N E W S
OF THE NATIONAL ACADEMY OF SCIENCES OF THE REPUBLIC OF KAZAKHSTAN
SERIES CHEMISTRY AND TECHNOLOGY
ISSN 2224-5286 **https://doi.org/10.32014/2019.2518-1491.73**
Volume 6, Number 438 (2019), 49 – 54

UDC 547.918:547.926

D. Usmanov[1], N.Sh. Ramazanov[1], U. Yusupova[1], K.Dzh. Kucherbayev[2,3]

[1]Institute of Chemistry of Plant Substances AS RUz, Tashkent, Uzbekistan;
[2]Regional Social-Innovational University, Shymkent, Kazakhstan;
[3]JSC «South-Kazakhstan Medical Academy», Shymkent, Kazakhstan

IRIDOIDS FROM *PHLOMIS SEVERTZOVII* AND ITS IMMUNOSTIMULATING AND ANTITOXIC ACTIVITY

Abstract. From the aerial parts of *Phlomis severtzovii* (*Lamiaceae*) growing in Uzbekistan, five known iridoid glycosides were isolated for the first time. On basis of UV, IR, ^{1}H and ^{12}C NMR spectroscopy the isolated substances were identified as 6β-hydroxypolamid (1), loganin (2), pulchelloside (3), shanshiside methyl ester (4) and phlorigidoside C (5). Additionally, noticeable immunostimulating and antitoxic activity of the sum of iridoids and 6β-hydroxypolamid (1) were revealed.

Keywords: *Phlomis severtzovii*, iridoid, 6β-hydroxypolamid, loganin, pulchelloside, shanshiside methyl ester, phlorigidoside C, immunostimulating activity, antitoxic effect.

Introduction. Among the various low-molecular biologically active substances synthesized by plants, iridoids occupy a prominent place. At present time, it is clear, that this type of compounds are widespread in plant world [1]. Presence of important biological activities (antitumor activity, antimicrobial activity, etc.) of these compounds is main perspective practical reason for further investigation [2].

Iridoid glycosides have great theoretical importance as in terms of chemistry and due to their participation as precursors in alkaloid biosynthesis [3].

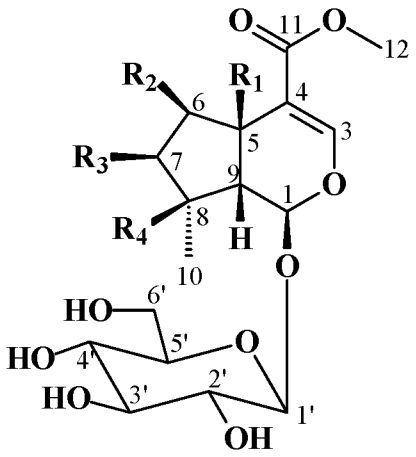
Therefore search of novel iridoidcontaining plant sources, development of rational scheme of isolation of these compounds, determination their chemical structure, revealing of physical, chemical parameters and useful properties of novel compounds are actual problems of the modern bioorganic chemistry [4].

The aim of our study is investigation of iridoids from *Phlomis severtzovii* (*Lamiaceae*) and their biological activity.

Materials and methods. *Phlomis severtzovii* (*Lamiaceae*) growing in Middle Asia, Tashkent and Fergana regions.

NMR spectra were acquired on a Bruker, 500 MHz, spectrometer using standard pulse sequence at ambient temperature. Chemical shifts are given in δ (ppm), and coupling constants are reported in Hz. The samples for NMR analysis were diluted in C_5D_5N.

Results and discussion. Five known iridoid glycosides were isolated from the aerial parts of *Phlomis severtzovii* collected during vegetation phase. Their chemical structure were determined on basis of analysis of UV, IR, ^{1}H and ^{13}C NMR spectra and identified as 6β-hydroxypolamid **1**, loganin **2**, pulchelloside **3**, shanshiside methyl ester **4** and phlorigidoside C **5** (Fig.1).



