ТЕХНОЛОГІЯ ЛІКАРСЬКИХ ПРЕПАРАТІВ

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The technological aspects for complex processing of hawthorn fruits

Considering the shortage of the plant raw material the issue of its complex processing is relevant, and it contributes to its rational use.

Aim. To develop the technology for complex processing of hawthorn fruits.

Materials and methods. The lipophilic complex (LC) extraction was carried out with chloroform in the raw material-extractant ratio of 1 : 10; the phenolic complex (PhC) was prepared with the ethyl acetate-alcohol mixture in the ratio of 8 : 2; WSPC was obtained by triple extraction of the fruits meal after preparing LC and PhC with hot purified water followed by concentration and precipitation with 96 % ethyl alcohol.

Results and discussion. It was found that the yield of LC in fruits of the hawthorn species studied was from 1 % to 10 %, the highest yield was determined in the fruits of *C. submollis* Sarg., *C. succulenta* Schrad. and *C. densiflora* Sarg. The yield of PhC was from 9 % to 18 %, *C. submollis* Sarg., *C. mollis* Sarg., *C. arnoldiana* Sarg. and *C. canadensis* Sarg. dominated. WSPC was from 15 % to 35 %, the highest yield was determined for fruits of *C. submollis* Sarg., *C. macracantha* Lodd, *C. mollis* Sarg., *C. arnoldiana* Sarg., *C.succulenta* Schrad.

Conclusions. The technology of the complex processing of hawthorn fruits – *C. submollis* Sarg., *C. mollis* Sarg., *C. arnoldiana* Sarg., *C. canadensis* Sarg., *C. oxyacantha* L., *C. monogyna* Jack., *C. sanguinea* Pall., *C. pentagyna* Waldst. Et Kit., *C. aronia* (L.) Bosc, *C. densiflora* Sarg., *C. succulenta* Schrad, *C. macracantha* Lodd., *C. flabellata* (*Bosc*) C. Koch., *C. pseudokyrtostylla* Klok., *C. ukrainica* A. Pojark., *C. ambiguae* C. A. M – has been developed. As a result of complex processing of the plant raw material such complexes as LC, PhC and WSPC have been obtained.

Key words: *hawthorn; fruits; complex processing; technology*

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Технологічні аспекти комплексної переробки плодів глоду

Зважаючи на дефіцит рослинної сировини, актуальним є питання комплексної переробки, що створює передумови для її раціонального використання.

Метою даної роботи є розробка технології комплексної переробки плодів глоду.

Матеріали та методи. Екстракцію ЛК із сировини проводили хлороформом у співвідношенні сировина-екстрагент 1 : 10; ФК отримували етилацетатно-спиртовою сумішшю у співвідношенні 8 : 2; ВРПС отримували трикратною екстракцією шроту плодів після одержання ЛК та ФК гарячою водою очищеною та подальшим концентруванням та осадженням спиртом етиловим 96 %.

Результати та їх обговорення. Встановлено, що вихід ЛК у плодах досліджених видів глодів становить від 1 % до 10 %, найбільший вихід визначено для плодів *C. submollis* Sarg., *C. succulenta* Schrad. та *C. densiflora* Sarg. Вихід ФК становить від 9 % до 18 %, найбільший вихід визначено для *C. submollis* Sarg., *C. mollis* Sarg., *C. arnoldiana* Sarg. та *C. canadensis* Sarg. ВРПС містяться від 15 % до 35 %, найбільший вихід ВРПС визначено для плодів *C. submollis* Sarg., *C. macracantha* Lodd, *C. mollis* Sarg., *C. arnoldiana* Sarg., *C.succulenta* Schrad.

Висновки. Розроблено технологію комплексної переробки плодів глодів: *C. submollis* Sarg., *C. mollis* Sarg., *C. arnoldiana* Sarg., *C. canadensis* Sarg., *C. oxyacantha* L., *C. monogyna* Jack., *C. sanguinea* Pall., *C. pentagyna* Waldst. Et Kit., *C. aronia* (L.) Bosc, *C. densiflora* Sarg., *C. succulenta* Schrad, *C. macracantha* Lodd., *C. flabellata* (*Bosc*) C. Koch., *C. pseudokyrtostylla* Klok., *C. ukrainica* A. Pojark., *C. ambiguae* C. A. M. У результаті комплексної переробки рослинної сировини були одержані ліпофільні (ЛК), фенольні (ФК) та водорозчинні полісахаридні (ВРПС) комплекси.

