www.ThePharmaJournal.com

The Pharma Innovation

ISSN: 2277- 7695
TPI 2016; 5(7): 47-49

www.thepharmajournal.com
Received: 09-05-2016
Accepted: 10-06-2016

LM Grytsyk
Ivano-Frankivsk National Medical University, Ukraine

AR Grytsyk
Ivano-Frankivsk National Medical University, Ukraine

IA Sas
Ivano-Frankivsk National Medical University, Ukraine

NI Legin
Ivano-Frankivsk National Medical University, Ukraine

TI Kolyadjin
Ivano-Frankivsk National Medical University, Ukraine

Correspondence
NI Legin
Ivano-Frankivsk National Medical University, Ukraine.

Study of amino acid composition of *Betonica* L., *Sanicula* L. and *Astrantia* L. Genera Species

LM Grytsyk, AR Grytsyk, IA Sas, NI Legin and TI Kolyadjin

Abstract
The results of research of qualitative and quantitative composition of amino acids in the herb of *Betonica peraucta* Klok., *Betonica brachydonta* Klok., *Astrantia major* L., and in the herb and roots with rhizomes of *Sanicula europaea* L. are represented in the article. Amino acid composition of the researched raw materials was studied by the method of ascending paper chromatography and amino acid analyzer AAA T-339 N. 17 amino acids, including 8 essential ones (threonine, valine, methionine, isoleucine, leucine, phenylalanine, histidine and lysine), were identified in the investigated samples.

Keywords: amino acids, *Betonica peraucta* Klok., *Betonica brachydonta* Klok., *Sanicula europaea* L., *Astrantia major* L.

Introduction
Amino acids are widespread in nature and are the structural units of proteins and the basis of the body structure. All organs of the human body are formed of proteins, which are also involved in the biochemical structure of hormones, enzymes and many other substances necessary for life [1].

More than 200 amino acids are discovered, but a human needs only 20 of them. Our body can synthesize 12 amino acids and the other 8 (threonine, valine, methionine, isoleucine, leucine, tryptophan, phenylalanine, lysine) can't be synthesized in the human body and therefore are called essential. These amino acids get into the human organism with food, as well as food additives and drugs. Amino acids play an important role in the human body, because each of them has its own special functions [1, 2].

Human's need of proteins is satisfied on 10 - 30% by animal proteins and on 70 - 90% by plant proteins [3]. That's why, the problem of search and study of plants containing a sufficient amount of protein origin substances, including amino acids, is topical.

The aim of our study was to investigate the amino acid composition of *Betonica* L., *Sanicula* L. and *Astrantia* L. genera species.

The objects of our study were the herb of *Betonica peraucta* Klok., *Betonica brachydonta* Klok., *Astrantia major* L., and the herb and roots with rhizomes of *Sanicula europaea* L., which had been harvested in Ivano-Frankivsk region in 2014 - 2015 years.

Betonica L. (*B.*) genus belongs to the *Lamiaceae* family and includes 15 species, among which only 3 species (*B. peraucta* Klok., *B. brachydonta* Klok., B. fusca Klok.) grow in Ukraine and are united in a row of *B. officinalis* L. In Ukraine *B. officinalis* is non-pharmacopoeial plant but is widely used in folk medicine. *B. officinalis* herb contains a variety of biologically active substances, including phenolic compounds and terpenoids; hydroxycinnamic acids and their derivatives (chlorogenic, neochlorogenic, izochlorogenic, *p*-coumaric acids); organic and ascorbic acids; flavonoids (apigenin, 7-methoxytricine, 7-glucoside of scutellarein); tannins (10 - 25%); vitamins C and K; triterpene saponins; steroids (0.01%); nitrogen-containing compounds (betaine (0.5%), betonicine, turicine); stachydrine alkaloid; essential oil [4 - 6].

Sanicula L. (*S.*) genus has about 50 species, spread almost all over the world. Three species of *Sanicula* L. genus grow on the territory of the CIS countries (*S. rubrifolia* Fr. Schmidt., *S. europaea* L., *S. chinensis* Bge.). Only *Sanicula europaea* L. grows in Ukraine. Underground and above-ground organs of *S. europaea* L. contain biologically active substances with various chemical structure: organic acids (malic, citric, malonic, oxalic); triterpene saponins; nitrogen-containing compounds (allantoin); hydroxycinnamic acids (chlorogenic, rosemary); ascorbic acid; flavonoids; tannins and bitter substances; essential oil [4, 7, 8].