1: R_1 = OH, R_2 = OH, R_3 = H, R_4 = OH
2: R_1 = H, R_2 = H, R_3 = OH, R_4 = H
3: R_1 = OH, R_2 = OH, R_3 = OH, R_4 = H
4: R_1 = H, R_2 = OH, R_3 = H, R_4 = OH
5: R_1 = H, R_2 = OH, $R_3 + R_4$ = O

Figure 1 – Chemical structures of iridoid glycosides isolated from the aerial parts of *Phlomis severtzovii*

6β-Hydroxypolamid (1). Compound **1** was isolated as pale yellow crystals, $C_{17}H_{26}O_{12}$, ESI-MS *m/z* 445.15 $[M + Na]^+$, mp 76°C, UV (C_2H_5OH) λ_{max} 235.5 nm. IR (KBr) ν_{max} 3388 (OH), 1639 (C=O), 1551 (C=C). Yield 0.031 %.

^{1}H NMR (Bruker 500 MHz, C_5D_5N): 6.05 (1H, s, H-1), 7.77 (1H, s, H-3), 5.55 (1H, d, H-6), 2.19 (1H, d, Ha-7), 2.55 (1H, d, Hb-7), 2.99 (1H, d, H-9), 1.44 (3H, s, H-10), 3.68 (3H, s, H-12), 5.67 (1H, d, H-1`), 4.34 (1H, t, H-2`), 4.29 (1H, t, H-3`),4.22 (1H, t, H-4`), 4.45 (1H, ddd, H-5`), 4.37 (1H, dd, , Ha-6`), 4.28 (1H, dd, Hb-6`), 6.52 (H, br.s., OH);

^{13}C NMR (126 MHz, C_5D_5N: 93.91 (C-1), 154.92 (C-3), 115.73 (C-4), 74.4 (C-5), 72.39 (C-6), 48.58 (C-7), 73.88 (C-8), 57.59 (C-9), 26.82 (C-10), 168.64 (C-11), 52.04 (C-12), 99.01 (C-1`), 74.49 (C-2`), 78.18 (C-3`), 71.81 (C-4`), 78.67 (C-5`), 63.65 (C-6`) [6].

Loganin (2). Compound 2 was isolated as white crystals. $C_{17}H_{26}O_{10}$, ESI-MS *m/z* 413.39 $[M + Na]^+$, mp 222.2°C, UV (C_2H_5OH) λ_{max} 234.72 nm. IR (KBr) ν_{max} 3440 (OH), 1642 (C=O), 1446 (C=C). Yield 0.021 %.

^{1}H NMR (Bruker 500 MHz, C_5D_5N): 5.87 (1H, d, H-1), 7.72 (1H, s, H-3), 3.59 (1H, dd, H-5), 2.07(1H, dd, Ha-6), 2.35 (1H, dd, Hb-6), 4.04 (1H, ddd, H-7), 2.59 (1H, m, H-8), 3.13 (1H, dd, H-9), 1.16 (3H, s, H-10), 3.60 (3H, s, H-12), 5.69 (1H, d, H-1`), 4.27 (1H, t, H-2`), 4.33 (1H, t, H-3`), 4.22 (1H, t, H-4`), 4.11 (1H, ddd, H-5`), 4.19 (1H, dd, Ha-6`), 4.34 (1H, dd, Hb-6`), 6.82 (H, br.s., OH);

^{13}C NMR (126 MHz, C_5D_5N): 96.04 (C-1), 151.64 (C-3), 111.46 (C-4), 32.71 (C-5), 37.71 (C-6), 74.57 (C-7), 40.42 (C-8), 43.36 (C-9), 14.18 (C-10), 168.69 (C-11), 51.73 (C-12), 100.72 (C-1`), 75.07 (C-2`), 78.60 (C-3`), 71.81 (C-4`), 78.40 (C-5`), 63.65 (C-6`) [7].

