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Study of Perfluoro Organic Solvent Novec 1230 Extraction Properties in Regards to Low Polar Substances from Plant Raw Materials

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***Abstract*—The purpose of this work is an experimental study of the extraction properties of Novec 1230 perfluoro organic solvent in regards to various types of low-polar biologically active substances from plant materials. For the study purposes, we used the following ground plant raw materials with particle size 0.1-0.5 mm: Anethum graveolens L. fruits, Eucalyptus viminalis Labill. leaves, Foeniculum vulgare Mill. fruits, Hypericum perforatum L. herb, Pastinaca sativa L. fruits, Pimpinella anisum L. fruits, Syzygium aromaticum L. buds (Clove buds), and Tumeric powder. For analytical purposes, we used the following standard samples: eugenol, carvone, trans-anethole, xanthotoxin, evcalimin, and spissum extract of Chlorophyllipt of the State Pharmacopoeia of Ukraine. Novec 1230 was used as a perfluoro organic solvent. RP HPLC and GC-MS were used as quantitative and qualitative analytical methods, respectively. For the extraction, we used the circulation method in "Soxhlet" extractor at the following conditions: plant raw material:extractant ratio 1:5 m/v, extraction time 1 and 3 h. It is found that in three hours of extraction with Novec 1230 perfluoro organic solvent, the low molecular weight components of essential oils are as follows: anethole and carvone (yield≥94%); eugenol is poorly extracted from Syzygium aromaticum L. buds (yield<50%); and chlorophylls, fatty oil, and a number of other low-polar biologically active substances are not extracted. The results obtained are comparable with the technologies for low-polar BAS extraction using liquefied gases or supercritical fluids. The extraction properties of Novec 1230 perfluoro organic solvent were studied experimentally for various types of low-polar biologically active substances from plant materials. It was found that a very limited number of low-polar biologically active substances are extracted into this type of perfluoro organic solvent, these are mainly components of essential oils, while triglycerides, chlorophylls, and a number of other low-polar biologically active substances are not extracted into this solvent. It is shown that the extraction properties of Novec 1230 perfluoro organic solvent are related to the polar part of TPSA parameter in low-polar BAS molecules. The limit value of TPSA was found in a molecule of low-polar BAS (LTPSA≤30 ± 5 Å²), above which BAS are not extracted into Novec 1230 perfluoro organic solvent.**

***Keywords*—perfluoro organic solvent Novec 1230, extraction properties, low-polar biologically active substances, limiting value of TPSA, circulation method in "Soxhlet" extractor.**

I. INTRODUCTION

Currently, the most promising technologies are the separation of low-polar biologically active substances (BAS) (fatty and essential oils, etc.) from plant materials using liquefied gases and supercritical fluids [1-5]. This is due to the fact that liquefied gases and supercritical fluids allow the extraction of more than 90% low-polar biologically active substances from plant materials within a few hours, and also allow the selective extraction of various low-polar substances depending on the extraction agent used and extraction conditions. In this case, the released biologically active substances are subjected to minimal thermal impact. However, these technologies have significant disadvantages: high cost of equipment, a need for high pressure and a refrigerator, and a need for additional energy costs at the stage of extractant regeneration.

Therefore, it is relevant to search for new solutions in the technology of selective separation of low-polar BAS from plant materials, which will allow avoiding the disadvantages described above.

Thus, a search for new solutions in the selective extraction technology of low-polar BAS from PRM that do not have these disadvantages seems to be actual.

One of such innovative solutions may be the use of new generation perfluoro organic solvents owing to their unique properties.

As a result of an information-patent search, the authors found that scientists from the USSR in the 80-90s, as well as from the EU and the USA in 2000-2006, experimentally discovered unique extraction properties in some perfluoro organic solvents [2, 6-10]. Among the solvents, perfluorocyclobutane (freon RC318), pentafluorobutane, methanol perfluorobutyl ether, ethanol perfluorobutyl ether, decafluoropentane, and perfluorohexane are mentioned.

In the Russian Federation, Novec 1230 perfluoro organic solvent is available on an industrial scale; it is used as a fire extinguishing agent (perfluoroketone, FK-5-1-12).