Astrantia L. (*A.*) genus includes about 10 species, which are widespread in Europe, the Caucasus and Asia.

In the CIS countries 5 species of *Astrantia* L. genus grow: *A. maxima* Pall., *A. colchica* Alb., *A. pontica* Aib., *A. major* L., *A. trifida* Hoffm. In Ukraine only the species of *A. major* L. is found. In the aerial part of *Astrantia major* tannins, ascorbic acid, salicylates, flavonoids (quercetin, kaempferol, kaempferitrin, nicotiflorin, astragalin, routine, izoquercetin), carbohydrates (sucrose), organic acids (malic, oxalic, citric, malonic), hydroxycinnamic acids (rosemary, chlorogenic) are identified. In Ukraine *A. major* is non-pharmacopoeial plant [4, 7, 8].

2. Materials and Methods. To identify amino acids aqueous extracts of the studied species were used. Equal volumes (about 2 ml) of the studied extract and 0.1% freshly prepared solution of ninhydrin were mixed and gently heated. The change of colour was observed after cooling.

Qualitative content of amino acids in the researched objects was determined by the method of ascending paper chromatography. Aqueous extracts obtained from the studied raw materials were chromatographed in a solvent systems of n-butanol-acetic acid-water (4:1:2) and n-butanol-acetic acid-water (18:2:5) on the paper of "Filtrak FN-1" brand. Solutions of amino acids in 0.1 N solution of hydrochloric acid were used as the samples for comparison. Chromatograms were sprinkled with 0.1 % solution of ninhydrin in ethanol and heated in the drying cabinet for a few minutes at 80 - 100 °C. Amino acids were coloured in purple or pink and purple [9].

The study of qualitative and quantitative content of amino acids in the researched raw materials was performed using the amino acid analyzer AAA T-339 M (Czech Republic). The analysis was carried out in comparison with the concentration of the standard amino acids hydrolysates according to State Standart of Ukraine ISO 13903:2005 "The components of animal feed - Determination of amino acids". To determine the amino acid composition of the investigated objects 100 mg of the sample were dried at 60 °C and placed in a test tube for hydrolysis, and after that equal amounts (5 ml) of purified water and concentrated hydrochloric acid were added and mixed. Hydrolysis was carried out at 120 °C for 15 min. Then the sample was neutralized with dry NaOH to pH = 11 and transferred to a porcelain cup for 1 hour in order to accelerate the evaporation of ammonia. After that, the solution of hydrochloric acid was added to the sample to obtain pH = 2.2, the sample was filtered, 0.1 - 0.5 ml of liquid was selected and its volume was adjusted to 2 ml by buffer solution with pH = 2.2 [10].

3. Results and Discussion. As a result of reaction with 0.1% solution of ninhydrin in ethanol all extracts of the studied materials got purple color, which indicates the presence of amino acids in all selected objects of research.

According to the results of chromatographic study of amino acid composition 17 amino acids, including 8 essential ones (threonine, valine, methionine, isoleucine, leucine, phenylalanine, histidine and lysine) were identified in the herb of *Betonica peraucta* Klok., *Betonica brachydonta* Klok., *Astrantia major* L., and in the herb and roots with rhizomes of *Sanicula europaea* L.

Results of the study of qualitative and quantitative content of amino acids in the researched raw materials are presented in Table 1.

Table 1: The chromatographic analysis of amino acid composition of the studied raw materials

Medicinal plant material	Herb of *Betonica peraucta* Klok.	Herb of *Betonica brachydonta* Klok.	Herb of *Astrantia major* L.	Herb of *Sanicula europaea* L.	Roots with rhizomes of *Sanicula europaea* L.
Amino acid	**The content of free amino acids, mg/100 g**				
Essential					
Threonine	0,37	0,48	3,76	3,25	2,88
Valine	0,48	0,7	7,08	6,43	6,92
Methionine	0,07	0,10	1,37	1,42	0,23
Isoleucine	0,36	0,58	2,48	2,62	1,50
Leucine	0,7	1,07	5,23	5,01	1,31
Phenylalanine	0,39	0,58	4,99	5,23	0,38
Non-essential					
Histidine	0,32	0,44	1,11	1,31	0,77
Lysine	0,43	0,65	6,32	7,41	1,27
Aspartic acid	0,95	1,31	13,08	12,42	10,77
Serine	0,43	0,55	7,95	7,63	10,15
Glutamic acid	1,11	1,52	6,10	5,67	5,19
Proline	0,63	1,01	4,14	5,23	11,54
Cystine	0,09	0,11	3,37	4,36	0,38
Glycine	0,49	0,64	12,75	12,21	5,77
Alanine	0,53	0,81	9,15	9,81	10,77
Tyrosine	0,26	0,41	0,81	0,97	3,85
Arginine	0,48	0,69	10,25	9,12	25,38