Pulchelloside (3). Compound **3** was isolated as yellow crystals. $C_{17}H_{26}O_{12}$, ESI-MS *m/z* 445.13 $[M + Na]^+$, mp. 124,6°C, UV (C_2H_5OH) λ_{max} 235.76 nm. IR (KBr) ν_{max} 3437 (OH), 1640 (C=O), 1443 (C=C). Yield 0.073 %.

^{1}H NMR (Bruker 500 MHz, C_5D_5N): 5.86 (1H, s, H-1), 7.77 (1H, s, H-3), 4.48 (1H, d, H-6), 3.72 (1H, dd, H-7), 2.41 (1H, dd, H-8), 2.90 (1H, dd, H-9), 1.22 (3H, d, H-10), 3.72 (3H, s, H-12), 5.70 (1H, d, H-1`), 4.32 (1H, dd, H-2`), 4.32 (1H, d, H-3`), 4.22 (1H, dd, H-4`), 4.46 (1H, ddd, H-5`), 4.36 (1H, dd, Ha-6`), 4.21 (1H, dd, Hb-6`), 6.57 (H, br.s., OH);

^{13}C NMR (126 MHz, C_5D_5N): 97.29 (C-1), 152.93 (C-3), 112.78 (C-4), 73.54 (C-5), 77.19 (C-6), 75.39 (C-7), 38.22 (C-8), 51.35 (C-9), 14.44 (C-10), 168.84 (C-11), 52.35 (C-12), 100.91 (C-1`), 74.94 (C-2`), 78.55 (C-3`), 71.81 (C-4`), 78.38 (C-5`), 63.65 (C-6`) [8].

Shanshiside methyl ester (4). White crystals, $C_{17}H_{26}O_{11}$, ESI-MS *m/z* 429.38 $[M + Na]^+$, mp 94.8°C, UV (C_2H_5OH) λ_{max} 236.97 nm. IR (KBr) ν_{max} 3437 (OH), 1642 (C=O), 1438 (C=C). Yield 0.013 %.

^1H NMR (Bruker 500 MHz, C_5D_5N) 6.28 (1H, s, H-1), 7.81 (1H, s, H-3), 3.33 (1H, dd, 6.48, H-5), 4.51 (1H, ddd, H-6), 2.20 (1H, dd, Ha-7), 1.80 (1H, dd, Hb-7), 2.64 (1H, dd, H-9), 1.51 (3H, s, H-10), 3.60 (3H, s, H-12), 5.77 (1H, d, H-1`), 4.30 (1H, dd, H-2`), 4.28 (1H, t, H-3`), 4.22 (1H, t, H-4`), 4.44 (1H, ddd, H-5`), 4.30 (1H, dd, Ha-6`), 4.38 (1H, dd, Hb-6`), 6.28 (1H, br.s., 6-OH);

^{13}C NMR (126 MHz, C_5D_5N) 93.62 (C-1), 152.48 (C-3), 111.72 (C-4), 41.08 (C-5), 70.95 (C-6), 48.65 (C-7), 78.79 (C-8), 49.87 (C-9), 26.95 (C-10), 168.69 (C-11), 51.78 (C-12), 100.92 (C-1`), 74.81 (C-2`), 78.54 (C-3`), 71.81 (C-4`), 78.35 (C-5`), 63.65 (C-6`) [9].

Phlorigidoside C (5). Colourless, $C_{17}H_{24}O_{11}$, ESI-MS *m/z* 427.11 $[M + Na]^+$, mp 66°C, UV (C_2H_5OH) λ_{max} 238.3 nm. IR (KBr) ν_{max} 3418 (OH), 1637 (C=O), 1549 (C=C). Yield 0.028 %.

