УДК 615.014.07:547.466:582.736.302
DOI https://doi.org/10.11603/2312-0967.2019.1.9949

AMINO ACID PROFILE OF PHASEOLUS VULGARIS PODS AND DRY EXTRACT PREPARED OF THEM

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The aim of the work. To study the amino acid profile of the phaseolus vulgaris pods and extract prepared of them.
Materials and Methods. Five samples of raw material of phaseolus vulgaris pods (erect herbaceous bushes varieties with white seeds) were collected in the Ternopil and Volyn regions, dry extract of phaseolus vulgaris pods was obtained according to previously developed technology. The study of amino acid composition of the raw materials of phaseolus vulgaris pods and extract prepared of them was carried out using a thin-layer chromatography (TLC) and a high-performance liquid chromatography (HPLC).
Results and Discussion. The better separation of amino acids in TLC-research of the raw material of phaseolus vulgaris pods was observed in the system of solvents isopropanol – formic acid – water (40: 2: 10). As a result of the study, aspartic and glutamic acids, glycine, valine, tyrosine and leucine were identified.
The amino acid profile of the studied samples of raw material is quite homogeneous in composition: 7 essential amino acids (histidine, threonine, valine, methionine, phenylalanine, isoleucine and leucine) and 8 non-essential amino acids (aspartic and glutamic acids, arginine, serine, glycine, alanine, tyrosine and proline); lysine was found among the bound amino acids in the 4th sample of raw material. Proline predominates in all samples of raw materials among free amino acids. Among the bound amino acids the content of glutamic acid, which is the product of the glutamine hydrolysis, is the highest. The content of glycine, serine and alanine is also high. Among the essential amino acids, leucine, phenylalanine, histidine, threonine, isoleucine, valine were determined in content descending order.
The quantitative determination of amino acids in the extract of phaseolus vulgaris pods proved that the content of proline was the highest (12.47 mg/g); the content of some compounds was also high: aspartic (5.41 mg/g) and glutamic (3.41 mg/g) acids, arginine (5.10 mg/g; both in free and bound forms), glycine (1.02 mg/g) and serine (1.04 mg/g). Among the essential amino acids in the extract, the amount of valine (0.80 mg/g), phenylalanine (0.67 mg/g), threonine (0.66 mg/g), leucine (0.63 mg/g) and isoleucine (0.58 mg/g) was a little different. The total content of amino acids in the extract was 3.2 %.
Conclusions. The amino acid profile of five samples of phaseolus vulgaris pods was studied by the HPLC method. It was established that the composition is quite homogeneous, and the total content varies within 0.7–1.1 %.
In the dry extract of phaseolus vulgaris pods the content of 5 essential and 7 non-essential amino acids was determined. The content of free amino acids in the extract is 0.52%; the total content of free and bound amino acids is 3.2 %.
When studying the stability and establishing the shelf life of the dry extract of phaseolus vulgaris pods, it is necessary to take into account the presence of free amino acids and protein substances.

Key words: phaseolus vulgaris pods; raw material; extract; amino acids; HPLC; quantitative determination.

Introduction. The study of the amino acid composition of the herb of five types of phaseolus vulgaris (black, lemon bean, multiflorous bean, red mash bean, green mash bean) was carried out in the research [1]; according to it, other varieties were fancied. The authors identified 16 amino acids; aspartic and glutamic acids, phenylalanine, valine, methionine, alanine, glycine, leucine were found in the largest amounts. 17 amino acids were identified in the lyophilisate of the homogenized fresh beans of green beans; aspartic and glutamic acids, arginine, alanine, lysine, serine, leucine, valine and proline were of the highest content [2].

The phaseolus vulgaris pods are medicinal plant raw material, which is used in mild diabetes because of hypoglycemic and hypolipidemic effect. Previously, the conditions for obtaining the dry extract of phaselous vulgaris pods were studied; its phenolic composition as well as hypoglycemic effect [3–6] was studied. For standardization of the extract is important to study the various classes of biologically active substances (BAS), which alone or in combination with other BAS cause a pharmacological effect. The study of BAS composition of the extract is also important in terms of study of its stability: in determining the conditions of its storage and shelf life, the chemical activity of the compounds, which are the most labile to the environmental factors (light, humidity, temperature), should be taken into account.