Ключові слова: *глід; плоди; комплексна переробка; технологія*

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Технологические аспекты комплексной переработки плодов боярышника

Учитывая дефицит растительного сырья, актуальным является вопрос его комплексной переработки, что создает предпосылки для его рационального использования.

Целью данной работы является разработка технологии комплексной переработки плодов боярышника.

Материалы и методы. Экстракцию ЛК из сырья проводили хлороформом в соотношении сырье-экстрагент 1 : 10; ФК получали этилацетатно-спиртовой смесью в соотношении 8 : 2; ВРПС получали трехкратной экстракцией шрота плодов после получения ЛК и ФК горячей водой очищенной с последующим концентрированием и осаждением спиртом этиловым 96 %.

Результаты и их обсуждение. Установлено, что выход ЛК в плодах исследованных видов боярышников составил от 1 % до 10 %, наибольший выход определен в плодах *C. submollis* Sarg., *C. succulenta* Schrad. и *C. densiflora* Sarg. Выход ФК составил от 9 % до 18 %, преобладают *C. submollis* Sarg., *C. mollis* Sarg., *C. arnoldiana* Sarg. и *C. canadensis* Sarg. ВРПС содержатся от 15 % до 35 %, наибольший выход ВРПС установлен для плодов *C. submollis* Sarg., *C. macracantha* Lodd, *C. mollis* Sarg., *C. arnoldiana* Sarg., *C. succulenta* Schrad.

Выводы. Разработана технология комплексной переработки плодов боярышников: *C. submollis* Sarg., *C. mollis* Sarg., *C. arnoldiana* Sarg., *C. canadensis* Sarg., *C. oxyacantha* L., *C. monogyna* Jack., *C. sanguinea* Pall., *C. pentagyna* Waldst. Et Kit., *C. aronia* (L.) Bosc, *C. densiflora* Sarg., *C. succulenta* Schrad, *C. macracantha* Lodd., *C. flabellata* (*Bosc*) C. Koch., *C. pseudokyrtostylla* Klok., *C. ukrainica* A. Pojark., *C. ambiguae* C. A. M. В результате комплексной переробки растительного сырья были получены липофильные (ЛК), фенольные (ФК) и водорастворимые полисахаридные (ВРПС) комплексы.

***Ключевые слова**: боярышник; плоды; комплексная переработка; технология*

Lately herbal drugs are gaining popularity at the pharmaceutical market of Ukraine. Together with this, reduction of natural resources of wild species of the plant raw material and increase of prices for the imported raw material should be taken into account. The topical problem of modern phytochemical production is ineffective use of the raw material [1].

In conditions of the raw material shortage of caused by various factors, including the environmental situation and urbanization, the issue of complex processing of the raw material is of current interest, which creates prospects for its rational use.

When working with the raw material in many cases the emphasis is made on extraction of certain classes of biological active substances (BAS), but the meal is not used although the rational use of wastes will allow obtaining BAS that differ by their physicochemical properties and pharmacological activity [2].

One of the directions of the rational use of the raw material and the cost reduction of herbal drugs is development of the rational technology for complex processing of the raw material, which allows obtaining several pharmacologically active substances from a single plant object. The extraction of BAS is carried out consistently with solvents of different polarity [2].

The aim of the work is develop the technology for complex processing of hawthorn fruits for obtaining BAS of different chemical nature and pharmacological activity.

Materials and methods

The objects of the study were fruits of hawthorn, which belong to different botanical sections by their morphological structure and differ in size, the nature of the pulp and number of seeds (from 1 to 5): *C. submollis* Sarg., *C. mollis* Sarg., *C. arnoldiana* Sarg., *C. canadensis* Sarg., *C. oxyacantha* L., *C. monogyna* Jack., *C. sanguinea* Pall., *C. pentagyna* Waldst. Et Kit., *C. aronia* (L.) Bosc, *C. densiflora* Sarg., *C. succulenta* Schrad, *C. macracantha* Lodd., *C. flabellata* (Bosc) C. Koch., *C. pseudokyrtostylla* Klok., *C. ukrainica* A. Pojark., *C. ambiguae* C. A. M. [3, 4, 5, 6].

The raw material was collected in August-September of 2016 in the Kharkiv, Poltava, Kyiv regions and the Botanical Garden of V. N. Karazin National University. Hawthorn fruits were gathered in the phase of full ripeness. The dry raw material was used.