^1H NMR (Bruker 500 MHz, C_5D_5N) 6.08 (1H, d, H-1), 7.83 (1H, s, H-3), 2.81 (1H, dd, H-5), 4.17 (1H, dd, H-6), 3.48 (1H, s, H-7), 2.43 (1H, dd, H-9), 1.49 (3H, s, H-10), 3.60 (3H, s, H-12), 5.45 (1H, d, H-1`), 4.28 (1H, d, H-2`), 4.29 (1H, dd, H-3`), 4.19 (1H, dd, H-4`), 4.44 (1H, ddd, H-5`), 4.28 (1H, dd, Ha-6`), 4.38 (1H, dd, Hb-6`), 6.08 (1H, br.s., OH);

^{13}C NMR (126 MHz, C_5D_5N) 93.07 (C-1), 152.55 (C-3), 106.18 (C-4), 38.20 (C-5), 73.09 (C-6), 64.23 (C-7), 65.05 (C-8), 46.80 (C-9), 16.67 (C-10), 168.47 (C-11), 51.92 (C-12), 101.46 (C-1`), 74.78 (C-2`), 78.45 (C-3`), 71.81 (C-4`), 78.35 (C-5`), 63.65 (C-6`) [10].

Investigation of the biological activity. Experiments were set on white outbred mice (18-20 g). Reaction of Jerne, Nordin was used for evaluation of the preparation influence to the humoral immunity [11]. Mice were immunized with red blood cells (RBCS) of sheep and in the same day of immunization, in biological active dose the water-alcohol solutions of the iridoids sum were injected intragastric (10 мг/кг. Mice of the control group received the water-alcohol solution in the same volume. On the 5th day after immunization, the number of antibody-producing cells (APC) was determined in the spleens of mice.

Studies were performed on intact mice and mice subjected to stress. The state of stress was caused by the N. Selye "swim stress" model. To do this, mice for 45 min were placed in a tank with water, so that the water level did not allow mice to jump out.

The research results are presented in table 1.

The table shows that the SI in mice causes a pronounced ($p<0.001$) enhancement of the process of antibody formation in response to sheep RBCS.

In the control group of immunized mice, 11,800 ± 928.08 APC accumulate on the spleen and 132.8 ± 2.49 APC per 1 million nucleated spleen cells (NSC). The SI increases the number of APC on the spleen to 21,400 ± 1,369.18, the number of APC per 1 million of NSC - to 242.0 ± 2.83.

Table 1 – The effect of sum of iridoids from the plant *Phlomis severtzovii* on immunity indicators

Group	Control	SI
Immunized mice		
APC on spleen	11800±928,08	21400± 1369,18
APC on 1 million of NSC	132,8±2,49	242,0±2,83
Immunized mice + stress		
APC on spleen	6400±314,11	13200±461,88
APC on 1 million of NSC	75,7±3,33	151,0±2,67

Under the influence of "swimming" stress, the sum of APC in mice decreases to 6400 ± 314.11 per spleen and to 75.7 ± 3.33 per 1 million NSC. The sum of iridoids increases the number of APC per spleen to 13,200 ± 461.88, by 1 million splenic cells - to 151.0 ± 2.67.

Under the influence of the SI, there is a tendency to increase the mass and cellularity (total cell content) of the spleen, the mass of the thymus and lymph nodes, a significant increase in the cellularity of the thymus and lymph nodes.

As a result, it was shown that the SI obtained from the plant *Phlomis severtzovii* possesses immunostimulating activity, and is not immunotoxic.

It was previously established, that iridoids obtained from the aerial part of *Phlomis severtzovii* have hepatoprotective properties and a positive effect on the functional state of the liver [12]. Given this fact,

we investigated the effect of the SI and iridoid 6β-hydroxypolamide on acute alcohol intoxication. Experiments were performed on white outbred mice - females weighing 20-22 g. Ethanol in the form of a 24% solution was injected intraperitoneally at a narcotic dose (4.8 g/kg). The test substances were administered orally using an atraumatic metal probe at doses of 10-25-50 mg/kg 60-70 minutes before the introduction of ethanol. Evaluation of the effectiveness of the compounds was determined by the duration of the lateral position of the animals. The control group of animals under similar conditions of experience instead of the drug was injected with sterile distilled water. The results of the research are presented in table 2.

