content in different parts of sweet basil (*Ocimum basilicum* L.) cv 'Cinnamon'
Tabela 1. Zawartość olejku eterycznego w różnych częściach bazylii pospolitej (*Ocimum basilicum* L.) odmiany Cinnamon

Distillation method Metoda destylacji	Oil content – Zawartość olejku [%] (w/w)								
	2010			2011			2010–2011		
	A	B	mean średnia	A	B	mean średnia	A	B	mean średnia
Hydrodistillation Hydrodestylacja	1.80	0.80	1.30	2.20	0.90	1.55	2.00	0.85	1.43
Water-steam distillation Destylacja z parą wodną	1.80	1.00	1.40	2.40	1.20	1.80	2.10	1.10	1.60
Mean – Średnia	1.80	0.95	1.35	2.30	1.05	1.68	2.05	0.98	1.51
$LSD_{\alpha=0.05}$ – $NIR_{\alpha=0.05}$									
Distillation method Metoda destylacji		n.s.			n.s.			n.s.	
Plant part Część rośliny		0.091			0.043			0.185	
Interaction Interakcja		0.129			0.061			n.s.	

A – inflorescences – kwiatostany.
B – leaves with stems – liście z łodygami.
n.s. – not significant differences – różnice nieistotne.

On the base of obtained results of the study it was proved that the distillation method did not significantly affect the essential oil content. However, the part of plant had a significant effect on the content of essential oil. In both years of the study significantly higher content of oil was found in inflorescences. It was higher on average by 1.07%, in comparison with the leaves with stems.

Similarly, the higher content of essential oil in flowers, compared to leaves and stems, was found by Tahira et al. (2013) in lemon basil (*Ocimum basilicum var.citriodorum*).

The relative amounts of the volatile components identified in the essential oils are listed in Table 2, in order of their elution from a HP-5MS column.

Table 2. Percentage composition of essential oils of sweet basil (*Ocimum basilicum* L.) cv 'Cinnamon' in dependence of distillation method
Tabela 2. Skład procentowy olejków eterycznych z bazylii pospolitej (*Ocimum basilicum* L.) odmiany 'Cinnamon' w zależności od metody destylacji

Components – Składniki	RI	Inflorescences – Kwiatostany				Leaves with stems – Liście z łodygami			
		HD		WSD		HD		WSD	
		2010	2011	2010	2011	2010	2011	2010	2011
Eucalyptol (1,8-cineole)	1032	0.70	1.20	0.72	1.28	1.20	1.97	1.75	1.83
(E)-β-Ocimene	1048	0.09	0.10	0.07	0.09	–	–	–	–
cis-β-Terpineol	1069	–	–	–	–	0.06	0.14	0.12	0.13
(E)-Linalool oxide	1073	0.54	0.24	0.39	0.12	0.46	0.33	0.56	0.19
Fenchone	1089	0.53	0.25	0.39	0.15	0.48	0.42	0.63	0.26
Linalool	1105	23.33	44.44	22.23	37.88	21.61	35.12	23.76	35.52
4-Acetyl-1-methylcyclohexene	1134	–	–	–	–	0.37	0.28	0.22	0.18
Camphor	1149	0.29	0.23	0.18	0.21	0.05	0.61	0.05	0.47
Borneol	1171	0.56	0.31	0.30	0.33	0.16	0.27	0.16	0.20
Terpinen-4-ol	1181	1.08	0.91	0.62	0.79	0.61	1.01	0.62	0.91
α-Terpineol	1195	0.48	0.30	0.26	0.27	0.38	0.46	0.36	0.35
Estragole (methyl chavicol)	1202	1.71	7.91	0.98	8.26	1.11	5.80	1.48	6.22
Isoestragole	1216	–	–	–	–	0.11	0.28	0.04	0.04
trans-Geraniol	1258	0.14	0.05	0.09	0.07	–	–	–	–
Chavicol	1270	–	–	–	–	0.71	0.90	0.25	0.30
Bornyl acetate	1289	0.21	0.32	0.22	0.37	0.05	0.26	0.06	0.30
(Z)-Methyl cinnamate	1310	3.37	1.68	2.30	1.47	7.52	4.76	5.74	4.16
Carvacrol	1322	0.30	0.08	–	–	–	–	–	–
exo-2-Hydroxycineole acetate	1345	–	–	–	–	0.08	0.15	0.12	0.16
α-Cubebene	1355	0.11	0.11	0.14	0.11	–	–	–	–
Eugenol	1365	8.16	0.82	7.05	0.58	4.84	1.32	2.63	1.06
Isoeugenol	1376	1.67	0.05	-	-	3.15	0.21	2.40	0.14
α-Copaene	1384	0.40	0.38	0.33	0.28	0.10	0.15	0.18	0.11
(E)-Methyl cinnamate	1403	36.17	22.27	31.36	26.89	45.58	28.19	35.53	26.16
β-Caryophyllene	1440	0.42	0.43	0.45	0.33	0.18	0.24	0.31	0.22

