

Chemical composition of essential oils from *plantago lanceolata* L. leaves extracted by hydrodistillation

Tomáš Bajer¹ · Václav Janda¹ · Petra Bajerová¹ · Daniel Kremr¹ · Aleš Eisner¹ · Karel Ventura¹

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Abstract Extensive traditional use of medical plants leads to research dealing with chemical composition of essential oils. The aim of this work was evaluation of quality of the essential oil and extending of the knowledge about chemical composition of essential oil from ribwort (*Plantago lanceolata* L.) and proportional representation of compounds. Extractions of essential oils from samples of ribwort were performed by hydrodistillation. GC-MS and GC-FID techniques were used for investigation of the qualitative and semi-quantitative content of aromatic compounds in the essential oils, respectively. Major aroma constituents of ribwort leaves were groups of fatty acids 28.0–52.1 % (the most abundant palmitic acid 15.3–32.0 %), oxidated monoterpenes 4.3–13.2 % (linalool 2.7–3.5 %), aldehydes and ketones 6.9–10.0 % (pentyl vinyl ketone 2.0–3.4 %) and alcohols 3.8–9.2 % (1-octen-3-ol 2.4–8.2 %). In relative high amount were identified apocarotenoids (1.5–2.3 %) which are important constituents because of their intense fragrant. The importance is in potential manufacture control of feedstocks before producing of food supplements.

Keywords *Plantago lanceolata* L. · Essential oil · Hydrodistillation · GC-MS · GC-FID

Research highlights

- Hydrodistillation was used for extraction of essential oil from ribwort
- Separation of volatiles was performed by gas chromatography
- Totally, 153 compounds were identified using mass spectra and retention indices
- Semi-quantitative determination was performed by GC-FID

✉ Tomáš Bajer
Tomas.Bajer@upce.cz

¹ Faculty of Chemical Technology, Department of Analytical Chemistry, University of Pardubice, Studentska 573, 532 10 Pardubice, Czech Republic

Introduction

Plantago lanceolata L., a perennial herb belonging to the family *Plantaginaceae*, usually grows to a high about 30–60 cm and produces a rosette of leaves 10–25 cm, sometimes up to 40 cm, long (Kubát 2002). The size of the plant depends on the growth habitats. It occurs as a wild plant in field crops, but due to the large consumption in the pharmaceutical industry is also cultivated.

The plant belongs historically to Eurasia continent, but gradually expanded around the world together with the colonizers from Europe. Plantain has been proclaimed for medicinal usage dating back to the ancient Greeks and Romans, who used it frequently on herpes, skin infections but also as an antidote for rabies. Notable pharmacological properties of *Plantago lanceolata* L. have been reported, such as anti-inflammatory (Beara et al. 2010), antimicrobial activity (Nostro et al. 2000), antioxidant and cytotoxic activity (Beara et al. 2012), anti-tumoural activity (Herbert et al. 1991), and antispasmodic effect (Fleer and Verspohl 2007). Furthermore, *Plantago lanceolata* L. is also included in the diet as herbal tea, which is used in folk medicine for treatment of disorders of the respiratory tract (Fons et al. 2008).

Many constituents of ribwort have been reported, such as phenolic compounds (phenolic acids, flavonoids and coumarins) (Beara et al. 2012; Fons et al. 1999), iridoid glycosides (e.g. catalpol, aucubin) and phenylethanoid glycosides (e.g. verbascoside, plantamajoside) (Adler et al. 1995), volatile components (Fons et al. 1998), etc. But no published study has previously dealt with the chemical composition of the essential oil extracted by hydrodistillation from leaves of *Plantago lanceolata* L.

