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The study of *Hosta lancifolia* rhizomes with roots, leaves and flowers volatile components

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ABSTRACT

The component composition of volatile compounds of rhizomes with roots, leaves and flowers of Hosta lancifolia was investigated by the gas chromatography technique. As the result of the research, 62 volatile compounds collectively were identified in the all types of studied plant raw material. 27 compounds were found in leaves. 34 and 32 compounds were determined in flowers and rhizomes with roots respectively. The total content of volatile compounds in the rhizomes with roots is 3635.5 mg/kg. Volatile compounds contained in leaves and flowers in an amount of 3204.9 mg/kg and 654.2 mg/kg respectively. Carboxylic acids and their esters were dominant in rhizomes with roots in the qualitative composition and quantitative content. The terpene compounds dominated in leaves. The major groups of organic substances that were found in Hosta lancifolia flowers were aldehydes, ketones and paraffin hydrocarbons.

Key words: gas chromatography, volatile compounds, *Hosta lancifolia*.

INTRODUCTION

Plants of the genus Hosta (Hostaceae Mathew.) are popular garden plants all over the world since the time they were introduced in the nineteenth century from South-East Asia. However, in their homeland, in Japan and China, these plants are not only used for decorative purposes, but also in folk medicine. Upper respiratory tract and pelvic organs inflammatory diseases are successfully treat with the help of some Hosta species.

Literary data testify to a rich and varied chemical composition and the antibacterial, antifungal, antiviral and anticancer properties among members of this genus [1, 2, 3]. *Hosta lancifolia* is an ornamental deciduous perennial plant. This is one of the most common cultivated Hosta species in Ukraine. According to the literature data, flavonoids, polysaccharides, vitamins, in particular ascorbic acid and saponins are accumulated in aboveground and underground parts of *Hosta lancifolia* [1, 4, 5]. However, reliable data on the qualitative composition and quantitative content of *Hosta lancifolia* essential oil are absent in the literature.

Essential oils are of particular importance among the main classes of biologically active compounds. Essential oils are a mixture of volatile organic compounds which define the plant odor. Component composition of essential oils may be represented by hydrocarbons, alcohols, esters, aldehydes, ketones, aromatic substances. Nevertheless, terpenoid compounds are dominant in such a mixture. Essential oils components show a broad spectrum of biological activity [6]. In particular, according to the literature sources, acyclic monoterpenoid linalool and monocyclic monoterpenoid menthol exhibit antispasmodic and antiviral properties [6-10]. Linalool and β-nerolidol which is an acyclic sesquiterpene alcohol have anti-inflammatory, wound healing and antifungal activity [6, 8, 9, 11]. Eugenol which refers to aromatic compounds and menthol has anesthetic and analgesic properties [6, 9, 12-14]. In addition, eugenol exhibits anti-inflammatory, sedative, vasodilator, cardioprotective, hypotensive and hepatoprotective pharmacological activity [8, 13, 14]. Monocyclic monoterpenoid α-terpineol has antifungal and antioxidant effect on the body [6-9]. Acyclic monoterpene geraniol has a strong antiviral and antioxidant action [14]. Aromatic compound ar-tumeron has anti-parasitic properties [15]. Phenolic compound 2-methoxy-4-vinylphenol exhibits antiseptic properties [6]. Diterpenoid geranilaceton has cytoprotective effects [13].

The acyclic monounsaturated diterpenoid alcohol phytol is able to reduce insulin resistance, regulate metabolic disorders and inhibit the synthesis of glucose in the liver. It is also involved in the chlorophyll and vitamins E and K1 synthesis [4, 8, 10]. It is proved that cyclic norterpenoid β-ionone, which is the product of carotene enzymatic breakdown, reduces tumor growth and slows metastasis development [13]. And the 13-epimanool is considered a precursor to some diterpenoids and exhibits pronounced antiviral and antitumor activity [9].Thus, the antibacterial activity inherent in all the above-mentioned compounds [2, 4, 6, 7, 9-11, 13-18].

