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EFFECT OF PLANTATION ESTABLISHMENT AND RAW MATERIAL STABILIZATION ON THE USEFULL TRAITS OF LOVAGE LEAVES (*Levisticum officinale* Koch.)

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Abstract. Lovage is a popular medicinal and spice plant. Its usable organs are roots, fruits and leaves. The aim of this study was to evaluate the influence of plantation establishment and different methods of lovage leaves stabilization on its functional characteristics, including the essential oil content, chemical composition and quality of odor. Higher content of essential oil was determined in leaves harvested from plantations established with seedlings in comparison to the one from late fall sowing (respectively: 0.890 ml·100 g^{-1}, 0.755 ml·100 g^{-1}). Freezing, oven-drying and freeze-drying resulted in a decrease of this substance content. Compounds present in the tested essential oils were monoterpenes and their oxygen-containing derivatives (an average of 80.2%). The main component in the analysed essential oils was the α-terpinyl acetate (average 36.6%). Percentage of α-terpinyl acetate, (Z)-ligustylid, (Z)-butylidenoftalid, (E)-butylidenoftalid and (E)-ligustylid increased under the used stability methods. Appearance in the oils collected from stabilized raw materials trans-p-menth-2-en-1-ol, cis-β-elemene, and butylophtalide (compounds that were absent in the essential oil from fresh raw material) was also observed.

Key words*:* lovage, freezing, oven-drying, freeze-drying, essential oil, sensory analysis

INTRODUCTION

Lovage (*Levisticum officinale* Koch.) is a popular plant from Apiaceae family with different utility values. Its roots and leaves have been used in herbal medicine as well as pharmaceutical, cosmetic and food industries in many countries for a long time. It is also often used in traditional Polish cuisine [Justensen and Knuthsen 2001, Andruszczak

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2004, Ezz El-Din and Hendawy 2010]. Its addition to meals regulates the digestive processes and the activation of the appropriate enzymes to accelerate nutrient absorption. The object of this work are lovage leaves. Many authors consider that in this organ there are many biological active compounds among which essential oil is indicated as the most important component [Bylaite et al. 2000, Novák and Németh 2002, Majchrzak and Kamiński 2004, Rizi et al. 2007, Wojdyło et al. 2007]. Unfortunately this raw material is characterized by low capacity of the storage. In Poland massive supply of lovage leaves from May to October is observed. Therefore, in order to ensure access to it through all the year, different methods to extend the ability of a consumer are used. The easiest way of stabilization treatment of herbs, including spices, used for years, is a drying method. As a result of this process the water content in the product is reduced (up to about 10–12%), which protects them against microbial and mold growth, and inhibits the enzymatic and non-enzymatic type of conversion. However, even in strongly dehydrated raw materials adverse changes occur (oxidation, a gradual denaturation of protein, fiber, crystallization, etc.) lowering their quality so opportune method for stability of fresh plants is looked for, that would help solve the above problem. Very interesting seems to be the method of stability through freeze-drying and freezing. Studies on the effects of freeze-drying and freezing in the herbal spices stabilization mainly present knowledge on the evaluation of that methods of color and vitamin C content as well as nitrates, nitrites, macro-and micronutrients content [Giannakourou and Taoukis 2003, Ali Sahari et al. 2004, Słupski et al. 2005]. However there is little information how these ways of plant stabilization affected on the essential oil content, especially in the case of the Apiaceae family.

The aim of this study was to assess the effect of growing conditions and method of the lovage leaves stabilization on the functional characteristics of this raw material, including the essential oil content and its chemical composition as well as its sensory characteristics.

MATERIAL AND METHODS

Materials and establishing of experiment. The object of researches were aboveground parts of lovage (leaves blades and petioles). The experiment was performed on the mapped out experimental plots of the Department of Vegetable and Medicinal Plants, on the heavy alluvial soil with a 1.9–2.3% content of organic matter, 2008 and 2009. During the cultivation, fertilization at the rate of 70 kg N, 40 kg P_2O_5 and 150 kg K_2O per hectare was applied. In both years of researches two methods of plantation establishing were used:

A) sowing of seeds directly to the field,

B) seedlings transplanting.

Ad A) – the seeds were sown in the second decade of November (late fall sowing) on plots of 10 m^2 at the spacing of 20 cm. In each plot 100 g of seeds were sown. After germination the plants was thinning, leaving about 300 plants per plot.

Ad B) – to prepare seedlings, seeds were sown in greenhouse at the beginning of March into multiplates filled with peat substrate. When the seedlings had 2–3 regular leaves, they were planted on the plots of 10 m^2 at 25 × 25 cm spacing. In both years of the experiment plants were transplanted in the second decade of April.