To compare some parameters of Novec 1230 perfluoro organic solvent and liquefied gases, their indicators are presented in Table I.

As can be seen from the data in Table 1, in many physicochemical, toxicological, and even environmental parameters, Novec 1230 perfluoro organic solvent is almost identical, and in some parameters, it is even superior to those of liquefied gases. From literature data, it is also known that perfluoro organic solvents, to one degree or another, have both, hydrophobic and lipophobic properties [7]. This fact, along with the above physicochemical, toxicological, and environmental indicators, suggests a high potential for the use of Novec 1230 perfluoro organic solvent in phytotechnology for selective extraction of low-polar substances.

Thus, based on an information-patent search, the authors suggest that Novec 1230 perfluoro organic solvent can be used to extract low-polar substances from plant materials.

The purpose of this work is an experimental study of the extraction properties of Novec 1230 perfluoro organic solvent in relation to various types of low-polar biologically active substances from plant materials.

II. EXPERIMENTAL

Plant raw material

For the study purposes, we used the following ground plant raw materials with particle size 0.1-0.5 mm: Anethum graveolens L. fruits, LLC Pharmaceutical shop “Medicinal plants”, Kharkiv, Ukraine, lot No. 981117, best before 09/2020; Eucalyptus viminalis Labill. leaves, “Krasnogorskleksredstva” company, Krasnogorsk, Russia, batch No. 100917, best before 10/2020; Foeniculum vulgare Mill. fruits, LLC Pharmaceutical shop “Medicinal plants”, Kharkiv, Ukraine, lot No. 135117, best before 08/2020; Hypericum perforatum L. herb, LLC Pharmaceutical shop “Medicinal plants”, Kharkiv, Ukraine, lot No. 120717, best before 07/2019; Pastinaca sativa L. fruits Globular cultivar, “Pnsemena” company, Ukraine, lot No. 454124, best before 12/2021; Pimpinella anisum L. fruits, LLC Pharmaceutical shop “Medicinal plants”, Kharkiv, Ukraine, lot No. 135117, best before 08/2020; Syzygium aromaticum L. buds (Clove buds), “Lechec” firm, Kharkiv, Ukraine, lot No. 002, best before 08/2020; and Tumeric powder “Goldiee”, Nayaganj, Kanpur, India, lot No 827, best before 01/2020.

Chemicals

Eugenol (CAS 97-53-0); Carvone (CAS 6485-4D-1); Trans-anethole (CAS 4180-23-8), Xanthotoxin (CAS 298-81-7), Sigma-Aldrich, Merck, with content ≥98.0 %;

TABLE I. SOME PHYSICOCHEMICAL, ENVIRONMENTAL, AND TOXICOLOGICAL PARAMETERS OF NOVEC 1230 PERFLUORO ORGANIC SOLVENT AND LIQUEFIED GASES [1]

Parameter	Extractant				
	Novec 1230	**Freon R22**	**Freon R227ea**	**Freon RC318**	**Carbon dioxide**
1. Empirical formula	$C_6F_{12}O$	$CHClF_2$	C_3F_7H	cC_4F_8	CO_2
2. Molecular weight, g/mol	316	86.5	170.0	200.0	44.0
3. Density at 20 °C, kg/m³	1,600	1,200	1,407	1,517	930
3. Pressure 20 °C, MPa	0.040	0.91	0.391	0.272	6.0
4. Boiling point, °C	49	-40.8	-16.4	-5.8	-78.5
5. Heat of vaporization, kJ/kg	88	232	133	117	155
6. Dielectric constant	1.8-2.3	3.1	2.0	1.0	1.6
6. Global warming potential	1	1,760	3,300	10,300	1
7. Ozone Depletion Potential	0	0.055	0	0	0
8. Toxicity, ppm	4,000	3,000	3,000	3,000	5,000

* Note, “-” sign means that no data was found.

Evcalimin standard sample of the All-Russian Scientific Research Institute of Medicinal and Aromatic Plants (a sum of macrocarpales); Spissum extract of Chlorophyllipt of the State Pharmacopoeia of Ukraine (a sum of cupric chlorophylls, euglobals with monoterpenoid structure and macrocarpales), were used as reference substances.

Novec 1230 (3M company, the USA) with content ≥99.0 % was used as perfluoro organic solvent.