Therefore, the aim of the research was to study the amino acid profile of phaseolus vulgaris pods and the extract prepared of them.

Materials and Methods. Five samples of the raw material of phaseolus vulgaris pods (erect herbaceous bushes varieties with white seeds: Laura (1), Eureka (2), Golden Saxa (3), Polka (4), Olga (5)) were collected in Ternopil and Volyn regions, the dry extract of phaseolus vulgaris pods was obtained according to the technology described previously [3]. The study of amino acid composition of the raw material of phaseolus vulgaris pods and the extract prepared of them was carried out by means of thin-layer chromatography (TLC) and high-performance liquid chromatography (HPLC).

Silica gel 60 F_{254} chromatographic plates and chromatographic chambers (Merck, Germany), and device for drawing samples Linomat 5 (CAMAG, Switzerland) were used for the TLC-study. The test solutions were prepared by boiling of the raw material (4 g) and 25 ml of ethanol (10 %, v/v) under a reflux condensor for 1 hour in water bath. The test solution of the extract was prepared by dissolving 0.4 g of dry extract in 10 ml of ethanol (10 % v/v). To prepare the witness standard solution 5 mg of glycine, valine, leucine, tyrosine, aspartic and glutamic acids, qualified as 'chemically pure', were dissolved in ethanol (10 % v/v) at 30 °C for 10 min in ultrasonic bath. 15–20 µl of the test solutions and 5 µl of witnessing standard solutions were put on the plate. The study of qualitative composition of amino acids was carried out in two mobile phases: isopropanol – formic acid – water (40: 2: 10), acetone – water (3: 2); development – over a path of 10 cm. Detection of chromatograms was performed by the solution of 2 g/L ninhydrin (acetic acid – n-butanol (5:95)), followed by heating at 100–105 ºC for 5 minutes. Analysis of the chromatograms was performed in visible light.

The liquid chromatograph Agilent 1200 (Agilent technologies, USA) and G1315A fluorescence detector (Agilent Technologies, USA) as well as 1313A autosampler (Agilent Technologies, USA) were used to determine the composition and content of amino acids by the HPLC method.

The preparation of the solution for determination of free amino acids was carried out by keeping the raw material (0.16 g) or the extract (0.16 g) and 4.0 ml of 0.1 mol/L hydrochloric acid solution in a hermetically sealed vial at 50 ºC in ultrasound bath for 3 hours. The total removal of free and bound amino acids was carried out under more tough conditions: the raw material (15 mg) or extract (15 mg) was placed in a vial, 0.5 ml of 6 mol/L hydrochloric acid solution was added, sealed, and kept in a thermostat for 24 h at 110 ºC for hydrolysis; the hydrolyzate was diluted with water for chromatography to 4.0 ml.

0.5 ml of each obtained extract was evaporated on a rotary evaporator to dry at 50 °C, 0.5 ml of water for chromatography was added and evaporated again, the procedure was performed two more times to remove hydrochloric acid. The dry residue was dissolved by stirring it in 0.5 ml of water for chromatography and filtered through syringe micro-filters of regenerated cellulose with a pore size of 0.22 µm. The obtained filtrate was put into the conical insert of the vial, which was inserted into the autosampler.

The derivatization of amino acids was carried out in an automated online mode of the autosampler according to the described scheme [7]. The derivatization was performed using o-phthalaldehyde reagent (OPA, Agilent 5061–3335) for primary amino acids and 9-fluorenylmethyl chloroformate reagent (FMOC, Agilent 5061–3335) for secondary amino acids.

To identify the amino acids and build the calibration curves, the solutions of amino acids standards were prepared by dilution / dissolution of the standards mixture (PN 5061-3334, PN 5062-2478, Agilent Technologies, USA).

Chromatographic conditions [7]:

Column: Zorbax Eclipse AAA 4.6x150 mm (3 µm).

Column temperature: 40 ºC.

Mobile phase A: 40 mmol/L solution of Na_2HPO_4 and pH 7.8 (5.5 g Na_2HPO_4, monohydrate was dissolved in 1 litre of water for chromatography, adjusted to pH 7.8 with 10 mol/L sodium hydroxide solution).