The aim of the study is the identification and quantification of volatile components in *Hosta lancifolia* plant raw material.

MATERIALS AND METHODS

2.1 Plant Material and Chemicals

Rhizomes with roots, leaves and flowers of *Hosta lancifolia* were selected as the objects of the study. The plant raw material was harvested in 2014-2015 in Kharkiv region, Ukraine.

2.2 Chromatographic conditions

Identification of volatile compounds and their quantitative content determination was performed by gas chromatography technique using an Agilent Technologies HP6890 chromatograph with mass spectrometric detector 5973 [10, 16, 17]. A sample preparation procedure was carried out according to the following protocol. 0.5 g of the plant raw material tested was placed in a 20 ml vial with the further addition of the internal standard. Tridecan at the rate of 50 µg on a weighed substance was taken as the internal standard. That was done with the following recalculation of the obtained concentration of the internal standard, which was used then for further calculations [10, 16, 17].

10 ml of distilled water was added to the sample. Volatile compounds were distilled from the samples with water vapor for 2 hours by using a reverse refrigerator air cooling [10, 16, 17]. Volatile compounds were adsorbed on the inner surface of the reverse refrigerator during the distillation process. Adsorbed compounds were flushed by the slow addition of 3 ml of especially pure pentane in a 10 ml dry vial after cooling. After washing compounds were concentrated by a high purity nitrogen purge (100 ml/min) to a residual volume of extract 10 µl which was fully taken up by a chromatographic syringe. The further concentration of the sample was conducted in the syringe up to a 2 ml volume [10, 16, 17]. A chromatographic capillary column DB-5 with an internal diameter of 0.25 mm and a length of 30 m was used for the analysis. Helium was chosen as a carrier gas. The rate of carrier gas was 1.2 ml/min. 2 µl sample volume was injected split less, i.e. without separation of flow, at a rate of 1.2 ml/min for 0.2 min. The thermostat temperature was programmed from 50˚ C to 320˚ C at a rate of 4˚ C/min. The temperature of input heater of the samples was 250˚ C [10, 16, 17]. The mass spectra library NIST05 and WILEY 2007 with more than 470000 total numbers of spectra in a combination with AMDIS and NIST identification programs was used to identify the components [10,16, 17]. The method of the internal standard was used for quantitative calculations.

The components concentration calculation was performed according to the formula: $C=K1*K2$, mg/kg

Where:-
$K1=P1/P2$ (P1 is the peak area of the analyzed substance; P2 is the peak area of the standard).
$K2=50/M$ (50 – mass of internal standard (µg), input into the sample, M – the weighed sample (g)) [10, 16, 17].

RESULTS AND DISCUSSION

As a result of research 62 volatile compounds were collectively identified in all types of studied *Hosta lancifolia* plant raw material. In particular, 32 compounds were identified in the rhizomes with roots, 15 of which are characteristic only for this plant raw material. 27 compounds were identified in *Hosta lancifolia* leaves, 12 of which were found only in this plant raw material. The studied plant flowers contained 34 compounds which were identified. 15 compounds of 34 determined were found only in this plant raw material. Substances belonging to carboxylic acids, paraffin hydrocarbons, alcohols, aldehydes, ketones and terpenoids were dominant among the detected compounds.

Gas chromatograms of volatile compounds of *Hosta lancifolia* rhizomes with roots, leaves and flowers are presented in:- Figure1, Figure2, Figure3