Table 2. Percentage composition of essential oils of sweet basil (*Ocimum basilicum* L.) cv 'Cinnamon' in dependence of distillation method (cont.)
Tabela 2. Skład procentowy olejków eterycznych z bazylii pospolitej (*Ocimum basilicum* L.) odmiany 'Cinnamon' w zależności od metody destylacji (cd.)

Components – Składniki	RI	Inflorescences – Kwiatostany				Leaves with stems – Liście z łodygami			
		HD		WSD		HD		WSD	
		2010	2011	2010	2011	2010	2011	2010	2011
α-Guaiene	1446	1.06	1.00	1.19	0.76	0.26	0.52	0.37	0.49
(Z)-β-Farnesene	1459	0.57	0.37	0.80	0.37	0.15	0.47	0.45	0.37
α-Caryophyllene	1465	0.56	0.38	0.62	0.34	0.13	0.27	0.32	0.20
Epi-bicyclosesquiphellandrene	1474	0.18	0.59	0.81	0.46	0.16	0.44	0.42	0.32
Germacrene D	1486	0.36	0.26	0.57	0.25	–	–	–	–
β-Selinene	1493	3.47	3.46	4.14	3.30	0.43	1.07	1.82	1.36
α-Selinene	1497	1.23	1.89	3.07	1.93	0.23	0.31	0.73	0.50
Bicyclogermacrene	1508	1.15	1.26	1.89	1.14	0.29	0.51	1.01	0.61
α-Bulnesene	1516	2.10	1.28	2.40	1.21	0.29	0.48	0.75	0.48
γ-Cadinene	1525	0.80	1.15	1.34	1.03	0.40	0.93	1.40	1.10
δ-Cadinene	1533	0.23	0.43	0.47	0.35	0.09	0.23	0.30	0.22
Nerolidol	1570	1.03	0.66	2.26	1.23	0.51	0.42	1.01	0.62
Spathulenol	1587	0.13	0.05	0.11	0.08	–	–	–	–
Virdiflorol	1592	0.35	0.12	0.49	0.18	0.38	0.37	0.03	0.36
Caryophyllene oxide	1597	0.13	0.15	0.14	0.19	0.10	0.16	0.41	0.16
Humulene epoxide II	1608	0.09	0.17	–	–	0.07	0.12	0.12	0.14
α-Gurjunene	1630	0.10	0.36	–	–	0.58	0.73	0.98	0.92
β-Eudesmol	1656	2.73	2.03	4.41	3.77	4.71	4.51	5.60	5.92
α-Cadinol	1670	0.59	0.42	0.12	0.91	0.86	0.95	0.77	0.97
1-Tetradecanol	1674	0.24	0.09	1.09	0.87	–	–	–	–
α-Bisabolol	1687	0.20	0.15	0.11	0.09	–	–	–	–
(E,Z)-Farnesol	1742	0.45	0.30	0.97	0.05	–	–	–	–
Identified – Zidentyfikowano		98.01	98.65	95.08	97.99	98.45	95.36	93.46	93.65

– Not found – Nie wykryto.
RI – Retention index – Indeks retencji, HD – Hydrodistillation – Hydrodestylacja, WSD – Water-steam distillation – Destylacja z parą wodną.