Table 2 – The results of the influence of drugs on the narcotic action of ethanol

No	Name of preparations	dose	The duration of the lateral position in minutes	Effect in %
1	Control EtOH	4,8 g/kg	110	100%
2	SI +EtOH	10 mg/kg 4,8 g/kg	85	22,8%
3	SI + EtOH	25 mg/kg 4,8 g/kg	62,7	43%
4	SI + EtOH	50 mg/kg 4,8 g/kg	63,8	42%
5	6β-hydroxypolamid + EtOH	10 mg/kg 4,8 g/kg	72,4	34,2%
6	6β-hydroxypolamid + EtOH	25 mg/kg 4,8 g/kg	53,9	51%
7	6β-hydroxypolamid + EtOH	50 mg/kg 4,8 g/kg	60,5	45%

The results of the studies (table 2) showed that in the control group of animals after applying EtOH at a dose of 4.8 g/kg in all mice came the state of anesthesia (lateral position) with an average duration of 110 minutes. Preliminary application of the SI and 6β-hydroxypolamide at doses of 10-25-50 mg/kg shortened the state of the lateral position of the experimental mice by 22.8%-43%-42% and 34.2% -51%-45%, respectively, compared to with a control group of animals.

Consequently, the compounds studied, depending on the dose administered, have an antitoxic effect during acute alcohol intoxication.

Experimental. Air-dried and grinded plant material was extracted with MeOH three times at room temperature. After filtration, the solvent was partly removed by rotary evaporation and the extract residue was diluted with equal quantity of water. The formed precipitate in the extract was filtered. Residues of MeOH were removed by rotary evaporation. The obtained water part of the extract was extracted in consecutive order with $CHCl_3$ and *n*-BuOH. *n*-BuOH extract was evaporated on rotary evaporator and the *n*-BuOH fraction was obtained. The *n*-BuOH was set to column with silica gel and obtained several fractions. Re-chromatography isolation of these fractions by elution with solvent systems $CHCl_3$÷MeOH: 1) 100÷1; 2) 50÷1; 3) 40÷1; 4) 30÷1; 5) 20÷1; 6) 15÷1; 7) 9÷1 and 8) 4÷1 yield the individual compounds: 6β-hydroxypolamid **1**, loganin **2**, pulcheloside **3**, shanshiside methyl ester **4** and florigidoside C **5**.

In order to get novel immunomodulating preparations dried aerial parts of *Phlomis severtzovii* were extracted with EtOH at room temperature. EtOH extract was filtered and the solvent was removed on rotary evaporator. The obtained extract was diluted with water and the water mixture was treated at first with $CHCl_3$, then with *n*-BuOH five times. The solvents were removed on rotary evaporator. Yellow powder were get in yield. Enriched SI were obtained from *n*-BuOH fraction by treating with the solvent system $CHCl_3$ - EtOH 6÷1.

Conclusion. In this study, we investigated the iridoid constituent of *Phlomis severtzovii* growing in Uzbekistan and for first time we have isolated known iridoids identified on basis of UV, IR, ^{1}H and ^{13}C

NMR spectra as 6β-hydroxypolamid **1**, loganin **2**, pulchelloside **3**, shanshiside methyl ester **4** and phlorigidoside C **5**.

Immunostimulating activity of the sum of iridoids obtained from *Phlomis severtzovii* was revealed.

It was determined that the sum of iridoids and iridoid 6β-hydroxypolamide have an antitoxic effect during acute alcohol intoxication.

Acknowledgments - Authors would like to thank the ICPS (Uzbek Academy of Sciences) for providing all necessary equipment, chemicals and animals for the study.