Mobile phase B: acetonitrile-methanol-water (45:45:10, v / v / v).

Flow: 1.5 ml/min.

Stoptime: 26 min.

Post time: off.

Gradients: according to the program.

Time, min	Mobile phase A, %	Mobile phase B, %
0–1.9	100	0
1.9–18.1	100→43	0→57
18.1–18.6	43→0	57→100
18.6–22.3	0	100
22.3–23.2	0→100	100→0
23.2–26.0	100	0

Detection: fluorescence detector according to the program: up to 15 min – excitation wavelength 340 nm, emission wavelength 450 nm; after 15 min – excitation wavelength 266 nm, emission wavelength 305 nm.

Injection: 0.5 µl.

Results and Discussion

Better separation of amino acids in the TLC-study of the raw material of phaseolus vulgaris pods was observed in a mobile phase of isopropanol – formic acid – water (40:2:10). A picture of a chromatogram for a test solution of the 1st sample is presented in Figure 1.

At the chromatogram of the test solution, 8 zones were evidenced, six of which were identified as aspartic acid (R_f = 0.25), glycine (R_f = 0.33), glutamic acid (R_f = 0.39), valine (R_f = 0.53), tyrosine (R_f = 0.63), leucine (R_f = 0.65). Two weak zones of R_f = 0.09 of yellow-orange colour and of R_f = 0.32 of purple-pink colour were not

Fig. 1. The chromatograms in the conditions of amino acids identification. 1 – the 1st sample solution in ethanol (10% v / v); standard solutions of amino acids in ethanol (10% v / v): 2 – glycine, 3 – valine, 4 – leucine, 5 – tyrosine, 6 – aspartic acid, 7 – glycine and glutamic acid.

identified. Similar chromatograms were obtained for all samples of phaseolus vulgaris pods.

Thus, in the phaseolus vulgaris pods, the aspartic and glutamic acids, valine, glycine, tyrosine and leucine were identified by thin-layer chromatography, which were mentioned in the studies of various types of phaseolus vulgaris herb [1] and lyophilized fresh beans of green beans [2].

A detailed study of the amino acid profile of five samples of phaseolus vulgaris pods and extract prepared of them was made by means of the HPLC method. Under the described conditions of chromatography and derivatization, the chromatographic profiles of free and bound amino acids were obtained. The examples of the chromatograms obtained are presented in Figures 2 and 3.

It is established that phaseolus vulgaris pods of all types can be used in cases of diabetes. Previously, the studies of phenolic profile of various samples of bush phaseolus vulgaris pods with white seeds and the extract prepared of them proved the presence of rutin, isoquercitrin, quercitrin and ferulic acid [4, 6]. The amino acid profile of the studied samples of raw materials is also quite homogeneous in composition: 7 essential amino acids (histidine, threonine, valine, methionine, phenylalanine, isoleucine and leucine) and 8 non-essential amino acids (aspartic and glutamic acid, arginine, serine, glycine, alanine, tyrosine and proline). Additionally, lysine was found among the bound amino