In the oils obtained from inflorescences by hydrodistillation (HD) forty-two compounds were identified, representing 98.01–98.65% of the oil, while in the oils obtained by water--steam distillation (WSD) thirty-eight compounds were identified, representing 95.08–97.99% of the oil. A total of thirty-eight constituents, representing 93.46–98.45% of the oil, were identified in the oils isolated from leaves with stems by HD and WSD methods.

In the essential oils of inflorescences obtained by HD and WSD, main components were (E)-methyl cinnamate (36.17 and 22.27% for HD, 31.36 and 26.89% for WSD in 2010 and 2011, respectively), linalool (23.33 and 44.44%, 22.23 and 37.88%), estragole (methyl chavicol) (1.71 and 7.91%, 0.98 and 8.26%), eugenol (8.16 and 0.82%, 7.05 and 0.58%), β-selinene (3.47 and 3.46%, 4.14 and 3.30%), β-eudesmol (2.73 and 2.03%; 4.41 and 3.77%) and (Z)-methyl cinnamate (3.37 and 1.68%; 2.30 and 1.47%), respectively.

In the essential oils of leaves with stems (E)-methyl cinnamate (45.58 and 28.19% for HD, 35.53 and 26.16% for WSD in 2010 and 2011, respectively), linalool (21.61 and 35.12%, 23.76 and 35.52%), (Z)-methyl cinnamate (7.52 and 4.76%, 5.74 and 4.16%), β-eudesmol (4.71 and 4.51%, 5.60 and 5.92%), estragole (1.11 and 5.80%, 1.48 and 6.22%) and eugenol (4.84 and 1.32%, 2.63 and 1.06%) were the major components obtained by two different distillation methods.

According to the results shown in Table 2, the major constituents of the essential oils obtained by hydrodistillation and water-steam distillation from inflorescences and leaves with stems were qualitatively identical, although the highest content of β-selinene was found in the oils isolated from inflorescences. Chavicol, cis-β-terpineol, 4-acetyl-1-methylcyclohexene, isoestragole and exo-2-hydroxycineole acetate were detected only in the oils from leaves with stems, while (E)-β-ocimene, trans-geraniol, carvacrol, α-cubebene, germacrene D, spathulenol, 1-tetradecanol, α-bisabolol and (E,Z)-farnesol were found only in the oils from inflorescences.

The differences between the two distillation methods are more clearly revealed in comparison of grouped contents of essential oils obtained by HD and WSD methods (Table 3).

Table 3. Main classes of compounds identified in *Ocimum basilicum* L. 'Cinnamon' essential oils
Tabela 3. Główne klasy związków zidentyfikowanych w olejkach eterycznych z *Ocimum basilicum* L. odmiany 'Cinnamon'

	Inflorescences – Kwiatostany				Leaves with stems – Liście z łodygami			
	HD		WSD		HD		WSD	
	2010	2011	2010	2011	2010	2011	2010	2011
Monoterpene hydrocarbons Węglowodory monoterpenowe	0.09	0.10	0.07	0.09	–	–	–	–
Oxygenated monoterpenes Utlenione monoterpeny	28.16	48.33	25.40	41.47	25.06	40.59	28.07	40.16
Sesquiterpene hydrocarbons Węglowodory seskwiterpenowe	12.74	13.35	18.22	11.86	3.29	6.35	9.04	6.90
Oxygenated sesquiterpenes Utlenione seskwiterpeny	5.70	4.05	8.61	6.50	6.63	6.53	7.94	8.17
Phenylpropanoids Fenylopropanoidy	51.08	32.73	41.69	37.20	63.02	41.46	48.07	38.08
Others Inne	0.24	0.09	1.09	0.87	0.45	0.43	0.34	0.34

Explanations see Table 2 – Objaśnienia zob. tab. 2.