Obtaining of Extracts

The lipophilic complex (LC). The accurate weights of the raw material (hawthorn fruits) were crushed to the particle size of 3 mm, separated from the dust through a sieve No. 18, placed in a filter paper bag and weighed on the analytical balance. The raw material prepared was subjected to exhaustive extraction in a Soxhlet apparatus with chloroform in the raw material-extractant ratio of 1 : 10 [7, 8].

The duration of extraction was 12-15 hours at a temperature of 60-65 °C. The extraction was carried out on a water bath until the extract was discolored in the discharge spout. The flask-receiver was weighed before and after extraction.

To remove the extractant the flask was dried at a temperature of 60 °C for 30 min. The samples of the complexes obtained had the appearance of a resinous liquid of a yellow-brown color.

The phenolic complex (PhC). The meal dried in the air at room temperature was weighed on the analytical balance and extensively extracted in a closed cycle with the ethyl acetate – alcohol mixture (8 : 2) in the general ratio of the raw material-extractant (1 : 10) for 2 hours at a constantly maintained temperature of 75-80 °C. After that the extractant was evaporated, the content of the flask was dried and weighed.

The samples of phenolic complexes obtained were resinous compounds from light brown to dark brown color with a characteristic odor [9].

The water soluble polysaccharide complex (WSPC). The meal dried in the air at room temperature after obtaining the phenolic complex was weighed on the analytical balance, placed in a round-bottomed flask and poured with hot purified water in the ratio of 1 : 5 and kept on a water bath at a temperature of 90 °C for 30 min.

Extraction with water was conducted three times. To do this, each extraction was poured into a separate flask and filled with a new portion of hot purified water.

The aqueous extracts obtained were combined, concentrated first to the volume corresponding to the mass of the raw material and after up to 1/3 of the previous volume.

For concentration a rotary evaporator was used. The resulting concentrate was added to a three-fold volume of 96 % ethyl alcohol while heating. The precipitate formed was separated by centrifugation. The samples of WSPC obtained were jelly-like substances.

The mixture of WSPC was dried in a dryer, and the quantitative content was determined by gravimetry. The complexes obtained are amorphous compounds from a yellow to yellow-brown color.

Results and discussion

Before determining the yield of BAS in the samples obtained the loss on drying of the fruits of the hawthorn species studied was identified. The moisture of the raw material was determined according to the pharmacopoeial method in five replicates [10], the results were statistically processed (Tab. 1).

As a result of complex processing of hawthorn fruits lipophilic, phenolic and water soluble polysaccharide complexes with a significant yield of BAS were obtained. The flowchart of complex processing of hawthorn fruits of different species is shown in Fig.

Table 1

The results of the loss on drying determination in hawthorn fruits (n = 5)

Hawthorn species	Loss on drying, %
C. submollis Sarg.	10.1512 ± 0.1423
C. mollis Sarg.	11.2513 ± 0.3712
C. arnoldiana Sarg.	9.1705 ± 0.1234
C. canadensis Sarg.	7.2512 ± 0.0623
C. oxyacantha L.	6.1321 ± 0.1314
C. monogyna Jack.	6.2500 ± 0.1314
C. sanguinea Pall.	7.1810 ± 0.2516
C. pentagyna Waldst. Et Kit.	6.8512 ± 0.2543
C. aronia (L.) Bosc	7.5643 ± 0.5412
C. densiflora Sarg	9.1325 ± 0.2612
C. succulenta Schrad	8.4500 ± 0.4532
C. macracantha Lodd	8.1005 ± 0.1652
C. flabellata (Bosc) C. Koch	7.8521 ± 0.5210
C. pseudokyrtostylla Klok.	10.2215 ± 0.3542
C. ukrainica A. Pojark.	9.1214 ± 0.2235
C. ambiguae C. A. M.	8.7505 ± 0.4710