The field experiment was carried out by randomized blocks design with four replications.

Raw material collecting. The herb was collected, for both method of cultivation, when plants reached a height of 20–30 cm. Plants were cut at a height about 5–10 cm above ground. For plantation established from late fall sowing the leaves harvest was carried out in second decade of June, for plantation established from sedlings – in first decade of July.

Stabilization method used. After harvesting the raw materials of the four plots were mixed and then divided for four parts. First part was subjected to chemical analysis directly after collecting (fresh matter). The second part of raw material was dried in dryer chamber at temperature of 35–40°C (until a 14% humidity). Till the moment of performing of quality analysis (about two months) it was stored in closed paper bags in airy dry place.

The third part of raw material was freeze-dried. Directly after the harvest leaves were deep-frozen at -70°C (24 hours). The frozen raw material was placed in a AGA Analytical dehydreter and subjected to freeze-drying process at temperature -49°C, pressure 0.0043 hPa. Freeze-dried raw material was placed in plastic bags for two months.

The fourth part was frozen. Freezing was performed in two stages: raw material directly after collecting was chopped, placed in plastic bags and held at temperature of -75°C by the period of 48 hours, next the bags were moved to a freezer where temperature -18°C to -22°C was kept. Evaluation of frozen herb was carried out after three months from the moment of raw material collecting.

Leaves quality evaluation. In investigated samples content of essential oil and its chemical composition were determined. Sensory evaluation of plant material using quantitative descriptive analysis (QDA) method was also carried out.

The content of essential oil. Essential oil content was determined by steam distillation according to a method described in Polish Pharmacopoeia, 6th ed. (2002). 100 g of fresh and frozen leaves and 50 g of dried and freeze-dried raw material for the distillation were used.

Chromatographic analysis. Essential oils were analyzed by GC-MS-FID. The analyses were performed on a Trace GC Ultra coupled with DSQII mass spectrometer (Thermo Electron). A simultaneous GC-FID and MS analysis was performed using a MS-FID splitter (SGE Analytical Science). Mass range was 33–550 amu, ion source-heating: 200°C, ionization energy: 70eV. Operating conditions: capillary column Rtx-1 MS (60 m × 0.25 mm i.d., film thickness 0.25 μm), temperature program: 60 (0.5 min) – 300°C (30 min) at 4°C/min. Injector and detector temperatures were 320°C and 310°C, respectively. Carrier gas was helium at a flow rate of 1.5 mL/min.

Identification of volatile compounds. Identification of compounds from the essential oils was based on a comparison of their mass spectra with spectra stored in the computer library NIST 98.1, Wiley Registry of Mass Spectral Data, 8th edn, and MassFinder 3.1, along with relative retention indices (RI, non-polar column). Qualitative analysis was performed for the essential oils from raw materials harvested in 2008.

Quantitative descriptive analysis. QDA method was used for odor of fresh, frozen, dry and freeze-dry lovage leaves. Before approaching to evaluation, the panelists made a list of attributes for investigated parameters (tab. 4). For all forms of raw material the same attributes were applied. During evaluation of odor the raw material was placed into plastic containers of capacity 50 ml. Weight of sample was about 0.5 g, in terms the dry mass of raw material. Samples were covered with Petri dish and were given for evaluation after about 30 min from their preparing. The panel consisted of 15 persons. Analysis of odor was performed during 2 independent sessions in two repeats. The overall intensity of odor and intensity of each attribute were evaluated with scale of range 10 contractual units, including suitable boundary attributes [Baryłko-Pikielna and Matuszewska 2009].

Results concerning the content of essential oil and sensory analysis were subjected to statistical analysis using the program Statgraphics Plus 4.1. Significance of differences was evaluated using test HSD – Tukey at the level of significance $\alpha = 0.05$ for essential oil content and at the level of significance $\alpha = 0.001$ or 0.01 or 0.05 or 0.1 for sensory analysis.

RESULTS AND DISCUSSION

In leading herbal plantations, particular attention is paid to the quality of the raw material, which is estimated inter alia on the basis of the content of biologically active compounds present in it. In the case of lovage important component, from a qualitative point of view is the essential oil, which is present in all parts of this plant. In the green part of lovage, which was the object of presented research, content of that constituent ranges from 0.09% to 1.4% [Rizi et al. 2007, Raal et al. 2008, Mirjalili et al. 2010]. Such a large divergence of results, might result from differences in the age of plant, its stage of development and the growing conditions [Hogg et al. 2001, Novák and Németh 2002, Ezz El-Din and Hendawy 2010]. In the present study in the lovage leaves content of essential oil was on average $0.82\ \text{ml}\cdot 100\ \text{g}^{-1}$. The method of plantation establishment influenced the content of this component significantly. In the leaves harvested from plantation established with seedlings the higher essential oil content was obtained than the raw material harvested from plantation from late fall sowing (tab. 1).