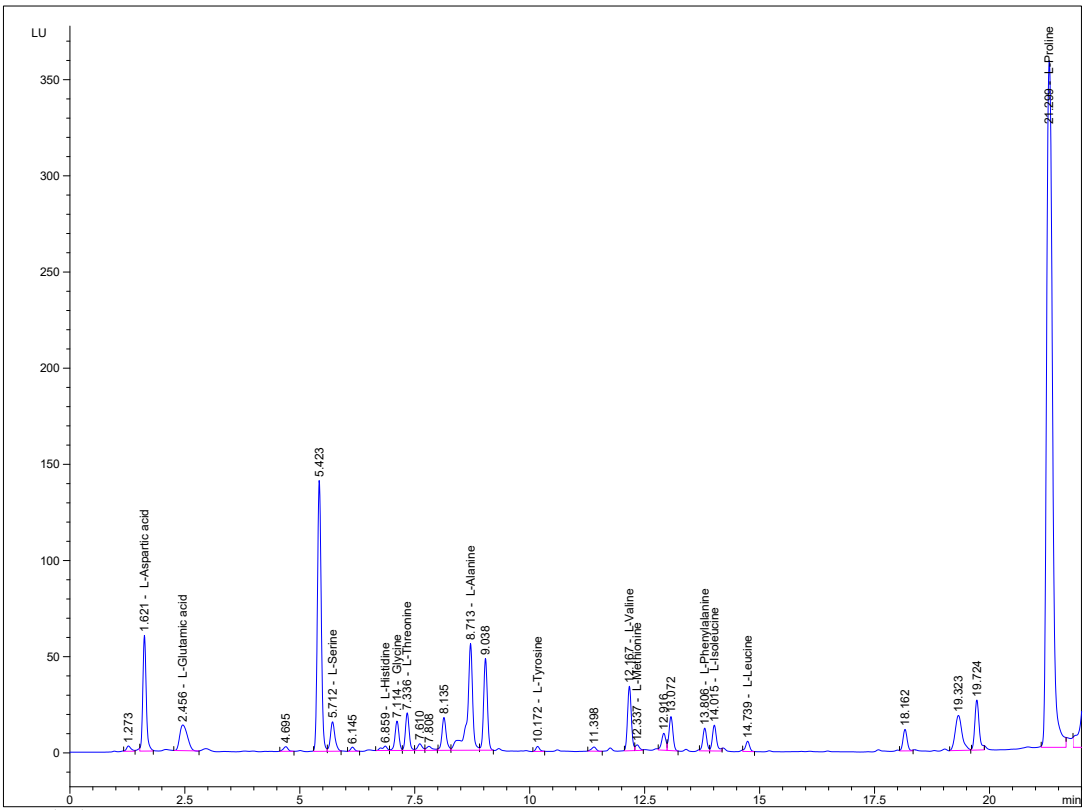

Fig. 2. Chromatogram of the test solution of the phaseolus vulgaris pods 3rd sample obtained in terms of quantitative determination of free amino acids content