Method of extraction

5.00 g sample of ground PRM (particle fraction of 0.1-0.5 mm) was put into Soxhlet extractor, then 25.0 ml of the solvent was added, and the process of extraction was carried out during the predefined period. The solution obtained was evaporated to obtain the extract, which was then blown by air for 5-10 min. The extract was diluted with ethanol 96 % v/v. The ethanolic solution was weighted, its density was determined, and BAS content was analyzed by the method of RP HPLC.

GC-MS method of analysis

Qualitative analysis of extracts was carried out by GC-MS method using equipment GCMS-QP2010 Ultra, by Shimadzu, Japan. Column: Zebron ZB-5MS, length 30 m, internal diameter 0.25 mm, film thickness 0.25 μm; liquid phase: 5% phenyl-arylene, 95% dimethylsiloxane; temperature settings: from 70 C° to 325/350 C°. Gas-carrier was helium with stable flow: 3.0 ml/min. The injection volume was 1 μl. Detection was carried out in full ion current regime (SCAN) in the range of m/z from 30 to 500 Da.

Reverse phase high performance liquid chromatography (RP HPLC)

Analyses of BAS were carried out by the method of RP HPLC on Agilent Technology chromatograph, Agilent 1200 Infinity model, the USA. The analyses were carried out under the following conditions: 1 % water solution of formic acid was used as mobile phase A and ethanol 96% v/v with linear gradient regime of elution was used as mobile phase B; the velocity of mobile phase was 0.5 ml/min; chromatography column: Supelco Ascentis express C18, length: 100 mm, internal diameter: 4.6 mm, particle size: 2.7 μm; temperature of chromatography column: + 35°C; sample volume: 1 μl. The conditions of RP HPLC analyses were similar to those described in the article by Zhilyakova et al. [11]. Analytical wavelengths: 240, 275, 280, 284, and 302 nm.

Theoretical part

To allow mathematical formalization and quantitative description of the extraction properties of Novec 1230 perfluoro organic solvent, the authors used a molecular descriptor: the size of the topological polar surface (TPSA) of a low-polar BAS molecule, since this parameter is associated with the surface energy and the interaction energy between the molecules of the perfluoro organic solvent and BAS, which affect solubility of the substance in the solvent and, accordingly, its extraction properties.

TPSA parameter was found in on-line base PubChem Database [12].

III. Results and Discussion

At the first stage of our studies, we obtained experimental data of the yield of low-polar BAS from PRM at their extraction by the circulation method in Soxhlet extractor using perfluoro organic solvent Novec 1230. The results of our studies are presented in Table II.

As can be seen from the data in Table 2, in three hours of extraction with Novec 1230 perfluoro organic solvent, the following low molecular weight components of the essential oils are extracted perfectly: anethole and carvone (yield≥94%); eugenol is extracted poorly from Syzygium aromaticum L. buds (yield <50%); and chlorophylls, fatty oil, and a number of other low-polar biologically active substances are not extracted.

For mathematical formalization and quantitative description of the extraction properties of Novec 1230 perfluoro organic solvent relative to various types of low-polar BAS, the authors used TPSA molecular parameter for low-polar BAS. The obtained theoretical data and

TABLE II. Systematic Results of Studies on the Extraction of Low-Polar BAS from PRM by the Circulation Method in Soxhlet Extractor Using Perfluoro Organic Solvent Novec 1230

PRM	BAS	Yield of BAS by the 1st h, %*	Yield of BAS by the 3rd h, %	Note
1. *Anethum graveolens* L. fruits	Carvone	93±5	97±5	Triglycerides were undetected
2. *Eucalyptus viminalis* Labill. leaves	Euglobals of monoterpenoid structure, chlorophylls and macrocarpales	None	None	Essential oil was detected (eucalyptol).
3. *Foeniculum vulgare* Mill. fruits	Anethole	71±4	94±5	Triglycerides were undetected
4. *Hypericum perforatum* L. herb	Hyperforin	None	None	Chlorophylls were undetected
5. *Pastinaca sativa* L. fruits	Xanthotoxin	None	None	Octyl butyrate was detected. Triglycerides were undetected
6. *Pimpinella anisum* L. fruits	Anethole	71±4	100±5	Triglycerides were undetected
7. *Syzygium aromaticum* L. buds	Eugenol	Less than 50		-
8. Turmeric powder	Curcuminoids	None	None	Essential oil was detected (curmene, zingiberene, curlone, ar-tumerone etc.)