It is consistent with the results obtained by Novák and Németh [2002], who claimed that delaying harvest positively influences on this parameter. Results obtained in this experiment showed that content of essential oil in lovage leaves depended also on the method of raw material stabilizing. Fresh raw material was characterized the highest content of essential oil. Taking into account only preserved plant material, the highest content of essential oil was in oven-dried leaves (an average of $1.63\ \text{ml}\cdot 100\ \text{g}^{-1}$). Lower content of this substance, in comparison with oven-dried leaves, in freeze-drying material was characterized ($1.28\ \text{ml}\cdot 100\ \text{g}^{-1}$), the least in frozen one was determined ($1.11\ \text{ml}\cdot 100\ \text{g}^{-1}$) (tab. 2).

Many studies concerning the effect of plant conservation on the content of volatiles in spices leaf show similar relations [Asekun et al. 2007, Rosłon et al. 2009, Sellami et al. 2011]. An important parameter of essential oil quality is its chemical composition.

Table 1. Influence of plantation establishment on essential oil content in fresh lovage leaves (ml·100 g^{-1})
Tabela 1. Wpływ sposobu zakładania plantacji na zawartość olejku eterycznego w świeżych liściach lubczyku ogrodowego (ml·100 g^{-1})

Method of plantation establishment Sposób założenia plantacji	Year of investigations Rok badań		Mean for method of plantation establishment Średnia dla sposobu założenia plantacji
	2008	2009	
Late fall sowing Siew późnojesienny	0.720 d	0.790 c	0.755 B
Seedlings planting Wysadzanie rozsady	0.860 b	0.920 a	0.890 A
Mean for the year of cultivation Średnia dla roku uprawy	0.790 B	0.855 A	

Means followed by the same letters do not differ significantly – Dane oznaczone taką samą literą nie różnią się istotnie

Table 2. Influence of method of stabilization on essential oil content in lovage leaves (ml·100 g^{-1} dry mass). Results are the mean of two investigation years
Tabela 2. Wpływ metody stabilizacji na zawartość olejku eterycznego w liściach lubczyku ogrodowego (ml·100 g^{-1} bsm). Wyniki stanowią średnią z dwóch lat badań

Method of plantation establishment Sposób założenia plantacji	Form of raw material – Forma surowca				Mean for plantation establishment Średnia dla sposobu założenia plantacji
	fresh świeży	freezed mrożony	oven-dried suszony termicznie	freeze-dried liofilizowa-ny	
Late fall sowing Siew późnojesienny	3.56 b	1.03 g	1.69 c	1.40 e	1.92 B
Seedlings planting Wysadzanie rozsady	4.16 a	1.18 f	1.56 d	1.16 f	2.02 A
Mean for form of raw material Średnia dla formy surowca	3.86 A	1.11 D	1.63 B	1.28 C	

Means followed by the same letters do not differ significantly – Dane oznaczone taką samą literą nie różnią się istotnie

Mirjalili et al. [2010] report that the number of identified compounds in essential oil from different parts of lovage ranged from 26 (oil from inflorescences) to 31 (ripening green fruit). In our researches the total of 44 compounds were identified. The tested oils contained from 27 to 41 compounds, which constituted from 93.7% to 99.9% of all the volatile ingredients. The volatile compounds present in investigated oils could be grouped in the following main chemical groups: monoterpenes and oxygenate monoterpenes (they accounted for about 56% to 93% of the identified components) and

Table 3. Composition of essential oil from lovage leaves in dependence of plantation establishment and method of raw material stabilization
Tabela 3. Skład chemiczny olejku eterycznego z liści lubczyku ogrodowego w zależności od sposobu założenia plantacji i metody stabilizacji surowca

