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THE COMPOSITION OF VOLATILE SUBSTANCES OF COWBERRY LEAVES

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Key words: cowberry; leaves; volatile substances; gas chromatography

*The qualitative composition and quantitative content of volatile substances of cowberry leaves (*Vaccinium vitis-idaea* L.) has been studied by gas chromatography. Identification of substances has been performed based on comparison of the mass spectra obtained with the data of the NIST05-WILEY library (approximately 500000 mass spectra). The retention indices of the components have been calculated according to the results of control tests of substances with addition of the mixture of normal alkanes (C_{10} - C_{18}). In cowberry leaves 51 substances have been found, 50 of them have been identified; squalene, palmitic acid, ethyl palmitate, oleic acid prevail. It has been determined that the content of volatile substances is 0.025%. Monoterpenes are presented by 14 substances, derivatives of linalool, geraniol, thymol and eugenol are prevalent. Caryophyllene oxide and farnesyl acetone C prevail among sesquiterpenes. Among organic and fatty acids the predominant substances are lauric, myristic, pentadecanoic, palmitoleic, palmitic, linoleic and linolenic acids.*

In the world there is a tendency of growing interest in phytotherapy. Microorganisms have developed resistance to many existing antibacterial agents; therefore, the use of the medicinal plant raw material for creating antimicrobial drugs is an urgent direction of development of pharmaceutical science. The promising plant to create a new herbal medicine is cowberry (*Vaccinium vitis-idaea* L.), its resources are sufficient in Ukraine.

In traditional and official medicine a decoction of cowberry leaves is used as a highly effective medicine for the treatment of diseases of the kidney and the urinary tract. However, this dosage form has a number of disadvantages: difficulty to prepare, the absence of standardization, impossibility of long-term storage, continuous use of the medicinal plant raw material. Thus, creation of a new standardized medicine based on biologically active substances of cowberry leaves is topical [5].

It is commonly known that phenolic compounds are the main group of substances with the pharmacological action of this medicinal plant raw material. Previously, we reported on the qualitative and quantitative chemical determination of some BAS classes in leaves and extracts of cowberry. They are: simple phenols, derivatives of hydroxycinnamic acid, flavonoids, polyphenolic compounds and organic acids [1, 2, 3]. Continuing the studies of BAS of cowberry leaves and products of their processing our attention was drawn to the fact that the composition of the volatile substances has not been studied yet. The raw material has a specific odour; therefore, the aim of our work was to study the chemical composition of the volatile fraction of cowberry leaves since their qualitative and quantitative composition may affect the overall pharmacological effect of the drug created on its basis.

Materials and Methods

The object of research was cowberry leaves bought in the chemist's shop (batch 0715, manufactured by "Phyto svit" firm).

To obtain the essential oil from the raw material studied the method allowing to isolate the essential oil from a small amount of the plant raw material was used [6]. For distillation 22 ml "Agilent" vials (part number 5183-4536) with open lids and silicone seal were used. The weighed quantity of 2.0-3.0 g of the plant material was placed in a vial and filled with water to half the volume. The vial was closed with a lid with an air refrigerator and boiled on a sand bath for an hour. To avoid losses the trace amounts of the essential oil adsorbed on the inner surface of the refrigerator were washed twice with 1-2 ml of petroleum ether; the washings were collected to the vial.

The qualitative composition and quantitative content of terpenoids were determined by gas chromatography using an Agilent Technology 6890 gas chromatograph (GC) with a mass spectrometric detector 5973 (MS). For analysis the HP-5 chromatographic column with the length of 30 m and the internal diameter of 0.25 mm was used. The analysis was carried out under the following conditions: the thermostat temperature was programmed from 50°C to 250°C at the rate of 4°C/min; the temperature of the sample injection heater was 250°C; the carrier gas was helium; the flow rate – ml/min; the flow from GC to MS was heated to 230°C; the source temperature was maintained at 200°C; electron ionization was carried out at 70 eV in the mass ranging from m/z 29 to 450. Identification was performed based on comparison of the mass spectra obtained with the data of the NIST05-WILEY library (approximately 500000

Table

Continuation of Table

The chemical composition of volatile substances of cowberry leaves

No.	Retention time	The substance identified	The content of volatile substances (mg/kg)
1	2	3	4
1	8.886	<i>trans</i> -linalool oxide	0.74
2	9.333	<i>cis</i> -linalool oxide	0.49
3	9.803	linalool	5.03
4	12.124	terpinene-4-ol	0.78
5	12.217	p-cumene-8-ol	0.30
6	12.556	p-ment-1-en-8-ol	14.05
7	15.301	geraniol	2.03
8	16.033	thymol	3.64
9	17.637	eugenol	2.84
10	19.002	caprylic acid	1.55
11	19.241	1,2,3,4-tetrahydro-2,5,8-trimethylnaphthalene-1-ol	0.35
12	19.557	oct-7-en-2-ol	0.26
13	19.942	tetradecane	0.59
14	20.451	*	0.46
15	20.79	geranyl acetone	5.10
16	21.577	<i>trans</i> - β -ionone	0.64
17	21.662	<i>cis</i> - β -ionone	1.11
18	22.541	α -muurolene	0.87
19	22.826	γ -cadinene	0.92
20	23.042	α -bisabolol	0.83
21	24.514	caryophyllene oxide	4.15
22	24.807	lauric acid	2.03
23	25.185	benzophenone	3.06