The hydrodistillation method offered higher percentage amounts of phenylpropanoids (51.08 and 32.73% for inflorescences; 63.02 and 41.46% for leaves with stems in 2010 and 2011, respectively) than the water-steam distillation method (41.69 and 37.20% for inflorescences; 48.07 and 38.08% for leaves with stems), while the water-steam distillation method offered higher amounts of sesquiterpene hydrocarbons (18.22 and 11.86% for inflorescences; 9.04 and 6.90% for leaves with stems) and oxygenated sesquiterpenes (8.61 and 6.50% for inflorescences; 7.94 and 8.17% for leaves with stems).

Inflorescences and leaves with stems of *O. basilicum* 'Cinnamon' mainly consisted of phenylpropanoids, followed by oxygenated monoterpenes and sesquiterpene hydrocarbons. The essential oils isolated from inflorescences were richer in oxygenated monoterpenes and sesquiterpene hydrocarbons while the oils obtained from leaves with stems were richer in phenylpropanoids. Moreover, monoterpene hydrocarbons were present in trace (0.07–0.10%) only in the inflorescence oils.

The statistical analysis of the results presented in Table 4 shows significant differences between the content of essential oil constituents.

Table 4. Content of main essential oil constituents in inflorescences of *Ocimum basilicum* L. 'Cinnamon'
Tabela 4. Zawartość głównych składników olejku eterycznego z kwiatostanów bazylii pospolitej odmiany 'Cinnamon'

Essential oil constituent (factor I) Składnik olejku eterycznego (czynnik I)	Distillation method (factor II) – Metoda destylacji (czynnik II)								
	2010			2011			2010–2011		
	HD	WSD	mean średnia	HD	WSD	mean średnia	HD	WSD	mean średnia
Eucalyptol	0.70	0.72	0.71	1.20	1.28	1.24	0.95	1.00	0.98
Linalool	23.33	22.23	22.78	44.44	37.88	41.16	33.89	30.06	31.97
Terpinen-4-ol	1.08	0.62	0.85	0.91	0.79	0.85	1.00	0.71	0.85
Estragole	1.71	0.98	1.34	7.91	8.26	8.09	4.81	4.62	4.72
(Z)-Methyl cinnamate	3.37	2.30	2.83	1.68	1.47	1.58	2.53	1.89	2.21
Eugenol	8.16	7.05	7.61	0.82	0.58	0.70	4.49	3.82	4.15
Isoeugenol	1.67	–	0.84	0.05	–	0.03	0.86	–	0.43
(E)-Methyl cinnamate	36.17	31.36	33.77	22.27	26.89	24.58	29.22	29.13	29.17
α-Guaiene	1.06	1.19	1.13	1.00	0.76	0.88	1.03	0.98	1.00
β-Selinene	3.47	4.14	3.81	3.46	3.30	3.38	3.47	3.72	3.59
α-Selinene	1.23	3.07	2.15	1.89	1.93	1.91	1.56	2.50	2.03
Bicyclogermacrene	1.15	1.89	1.52	1.26	1.14	1.20	1.21	1.52	1.36
α-Bulnesene	2.10	2.40	2.25	1.28	1.21	1.25	1.69	1.81	1.75
γ-Cadinene	0.80	1.34	1.07	1.15	1.03	1.09	0.98	1.19	1.08
Nerolidol	1.03	2.26	1.65	0.66	1.23	0.94	0.85	1.75	1.30
β-Eudesmol	2.73	4.41	3.57	2.03	3.77	2.90	2.38	4.09	3.24
α-Cadinol	0.59	0.12	0.36	0.42	0.91	0.66	0.51	0.52	0.51
Mean – Średnia	5.32	5.06	5.19	5.44	5.44	5.44	5.38	5.25	5.31
$LSD_{\alpha = 0.05}$ for factor I $NIR_{\alpha = 0.05}$ dla czynnika I	0.323			0.302			0.212		
$LSD_{\alpha = 0.05}$ for factor II $NIR_{\alpha = 0.05}$ dla czynnika II	0.046			n.s.			0.041		
$LSD_{\alpha = 0.05}$ for interaction I x II – $NIR_{\alpha = 0.05}$ dla interakcji I x II	0.191			0.245			0.168		

Explanations see Table 1, 2 – Objaśnienia zob. tab. 1, 2.