Składnik olejku Essential oil component	RI* lit. RI ref.	Fresh leaves Liście świeże				Frozen leaves Liście mrożone				Oven-dried leaves Liście suszone termicznie				Freeze-dried leaves Liście liofilizowane			
		late fall sowing siew późnojesienny		seedlings planting wysadzanie rozsady		late fall sowing siew późnojesienny		seedlings planting wysadzanie rozsady		late fall sowing siew późnojesienny		seedlings planting wysadzanie rozsady		late fall sowing siew późnojesienny		seedlings planting wysadzanie rozsady	
		(%)	RI exp. RI dośw.	(%)	RI exp. RI dośw.	(%)	RI exp. RI dośw.	(%)	RI exp. RI dośw.	(%)	RI exp. RI dośw.	(%)	RI exp. RI dośw.	(%)	RI exp. RI dośw.	(%)	RI exp. RI dośw.
1. Nonane 1. Nonan	900	0.0	-	0.0	-	0.0	-	0.0	-	t	900	0.0	-	0.0	-	0.0	-
2. Tricyclene 2. Tricyklen	927	0.0	-	t**	922	0.0	-	t	922	t	922	t	920	0.0	-	t	920
3. α-Thujene 3. α-Tujen	932	0.1	930	0.1	928	0.1	928	0.1	928	0.1	928	0.1	928	t	928	t	925
4. α-Pinene 4. α-Pinen	936	1.9	933	1.1	932	0.9	933	1.2	932	1.4	932	1.6	932	0.4	932	0.7	932
5. Fenchene 5. Fenchen	941	0.0	-	0.0	-	0.0	-	0.0	-	t	938	0.0	-	0.0	-	0.0	-
6. Camphene 6. Kamfen	950	0.3	946	0.1	946	0.1	946	0.2	946	0.3	946	0.3	947	0.1	946	0.1	946
7. Sabinene 7. Sabinen	973	1.2	968	0.8	968	0.8	968	0.9	968	0.9	968	0.8	968	0.2	968	0.6	968
8. β-Pinene 8. β-Pinen	978	0.4	973	0.3	973	0.2	973	0.3	973	0.4	973	0.3	973	0,1	973	0,2	973
9. 2,3-Dehydro-1,8-cyneole 9. 2,3-Dehydro-1,8-cyneol	980	0.1	981	0.0	-	0.0	-	t	981	0.1	981	t	981	0.4	981	0.2	981
10. Myrcene **10. Myrcen**	987	**5.8**	983	**4.8**	983	**5.0**	983	**6.2**	983	**4.2**	984	**3.7**	983	**1.3**	983	**3.1**	983

11. α-Phellandrene 11. α-Felandren	1002	1.8	999	1.2	999	0.7	999	1.1	999	1.5	999	1.0	999	0.2	999	0.5	999
12. δ-Car-3-ene 12. δ-Car-3-en	1010	0.0	-	t	1007	t	1007	t	1007	t	1007	t	1006	0.0	-	t	1006
13. α-Terpinene 13. α-Terpinen	1013	t	1011	t	1012	t	1012	0.1	1012	0.1	1012	t	1011	0.1	1011	t	1011
14. p-Cymene 14. p-Cymen	1015	0.1	1015	0.1	1015	0.1	1015	0.1	1015	0.2	1015	0.1	1015	0.3	1015	0.1	1019
15. β-Phellandrene **15. β-Felandren**	1023	**44.0**	1023	**29.8**	1024	**24.8**	1023	**28.2**	1026	**39.0**	1029	**27.0**	1023	**9.6**	1024	**16.6**	1024
16. (Z)-β-Ocymene 16. (Z)-β-Ocimen	1029	6.2	1028	4.5	1028	5.9	1028	4.1	1028	0.3	1040	4.4	1028	1.1	1027	2.4	1027
17. (E)-β-Ocymene 17. (E)-β-Ocimen	1041	1.4	1031	0.9	1039	1.1	1031	0.7	1039	0.6	1043	1.0	1031	0.6	1039	0.5	1031
18. γ-Terpinen 18. γ-Terpinen	1051	0.7	1052	0.2	1052	0.3	1052	0.5	1052	0.6	1053	0.4	1052	0.2	1052	0.1	1052
19. p-Menth-2,4(8)-diene 19. p-Menta-2,4(8)-dien	1077	0.4	1080	0.3	1081	0.7	1082	0.4	1082	0.2	1082	0.4	1082	0.3	1082	0.3	1082
20. Terpinolene 20. Terpinolen	1082	t	1087	0.1	1086	0.1	1086	0.1	1086	0.1	1086	t	1087	0.2	1086	0.1	1086
21. *cis*-p-Menth-2-en-1-ol 21. *cis*-p-Menta-2-en-1-ol	1108	0.0	-	t	1111	0.0	-	t	1111	t	1111	0.0	-	t	1111	t	1111
22. (4E,6Z)-allo-Ocymene 22. (4E,6Z)-allo-Ocimen	1121	1.1	1121	0.9	1120	1.2	1121	1.1	1120	1.1	1121	0.7	1121	0.2	1120	0.5	1120
23. *trans*-p-Menth-2-en-1-ol 23. *trans*-p-Menta-2-en-1-ol	1123	0.1	1124	0.1	1124	0.0	-	t	1128	0.1	1128	0.0	-	t	1128	0.1	1128
24. n-Pentylbenzene 24. n-Pentylbenzen	1150	0.0	-	t	1148	0.0	-	t	1148	t	1148	0.0	-	t	1148	t	1148
25. Dictyotene 25. Diktyoten	1155	0.3	1154	0.3	1153	0.1	1153	0.4	1153	0.3	1153	t	1153	0.1	1153	0.3	1153
26. Cryptone 26. Krypton	1160	0.0	-	0.1	1163	0.0	-	0.1	1163	0.1	1163	0.0	-	0.3	1163	0.2	1163