1	2	3	4
24	26.218	tau-muorol	1.65
25	26.519	α -cadinol	1.26
26	27.136	vinyl laurate	0.37
27	27.622	6,10-methylundecane-2-one	0.22
28	29.194	myristic acid	9.11
29	29.472	ethyl myristate	1.39
30	30.297	6,10,14-pentadec-2-one	4.24
31	30.736	pentadecanoic acid	5.47
32	31.222	farnesyl acetone C	3.03
33	31.901	palmitoleic acid	19.14
34	32.34	palmitic acid	49.39
35	32.464	ethyl palmitate	11.02
36	32.626	14- β -pregnane	2.36
37	34.144	linoleic acid	4.29
38	34.299	linolenic acid	5.99
39	34.345	oleic acid	10.14
40	34.43	ethyl linoleate	0.98
41	34.476	ethyl linoleate	0.52
42	34.538	ethyl oleoate of stearic acid	1.05
43	34.615	ethyl stearate	4.32
44	34.885	tricosane	0.48
45	36.226	tetracosane	0.91
46	36.789	pentacosane	0.25
47	37.259	hexacosane	0.69
48	38.269	heptacosane	4.10
49	40.128	squalene	2.74
50	41.168	nonacosane	47.70
51	41.862	<i>trans</i> -linalool oxide	2.22

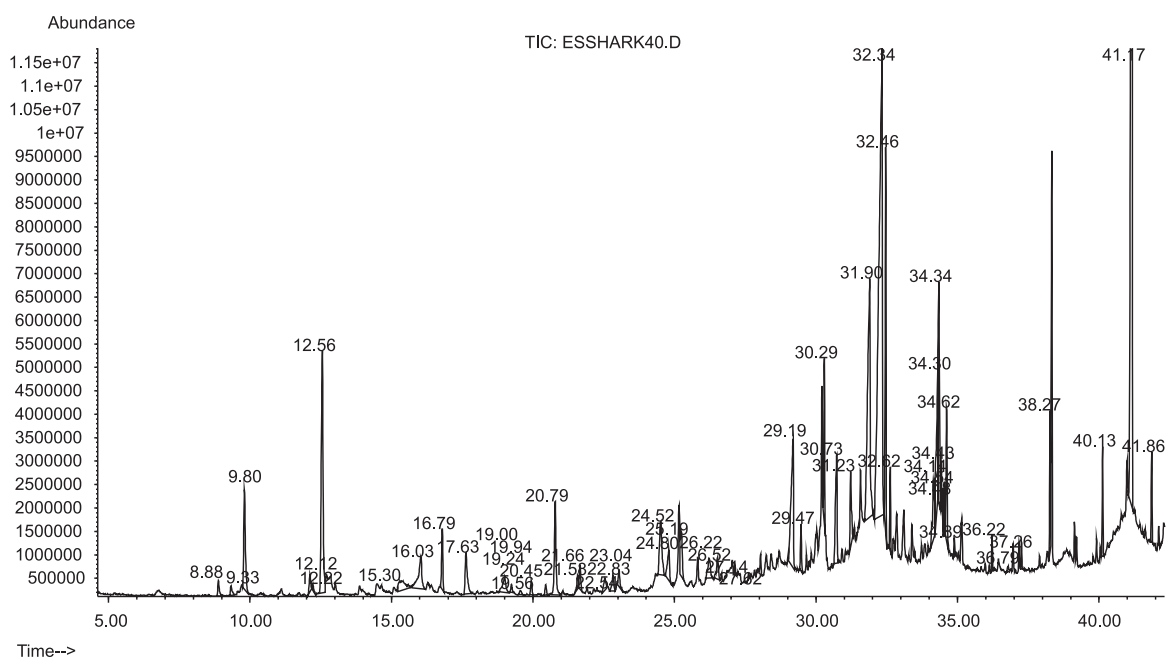


Fig. The chromatogram of volatile substances of cowberry leaves.

mass spectra). The retention indices of the components were calculated according to the results of control tests of substances with addition of the mixture of normal alkanes (C_{10} - C_{18}). The quantitative content of each component (%) was determined by the method of internal normalization [4].

Results and Discussion

The yield of volatile substances was calculated from the sum of all areas on the chromatogram. The content of volatile substances is 0.025%.