In both years of the study the highest concentration was noted for (E)-methyl cinnamate and linalool. Moreover, mean results of both years of the experiment proved, that the distillation method had a significant effect on the content of essential oil constituents. Higher amounts were found for hydrodistillation in comparison with water-steam distillation method, on average by 0.13%. Statistical analysis also showed the significance of the interaction between both of the experimental factors. According to the method of hydrodistillation the highest values (mean for the two years) were noted for linalool, (E)-methyl cinnamate, estragole (methyl chavicol), eugenol, β-selinene, β-eudesmol, (Z)-methyl cinnamate and α-selinene. Similar results were obtained using water-steam distillation method. However, comparing both methods of distillation, the content of linalool, (E)-methyl cinnamate, estragole, eugenol and (Z)-methyl cinnamate was higher for HD method, while the content of β-selinene, β-eudesmol and α-selinene – for WSD method.

The statistical analysis of the results given in Table 5 shows significant differences between content of essential oil constituents.

Table 5. Content of main essential oil constituents in leaves with stems of *Ocimum basilicum* L. 'Cinnamon'
Tabela 5. Zawartość głównych składników olejku eterycznego z liści i łodyg bazylii pospolitej odmiany 'Cinnamon'

Essential oil constituent (factor I) Składnik olejku eterycznego (czynnik I)	Distillation method (factor II) – Metoda destylacji (czynnik II)								
	2010			2011			2010–2011		
	HD	WSD	mean średnia	HD	WSD	mean średnia	HD	WSD	mean średnia
Eucalyptol	1.20	1.75	1.47	1.97	1.83	1.90	1.59	1.79	1.69
Linalool	21.61	23.76	22.68	35.12	35.52	35.32	28.37	29.64	29.00
Terpinen-4-ol	0.61	0.62	0.61	1.01	0.91	0.96	0.81	0.77	0.79
Estragole	1.11	1.48	1.29	5.80	6.22	6.01	3.46	3.85	3.65
(Z)-Methyl cinnamate	7.52	5.74	6.63	4.76	4.16	4.46	6.14	4.95	5.55
Eugenol	4.84	2.63	3.73	1.32	1.06	1.19	3.08	1.85	2.46
Isoeugenol	3.15	2.40	2.78	0.21	0.14	0.17	1.68	1.27	1.48
(E)-Methyl cinnamate	45.58	35.53	40.55	28.19	26.16	27.17	36.89	30.85	33.87
β-Selinene	0.43	1.82	1.13	1.07	1.36	1.21	0.75	1.59	1.17
α-Selinene	0.23	0.73	0.48	0.31	0.50	0.41	0.27	0.62	0.44
Bicyclogermacrene	0.29	1.01	0.65	0.51	0.61	0.56	0.40	0.81	0.61
α-Bulnesene	0.29	0.75	0.52	0.48	0.48	0.48	0.39	0.62	0.50
γ-Cadinene	0.40	1.40	0.90	0.93	1.10	1.02	0.67	1.25	0.96
Nerolidol	0.51	1.01	0.76	0.42	0.62	0.52	0.47	0.82	0.64
α-Gurjunene	0.58	0.98	0.78	0.73	0.92	0.82	0.66	0.95	0.80
β-Eudesmol	4.71	5.60	5.15	4.51	5.92	5.21	4.61	5.76	5.19
α-Cadinol	0.86	0.77	0.81	0.95	0.97	0.96	0.91	0.87	0.89
Mean – Średnia	5.25	4.90	5.08	4.95	4.93	4.94	5.11	4.92	5.01
$LSD_{\alpha = 0.05}$ for factor I $NIR_{\alpha = 0.05}$ dla czynnika I	0.996			2.087			1.063		
$LSD_{\alpha = 0.05}$ for factor II $NIR_{\alpha = 0.05}$ dla czynnika II	0.209			n.s.			n.s.		
$LSD_{\alpha = 0.05}$ for interaction I x II – $NIR_{\alpha = 0.05}$ dla interakcji I x II	0.887			n.s.			0.816		

Explanations see Table 1, 2 – Objaśnienia zob. tab. 1, 2.