* The mean value and its confidence interval (Mean±SEM) are calculated with repeat counts n=3 and significance level P=0.95.

experimental results are presented in Table III. As can be seen from the data in Table III, when low-polar BAS are arranged in a row according to their molecular parameter TPSA and compared with the experimentally discovered fact of their extraction, the limiting value of the molecular parameter TPSA (LTPSA ≤30±5 Å²), which characterizes the extraction properties of perfluoro organic solvent Novec 1230. At the same time, a very limited number of low-polar BAS, which have small TPSA ≤30 Å², are extracted into this type of perfluoro organic solvent. Basically, these are low molecular weight BAS, which are characteristic of essential oils, while higher molecular weight lipophilic substances, such as triglycerides, chlorophylls, etc., are not extracted into this solvent.

IV. DISCUSSION

The obtained experimental results are consistent with the hypothesis that the extraction properties of Novec 1230 perfluoro organic solvent should be related to the polar part of TPSA parameter in low-polar BAS molecules. Moreover, the limiting value of this molecular parameter was found, above which BAS can no longer overcome the hydrophobic repulsive forces between the solvent molecules and, accordingly, cannot be extracted from plant materials.

These results make it possible to predict the possibility of extracting of low-polar biologically active substances from plant materials using this type of solvent according to the magnitude of the polar part of TPSA in the biologically active substance. Moreover, for successful extraction of BAS molecules from plant materials using Novec 1230 perfluoro organic solvent, the condition TPSA≤30±5 Å² should be fulfilled.

This allows developing a method for selective extraction of certain types of low-polar BAS from plant materials. So, for example, we have patented in the Russian Federation methods for extracting the components of essential oil from fennel and dill fruit using this solvent and using the circulating extraction method in “Soxhlet” [13, 14].

Moreover, in case of dill fruit, the yield of carvone was achieved (93÷99)±4%, the yield of carvone in relation to the weight of plant material was (1.38÷1.48)±0.05% by weight, and the yield of lipophilic substances in relation to the weight of plant material was (3.6 ÷ 3.7)±0.2% by weight. Extraction conditions: the ratio plant material: extractant 1: (3÷6) wt./vol., extraction time 1÷4 hours.

In case of fennel fruit, the yield of anethole was (71÷94)±4%, the yield of anethole in relation to the weight of plant material was (1.60÷2.10)±0.06% by weight, and the yield of lipophilic substances in relation to the weight of plant material was (2.4÷2.7)±0.1% by weight. Extraction conditions: the ratio plant material extractant 1:(3÷6) wt./vol., extraction time 1÷3 hours. The results described above are comparable with the technologies for the extraction of low-polar biologically active substances using liquefied gases or supercritical fluids [1, 2, 4, 5].

On the whole, the results show a high potential for the use of perfluoro organic solvents (including Novec 1230) in phytotechnology for the selective extraction of low-polar BAS from plant materials. Moreover, the use of perfluoro organic solvents may become a new direction in the technology for the extraction of low-polar biologically active substances from plant materials.

TABLE III. TPSA OF LOW-POLAR BAS AND POSSIBILITY OF THEIR EXTRACTION BY NOVEC 1230

	BAS	TPSA, Å²	Possibility of extraction*
1.	Curmene, Zingiberene	0	+
2.	Anethole, Eucalyptol	9	+
3.	Ar-tumerone, Carvone, Curlone	17	+
4.	Octyl butyrate	26	+
5.	Eugenol	29	+/-
6.	Xanthotoxin	49	-
7.	Xanthotoxol	60	-
8.	Hyperforin	71	-
9.	Triglycerides	79	-
10.	Euglobal IIb	84	-
11.	Curcumin	93	-
12.	Macrocarpal C	95	-
13.	Chlorophyll *a*	96	-
14.	Chlorophyll *b*	114	-
15.	Macrocarpal A	115	-
16.	Limiting value of TPSA of low-polar BAS molecules for the solvent (*LTPSA*), Å²		30±5

Note. * (+) means that BAS is extracted by the solvent; (-) means that BAS is not extracted by the solvent; (+/-) means that BAS is poorly extracted by the solvent; ** n/a means that data is not available.