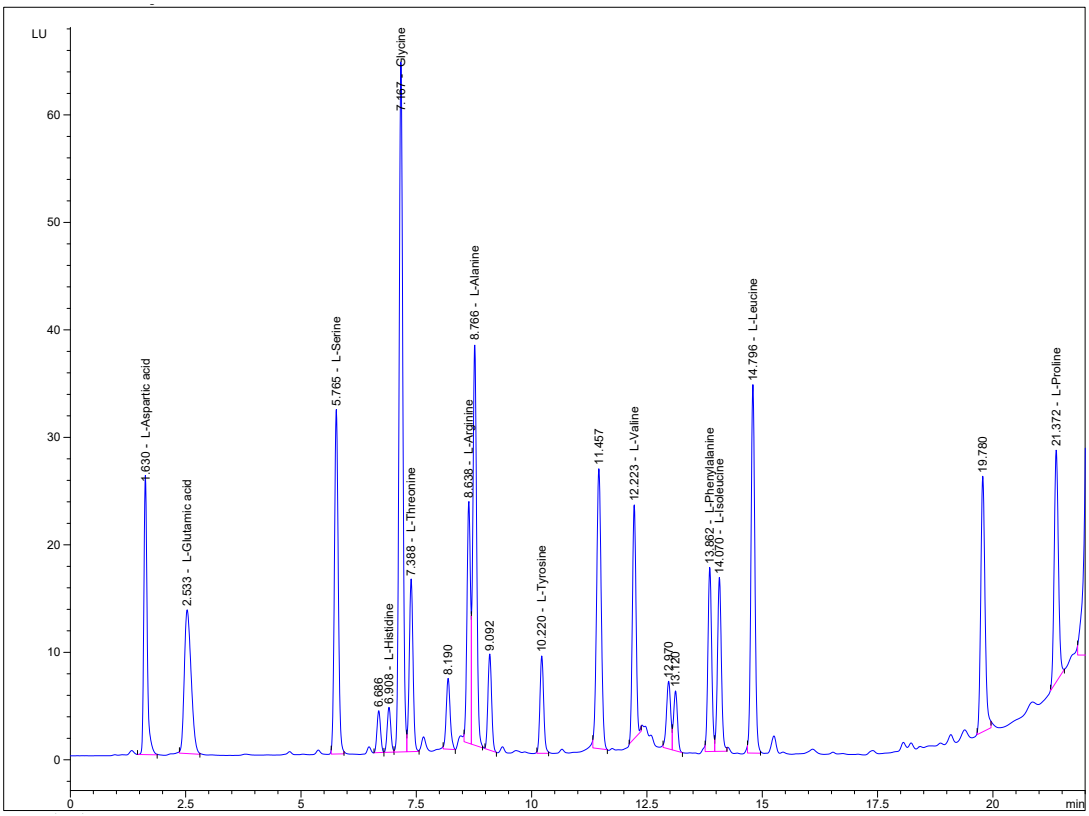

Fig. 3. Chromatogram of the test solution of the phaseolus vulgaris pods 3rd sample obtained in terms of quantitative determination of total amino acids content

acids in the 4th sample of raw materials; methionine was not identified in samples 4 and 5; in the 2nd and 5th samples, free histidine was not found as well; no free tyrosine was identified in samples 2, 4 and 5; arginine was present only in a bound form in all samples. The comparison of the composition of amino acids in the studied samples of phaseolus vulgaris pods and that in the literature is presented in Table 1.

The comparison of the chromatographic profiles of amino acids of green beans [2], which are immature beans with immature seeds, and pods of phaseolus vulgaris, proves that in the green beans and pods there are identical amino acids, the exception is hydroxyproline: it is not found in the pods.

Histidine and alanine, which are present in the raw material of phaseolus vulgaris pods, are not found in the extract. There were no lysine and methionine in the extract as well, which are identified in some samples of raw materials. The technology of the extract, that is the concentration of alcohol in the extractant [8], the method

Table 1. The amino acid composition of the phaseolus vulgaris

Raw material	Component composition	Method and conditions of determination	Sourse
Phaseolus vulgaris pods	His, Thr, Arg, Val, Met, Phe, Ile, Leu, Asp, Glu, Ser, Gly, Ala, Tyr, Pro, Lys (only in the fourth sample)	Acid hydrolysis, online derivatization using o-phthalaldehyde (OPA) for primary amino acids and 9-fluorenylmethyl chloroformate (FMOC) for secondary amino acids, HPLC with fluorescence detection	own results
5 species of phaseolus vulgaris grass	Asp, Glu, Thr, Ser, Pro, Gly, Ala, Val, Met, Ile, Leu, Tyr, Phe, His, Lys, Arg	Acid hydrolysis, post-column derivatization using ninhydrin, amino acid analyzer (ion-exchange using ion exchange resin DCGA) with visible region detection	[1]
Green beans	Asp, Glu, H-Pro, Ser, Gly, His, Arg, Thr, Ala, Pro, Tyr, Val, Met, Ile, Leu, Phe, Lys	Acid hydrolysis, derivatization using Phenylisothiocianate for primary and secondary amino acids, HPLC with ultraviolet detection	[2]

of thickening and drying, may cause it. It should be also noted that some reactions may take place during the hydrolysis in preparation of the sample: peptide with valine and isoleucine are ruptured toughly; threonine, tyrosine and serine are decomposed slowly partially, and tryptophan and cysteine are degraded in the process of acid hydrolysis, which leads to efficiency; methionine is partially oxidized in hydrolysis; asparagine and glutamine come to be asparagine and glutamic acids, and some amino acids such as glycine and serine are common contaminants [9–12].

The results of quantitative determination of amino acids in the studied samples of raw material and extract are presented in Table 2.

The presented results prove that proline predominates among free amino acids in all samples. Among the bound amino acids, the content of glutamic acid, which is the product of glutamic hydrolysis, is the highest. The level of glycine and serine is also high, which may be excessive due to the prevalence of these amino acids; the content of alanine is also high. Leucine, arginine, phenylalanine, histidine, threonine, isoleucine, valine are identified among the important amino acids in content descending order. Methionine, an essential amino acid, is found in three of five samples, probably because of both the quality of the raw material and the chosen conditions of sample preparation. The numerical value of the content of some amino acids is quite homogeneous in all samples, and the total content in the studied samples of raw materials varies within 7–11 mg/g (0.7–1.1 %).

The content of amino acids in the 4th sample of the raw material was the highest, so the results of the analysis of this samples were compared with those of quantitative determination of amino acids in green beans according to the research [2] (Fig. 4).