The highest values were noted for (E)-methyl cinnamate and linalool. The significance of the distillation method was observed only in the first year of the study – significantly higher content of the main constituents of essential oil was noted for hydrodistillation in comparison with water-steam distillation method. Moreover, mean results of both study years shows the significance of the interaction between both of the experimental factors. According to the method of hydrodistillation the highest values (mean for the two years) were noted for (E)-methyl cinnamate, linalool, (Z)-methyl cinnamate, β-eudesmol and estragole. Similar results were obtained using water-steam distillation method. However, comparing both methods of distillation, the content of (E)-methyl cinnamate and (Z)-methyl cinnamate was higher for HD method, while the content of linalool, β-eudesmol and estragole – for WSD method.

DISCUSSION

The composition of the essential oils isolated from aerial parts of *O. basilicum* 'Cinnamon' has been previously investigated by several authors. Basil 'Cinnamon'cultivated in Colombia contained mainly (E)-methyl cinnamate (74.52%), methyl chavicol (estragole) (12.33%), (Z)-methyl cinnamate (5.27%) and linalool (4.44%) – Viña and Murillo (2003). De Masi et al. (2006) found linalool (40.65%), methyl cinnamate (23.91%), methyl chavicol (10.32%), 1,8-cineole (eucalyptol) (3.93%) and eugenol (2.64%) as the main components of *O. basilicum* 'Cinnamon' from Italy. (E)-Methyl cinnamate (29.90 and 53.78% in 2005 and 2006, respectively), linalool (26.54 and 18.89%) and methyl chavicol (7.79%) were reported as the main components of basil 'Cinnamon'cultivated in south-eastern Poland by Nurzyńska-Wierdak (2007). Lachowicz et al. (1997) found methyl cinnamate (28.11%), linalool (27.3%), methyl chavicol (6.8%) and eugenol (3.9%) as the main components of the same cultivar grown in Australia.

The comparison of our results with earlier studies showed that the percentage of methyl chavicol (estragole) in the oils isolated from inflorescences and leaves with stems of sweet basil (*Ocimum basilicum* L.) 'Cinnamon' using two distillation methods was lower than that reported by De Masi et al. (2006) and Viña and Murillo (2003). Although, the content of linalool found in the oils obtained from inflorescences by hydrodistillation and leaves with stems by water-steam distillation was higher compared to the results obtained by Lachowicz et al. (1997) and Viña and Murillo (2003). Similarly, the content of (E)-methyl cinnamate found in our oils isolated by hydrodistillation was higher compared to the results obtained by De Masi et al. (2006). The higher concentration of 1,8-cineole (eucalyptol) we found in the leaves-stems oils (both distillation methods), but it was lower compared to the results reported by De Masi et al. (2006).

Due to the high content of linalool and (E)-methyl cinnamate in the oils isolated from inflorescences and leaves with stems of basil 'Cinnamon' by two distillation methods, it can be stated that the studied oils may found commercial applications in perfumed hygiene products. Moreover, both the oils (from inflorescences and leaves with stems) may found wide industrial application.

According to the international standards, the product is commercially acceptable when its essential oil concentration is higher than 0.4% (Wogiatzi et al. 2011). Most of the essential oil of *O. basilicum* 'Cinnamon' is concentrated in the inflorescences (1.80–2.40%), although the content of oil in leaves with stems (0.80–1.20%) is also high, which made them suitable for industrial purposes.

Essential oil with linalool and methyl chavicol (estragole) as the main components, is often considered to have the finest flavor and the highest quality (Bowes and Zheljazkov 2004). Hydrodistillation, which offer essential oil with high percentage amounts of phenylpropanoids (methyl cinnamate, estragole, eugenol) and monoterpene alcohol – linalool, seems to be an excellent method of oil isolation from basil inflorescences. However, water-steam distillation is better in case of oil isolation from *O. basilicum* 'Cinnamon' leaves with stems, as it offer essential oil with high content of linalool and estragole (methyl chavicol).