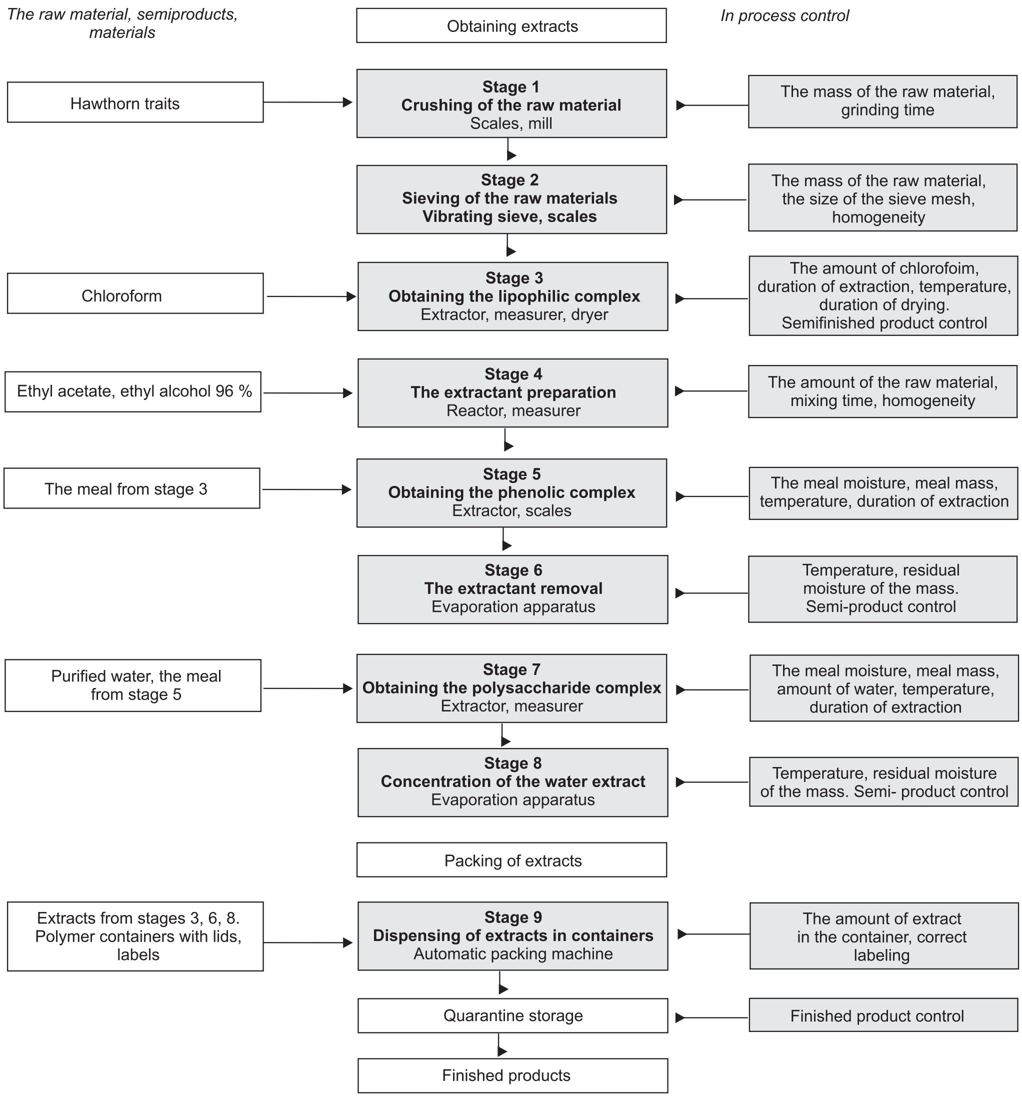

Fig. The flowchart of complex processing of hawthorn fruits

Table 2

The yield of lipophilic and phenolic complexes of hawthorn fruits

Hawthorn species	The yield of LC, %	The yield of PhC, %
C. submollis Sarg.	10.12	18.12
C. mollis Sarg.	6.75	18.45
C. arnoldiana Sarg.	4.25	18.62
C. canadensis Sarg.	5.12	17.45
C. oxyacantha L.	4.45	10.12
C. monogyna Jack.	3.65	9.25
C. sanguinea Pall.	4.15	10.24
C. pentagyna Waldst. Et Kit.	5.35	12.15
C. aronia (L.) Bosc	5.15	10.75
C. densiflora Sarg	8.94	14.15
C. succulenta Schrad	9.17	13.95
C. macracantha Lodd	2.85	13.10
C. flabellata (*Bosc*) C. Koch	2.15	12.76
C. pseudokyrtostylla Klok.	1.95	8.17
C. ukrainica A. Pojark.	1.85	9.25
C. ambiguae C. A. M.	1.75	8.75

As can be seen from Fig., the technological process of complete depletion of the hawthorn raw material consists of 9 stages: crushing of the raw material, sieving of the raw material, obtaining the lipophilic complex, the extractant preparation, obtaining the phenolic complex, the extractant removal, obtaining the polysaccharide complex, concentration of the water extract, dispensing of extracts in containers.