Our interest was not in the extraction process, but in the quality of the essential oil and extending of our knowledge about chemical composition of essential oil from *Plantago*

Table 1 List of plant material

Sample	Company/producer
<i>Plantago lanceolata</i> L. – sample 1 (crushed leaves)	Dr. Müller Pharma s.r.o. (Hradec Králové, Czech Republic)
<i>Plantago lanceolata</i> L. – sample 2 (crushed leaves)	Valdemar Grešík – Natura s.r.o. (Děčín, Czech Republic)
<i>Plantago lanceolata</i> L. – sample 3 (crushed leaves packed in tea bags)	Megafyt Pharma s.r.o. (Vrané n. Vltavou, Czech Republic)

lanceolata L. and proportional representation of compounds. Individual components of the essential oils are often used as food flavourings extracted from plant material. Potential use of essential oils are application as additives in many types of foods (beverages odourisers) and food supplements (like cough drops), with different organoleptic effects. The importance is in potential manufacture control of feedstocks before producing of food supplements. Separation of volatile compounds of essential oils and subsequent calculation of proportional representation has been carried out by gas chromatography with flame ionization detector (GC-FID). Identification of compounds has been carried out by gas chromatography/mass spectrometry system (GC-MS) and by comparing to linear retention indices.

Materials and methods

Plant material

Three samples of *Plantago lanceolata* L. (Table 1) were purchased from local companies in Czech Republic.

Chemicals

n-Hexane, and *n*-alkane mixture standard solutions C8-C20 and C21-C40 in concentrations of $40 \text{ mg}\cdot\text{l}^{-1}$ dissolved in *n*-hexane and in toluene, respectively, were purchased from Sigma-Aldrich (Prague, Czech Republic). Distilled water was purified using a Milli-Q® Water Purification System (Millipore SAS, Molsheim, France).

Sample preparation

Sample was not treated before extraction, treatment (drying and crushing) was done by producers. A 40 g of crushed dry sample were extracted with 600 mL of water in an apparatus (Kavalierglass a.s., Prague, Czech Republic) of Clevenger type for 5 h, until no more essential oil was obtained. The essential oil was collected into 1 ml of *n*-hexane and analysed. Each extraction was performed at least three times.

GC-MS analysis

A gas chromatograph, model GC-2010 Plus, coupled with mass spectrometry detector TQ-8030 and auto-sampler AOC-5000 Plus (all from Shimadzu, Kyoto, Japan) was used for analysis. Injections have been performed in split mode 1:5. The GC-MS system has been equipped with a capillary column SLB-5 ms with length 30 m, 0.25 mm inner diameter and 0.25 μm film thickness (Supelco, Bellefonte, PA, USA). Helium 5.0 (Linde Gas a.s., Prague, Czech Republic) was used as a carrier gas at a constant linear velocity of 30 cm/s. The injector and the interface temperature were maintained at 250 °C. The column temperature has been programmed as follows: the initial temperature was 40 °C (1 min) then increased at a rate of 2 °C/min up to 200 °C (10 min). The mass spectrometer was operated in the full scan mode over a mass