CONCLUSIONS

Based on the obtained data it was proved that the distillation method did not significantly affect the essential oil content, but affect its chemical composition. In case of oils obtained from inflorescences of sweet basil 'Cinnamon', the content of linalool, (E)-methyl cinnamate, estragole, eugenol and (Z)-methyl cinnamate was higher in the oil isolated by hydrodistillation, while the content of β-selinene, β-eudesmol and α-selinene was higher in the oil isolated by water-steam distillation. Similarly, in case of oils obtained from basil leaves with stems, the content of (E)-methyl cinnamate and (Z)-methyl cinnamate was higher in the oil obtained by hydrodistillation, while the content of linalool, β-eudesmol and estragole was higher in the oil isolated by water-steam distillation.

REFERENCES

Abduelrahman A.H.N., Elhussein S.A., Osman N.Al., Nour A.H. 2009. Morphological variability and chemical composition of essential oils from nineteen varieties of basil (*Ocimum Basilicum* L.) growing in Sudan. Int. J. Chem. Technol. 1(1), 1–10.

Adams R.P. 2007. Identification of essential oil components by gas chromatography/ mass spectrometry. 4th edition. Allured Publishing Corporation, Illinois, USA.

Bonnardeaux J. 1992. The effect of different harvesting methods on the yield and quality of basil oil in the Ord River irrigation area. J. Oil Res. 4, 65–69.

Bowes K.M., Zheljazkov V.D. 2004. Factors affecting yields and essential oil quality of *Ocimum sanctum* L. and *Ocimum basilicum* L. cultivars. J. Am. Soc. Hort. Sci. 129(6), 789–794.

Chalchat J.-C., Özcan M.M. 2008. Comparative essential oil composition of flowers, leaves and stems of basil (*Ocimum basilicum* L.) used as a herb. Food Chem. 110, 501–503.

Charles D.J., Simon J.E. 1990. Comparison of extraction methods for the rapid determination of essential oil content and composition of basil. J. Am. Soc. Hort. Sci. 115, 458–462.

Davis P. 2005. Aromatherapy an A-Z. London, Vermilion, 52.

De Masi L., Siviero P., Esposito C., Castaldo D., Siano F., Laratta B. 2006. Assessment of agronomic, chemical and genetic variability in common basil (*Ocimum basilicum* L.). Eur. Food Res. Technol. 223, 273–281.

Dube S., Upadhyay P.D., Tripathi S.C. 1989. Antifungal, physicochemical, and insect repelling activity of the essential oil of *Ocimum basilicum*. Can J. Bot. 67, 2085–2087.

Dzida K. 2010. Biological value and essential oil content in sweet basil (*Ocimum basilicum* L.) depending on calcium fertilization and cultivar. Acta Sci. Pol. Hort. Cult. 9(4), 153–161.

Gardner J.A. 1998. Herbs in bloom: a guide to growing herbs as ornamental plants. Portland, Timber Press, Inc., USA, 221–222.

Hassanpouraghdam M.B., **Gohari G.R., Tabatabaei S.J., Dadpour M.R.** 2010. Inflorescence and leaves essential oil composition of hydroponically grown *Ocimum basilicum* L. J. Serb. Chem. Soc. 75, 1361–1368.

Hussain A.I., Anwar F., Sherazi S.T.H., Przybylski R. 2008. Chemical composition, antioxidant and antimicrobial activities of basil (*Ocimum basilicum* L.) essential oils depends on seasonal variations. Food Chem. 108(3), 986–995.

Lachowicz K.J., Jones G.P., Briggs D.R., Bienvenu F.E., Palmer M.V., Mishra V., Hunter M.M. 1997. Characteristics of plants and plant extracts from five varieties of basil (*Ocimum basilicum* L.) grown in Australia. J. Agric. Food Chem. 45, 2660–2665.

Nur Ain A.H., Zaibunnisa A.H., Halimathon Zahrah M.S., Norashikin S. 2013. An experimental design approach for the extraction of lemongrass (*Cymbopogon citratus*) oleoresin using pressurized liquid extraction (PLE). Int. Food Res. J. 20(1), 451–455.