27. Terpinen-4-ol 27. Terpinen-4-ol	1164	0.1	1168	0.1	1168	0.2	1154	0.1	1168	0.1	1168	t	1168	0.3	1167	0.2	1168
28. α-Terpineol 28. α-Terpineol	1176	0.5	1178	0.9	1178	0.7	1178	0.8	1178	0.5	1179	0.5	1179	0.9	1178	0.8	1178
29. Geraniol 29. Geraniol	1235	0.0	-	t	1238	0.0	-	0.1	1241	0.1	1241	0.0	-	0.2	1238	0.1	1241
30. Bornyl acetate 30. Octan bornylu	1270	0.0	-	t	1274	0.1	1274	0.1	1274	0.1	1274	0.1	1274	0.1	1274	0.1	1274
30. Carvacrol 31. Karwakrol	1278	0.0	-	0.0	-	0.0	-	t	1284	0.0	-	0.0	-	0.1	1285	t	1285
31. α-Terpinyl acetate **31. Octan α-terpinylu**	1335	**26.1**	1337	**37.3**	1339	**39.8**	1337	**34.5**	1343	**34.3**	1346	**42.0**	1337	**38.5**	1340	**40.5**	1340
32. Geranyl acetate 33. Octan geranylu	1362	0.3	1362	0.5	1361	0.4	1362	0.8	1361	0.9	1362	0.4	1362	0.4	1361	0.5	1361
34. *cis*-β-Elemene 34. *cis*-β-Elemen	1381	0.0	-	t	1394	t	1394	0.1	1394	t	1391	0.0	-	t	1390	t	1391
35. γ-Elemene 35. γ-Elemen	1429	0.0	-	t	1434	0.0	-	t	1434	t	1434	0.0	-	t	1434	t	1430
36. α-Humulene 36. α-Humulen	1455	0.0	-	t	1461	0.0	-	0.0	-	t	1460	0.0	-	t	1460	0.0	-
37. Germacrene D 37. Germakren D	1479	0.1	1486	0.1	1486	0.1	1486	0.2	1479	0.2	1487	0.2	1486	0.1	1486	0.1	1486
38. Bicyclogermacrene 38. Bicyklogermakren	1494	0.0	-	t	1490	0.0	-	t	1491	0.0	-	0.0	-	t	1490	t	1490
39. Elemol 39. Elemol	1541	0.0	-	t	1537	0.0	-	t	1537	0.0	-	0.0	-	0.1	1537	t	1537
40. Butylphtalide 40. Butyloftalid	1616	0.0	-	t	1624	0.0	-	t	1620	t	1624	0.0	-	0.1	1624	t	1624
41. (Z)-Butylidenophtalide 41. (Z)-Butulidenoftalid	1644	0.1	1653	0.2	1651	0.2	1651	0.2	1651	0.2	1652	0.2	1651	2.3	1651	0.8	1651

42. (E)-Butylidenophtalide 42. (E)-Butylidenoftalid	1700	0.0	-	0.1	1697	0.1	1657	0.1	1699	0.1	1700	0.1	1698	1.3	1698	0.3	1698
43. (Z)-Ligustilide **43. (Z)-Ligustylid**	1732	**6.6**	1721	**14.0**	1724	**15.6**	1721	**16.2**	1721	**11.0**	1721	**13.9**	1721	**32.1**	1729	**26.7**	1728
43. (E)-Ligustilide 44. (E)-Ligustylid	1782	0.1	1779	0.4	1778	0.3	1779	0.5	1778	0.8	1779	0.6	1779	1.7	1776	0.8	1777
The sum of identified compounds (%) Suma zidentyfikowanych składników (%)		**99.8**		**99.4**		**99.9**		**99.5**		**99.9**		**99.9**		**93.7**		**97.6**	
Number of identified compounds Liczba zidentyfikowanych składników		**27**		**40**		**29**		**41**		**41**		**30**		**40**		**41**	
The sum of monoterpenes and their oxygenated derivatives (%) Zawartość monoterpenów i ich tlenowych pochodnych (%)		**92.6**		**84.1**		**85**		**82.7**		**87.2**		**85.5**		**55.7**		**68.4**	
The sum of sesquiterpenes and their oxygenate derivatives (%) Zawartość seskwiterpenów i ich tlenowych pochodnych (%)		**0.2**		**0.2**		**0.1**		**0.3**		**0.1**		**0.2**		**0.2**		**0.2**	
The sum of phtalide compounds (%) Zawartość ftalidów (%)		**6.8**		**14.7**		**16.2**		**17.0**		**12.1**		**14.8**		**37.6**		**28.6**	