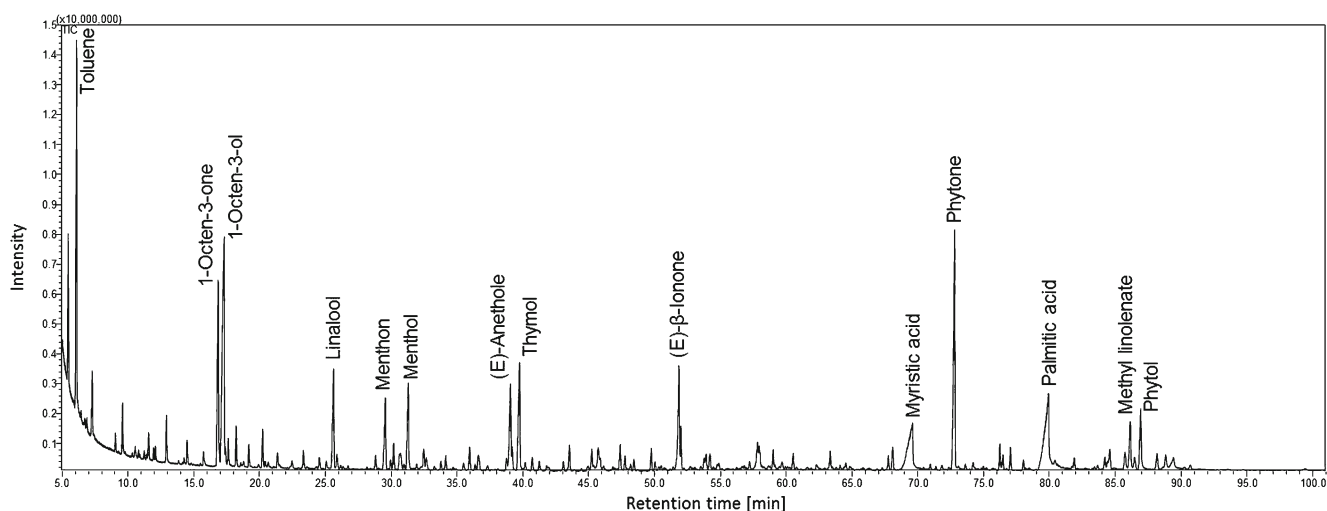


Fig. 1 GC-MS chromatogram of the essential oil (sample 3) with labeling of the most intensive peaks

Table 2 Comparison of volatile compounds presented in extracts obtained by hydrodistillation of three samples of *Plantago lanceolata* L., contents of individual compounds are expressed as average relative percent peak area of GC-FID after three replicates ($n = 3$), n.i. = not identified

Compound	CAS number	RI ^a	Sample 1 [% rel.]	Sample 2 [% rel.]	Sample 3 [% rel.]
Monoterpenes					
α -Pinene	80–56-8	934	n.i.	n.i.	0.05
δ -3-Carene	13,466–78-9	1010	n.i.	n.i.	<0.01
p-Cymene	99–87-6	1025	n.i.	0.05	0.07
Limonene	138–86-3	1029	0.09	0.14	0.76
β -cis-Ocimene	3338–55-4	1037	0.02	0.10	0.02
α -Ocimene	502–99-8	1047	0.47	0.70	0.27
γ -Terpinene	99–85-4	1058	0.03	0.08	0.02
α -Terpinolene	586–62-9	1086	0.04	0.04	0.02
Σ Identified monoterpenes			0.65	1.11	1.21
Oxidated monoterpenes					
1,8-Cineole	470–82-6	1033	0.01	0.04	0.07
Fenchone	1195–79-5	1089	n.i.	0.10	0.30
Linalool	78–70-6	1104	2.86	3.54	2.66
β -Thujone	33,766–30-2	1119	n.i.	0.05	0.07
Camphor	76–22-2	1147	n.i.	0.18	0.37
Menthone	10,458–14-7	1157	0.05	0.65	1.80
Isomenthon	491–07-6	1166	0.11	0.06	0.15
Neomenthol	3623–51-6	1172	n.i.	n.i.	0.95
Menthol	1490–04-6	1180	0.43	1.03	2.12
α -Terpineol	98–55-5	1197	0.34	0.58	0.36
Nerol	106–25-2	1228	0.28	0.17	0.35
Pulegone	89–82-7	1239	n.i.	<0.01	0.05
Carvone	99–49-0	1246	0.02	0.06	0.16
Geraniol	106–24-1	1255	0.04	0.03	0.02
Thymol	89–83-8	1298	0.15	0.02	3.75
Carvacrol	499–75-2	1306	n.i.	<0.01	<0.01
Σ Identified ox. monoterpenes			4.29	6.51	13.18
Sesquiterpenes					
β -Bourbonene	5208–59-3	1382	n.i.	n.i.	0.40
α -Gurjunene	489–40-7	1406	n.i.	n.i.	0.04
β -Caryophyllene	87–44-5	1418	0.14	0.07	0.05
(E)- α -bergamotene	13,474–59-4	1433	n.i.	0.14	0.26
(E)- β -Farnesene	18,794–84-8	1454	0.01	0.02	0.01
(E,E)- α -Farnesene	502–61-4	1505	n.i.	0.21	n.i.
β -Bisabolene	4891–79-6	1508	n.i.	0.03	0.08
β -Sesquiphellandrene	20,307–83-9	1524	n.i.	n.i.	0.28
Σ Identified sesquiterpenes			0.15	0.47	1.11
Oxidated sesquiterpenes					
Cabreuva oxide A	107,602–54-0	1440	n.i.	0.13	n.i.
Cabreuva oxide B	107,602–53-9	1458	0.30	0.47	0.44
Cabreuva oxide D	107,602–52-8	1475	n.i.	<0.01	0.02
(E)-Nerolidol	40,716–66-3	1563	0.03	0.01	0.03
Spathulenol	6750–60-3	1579	1.21	n.i.	n.i.
Viridiflorol	552–02-3	1595	n.i.	0.22	0.33
Fokienol	33,440–00-5	1599	1.29	0.66	0.32
Humulene epoxide II	19,888–34-7	1611	n.i.	0.01	0.05