Nurzyńska-Wierdak R. 2007. Evaluation of morphological and developmental variability and essential oil composition of selected basil cultivars. Herba Polon. 53(3), 256–261.

Ogden E. 1990. Growing & using basil. A Storey Country Wisdom Bull. A 119 ,13–15.

Raghavan S. 2007. Handbook of spices, seasonings and flavourings. 2nd edition. Boca Raton, USA, CRC Press, 70–73.

Rahmat A., Edrini S., Ismail P., Hin T.Y.Y., Abu Bakar M.F. 2006. Chemical constitu-ents, antioxidant activity and cytotoxic effects of essential oil from *Strobilanthes crispus* and *Lawsonia inermis*. J. Biol. Sci. 6(6), 1005–1010.

Reuveni R., **Fleischer A., Putievsky E.** 1984. Fungistatic activity of essential oils from *Ocimum basilicum* chemotypes. Phytopath. Z. 110(1), 20–22.

Sajjadi S.E. 2006. Analysis of the essential oils of two cultivated basil (*Ocimum basilicum* L.) from Iran. Daru J. Pharm. Sci. 14(3), 128–130.

Sellar W. 2005. The dictionary of essential oils. London, Vermilion, 20–21.

Suppakul P., Miltz J., Sonneveld K., Bigger S.W. 2003. Antimicrobial properties of basil and its possible application in food packaging. J. Agric. Food Chem. 51, 3197–3207.

Švecova E., Neugebauerova J. 2010. A study of 34 cultivars of basil (*Ocimum* L.) and their morphological, economic and biochemic characteristics, using standard descriptors. Acta Univ. Sapientiae, Ser. Alimentaria 3, 118–135.

Tahira R., Rehan T., Ata-ur-Rehman, Naeemullah M. 2013. Variation in bioactive compounds in different parts of lemon basil (*Ocimum Basilicum var. citriodorum*). The Experimental 17(2), 1184–1190.

Tajidin N.E., Ahmad S.H., Rosenani A.B., Azimah H., Munirah M. 2012. Chemical composition and citral content in lemongrass (*Cymbopogon citratus*) essential oil at three maturity stages. Afr. J. Biotechnol. 11(11), 2685–2693.

Telci I., Bayram E., Yilmaz G., Arci B. 2006. Variability in essentials oil composition of Turkish basils (*Ocimum Basilicum* L.). Biochem. Syst. Ecol. 34, 489–497.

Viňa A., Murillo E. 2003. Essential oil composition from twelve varieties of basil (*Ocimum* spp.) grown in Colombia. J. Braz. Chem. Soc.14(5), 744–749.

Wogiatzi E., Papachatzis A., Kalorizou H., Chouliara A., Chouliaras N. 2011. Evaluation of essential oil yield and chemical components of selected basil cultivars. Biotechnol. Biotechnol. 25, 2525–2527.

Zheljazkov V.O., Cantrell Ch.L., Tekwani B., Khan S.I. 2008. Content, composition, and bioactivity of the essential oils of three basil genotypes as a function of harvesting. J. Agric. Food Chem. 56, 380–385.

Abstract. The chemical composition of the essential oils isolated by hydrodistillation and water--steam distillation from inflorescences and leaves with stems of basil (*Ocimum basilicum* L.) 'Cinnamon' were investigated by GC-MS method. (E)-Methyl cinnamate, linalool and estragole were the dominating components in the all analyzed oil samples. Although, comparing both methods of distillation it was proved, that in case of oils obtained from inflorescences, the content of linalool, (E)-methyl cinnamate, estragole, eugenol and (Z)-methyl cinnamate was higher in the oil isolated by hydrodistillation, while the content of β-selinene, β-eudesmol, and α-selinene was higher in the oil isolated by water-steam distillation. Similarly, in case of oils obtained from leaves with stems, the higher content of (E)-methyl cinnamate and (Z)-methyl cinnamate was found in the oil isolated by hydrodistillation, while the content of linalool, β-eudesmol and estragole was higher in the oil isolated by water-steam distillation.