*RI – retention index: ref. – references, exp. – experimental; RI – indeks retencji: lit. – literaturowy, dośw. – doświadczalny
**t – trace amounts; t – ilości śladowe

Table 4. Influence of plantation establishment and method of stabilization on lovage leaves odor (uc)
Tabela 4. Wpływ sposobu założenia plantacji i metody stabilizacji na zapach liści lubczyku ogrodowego (ju)

Attributes of odor Wyróżniki zapachu	Fresh leaves Liście świeże		Frozen leaves Liście mrożone		Oven-dried leaves Liście suszone termicznie		Freeze-dried leaves Liście liofilizowane		Difference for plantation establishment Istotność różnic dla sposobu zakładania plantacji	Difference for leaves stabilization Istotność różnic dla sposobu stabilizacji
	A	B	A	B	A	B	A	B		
Overall intensity of odor Ogólna intensywność zapachu	6.37	6.28	6.44	6.57	5.54	5.87	5.28	6.28	a	a
Sharp, irritates odor Zapach ostry, drażniący	3.28	2.86	3.08	3.64	2.80	3.18	2.31	2.57	ns	ns
Spicy odor Zapach przyprawowy	3.88	4.03	3.25	4.67	4.02	4.13	4.80	4.55	ns	ns
Bitter odor Zapach gorzki	2.09	2.11	1.68	2.10	1.39	1.44	0.63	0.78	ns	xxx
Celery odor Zapach selerowy	3.71	3.83	2.21	4.77	3.54	3.44	4.74	4.54	a	x
Sweet odor Zapach słodki	2.00	2.06	2.36	2.44	2.51	2.25	1.02	2.21	ns	xx
Sour odor Zapach kwaśny	0.75	0.83	1.32	1.69	1.22	1.20	0.50	0.46	x	xxx
Hey odor Zapach sianowy	0.78	0.62	1.32	1.28	4.40	4.41	3.76	2.70	ns	xxx
Sallow odor Zapach ziemisty	0.82	1.07	1.07	0.99	1.67	1.50	0.88	0.32	ns	xx
Else odor Zapach inny	0.26	0.23	0.00	0.00	0.06	0.06	0.21	0.10	ns	x

A – Late fall sowing – siew późnojesienny; B – seedlings planting – wysadzanie rozsady
ns, a, x, xx, xxx, – non significant or significant at $\alpha = 0.1$ or 0.05 or 0.01 or 0.001 – różnice nieistotne lub istotne przy $\alpha = 0{,}1$ lub 0,05 lub 0,01 lub 0,001
(uc) – unit contractual; (ju) – jednostki umowne

phtalides (6.8–37.6%). Phtalides are characteristic ingredients of plants belonging to the *Apiaceae* family [Beck and Chou 2007]. Many studies related to phtalides indicate that they affect the specific smell of raw materials from *Apiaceae* family, they also have very distinct therapeutic properties. Especially is emphasized pharmacological activity of (Z)-ligustilide and (Z)-butylidenephalide, which presence was found, inter alia, in lovage [Gijbels et al. 1982, Jun-Rong, et al. 2007, Jing et al. 2009]. Besides monoterpenes with its oxygenate derivatives and phtalides a few sesquiterpenes and one oxygenate sesquiterpene were also identified.

All the factors used in the experiment affected the content of above chemical groups in the tested essential oils. A higher share of monoterpenes and their oxygenated derivatives was found in oil from the late fall sowing plantation in comparison with plantation established from sedlings. In assessing the influence of the stabilization process on this parameter more monoterpenes and their oxygenated derivatives in fresh raw material was noted, in comparison with the stabilized one (and therefore each process of conservation contributed to the loss of the most volatile components). It has been also observed that with the decreasing share of monoterpenes and oxygenate monoterpenes, phtalides fraction content was increased. Taking into account only the stabilized raw materials, the lowest share of monoterpenes and oxygenate monoterpenes (and thus the highest percentage of phtalide compounds) in oil from freeze-dried plant material was determined. In the case of frozen and oven-dried leaves results were similar and lower in comparison with freeze-dried material.