V. CONCLUSION

The extraction properties of Novec 1230 perfluoro organic solvent with various types of low-polar BAS from plant materials have been experimentally studied. It has been found that a very limited number of low-polar BAS are extracted into this type of perfluoro organic solvent; these are mainly components of essential oils, while triglycerides, chlorophylls and a number of other low-polar BAS are not extracted into this solvent. It has been shown that the extraction properties of Novec 1230 perfluoro organic solvent are related to the polar part of TPSA parameter in low-polar BAS molecules. The limiting value of TPSA was found in a molecule of low-polar BAS (LTPSA≤30±5 Å²), above which BAS are not extracted into Novec 1230 perfluoro organic solvent. Based on Novec 1230 perfluoro organic solvent, new methods for selective extraction of the components of essential oils from fennel and dill fruits using the circulating extraction method in "Soxhlet" have been developed.

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REFERENCES

[1] Technology and standardization of drugs: Collection of research papers. Edited by Georgievskiy V.P., Konev F.A.. Kharkiv: LLC RIREG; 2000.

[2] P.A. Dowdle, S.S.H. Merseyside, S. Corr, A. Cheshire, H. Harris, W. Cheshire, Solvent extraction process, Patent US 2004/0105899 A1. A61K 35/78. Pub. Date: Jun.3, 2004.

[3] G. Brunner, Gas Extraction. An introduction to fundamentals of supercritical fluids and the application to separation processes. New York: Springer, 1994.

[4] T. Diego, D.Y. Santos, M.A.A. Meireles, "Extraction of Volatile Oils by Supercritical Fluid Extraction: Patent Survey", Recent Patents on Engineering, vol. 5(1), pp. 17-22, 2011.

[5] M. Mukhopadhyay, Natural extracts using supercritical carbon dioxide. 1st ed., Boca Raton: CRC Press, 2000.

[6] F. Chemat, M.A.Vian, Alternative solvents for natural products extraction. Green chemistry and sustainable technology. Berlin Heidelberg: Springer-Verlag, 2014.

[7] Perfluoro organic chemistry. Principles and commercial applications, Edited by Banks R.E., Smart B.E., Tatlow J.C. New York: Springer, 1994.

[8] B. Lemaire, B. Mompon, I. Surbled, M. Surbled, Method for fractionating essential oils using at least fluorinated solvent, Patent EP 1307533B1. C11B9/025. Priority date 2000-06-22.

[9] B. Lemaire, M. Surbled, J. Zwegers, B. Mompon, Method for extracting and fractionating fats with solvent, using at least a hydrofluroether, Patent US 6673952 B2, C11B 1/00. Pub. Date: Jan.6, 2004.

[10] P. Dournel, M. Surbled, B. Mompon, Process for separating out at least one organic compound, Patent US 2006/0182689 A1. A61L 9/04, A61K 8/00, C07C 405/00, C07J 9/00, C07C 61/06. Pub. Date: Aug.17, 2006.

[11] E.T. Zhilyakova, O.O. Novikov, D.I. Pisarev, A.Y. Malyutina, N.N. Boyko, "Studying the polyphenolic structure of Laurus nobilis L. leaves", Indo Am J Pharm Sci, vol. 4(09), pp. 3066-3074, 2017.

[12] U.S. National library of medicine. National center for biotechnology information. PubChem Database. https://pubchem.ncbi.nlm.nih.gov (access date 06.08.2019).

[13] N.N. Bojko, E.T. Zhilyakova, D.I. Pisarev, O.O. Novikov, L.V. Nefedova, Method for production of essential oil of Dill, Patent RU 2696134 C1. C11B9/00. Pub. Date: 01.08.2019.

[14] N.N. Bojko, E.T. Zhilyakova, D.I. Pisarev, A.V. Bondarev, O.O. Novikov, Method for production of essential oil of common Fennel fruits. Patent RU 2697097 C1. C11B9/00. Pub. Date: 12.08.2019.