The yield of the complexes taking into account the moisture content of the raw material was calculated by the formula:

$$X = \frac{m_1 \cdot 100 \cdot 100}{m_2 \cdot (100 - W)},$$

where: m_1 – is the mass of the resulting complex, g; m_2 – is the accurate weight of the raw material, g; W – is the loss in mass when dried (moisture), %.

The yield of lipophilic and phenolic complexes of hawthorn fruits species is presented in Tab. 2.

As can be seen from Tab. 2, the highest yield of LC (%) was determined for fruits of *C. submollis* Sarg. (10.12), *C. succulenta* Schrad. (9.17) and *C. densiflora* Sarg. (8.94). The highest yield of PhC (%) was found for *C. submollis* Sarg. (18.12), *C. mollis* Sarg. (18.45), *C. arnoldiana* Sarg. (18.62) and *C. canadensis* Sarg. (17.45).

The yield of WSPC of hawthorn fruits is given in Tab. 3.

Table 3

The yield of water soluble polysaccharide complexes of hawthorn fruits

Hawthorn species	The moisture content of WSPC, (%)	The yield of WSPC, (%)
C. submollis Sarg.	7.32	35.75
C. mollis Sarg.	7.45	32.12
C. arnoldiana Sarg.	7.24	30.65
C. canadensis Sarg.	6.95	29.70
C. oxyacantha L.	5.64	25.12
C. monogyna Jack.	5.93	22.18
C. sanguinea Pall.	6.23	28.45
C. pentagyna Waldst. Et Kit.	5.13	27.89
C. aronia (L.) Bosc	4.95	20.15
C. densiflora Sarg	6.81	24.86
C. succulenta Schrad	6.32	30.15
C. macracantha Lodd	5.95	32.16
C. flabellata (*Bosc*) C. Koch	4.13	28.75
C. pseudokyrtostylla Klok.	5.71	15.12
C. ukrainica A. Pojark.	5.31	16.75
C. ambiguae C. A. M.	5.17	15.78

The data of Tab. 3 show that the highest yield (%) of WSPC was determined for fruits *C. submollis* Sarg. (35.75), *C. macracantha* Lodd (32.16), *C. mollis* Sarg. (32.12), *C. arnoldiana* Sarg. (30.65), *C. succulenta* Schrad (30.15).

CONCLUSIONS

1. Based on the experimental studies the technology of complex processing of fruits of 16 hawthorn species – *C. submollis* Sarg., *C. mollis* Sarg., *C. arnoldiana* Sarg., *C. canadensis* Sarg., *C. oxyacantha* L., *C. monogyna* Jack., *C. sanguinea* Pall., *C. pentagyna* Waldst. Et Kit., *C. aronia* (L.) Bosc, *C. densiflora* Sarg., *C. succulenta* Schrad, *C. macracantha* Lodd., *C. flabellata* (*Bosc*) C. Koch., *C. pseudokyrtostylla* Klok., *C. ukrainica* A. Pojark., *C. ambiguae* C. A. M. – has been developed.

2. For the purpose of complete depletion of the raw material the studies on isolation of hydrophilic substances after separation from the meal of the pulp of lipophilic substances were conducted. It has been found that 70 % ethyl alcohol is the optimal extractant, and the optimal extraction time is 5-5.5 hours.

3. As a result of the study such complexes as LC, PhC and WSPC have been obtained from the plant raw material.

4. It has been experimentally determined that the yield of LC is from 1 to 10 %; PhC – 8-18 %, WSPC – 15-35 % depending on the species of hawthorn.

Conflict of Interests: authors have no conflict of interests to declare.