Д. Усманов[1], Н.Ш. Рамазанов[1], У. Юсупова[1], К.Дж. Кучербаев[2,3]

[1]ӨР ҒА Өсімдік затларының химиясы институты, Ташкент, Өзбекстан;
[2]Аймақтық әлеуметтік-инновациялық университет, Шымкент, Қазақстан;
[3]«Оңтүстік-Қазақстан медицина академиясы» АҚ, Шымкент, Қазақстан

PHLOMIS SEVERTZOVII ИРИДОИДТАРЫ ЖӘНЕ ОЛАРДЫҢ ИМУНИТЕТТІ ЫНТАЛАНДЫРУШЫ ЖӘНЕ УЫТТЫЛЫҚҚА ҚАРСЫ БЕЛСЕНДІЛІКТЕРІ

Аннотация. Өзбекстанда өсетін *Phlomis severtzovii* (*Lamiaceae*) жер үсті бөліктерінен бірінші рет бес иридоид гликозидтері бөліп алынды. УК, ИҚ, ^{1}Н және ^{12}С ЯМР спектроскопия көмегімен олар 6β-гидроксиполамид (1), логанин (2), пулчелозид (3), шаншизид метил эфирі (4) және флоригидозид С (5) екендігі анықталды. Қосымша иридоидтар жыйынтығы және 6β-гидроксиполамидтың иммунитетті ынталандырушы және уыттылыққа қарсы белсенділіктері анықталды.

Түйін сөздер: *Phlomis severtzovii*, иридоид, 6β-гидроксиполамид, логанин, пулчелозид, шаншизид метил эфирі, флоригидозид С, иммунитетті ынталандырушы белсендік, уыттылыққа қарсы белсенділік.

Д.Усманов[1], Н.Ш.Рамазанов[1], У.Юсупова[1], К.Дж.Кучербаев[2,3]

[1] Институт химии растительных веществ АН РУз, Ташкент, Узбекистан;
[2]Региональный социально-инновационный университет, Шымкент, Казахстан;
[3]АО «Южно-Казахстанская медицинская академия», Шымкент, Казахстан

ИРИДОИДЫ ИЗ *PHLOMIS SEVERTZOVII* И ИХ ИММУНОСТИМУЛИРУЮЩАЯ И АНТИТОКСИЧЕСКАЯ АКТИВНОСТЬ

Аннотация. Впервые из надземных частей *Phlomis severtzovii* (*Lamiaceae*) произрастающей в Узбекистане выделены пять ранее известных иридоидных гликозидов. На основе УФ, ИК, ^{1}Н и ^{12}С ЯМР спектроскопии выделенные вещества были идентифицированы с 6β-гидроксиполамидом (1), логанин (2), пулчеллозид (3), метиловый эфир шаншизида (4) и флоригидозид С (5). Дополнительно, выявлены значительная иммуностимулирующая и антитоксическая активность суммы иридоидов и 6β-гидроксиполамида (1).

Ключевые слова: *Phlomis severtzovii*, иридоид, 6β-гидроксиполамид, логанин, пулчелозид, метиловый эфир шаншизида, флоригидозид С, иммуностимулирующая активность, антитоксический эффект.

Information about the authors:

Durbek Usmanov - Institute of Chemistry of Plant Substances AS RUz, Tashkent, Uzbekistan, durbekusmanov@bk.ru, https://orcid.org/0000-0002-7349-377X;

Ugiloy Yusupova - Institute of Chemistry of Plant Substances AS RUz, Tashkent, Uzbekistan, yusupovauyu@gmail.com , https://orcid.org/0000-0003-3401-2379;

Nurmurod Sheralievich Ramazanov - Institute of Chemistry of Plant Substances AS RUz, Tashkent, Uzbekistan, ramazonovn@list.ru, https://orcid.org/0000-0003-2563-626X;

Kamal Dzhamalovich Kucherbayev - Regional Social-Innovational University, Shymkent, Kazakhstan, JSC «South-Kazakhstan Medical Academy», Shymkent, Kazakhstan, kkamaljsci@gmail.com, https://orcid.org/0000-0003-1981-1260

REFERENCES

[1] Galves M, Martin-Cordero C, Houghton PJ, Jesus Ayoso M. (**2005**) Antioxidant activity of methanol extracts from Plantago species. *Agricultural and food chemistry*, **53**, 1927-1933.