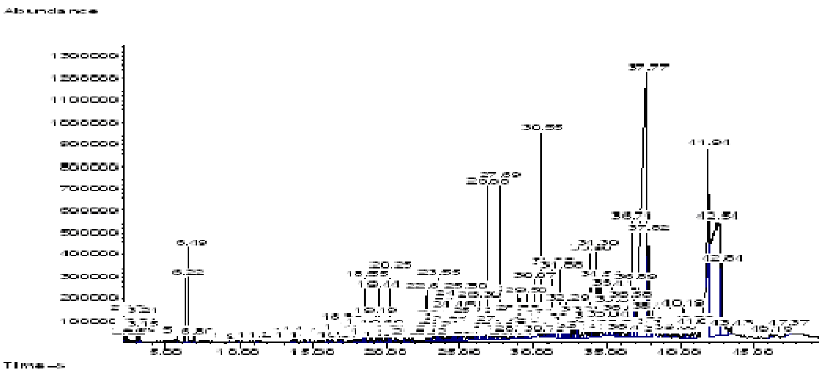

Figure 1: Gas chromatography of rhizomes with roots of *Hosta lancifolia* volatile compounds

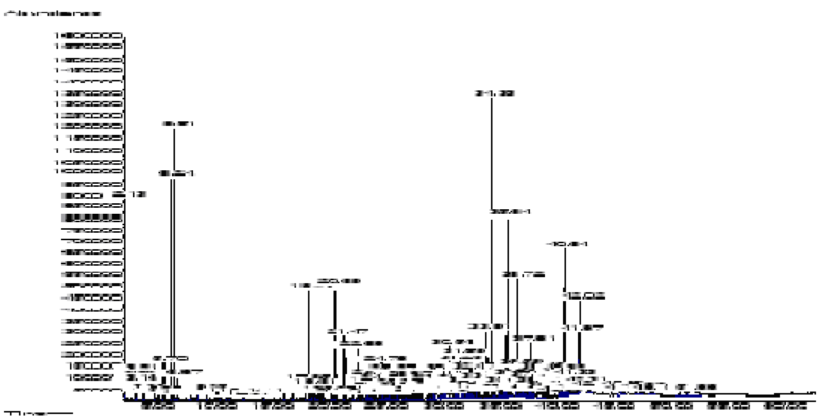

Figure 2: Gas chromatography of leaves of *Hosta lancifolia* volatile compounds

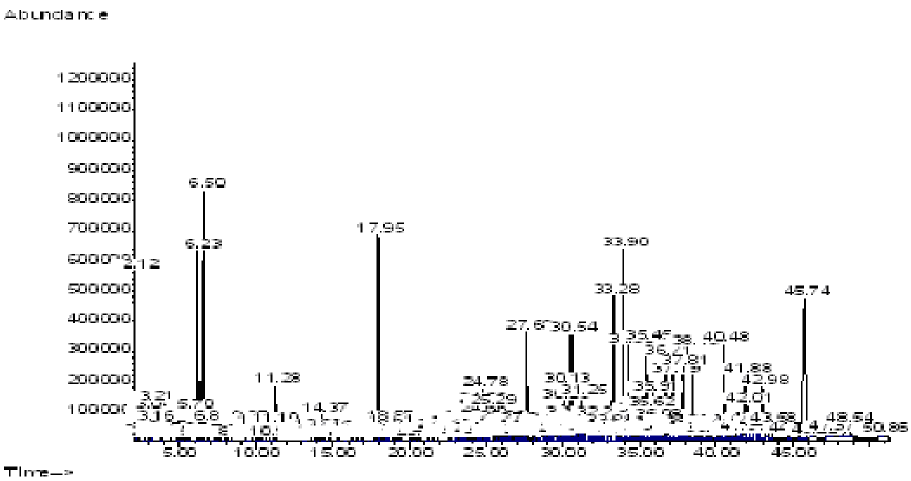

Figure 3: Gas chromatography of flowers of *Hosta lancifolia* volatile compounds

The total content of volatile compounds in the rhizomes with roots is 3635.5 mg/kg. For *Hosta lancifolia* leaves and flowers this index is consistent with 3204.9 mg/kg and 654.2 mg/kg on dry substance recalculation.