The comparison analysis proved that the total content of amino acids in the phaseolus vulgaris pods (3.2 %) was almost 4 times lower than that in the green beans (13.5 %), which in addition to practically immature pods also had immature seeds. For the most part, the amino acid profiles of phaseolus vulgaris pods and green beans were similar. The difference in the profiles was regarding the quantitative issue. The proportions of proline, glycine and glutamic acid in the total amount of amino acids were slightly higher compare to their proportions in green beans, while the proportion of aspartic acid was slightly lower. The amino acid profiles of phaseolus vulgaris pods and green beans regarding the issue of quantitative ratio of essential amino acids correlated slightly better: only particle of lysine was slightly lower in the pods. Those differences were

Table 2. The content of amino acids in the phaseolus vulgaris pods and extract prepared of them

Time, min	Substance	Sample								
		1			2			3		
		Content, mg / g								
		free	bound	**total**	free	bound	**total**	free	bound	**total**
1,63	L-Aspartic acid	0,098	0,398	**0,496**	0,155	0,380	**0,535**	0,249	0,551	**0,800**
2,498	L-Glutamic acid	0,133	0,593	**0,726**	0,097	0,741	**0,838**	0,169	0,991	**1,160**
5,731	L-Serine	0,061	0,516	**0,577**	0,069	0,689	**0,758**	0,062	0,777	**0,839**
6,878	L-Histidine	0,047	0,202	**0,249**	-	0,384	**0,384**	0,039	0,319	**0,358**
7,128	Glycine	0,030	0,617	**0,647**	0,017	0,687	**0,704**	0,040	1,198	**1,238**
7,355	L-Threonine	0,034	0,276	**0,310**	0,026	0,332	**0,358**	0,074	0,388	**0,462**
8,624	L-Arginine	-	0,458	**0,458**	-	0,484	**0,484**	-	0,829	**0,829**
8,734	L-Alanine	0,066	0,477	**0,543**	0,085	0,527	**0,612**	0,236	0,602	**0,838**
10,193	L-Tyrosine	0,024	0,236	**0,260**	-	0,315	**0,315**	0,015	0,372	**0,387**
12,191	L-Valine	0,040	0,262	**0,302**	0,036	0,311	**0,347**	0,091	0,340	**0,431**
12,433	L-Methionine	0,022	0,032	**0,054**	0,054	0,012	**0,066**	0,012	0,021	**0,033**
13,828	L-Phenylalanine	0,037	0,352	**0,389**	0,020	0,419	**0,439**	0,058	0,561	**0,619**
14,037	L-Isoleucine	0,035	0,250	**0,285**	0,021	0,295	**0,316**	0,052	0,393	**0,445**
14,760	L-Leucine	0,019	0,589	**0,608**	0,014	0,660	**0,674**	0,021	0,952	**0,973**
15,227	L-Lysine	-	-	**-**	-	-	**-**	-	-	**-**
21,342	L-Proline	1,243	0,002	**1,245**	1,255	0,029	**1,284**	1,370	0,170	**1,540**
Total		1,889	5,260	**7,149**	1,849	6,265	**8,114**	2,488	8,464	**10,952**

Note: sample 1-5 - the phaseolus vulgaris pods; sample 6 - the phaseolus vulgaris pods dry extract.

Continuation Table 2.