REFFERENCES

1. Ветров, П. П. Технология комплексной переработки и рациональное использование лекарственного растительного сырья / П. П. Ветров, С. В. Гарная, А. И. Русинов // Фітотерапія. Часопис. – 2005. – № 4. – С. 59–62.
2. Оптимізація технології екстракції ліпофільних комплексів з лікарської рослинної сировини / С. В. Гарна, П. П. Вєтров, О. І. Русинов, В. А. Георгіянц // Запорожский мед. журн. – 2010. – № 3. – С. 92–94.
3. Phipps, J. B. Biogeographic, Taxonomic and Cladistic Relationships between East Asiatic and North American Crataegus / J. B. Phipps // Annals of the Missouri Botanical Garden. – 1983. – Vol. 70, Issue 4. – P. 667–700. doi: 10.2307/2398984
4. Phipps, J. B. Crataegus : A Nomenclator for Sectional and Serial Names / J. B. Phipps // Taxon. – 1983. – Vol. 32, Issue 4. – P. 598–604. doi: 10.2307/1221729
5. Origin and evolution of subfam. Maloideae (Rosaceae) / J. B. Phipps, K. R. Robertson, J. R. Rohrer, P. G. Smith // Systematic Botany. – 1991. – Vol. 16, Issue 4. – 302 p. doi: 10.2307/2419283
6. Wells, T. C. Studies in Crataegus (Rosaceae, Maloideae) XX. Interserial hybridization between Crataegus monogyna (series Oxyacanthae) and Crataegus punctata (series Punctata) in southern Ontario / T. C. Wells, J. B. Phipps // Canadian J. of Botany. – 1989. – Vol. 67, Issue 8. – P. 2465–2472. doi: 10.1139/b89–316
7. Phytochemical research of Crataegus submollis Sarg. leaves lipophilic complex and study of its antibacterial activity / N. V. Sydora, A. M. Кovaleva, V. K. Yakovenko et al. // Der Pharmacia Lett. – 2016. – Vol. 8, Issue 21. – P. 19–23.
8. Chromato–mass–spectrometry identification of lipophilic compounds in plant extract Liliia / I. Vyshnevska, Ye. Degtiarova, M. Bavikina, E. Bisaga // The Pharma Innovation J. – 2015. – Vol. 311. – P. 72–74.
9. Олійник, С. В. Актуальність використання різних видів сировини Crataegus при розробці гомеопатичних препаратів : зб. наук. праць / С. В. Олійник, Л. І. Вишневська // Сучасні досягнення фармацевтичної технології і біотехнології. – Х., 2017. – С. 141–143.
10. Державна фармакопея України / Державне підприємство «Український науковий фармакопейний центр якості лікарських засобів». – 2–е вид. – Х. : Український науковий фармакопейний центр якості лікарських засобів, 2014. – 732 с.

REFERENCES

1. Vetrov, P. P., Garnaia, S. V., Rusinov, V. A. (2005). *Fitoterapia. Chasopys, 4*, 59–62.
2. Garna, S. V., Vietrov, P. P., Rusinov, V. A., Georgiiants, V. A. (2010). *Zaporozhskii meditcinskii zhurnal, 3*, 92 – 94.
3. Phipps, J. B. (1983). Biogeographic, Taxonomic, and Cladistic Relationships Between East Asiatic and North American Crataegus. *Annals of the Missouri Botanical Garden, 70 (4)*, 667. doi: 10.2307/2398984
4. Phipps, J. B. (1983). Crataegus: A Nomenclator for Sectional and Serial Names. *Taxon, 32 (4)*, 598. doi: 10.2307/1221729
5. Phipps, J. B., Robertson, K. R., Rohrer, J. R., Smith, P. G. (1991). Origins and Evolution of Subfam. Maloideae (Rosaceae). *Systematic Botany, 16 (2),* 303. doi: 10.2307/2419283
6. Wells, T. C., Phipps, J. B. (1989). Studies in Crataegus (Rosaceae: Maloideae). XX. Interserial hybridization between Crataegus monogyna (series Oxycanthae) and Crataegus punctata (series Punctatae) in southern Ontario. *Canadian Journal of Botany, 67 (8),* 2465–2472. doi: 10.1139/b89–316
7. Sydora, N. V., Кovaleva, A. M., Yakovenko, V. K. et al. (2016). Phytochemical research of Crataegus submollis Sarg. leaves lipophilic complex and study of its antibacterial activity. *Der Pharmacia Lettre, 8 (21)*, 19–23.
8. Vyshnevska, I., Degtiarova, Ye., Bavikina, M., Bisaga, E. (2015). Chromato–mass–spectrometry identification of lipophilic compounds in plant extract Liliia. *The Pharma Innovation Journal, 3 (11)*, 72–74.
9. Oliinyk, S. V., Vyshnevska, L. I. (2017). *Aktualnist vykorystannia riznykh vydiv syrovyny Crataegus pry rozrobtsi homeopatychnych preparativ.* Kharkiv, 141–143.
10. Derzhavna Farmakopeia Ukrainy (2-nd ed.). (2015). Kharkiv: Ukrainskyi naukovyi farmakopeinyi tsentr yakosti likarskykh zasobiv, 723.

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