Table 1. The qualitative composition and quantitative content of volatile components of the rhizomes with roots, leaves and flowers of *Hosta lancifolia*

N	Component	The molar mass in (a.m.u)	The retention time	Content in mg/kg in terms of dry plant material types		
				Rhizomes with roots	Leaves	Flowers
1	Benzeneacetaldehyde	120.15	9.3	1.5	-	4.4
2	1,3,6 –Octatriene,3,7-dimethyl-, (Z)-	136.24	9.3	-	18.7	-
3	1,6-Octadien-3-ol, 3,7-dimethyl (Linalool)	154.24	11.3	-	-	4.1
4	Nonanal	142.239	11.3	1.5	-	11.6
5	2,6,6-Trimethyl-2-cyclohexene-1,4-dione	152.1904	12.6	-	5.6	-
6	1-Nonanol	144.26	13.55	4.0	-	-
7	Menthol	156.27	13.85	2.0	-	2.1
8	α-Terpineol (3-Cyclo hexene-1 methanol,α, α, 4-trimethyl-)	154.25	14.4	4.0	10.3	6.1
9	Decanal	156.27	14.78	-	-	1.3
10	trans-Geraniol (2,6-octadien-1-ol, 3,7-dimethyl)	154.26	16.3	-	10.3	2.3
11	2,4-Decadienal isomer	152.2334	17.76	4.0	-	-
12	2-Methoxy-4-vinylphenol	150.177	18.22	-	242.9	-
13	2,4-Decadienal isomer	152.2334	18.57	-	-	4.5
14	2,6-Octadienoic acid, 3,7-dimethyl-, methyl ester	182.263	18,6	8.0	-	-
15	Eugenol	164.2	19.6	11.0	-	-
16	2(3H)-Furanone,dihydro-5-pentyl- or γ-nonalactone	156.222	19.9	3.5	-	-
17	2-Undecenal	170.28	20.1	5.5	-	-
18	n-decanoic acid	172.26	20.36	7.5	-	-
19	n-Tetradecane	198.39	21.22	-	-	1.7
20	Dodecanal	184.32	21.5	5.5	-	1.8
21	3-Buten-2-one, 4-(2,6,6-trimethyl-2-cyclohexen-1-yl)-, (E)- or a-Ionone	192.297	21.85	-	-	2.1
22	6,10-dimethyl-5, 9-undecadien-2-one	194.318	22.67	30.5	112.1	3.3
23	Dodecanol-1	186.34	23.5	-	29.0	-
24	3-Buten-2-one,4-(2,6,6-trimethl-1-cyclohexen-1-yl)- or β-Ionone	192.29	23.6	-	38.3	-
25	Tridecanal	198.345	24.7	32.5	-	-
26	Dodecanoic acid	200.318	26.37	55.0	-	-
27	n-Hexadecane	226.44	27.3	-	16.8	4.5
28	Tetradecanal	212.3715	27.7	90.0	59.8	25.5
29	Benzophenone	182.217	28.0	21.0	45.8	-
30	Ar-tumerone	216.3187	29.05	37.5	44.8	-
31	n-Heptadecane	240.48	30.15	-	-	14.1
32	2,3-dihydro-1,1,3-trimethyl-3-phenyl-1H-Indene	234.3355	30.22	-	-	5.1
33	Pentadecanal	226.3981	30.5	120.0	130.8	27.7
34	Tetradecanoic acid	228.37	31.87	90.0	-	-
35	n-Octadecane	254.5	32.85	-	-	7.6
36	Hexadecanal	240.4247	33.25	17.0	50.4	38.2
37	2-Pentadecanone,6, 10,14-trimethyl-(Hexahydrofarnesyl acetone)	268.4778	33.87	60.0	196.2	50.9
38	Pentadecanoic acid	219.23	34.5	60.0	-	-
39	6,10,14-trimethyl-,5 , 9,13-Pentadecatrien-2-one	262.4302	35.6	-	457.7	13.0
40	Heptadecanal	254.4513	35.9	-	-	13.7
41	Hexadecanoic acid, methyl ester	270.45	36.06	19.0	53.2	6.1
42	1-Hexadecen-3-ol,3,5,11,15-tetramethyl- or isophytol	296.5310	36.57	-	76.6	-
43	n-Hexadecenoic acid	254.4082	36.6	41.0	-	-
44	Z 7 - n-Hexadecenoic acid	256.4241	36.7	95.0	-	-
45	Z 7 - n-Hexadecenoic acid	256.43	37.5	1205.0	-	42.4
46	Hexadecanoic acid, ethyl ester	284.477	37.8	90.0	168.1	25.5
47	n-Eicosane	282.552	37.9	-	-	4.2
48	1H-Naphtho[2,1-b]pyran, 3-ethenyldodecahydro-3,4a,7,7, 10a-pentamethyl-, [3S-(3α, 4a, α,6a, β,10a, α,10b, β)]-or 13-epimanool	290.483	38.1	21.0	35.5	-
49	1,6,10-Dodecatrien-3-ol, 3,7,11-trimethyl-, [S-(Z)]- (β-Nerolidol)	226.36	38.4	-	102.7	-
50	Octadecanal	268.4778	38.5	-	-	32.5
51	9,12-octadecadienoic acid,methyl ester	294.48	40.1	28.0	53.2	4.2
52	9-octadecenoic acid,methyl ester	296.5	40.3	-	89.7	3.8
53	n-Heneicosane	296.57	40.7	-	82.2	41.0
54	Phytol	296.531	40.55	-	569.8	-
55	9,12-octadecadienoic acid	280.4472	41.5	-	-	32.5
56	9,12-octadecadienoic acid, ethyl ester	308.4986	41.8	135.0	252.2	-
57	9,12-Octadecadienoic acid, ethyl ester	292.4562	42.1	1200.0	-	-
58	9,12,15-Octadecatrienoic acid, ethyl ester, (Z,Z,Z)-	290.4403	41.9	-	252.2	-
59	n-Octadecanoic acid	284.48	42.3	130.0	-	-
60	n-Docosane	310.6027	43.5	-	-	35.4
61	n-Tricosane	324.6	46.4	-	-	161.2
62	n-Tetracosane	338.654	50.7	-	-	19.8
The total content				3635.5	3204.9	654.2