Table 2 (continued)

Compound	CAS number	RI ^a	Sample 1 [% rel.]	Sample 2 [% rel.]	Sample 3 [% rel.]
Isospathulenol	88,395–46-4	1631	0.67	n.i.	n.i.
α-Bisabolol oxide B	26,184–88-3	1656	n.i.	0.01	0.02
β-eudesmol	473–15-4	1657	0.19	n.i.	n.i.
Valeranone	1803–39-0	1675	0.18	n.i.	n.i.
α-Bisabolone oxide A	58,985–73-2	1682	n.i.	0.03	0.06
Valerenal	4176–16-3	1717	0.11	n.i.	n.i.
α-Bisabolol oxide A	22,567–36-8	1750	n.i.	0.09	n.i.
Xanthorhizol	30,199–26-9	1752	n.i.	n.i.	0.08
Σ Identified ox. sesquiterpenes			3.98	1.64	1.35
Oxidated diterpenes					
Phytone	502–69-2	1846	1.81	2.16	2.99
Isophytol	505–32-8	1947	n.i.	0.04	0.10
Geranyl linalool	1113–21-9	2022	0.08	<0.01	0.06
13-Epi-manool	1438–62-6	2050	n.i.	<0.01	<0.01
Phytol	150–86-7	2111	3.60	1.01	0.88
Σ Identified ox. diterpenes			5.49	3.21	4.03
Apocarotenoids					
β-Isophorone	471–01-2	1043	n.i.	n.i.	<0.01
α-Isophorone	78–59-1	1124	n.i.	<0.01	<0.01
Safranal	116–26-7	1200	0.34	0.58	n.i.
β-cyclocitral	432–25-7	1221	0.13	0.20	0.37
β-Damascenone	23,726–93-4	1380	0.79	0.40	0.39
Tetrahydrogeranylacetone	1604–34-8	1404	n.i.	n.i.	<0.01
Geranylacetone	3796–70-1	1450	0.01	0.02	0.02
(E)-β-Ionone	79–77-6	1482	0.11	0.12	0.05
5,6-Epoxy-β-ionone	23,267–57-4	1484	0.03	0.05	0.41
(E,E)-Pseudoionone	3548–78-5	1585	n.i.	0.77	0.77
Megastigmatrienon	38,818–55-2	1626	0.06	0.10	n.i.
(5E,9E)-Farnesyl acetone	1117–52-8	1912	0.07	0.01	0.03
Σ Identified apocarotenoids			1.54	2.25	2.04
Aldehydes and ketones					
Propyl vinyl ketone	1629–60-3	778	0.03	0.06	0.07
(3E,5E)-1,3,5-Heptatriene	17,679–93-5	785	0.16	n.i.	n.i.
Ethyl propyl ketone	589–38-8	789	0.06	0.05	0.05
Methyl butyl ketone	591–78-6	794	0.03	0.03	0.04
Capronaldehyde	66–25-1	805	1.00	0.44	0.69
(E)-2-hexenal	6728–26-3	854	1.27	0.70	0.89
Heptan-2-one	110–43-0	893	0.03	0.02	0.16
(Z)-4-Heptenal	6728–31-0	903	0.05	0.03	0.02
Heptanal	111–71-7	905	0.49	0.28	0.34
(E)-3-Hepten-2-one	5609–09-6	937	n.i.	0.01	n.i.
α-Ethylcaproaldehyde	123–05-7	949	n.i.	n.i.	0.05
(E)-2-Heptenal	18,829–55-5	958	0.37	0.01	0.02
Benzaldehyd	100–52-7	962	0.19	0.23	0.20
Pentyl vinyl ketone	4312–99-6	980	2.01	2.55	3.40
2,3-Octanedione	585–25-1	988	0.27	0.09	0.09
Caprylaldehyde	124–13-0	1006	0.12	0.06	0.06
(E,E)-2,4-Heptadienal	4313–03-5	1015	1.28	0.69	0.39