Many authors reported that among the constituents present in the essential oil from aerial parts of lovage the main compound is α-terpinyl acetate. They evaluate the share of this component from 40.5% to 70.0% [Bylaite et al. 1998, Novák and Németh 2002, Mirjalili et al. 2010]. According to the above authors the next components in respect the percentage share are β-phellandrene, α-pinene and β-pinene. Raal et al. [2008] said that in addition to the above compounds, a high share of that essential oil has (Z)-ligustilide. Rizi et al. [2007] in turn reported that the components with the highest percentage concentration of the oil from the lovage leaves were β-phellandrene, α-terpineole, *cis*-ocimene and dehydro-1,8-cineole. Component with the largest share of the analyzed essential oils was α-terpinyl acetate (an average of 36.6%). Another important components were β-phellandrene (9.6–44.0%), (Z)-ligustilide, (6.6–32.1%), myrcene (1.3–6.2%) and (Z)-β-ocimene (0.3–6.2%). The presence of *cis*-ocimene and dehydro-1,8-cineole was not revealed. The content of other compounds ranged from trace amounts (< 0.05%) to 1.9%. Among these components inter alia, nonane, tricyclene, *cis*-p-menth-2-en-1-ol, n-pentylbenzene, dictyotene and cryptone were determined, which presence in the oil from the lovage leaves was found for the first time.

In the fresh raw material the percentage content of majority components depended on the method of plantation establishment. Taking into account the most important components, the higher percentage share of α-terpinyl acetate and (Z)-ligustilide in oil from the plantation established from transplanted plants, as compared to oil from plants sown directly into field has been observed, while in case of β-phellandrene, myrcene and (Z)-β-ocimene, there was an inverse relationship. The way of the raw material stabilization also affected the chemical composition and content of the components determined in the investigated oils.

The content of such compounds as the α-phellandrene, β-phellandrene, myrcene, (Z)-β-ocimene, decreased in all stabilized raw materials, in comparison with fresh material. In the oils from the lovage leaves collected from plants sown late fall sowing very clear decline in the first three compounds was recorded in the essential oil from freeze-dried material. In the case of (Z)-β-ocimene the largest decline was recorded in the essential oil from oven-dried raw material. Under the influence of used stabilization methods the percentage of α-terpinyl acetate and (Z)-ligustilide increased. Particularly high, more than five times, growth of (Z)-ligustilide in oil from freeze-dried material compared with fresh material was observed. Also in the case of other phtalide compounds presented in the tested oils ((Z)- and (E)-butylidenophtalide, (E)-ligustilide) a very high growth of their content in the oil from freeze-dried raw material was reported. The presence of constituents in the oils from stabilized raw materials that were not present in the essential oil from fresh leaves (such as *trans*-p-menth-2-en-1-ol, *cis*-β-elemene, butylophtalide et al.) was also observed.

There were no observed clear changes in the content of α-phellandrene under the influence of the used stabilizing methods in the oils from the transplanted plants, with the exception of freeze-dried material, wherein the content of this component has considerably decreased. In the case of α-terpinyl acetate, freezing caused a decrease in the content of that constituent, other stabilizing methods have increased content of this component. The content of (Z)-ligustilide also increased due to the stabilization, only in essential oil from oven-dried raw material a slight decline in this component was recorded in comparison with fresh material. There were no noticeable changes in the content of essential oils components with a low share (up to about 1%) under the influence of applied experience factors. The results obtained in this experiment confirm the observations of many authors, who studied the effect of stabilization processes on the quality of volatile raw materials [Venskutonis 1997, Asekun et al. 2007, Figiel et al. 2010, Szumny et al. 2010, Sellami et al. 2011]. These authors emphasize that as a result of this processes changes in the composition and the percentage content of most chemical compounds present in essential oils occur and the smallest changes in frozen raw material are observed, while the oven-drying and freeze-drying bring about a decrease in the concentrations of many of the volatile components, particularly in the case of monoterpenes and their oxygenated derivatives (tab. 3).

Another aspect of the present study was to find the relationship between method of plantation establishment, methods of lovage leaves stabilization and the most important determinant of this spice quality which is the odor. The overall intensity of odor and intensity of all descriptors designated by the experts team, affecting this trait was determined (tab. 4).