Time, min	Substance	Sample								
		4			5			6		
		Content, mg / g								
		free	bound	**total**	free	bound	**total**	free	bound	**total**
1,63	L-Aspartic acid	0,135	0,474	**0,609**	0,112	0,346	**0,458**	0,681	4,737	**5,418**
2,498	L-Glutamic acid	0,117	0,817	**0,934**	0,153	0,558	**0,711**	0,296	3,115	**3,411**
5,731	L-Serine	0,076	0,718	**0,794**	0,038	0,508	**0,546**	0,161	0,887	**1,048**
6,878	L-Histidine	0,041	0,446	**0,487**	-	0,266	**0,266**	-	-	**-**
7,128	Glycine	0,028	0,959	**0,987**	0,018	0,829	**0,847**	0,098	0,924	**1,022**
7,355	L-Threo-nine	0,033	0,325	**0,358**	0,027	0,254	**0,281**	0,172	0,491	**0,663**
8,624	L-Arginine	-	0,689	**0,689**	-	0,468	**0,468**	1,640	3,457	**5,097**
8,734	L-Alanine	0,101	0,585	**0,686**	0,059	0,454	**0,513**	-	-	**-**
10,193	L-Tyrosine	-	0,322	**0,322**	-	0,250	**0,250**	0,027	0,269	**0,296**
12,191	L-Valine	0,048	0,313	**0,361**	0,028	0,253	**0,281**	0,157	0,644	**0,801**
12,433	L-Methio-nine	-	-	**-**	-	-	**-**	-	-	**-**
13,828	L-Phenylalanine	0,022	0,466	**0,488**	-	0,364	**0,364**	0,077	0,592	**0,669**
14,037	L-Isoleucine	0,027	0,305	**0,332**	0,016	0,239	**0,255**	0,074	0,510	**0,584**
14,760	L-Leucine	0,020	0,738	**0,758**	0,011	0,588	**0,599**	0,045	0,585	**0,630**
15,227	L-Lysine	-	0,314	**0,314**	-	-	**-**	-	-	**-**
21,342	L-Proline	1,240	0,474	**1,262**	1,270	0,059	**1,329**	1,806	10,668	**12,474**
Total		1,888	7,493	**9,381**	1,732	5,436	**7,168**	5,234	26,879	**32,113**

Note: sample 1-5 - the phaseolus vulgaris pods; sample 6 - the phaseolus vulgaris pods dry extract.

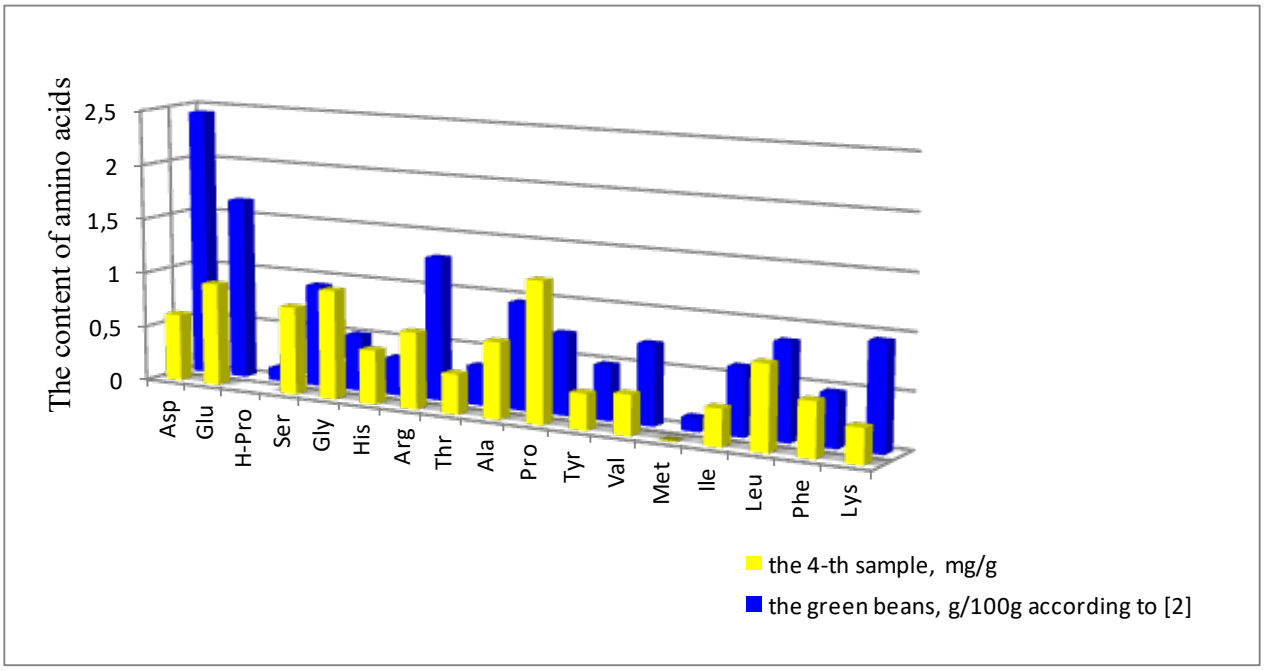

Fig. 4. Diagram of the amino acids content in the 4th sample of the paseolus vulgaris and in the green beans according to [2]

probably associated with different terms of storage of the comparable samples as well as peculiar features of accumulation of the amino acids and proteins in the pods and seeds.