Table 2 (continued)

Compound	CAS number	RI ^a	Sample 1 [% rel.]	Sample 2 [% rel.]	Sample 3 [% rel.]
2,2,6-Trimethylcyclohexanone	2408–37-9	1035	0.10	0.04	0.08
3-Octen-2-one	1669–44-9	1041	<0.01	<0.01	<0.01
Phenylacetaldehyde	122–78-1	1045	0.02	0.02	0.03
2-Octenal	2363–89-5	1060	0.29	0.12	0.14
Acetophenone	98–86-2	1067	n.i.	<0.01	<0.01
(E,E)-3,5-Octadien-2-one	30,086–02-3	1072	<0.01	<0.01	<0.01
(E,Z)-3,5-octadien-2-one	4173–41-5	1096	<0.01	<0.01	<0.01
Nonanal	124–19-6	1107	0.53	0.23	0.15
(E,E)-2,4-octadienal	30,361–28-5	1114	0.02	0.02	0.04
3-Nonen-2-one	14,309–57-0	1142	n.i.	n.i.	<0.01
(E,Z)-2,6-nonadienal	557–48-2	1156	0.59	0.21	0.35
(E)-2-nonenal	18,829–56-6	1163	0.30	0.06	0.05
p-Ethylbenzaldehyde	4748–78-1	1177	n.i.	n.i.	<0.01
p-Methylacetophenone	122–00-9	1187	0.08	0.13	0.24
Capraldehyde	112–31-2	1208	0.11	0.12	0.04
Undecanal	112–44-7	1310	n.i.	<0.01	<0.01
(E,E)-2,4-decadienal	25,152–84-5	1321	0.63	0.67	0.35
Benzophenone	119–61-9	1629	n.i.	n.i.	<0.01
Pentadecanal	2765–11-9	1717	n.i.	0.03	0.02
p-Methylbenzophenone	134–84-9	1758	n.i.	0.01	0.24
Palmitaldehyde	629–80-1	1819	n.i.	n.i.	0.03
Σ Identified aldehydes and ketones			10.03	6.91	8.23
Alcohols					
2-Hexanol	626–93-7	808	<0.01	<0.01	<0.01
(Z)-3-hexen-1-ol	928–96-1	859	n.i.	n.i.	<0.01
1-Octen-3-ol	3391–86-4	984	2.39	6.94	8.24
3-Octanol	589–98-0	1001	0.97	0.53	0.39
Octanol	111–87-5	1076	0.46	0.22	0.35
Tetradecanol	112–72-1	1674	n.i.	0.19	0.21
Σ Identified alcohols			3.82	7.88	9.19
Phenols and phenolic ethers					
p-Allylanisole	140–67-0	1200	n.i.	n.i.	0.36
(E)-anethole	4180–23-8	1289	<0.01	0.78	3.21
p-Vinylguaiaicol	7786–61-0	1313	0.01	0.12	0.22
Eugenol	97–53-0	1355	0.06	0.10	0.22
2,5-Di-tert-butylphenol	5875–45-6	1513	n.i.	n.i.	0.05
Σ Identified phenols and phenolic ethers		0.07	1.00	4.06	
Fatty acids					
Capric acid	334–48-5	1390	0.69	0.61	0.47
Myristic acid	544–63-8	1785	4.63	5.99	4.48
Pentadecanoic acid	1002–84-2	1875	n.i.	1.11	0.53
Palmitic acid	57–10-3	1990	27.86	31.97	15.26
Margaric acid	506–12-7	2071	0.15	0.16	0.11
Linoleic acid	60–33-3	2149	4.68	6.85	2.89
Linolenic acid	463–40-1	2160	6.29	5.36	4.23
Σ Identified fatty acids			44.30	52.05	27.97
Esters					
Methyl salicylate	119–36-8	1193	n.i.	0.10	n.i.