The sensory analysis carried out in this experiment showed that the method of plantation establishment significantly influenced only three determinants of odor quality: the overall odor intensity, intensity of celery odor and sour odor. Higher overall intensity of odor and intensity of celery odor (at significance level $\alpha = 0.1$) was characterized by the raw material harvested from plantations established with seedlings, which probably was associated with a higher content of essential oil in that raw material and a higher percentage share of phtalide components, in comparison with the leaves harvested from plantations established from the late fall sowing. The methods of stabilization used in this experiment in different ways influenced the sensory properties of the leaves of lovage. Fresh and frozen raw material was characterized by higher overall intensity of odor in the comparison with that oven- dried and freeze-dried, although frozen leaves contained less essential oil than both forms of dried materials. However freeze-dried and oven-dried leaves gained higher (compared with fresh and frozen raw material) notes for the celery odor as well as spice and hay odor (recognized by a panelist as positive traits) and significantly lower for the odor of bitter, sour and sharp, irritation (considered as negative traits). It is worth noting that both methods of drying (oven-drying and freeze-drying) clearly contributed to the increased perceptibility of hay smell in the raw materials, which can be associated with the formation of coumarin compounds under the influence of drying processes (tab. 4) [Yang et al. 2009].

SUMMARY

Agronomic methods used in the experiment suggest that, in the case of lovage leaves, better raw material (with distinctly higher content of essential oil and higher share of α-terpinyl acetate and (Z)-ligustilide) was obtained from plantations established with seedlings planted in spring.

Stabilization methods used in the experiment affected the quality of the lovage leaves, first of all essential oil content. All applied methods of stabilization cause a significant decrease of essential oil content. Oven-drying was the best method of preserve of lovage leaves. This raw material stabilized by above method characterized by higher essential oil content than the freeze-dried and frozen one. Preservation of lovage leaves also affect the chemical composition of essential oil: increase the percentage of α-terpinyl acetate and (Z)-ligustilide, important from a qualitative point of view components of analyzed essential oils, while decrease content of other important components such as α-phellandrene, β-phellandrene, myrcene, (Z)-β-ocimene. Chemical changes occurring under the influence of the used method of stabilization affect flavour of lovage leaves.

The highest overall intensity of odor have frozen lovage leaves, although they contain less essential oil in comparison with fresh, oven-dried and freeze-dried raw material. On the other hand they received high notes of sharp, irritating and sour odor, which may affect such result. Higher overall intensity of odor have fresh raw material in comparison with oven-dried and freeze-dried one, which is associated with a higher content of essential oil in fresh leaves. Oven-dried and freeze-dried leaves characterized by a higher intensity of celery odor as well as spice and hay odor

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WPŁYW SPOSOBU ZAKŁADANIA PLANTACJI I SPOSOBU STABILIZACJI SUROWCA NA CECHY UŻYTKOWE LIŚCI LUBCZYKU OGRODOWEGO (*Levisticum officinale* Koch.)

Streszczenie. Lubczyk ogrodowy jest popularną rośliną leczniczą i przyprawową, której organami użytkowymi są korzenie, owoce i liście. Celem przeprowadzonych badań była ocena wpływu różnych metod uprawy i stabilizacji liści lubczyku na ich cechy funkcjonalne, w tym zawartość olejku eterycznego, jego skład chemiczny oraz jakość zapachu. Wyższą zawartość olejku eterycznego oznaczono w liściach z plantacji założonej z rozsady, w porównaniu z surowcem zebranym z plantacji z siewu pod grudę (odpowiednio: 0,890 ml·100 g^{-1}, 0,755 ml·100 g^{-1}). Mrożenie, suszenie termiczne i liofilizacja wpłynęły na obniżenie się zawartości tej substancji. Najważniejszymi składnikami obecnymi w testowanych olejkach były monoterpeny i ich tlenowe pochodne (średnio 80,2%). Głównym składnikiem w analizowanych olejkach okazał się octan α-terpinylu (średnio 36,6%). Zawartość octanu α-terpinylu, (Z)-ligustylidu, (Z)-butylidenoftalidu, (E)-butylidenoftalidu i (E)-ligustylidu wzrosła pod wpływem zastosowanych metod stabilizacji, zaobserwowano także pojawienie się w surowcu stabilizowanym *trans*-p-menta-2-en-1-olu, *cis*-β-elemenu i butyloftalidu – związków, które nie były obecne w surowcu świeżym.

Słowa kluczowe: lubczyk, mrożenie, suszenie termiczne, liofilizacja, olejek eteryczny, analiza sensoryczna

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