Thus, according to the obtained results of the researched objects amino acid composition study it was found out that for *Betonica peraucta* Klok. herb the dominant amino acids are glutamic and aspartic acids, leucine, proline, alanine; for *Betonica brachydonta* Klok. herb – glutamic and aspartic acids, leucine, alanine, valine; for *Astrantia major* L. herb – aspartic acid, glycine, arginine, alanine, serine; for *Sanicula europaea* L. herb – aspartic acid, glycine, alanine, arginine, serine; for *Sanicula europaea* L. roots with rhizomes – arginine, proline, alanine, aspartic acid, serine.

4. Conclusions

1. The qualitative and quantitative content of amino acids in the herb of *Betonica peraucta* Klok., *Betonica brachydonta* Klok., *Astrantia major* L., and in the herb and roots with rhizomes of *Sanicula europaea* L. was

analysed for the first time. The presence of 17 amino acids, including 8 essential ones (threonine, valine, methionine, isoleucine, leucine, phenylalanine, histidine and lysine) was identified.
2. The dominant essential amino acids in the investigated samples of raw materials in quantitative proportion are valine, leucine, phenylalanine, isoleucine; the dominant non-essential amino acids are arginine, aspartic and glutamic acids, glycine, proline, serine, alanine.
3. It was established that the investigated plants of *Asteraceae* family accumulate more amino acids comparing with *Betonica* L. genus species, which belong to *Lamiaceae* family.
4. The results of the conducted research indicate that further phytochemical study of *Betonica peraucta* Klok., *Betonica brachydonta* Klok., *Astrantia major* L. and *Sanicula europaea* L. raw materials is promising.

5. References

1. Грицик А.Р. Мельник М.В. Дослідження амінокислотного складу та жирно кислотного складу трави рути садової. Фармацевтичний часопис. 2014; № 4: 24 – 26.
2. Тернинко І.І. Вивчення амінокислотного складу мальви лісової (*Malvae sylvestris* L.). Фармацевтичний журнал. 2012; № 5: 81 – 84.
3. Бурцева О.В. Дослідження амінокислотного складу трави та зерна вівса посівного. Український журнал клінічної та лабораторної медицини. 2009; Том 4, № 4: 50 – 52.
4. Лікарські рослини: Енциклопедичний довідник / Відп. ред. А.М. Гродзінський. – Київ : Голов. ред. УРЕ, 1990; 544.
5. Флора УРСР IX / Під ред. члена АН УРСР Д.К. Зерова – Київ : В-во Академії наук УРСР, 1955; 184 – 194.
6. Растительные ресурсы СССР: Цветковые растения, их химический состав, использование; Семейства *Hippuridaceae – Lobeliaceae*. — Санкт-Петербург : Наука, 1991; 15 – 17.
7. Флора УРСР VII / Під ред. члена АН УРСР Д.К. Зерова – Київ : В-во Академії наук УРСР, 1955; 670.
8. Растительные ресурсы СССР: Цветковые растения, их химический состав, использование; Семейства Rutaceae – Elaeagnaceae. – Ленинград : Наука, 1988; 357.
9. Грицик Л.М. Легінь Н.І., Грицик А.Р. Ідентифікація деяких біологічно активних речовин у надземних і підземних органах підлісника європейського *Sanicula europaea* L. Матеріали IV науково-практичної конференції з міжнародною участю [“Сучасні досягнення фармацевтичної технології”] (16 – 17 жовтня 2014 р.). – Харків : Вид-во НФаУ, 2014; 88.
10. Ионов И.А., Шаповалов С.О., Руденко Е.В., Долгая М.Н., Ахтырский А.В., Зозуля Ю.А.и др. Критерии и методы контроля метаболизма в организме животных и птиц. – Харьков: Институт животноводства НААН, 2011; 378.