Table 2 (continued)

Compound	CAS number	RI ^a	Sample 1 [% rel.]	Sample 2 [% rel.]	Sample 3 [% rel.]
Linalyl acetate	115–95-7	1252	n.i.	0.13	0.51
Bornyl acetate	92,618–89-8	1285	n.i.	n.i.	0.13
Menthyl acetate	16,409–45-3	1291	0.05	0.19	0.27
α-Terpinyl acetate	80–26-2	1348	n.i.	0.03	0.02
Neryl acetate	141–12-8	1361	n.i.	n.i.	0.03
2,2,4-Trimethyl-1,3-pentanediol diisobutyrate	6846–50-0	1587	n.i.	0.64	n.i.
Isopropyl laurate	10,233–13-3	1628	n.i.	0.04	n.i.
Methyl myristate	124–10-7	1727	n.i.	n.i.	0.06
Methyl palmitate	112–39-0	1928	0.05	0.05	0.21
Methyl linoleate	112–63-0	2092	0.32	0.38	0.25
Methyl linolenate	301–00-8	2099	0.06	0.07	0.26
Ethyl linoleate	544–35-4	2161	n.i.	n.i.	0.08
Σ Identified esters			0.48	1.63	1.82
Aliphatic hydrocarbons					
Dodecane	112–43-3	1201	n.i.	0.12	n.i.
Tridecane	629–50-5	1301	n.i.	<0.01	n.i.
Tetradecane	629–59-4	1401	n.i.	0.03	0.02
Pentadecane	629–62-9	1501	n.i.	0.04	0.05
Cetane	544–76-3	1601	n.i.	n.i.	0.32
Heptadecane	629–78-7	1701	n.i.	0.08	0.05
Octadecane	593–45-3	1801	n.i.	<0.01	<0.01
Nonadecane	629–92-5	1893	n.i.	<0.01	0.02
Σ Identified aliphatic hydrocarbons		n.d.	0.27	0.46	
Aromatic hydrocarbons					
Toluene	108–88-3	765	0.21	0.31	6.92
1,3-Diisopropylnaphthalene	57,122–16-4	1667	n.i.	0.15	n.i.
1,4-Diisopropylnaphthalene	24,157–79-7	1714	n.i.	0.01	n.i.
2,7-Diisopropylnaphthalene	40,458–98-8				
1,6-Diisopropylnaphthalene	51,113–41-8	1719	n.i.	0.02	n.i.
2,6-Diisopropylnaphthalene	24,157–81-1	1724	n.i.	<0.01	n.i.
Diisobutyl phthalate	84–69-5	1862	0.07	0.05	0.01
Dibutyl phthalate	84–74-2	1957	0.08	0.04	0.13
Σ Identified aromatic hydrocarbons		0.36	0.58	7.06	
Others					
Dimethylfulvene	2175–91-9	870	n.i.	0.03	0.04
Cyclooctatetraene	629–20-9	896	n.i.	0.06	0.20
Dimethyl trisulfide	3658–80-8	967	n.i.	0.23	n.i.
2-Amylfuran	3777–69-3	992	0.77	0.44	0.54
Σ Identified other compounds			0.77	0.76	0.78
Number of identified compounds			85	125	131

^a RI, retention index on capillary column SLB-5 ms, $RI = 100 \cdot n + 100 \cdot (t_x - t_n) / (t_{n+1} - t_n)$; n, the number of carbon atoms in the alkane; t_x , retention time of peak of unknown compound; t_{n+1} and t_n , the retention time of alkane with $n + 1$ and n carbon atoms

range of m/z 45–500 and in the electron ionization (EI) mode (70 eV). The mixtures of n -alkanes (C8–C20, C21–C40) were injected using the above temperature program in order to calculate the retention index (RI) for each peak. Identification of

the components was done by comparison of mass spectral fragmentation patterns stored in MS data libraries (NIST 11, FFNSC 2), and verified by comparison of retention indices (RIs) of identified compounds with published index data

(Adams 2007; Goodner 2008; Nijssen et al. 1963–2015; NIST 2011) and RIs from MS data library FFNSC 2.