As can be seen from table 1, carboxylic acids and their esters dominated in the rhizomes with roots for qualitative composition and quantitative content. Generally, 13 compounds in this group of organic substances were identified. Their total content in the plant raw material equaled 3155.5 mg/kg. Palmitic and stearic acid and ethyl ester of linolenic acid in the rhizomes and roots quantitatively dominated among all identified substances. In addition, heterocyclic ketone – γ-nonalactone was present in this type of raw material.

Terpenoid compounds dominated in a leaves. Their total amount was 1262.0 mg/kg. Phytol, β-nerolidol, 6,10-dimethyl-5,9-undecadien-2-one dominated among the 11 identified substances of this group. β-ionone, ar-tumeron and 13-epimanool were accumulated in a less amount. The total content of the 8 identified aldehydes and ketones in this of plant raw material equaled 975.3 mg/kg. Paraffin hydrocarbons of Hosta leaves were accumulated in a small amount. In addition, the compounds of aromatic nature were detected in this type of raw material – 2-Methoxy-4-vinylphenol in the amount 242.9 mg/kg.

Major groups of organic substances that are found in *Hosta lancifolia* flowers are paraffinic hydrocarbons (289.5 mg/kg) and aldehydes and ketones, the general content of which was 225.1 mg/kg. Wherein, 9 paraffin hydrocarbons, 10 aldehydes and 2 ketones were identified in this plant raw material. Hexahydrofarnesyl acetone, octadecanal and n-tricosane quantitatively dominated among the identified substances belonging to these groups of organic compounds. Polycyclic aromatic hydrocarbon 2,3-dihydro-1,1,3-trimethyl-3-phenyl-1H-Indene was found in this plant raw material in a small amount.