[2] Yang L, Jia Z–J, Su B–N. (**1997**) Neolignan, phenylpropanoid and iridoid glycosides from Pedicularis verticillata. *Phytochemistry*, **45**, No 6, 1271-1273.

[3] Lucjan Swigtek. Glukozydy iridoidowe w rodzinie Scrophullariaceae (**1973**) *Acta Polon Pharm.* **XXX**, No2, 203-212.

[4] Povodish MN. (**2006**) Diversity of iridoids in Lamiaceae family and their biological activity. *Plant resources* [In Russian], **42**, 2, 131-149.

[5] Maksudov MS. (**1996**) Iridoids from plants of Scrophulariaceae, Bignoniaceae and Lamiaceae family. *Dissertation of PhD on chemistry* [In Russian], Tashkent, 127.

[6] Stermitz FR, Abdel-Kader MS, Foderaro TA, Pomeroy M. (**1994**) Iridoid glycosides from some butterflies and their larval food plants. *Phytochemistry*, **37**, 4, 997-9.

[7] Taguchi, H, Yokokawa Y, Endo T. (**1973**) Constituents of Patrinia villosa. *Yakugaku Zasshi*, **93**, 5, 607-11.

[8] Milz S, Rimpler H. (**1978**) Pulchelloside I, a new iridoid from Verbena pulchella Sweet. *Tetrahedron Letters*, **10**, 895-8. CODEN:TELEAY ISSN:0040-4039.

[9] Yoshio T, Hiroshi N, Hiroyuki I. (**1977**) Two new iridoid glucosides from Messaenda parviflora and Mussaenda shikokiana. *Phytochemistry*, **16**, 9, 1401-1404.

[10]Kobayashi S, Mima A, Kihara M, Imakura Y. (**1986**) *Chem.Pharm.Bull.*, **34**, 876.

[11]Jerne NK, Nordin AA. (**1963**) Plague formation in agar by single antibody producing cells. *Science*. **140**, 405.

[12] Nabiev AN, Khushbaktova ZA, Zakhidov UV, etc. (**1999**) Hepatoprotective properties of iridoid glycosides in acute toxic liver damage by heliotrin in mice. *Chemical-pharm. Journ.*, **8**, 11.

[13]A.Zh. Kaldybekova, A.T. Amangazyeva, Z.B. Halmenova, A.K. Umbetova. Development of technology for the complex isolation of biological active substances from plants of the genus haplophyllum A.juss., News of NAS RK. Series of Chemistry and technology. Volume 5, Number 431 (2018), 74 – 81 https://doi.org/10.32014/2018. 2518-1491.10 ISSN 2518-1491 (Online), ISSN 2224-5286 (Print).

[14]A.A. Janibekov, T.X. Naubeev, N.Sh. Ramazanov, G.A. Sabirova, K.Dzh. Kucherbayev. Determination of chemical structure of cyclolehmanoside A from *Astragalus lehmannianus.* News of NAS RK. Series of Chemistry and technology. Volume 2, Number 434 (2019), 50 – 54 https://doi.org/10.32014/2019.2518-1491.18 ISSN 2224-5286 (Online), ISSN 2224-5286 (Print).

[15]Kenzhaliyev B. K. et al., Development of technology for chromite concentrate from the slurry tailings of enrichment.News of the National Academy of Sciences of the Republic of Kazakhstan series of geology and technical sciences. Volume 3, Number 429 (2018), 182 – 188.

[16]Presnyakov A. A., Kenzhaliev B. K., Kozhakhmetov S. M., Panichkin, A. V., &Ponomareva, S. V. (2002). Metal Science and Heat Treatment, 44(1/2), 80–80. https://doi.org/10.1023/a:1015341013948

[17]Kenzhaliev B.K., Tashuta G.N., Valutskaya T.A. et al. Potentiometric Determination of Mercury with Iodide-Selective Electrodes. Journal of Analytical Chemistry (2002) 57: 261. https://doi.org/10.1023/A:1014456602075