GC-FID analysis

GC-FID analysis was accomplished on a Shimadzu GC2010 equipped with a flame ionization detector (FID). The detector temperature was 220 °C. The other analytic conditions including the column type and column temperature, the injector temperature, carrier gas and the linear velocity were the same as those of GC–MS analysis.

Results and discussion

Totally, up to 236 peaks were observed in chromatograms of the extracted compounds from ribwort. However, relatively large number of the peaks was not identified, often due to absence of appropriate mass spectrum in libraries or absence of retention indexes calculated for given column. Figure 1 depicts representative chromatogram. The identified peaks are listed in Table 2. Semi-quantitative data were obtained from the integration of the FID peak areas.

As is seen in Fig. 2 showing aroma profiles of ribwort using bubbles whose sizes correspond to the representation of the individual compounds, a composition of all three samples is very similar. The biggest portion of the essential oils is generated by fatty acids (28.0–52.0 %), particularly myristic acid (RI 1785), palmitic acid (RI 1990) linoleic acid (RI 2145) and linolenic acid (RI 2163). Fatty acids are common components of the essential oils (Clarke 2008), for instance palmitic acid was found in ribwort leaves by Fons (Fons et al. 1998). Furthermore, in the present work, methyl esters of some fatty acids were also identified.

Oxidated monoterpenes, which commonly generate a large part of the essential oils contented in aromatic herbs, made up 4.3–13.2 %. From non-oxidated monoterpenes only 8 compounds were identified, the biggest part was created by limonene (0.8 % in sample 3) and α -ocimene (0.7 % and 0.5 % in

sample 2 and 3, respectively). Similar representation of non-oxidated compounds was observed also in sesquiterpenes. Sixteen oxidated sesquiterpenes were identified in all three samples. It is the same as in the case of oxidated monoterpenes, but amount in percents was lower, 1.4–4.0 %.

One group of the main identified components of the essential oils was oxidated diterpenes (particularly phytol (RI 2111) and phytone (RI 1846)), which generated 3.2–5.5 %. Free phytol is created by chlorophyll hydrolysis by an enzyme chlorophyllase as an integral part of plants catabolism in fruits ripening or leave yellowing, and it does not have any influence to plant aroma (Velišek et al. 2009). Phytone is created by its oxidation which was most likely invoked during distillation.

Unlike Fons (Fons et al. 1998) who identified great amount of apocarotenoids in ribwort, namely (E)-9-hydroxymegastigma-4,7-dien-3-one and 3-hydroxy-5,6-epoxy- β -ionol, in the present work these compounds were not found. Nevertheless, some other apocarotenoids were observed in the aroma profile of ribwort and generated 1.5–2.3 % of aroma. Moreover in this work, (E)- β -damascenone (RI 1380) was identified. It is a compound belonging among the most aromatic ones (Velišek and Hajšlová 2009), which is ordinarily used in perfumes production (Pybus and Sell 1999).

Regarding the content of phenolic compounds, quite big differences among individual samples appeared. They were most abundantly contained in sample 3 (4.1 %). In samples 1 and 2 only 0.1 % and 1.0 %, respectively, were identified. These differences are obvious in the Fig. 2, showing comparison of aroma profiles using bubbles corresponding to (E)-anethol (RI 1289).

Oxidated compounds as alcohols, aldehydes, and ketones (except oxidated terpenes) were the second most abundant family of compounds generating together 14.1–17.4 % (in that the carbonyl compounds 6.9–10.0 %). High content of 1-octen-3-ol (up to 8.2 %) is in the compliance with the work of Fons (Fons et al. 1998).