The biggest amount of carboxylic acids and their esters was accumulated in the rhizomes with roots. In *Hosta lancifolia* leaves they were found in 3.6 times less than in underground part in the amount of 868.6 mg/kg. Only 5 compounds of this group of substances were found in flowers, the total content of which was 82 mg/kg. Compounds of terpenoid nature prevailed in Hosta leaves. Their content is significantly lower in rhizomes with roots and equals 114 mg/kg, and in the flowers these compounds content is small – only 20 mg/kg. Aldehydes and ketones in quantitative content prevailed in the leaves of the studied parts of plant. As for qualitative composition they prevailed in the flowers and rhizomes with roots, where were identified 12 substances from this group. Their content was 362.5 mg/kg in rhizomes with roots and 225.1 mg/kg in flowers. Mainly paraffinic hydrocarbons were concentrated in the *Hosta lancifolia* flowers. They were not detected in Hosta rhizomes and roots. Only 2 paraffin hydrocarbons which had the total content of 99.0 mg/kg were identified in leaves. Furthermore, in rhizomes with roots 3.5 mg/kg of the heterocyclic lactone – γ-nonalactone was detected. Polycyclic aromatic hydrocarbons were identified in all types of the plant raw material tested. Polycyclic aromatic hydrocarbon 2,3-dihydro-1,1,3-trimethyl-3-phenyl-1H-Indene was detected in the flowers of *Hosta lancifolia*.

CONCLUSION

1. The rhizomes with roots, leaves and flowers of *Hosta lancifolia* were studied by gas chromatography. As the results of the analysis 62 volatile compounds collectively were identified in all types of studied plant raw material. 27 compounds were found in leaves. 34 and 32 compounds were identified in flowers and rhizomes with roots respectively.
2. The total content of volatile compounds in rhizomes and roots was 3635.5 mg/kg. For leaves and flowers of *Hosta lancifolia* this index was 3204.9 mg/kg and 654.2 mg/kg respectively.
3. Carboxylic acids and their esters dominated in the rhizomes with roots for qualitative composition and quantitative content. Compounds of terpenoid nature dominated in the leaves. Major groups of organic substances found in *Hosta lancifolia* flowers were aldehydes, ketones, and paraffin hydrocarbons.
4. The biggest number of carboxylic acids and their esters was accumulated in *Hosta lancifolia* rhizomes with roots.
5. Compounds of terpenoid nature prevailed in the leaves of this plant. Their content was much lower in rhizomes with roots. These compounds were detected in small quantities in Hosta flowers.
6. Aldehydes and ketones prevailed in the leaves of the studied plants by their quantitative content, and the flowers and rhizomes with roots were richer in diversity of these classes of volatile compounds.
7. Paraffinic hydrocarbons were concentrated mainly in *Hosta lancifolia* flowers. Compounds of this group were absent in the rhizomes with roots, while only 2 paraffin hydrocarbons were identified in the leaves.
8. Terpene compounds have better expression of pharmacological activity among the compounds found in *Hosta lancifolia*. The leaves of this plant have the most diverse composition of terpenes. phitol, β-nerolidol, ocimene and isophytol were present in great quantities in this type of plant material. Ar-tumerone and 13-epimanool were present in the rhizomes with roots and leaves of *Hosta lancifolia* in large quantities. The content of α-terpineol and trans-geraniol in the leaves of *Hosta lancifolia* was identical and their amount was 10.3 mg/kg. α-Terpineol was accumulated in small quantities in the rest of the plant material studied, and trans-geraniol was present only in the leaves and flowers. The flowers of this plant tend to contain up to 4.1 mg/kg of linalool and 2.1 mg/kg ά-ionone. It was determined that the rhizomes and roots of *Hosta lancifolia* contained 11.0 mg/kg of eugenol, and menthol was present in small quantities in the rhizomes with roots and flowers of the investigated plants.
9. Obtained data can be further used in the development of quality control methods for medicinal plant raw material and the development of phyto-remedies on its basis.

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