The results of the study of the chemical composition of the volatile fraction of cowberry leaves are presented in Table and Fig.

In cowberry leaves 50 volatile substances have been identified, identification of 1 substance has not been successful. Monoterpenes are presented by 14 substances,

derivatives of linalool, geraniol, thymol and eugenol are prevalent. Caryophyllene oxide and farnesyl acetone C prevail among sesquiterpenes. Among organic and fatty acids the predominant substances are lauric, myristic, pentadecanoic, palmitoleic, palmitic, linoleic and linolenic acids.

CONCLUSIONS

Using the method of gas chromatography the qualitative composition and quantitative content of volatile substances of cowberry leaves have been studied. Their content is 0.025%.

In cowberry leaves 51 substances have been found, 50 of them have been identified. The predominant substances are monoterpenes, fatty acids and triterpenes presented by p-ment-1-en-8-ol, palmitic acid, ethyl palmitate, oleic acid, and squalene.

REFERENCES

1. Комісаренко М.А., Гейдеріх А.С., Ковальова А.М., Кошовий О.М. // Укр. журн. клін. та лабораторної медицини. – 2012. – №2. – С. 24-26.
2. Комісаренко М.А., Кошовий О.М., Зайцев Г.П., Ковальова А.М. // Зб. наук. праць співробітників НМАПО ім. П.Л.Шупика. – 2015. – Вип. 24, кн. 5. – С. 127-131.
3. Комісаренко М.А., Кошовий О.М., Ковальова А.М., Сидора Н.В. // Зб. наук. праць співробітників НМАПО ім. П.Л.Шупика. – 2014. – Вип. 23, кн. 4. – С. 291-295.
4. Кошовий О.М., Комісаренко М.А., Ковальова А.М., Виноградов Б.А. // Фармац. журн. – 2012. – №4. – С. 101-104.
5. Фролов В.М., Гарник Т.П., Гришина В.С. // Фітотерапія. – 2006. – №4. – С. 32-35.
6. Черногород Л.Б., Виноградов Б.А. // Растит. ресурсы. – 2006. – Т. 42, вып. 2. – С. 61-68.

СКЛАД ЛЕТКИХ СПОЛУК ЛИСТЯ БРУСНИЦІ ЗВИЧАЙНОЇ

М.А.Комісаренко

Ключові слова: брусниця; листя; леткі речовини; газова хроматографія; терпени

Вивчення якісного складу та кількісного вмісту летких сполук листя брусниці звичайної проводили методом газової хроматографії. Ідентифікацію речовин проводили на основі порівняння отриманих мас-спектрів з даними бібліотеки NIST05-WILEY (близько 500000 мас-спектрів). Індокси утримання компонентів розраховували за результатами контрольних аналізів сполук з додаванням суміші нормальних алканів (C_{10} - C_{18}). У листі брусниці виявлено 51 речовину, 50 з яких ідентифіковано; домінуючими речовинами є сквален, пальмітинова кислота, етилпальмітат, олеїнова кислота. Встановлено, що вміст летких речовин складає 0,025%. Монотерпени представлені 14 речовинами, домінуючими є похідні леналоолу, гераніолу, тимол та евгенол. Домінуючими речовинами серед сесквітерпенів є каріофіленоксид та фарнезилу ацетон С. Переважаючими речовинами серед органічних та жирних кислот є лауринова, міристинова, пентадеканова, пальмітолеїнова, пальмітинова, лінолева та ліноленова кислоти.

СОСТАВ ЛЕТУЧИХ СОЕДИНЕНИЙ ЛИСТЬЕВ БРУСНИКИ ОБЫКНОВЕННОЙ

Н.А.Комиссаренко

Ключевые слова: брусника; листья; летучие вещества; газовая хроматография

Изучение качественного состава и количественного содержания летучих соединений листьев брусники обыкновенной проводили методом газовой хроматографии. Идентификация веществ проводилась на основе сравнения полученных масс-спектров с данными библиотеки NIST05-WILEY (около 500000 масс-спектров). Индексы удерживания компонентов рассчитывали по результатам контрольных анализов веществ с добавлением смеси нормальных алканов (C_{10} - C_{18}). В листьях брусники обнаружено 51 вещество, 50 из которых идентифицированы; доминирующими веществами являются сквален, пальмитиновая кислота, этилпальмитат, олеиновая кислота. Установлено, что содержание летучих веществ составляет 0,025%. Монотерпены представлены 14 веществами, доминирующими являются производные леналоола, гераниола, тимол и эвгенол. Доминирующими веществами среди сесквитерпенов являются карофилленоксид и фарнезил ацетон С. Преобладающими веществами среди органических и жирных кислот являются лауриновая, миристиновая, пентадекановая, пальмитолеиновая, пальмитиновая, линолевая и линоленовая кислоты.