Also some aromatic compounds were identified, particularly toluene, diisobutyl phthalate or dibutyl phthalate.

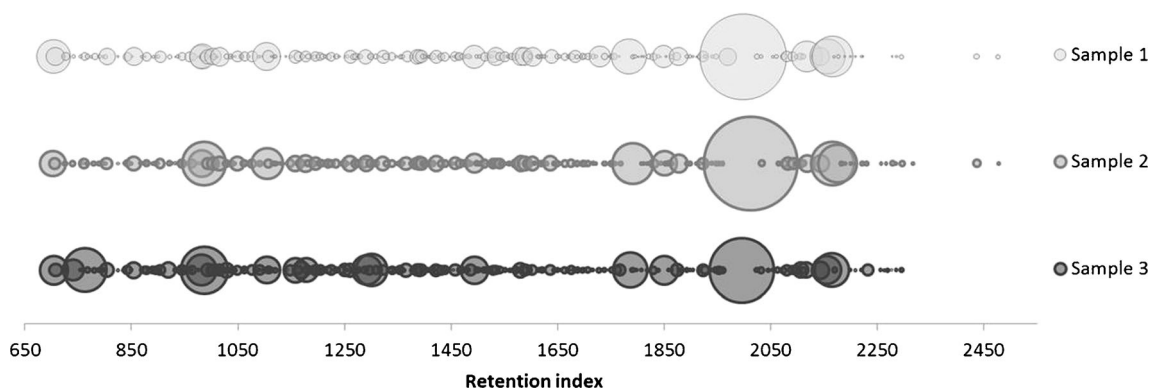


Fig. 2 Comparison of three hydrodistillation extracts of ribwort according to RI; area of the bubbles represents the relative peak area of GC-FID

However, those were probably contaminants. Equally, Fons (Fons et al. 1998) identified in the sample of ribwort toluene and *o*-, *m*- and *p*-xylene, which were also stated as contaminants. The highest content of toluene (6.9 %) was presented in sample 3. This sample was finely ground and packed in teabags individually closed by plastic packaging. The other two samples were packed as whole leaves (sample 1) and coarsely ground leaves (sample 2) into a paper back. Therefore, it is expected toluene most likely appeared from the plastic packaging. Moreover, isomers of diisopropyl-naphthalene (DIPN) were identified in essential oil obtained from sample 2 in amount less than 0.2 %. DIPNs are used as a solvent in the manufacture of certain printing materials and as plant growth regulator in agriculture (Brzozowski et al. 2002). DIPNs probably originated from recycled paper packaging, which were dried plants packed. A carton was evaluated as a source of DIPN contamination also by study examining contaminants in dried foods (Großmann-Kühnau 2011).

Different composition of individual samples could be affected by growing environment, time and conditions of collection, method of drying or storage conditions. These effects were described for various plants (Figueiredo et al. 2008; Gil et al. 2002; Mahmoodi Sourestani et al. 2014; Orav et al. 2004; Rowshan et al. 2013). Moreover, it is expected presence of different chemotypes of ribwort which differ in quantitative as well as in qualitative content of aromatic compounds.

Conclusion

In this work, the essential oil's analysis of three ribwort samples (*Plantago lanceolata* L.) were conducted in order to observe qualitative and semi-quantitative composition. Hydrodistillation enabled to gain great amount of volatile compounds, which is obvious from a number of peaks (up to 236) occurred in chromatograms. The most abundant family of compounds were fatty acids. There were also identified methyl esters of fatty acids and great amount of aliphatic alcohols, aldehydes and ketones. Significant constituents of essential oils contented in ribwort showed to be apocarotenoids that belong among the most intense fragrant compounds. On the other hand, non-oxidated terpenes were found in relatively small amount and most of terpenes were oxidated as monoterpenes, sesquiterpenes and diterpenes. Novelty of the study up to now, there is no published study deals with the chemical composition of the essential oil extracted by hydrodistillation from leaves of *Plantago lanceolata* L. The importance is in potential manufacture control of feedstocks before producing of food supplements, too.

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