The quantitative determination of amino acids in the extract of phaseolus vulgaris pods proved that the content of proline was the highest (12.47 mg/g); the content of aspartic (5.41 mg/g) and glutamic (3.41 mg/g) acids, arginine was the most numerous (5.10 mg/g) (both in free and bound forms), glycine (1.02 mg/g) and serine (1.04 mg/g) was also high. Among the essential amino acids the amounts of valine (0.80 mg/g), phenylalanine (0.67 mg/g), threonine (0.66 mg/g), leucine (0.63 mg/g), and isoleucine (0.58 mg/g) slightly differed. The total amino acid content of the extract was 32 mg/g or 3.2 %.

Conclusions. 1. The amino acid profile of five samples of phaseolus vulgaris pods was studied by means of the HPLC method. It was established that the composition is quite homogeneous for all samples, and comprises 7 essential amino acids (histidine, threonine, valine, methionine, phenylalanine, isoleucine and leucine) and 8 non-essential amino acids (aspartic and glutamic acids, arginine, serine, glycine, alanine, tyrosine and proline); the total content varies within 0.7–1.1 %.

2. In the dry extract of phaseolus vulgaris pods the content of 5 essential (threonine, valine, phenylalanine, isoleucine and leucine) and 7 non-essential amino acids (asparagine and glutamic acid, serine, arginine, glycine, tyrosine and proline) was determined. The content of free amino acids in the extract is 0.52 %; the total content of free and bound amino acids is 3.2 %.

3. When studying the stability and establishing the shelf life of the dry extract of phaseolus vulgaris pods, it is necessary to take into account the presence of free amino acids and protein substances.

Конфлікт інтересів: відсутній.

Conflicts of interest: author has no conflict of interest to declare.

АМІНОКИСЛОТНИЙ ПРОФІЛЬ СТУЛОК КВАСОЛІ ЗВИЧАЙНОЇ ТА СУХОГО ЕКСТРАКТУ НА ЇХНІЙ ОСНОВІ

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Мета роботи. Вивчення амінокислотного профілю стулок квасолі звичайної та екстракту на їхній основі.
Матеріали і методи. П'ять зразків сировини стулок квасолі звичайної (кущові сорти із білим насінням) зібрали у Тернопільській і Волинській областях, сухий екстракт стулок квасолі отримали згідно з розробленою раніше технологією. Вивчення амінокислотного складу сировини стулок квасолі та екстракту на їхній основі здійснювали методами тонкошарової хроматографії (ТШХ) і високоефективної рідинної хроматографії (ВЕРХ).
Результати й обговорення. Краще розділення амінокислот при ТШХ-дослідженні сировини стулок квасолі звичайної спостерігали у системі розчинників ізопропанол – мурашина кислота – вода (40 : 2 : 10). У результаті дослідження ідентифіковано аспарагінову і глутамінову кислоти, гліцин, валін, тирозин і лейцин.
Амінокислотний профіль досліджуваних зразків сировини є достатньо однорідний за складом: 7 незамінних амінокислот – гістидин, треонін, валін, метіонін, фенілаланін, ізолейцин і лейцин та 8 замінних – аспарагінова і глутамінова кислоти, аргінін, серин, гліцин, аланін, тирозин і пролін; лізин було виявлено серед зв'язаних амінокислот в 4 зразку сировини. В усіх зразках сировини серед вільних амінокислот пролін домінує. Серед зв'язаних амінокислот найбільший вміст глутамінової кислоти, яка є продуктом гідролізу глутаміну. Також високим є вміст гліцину і серину, аланіну. Серед незамінних амінокислот, в порядку зменшення вмісту, визначено лейцин, фенілаланін, гістидин, треонін, ізолейцин, валін.
Кількісне визначення амінокислот в екстракті стулок квасолі підтвердило найвищий вміст проліну – 12,47 мг/г, а також високий вміст аспарагінової (5,41 мг/г) і глутамінової (3,41 мг/г) кислот, аргініну (5,10 мг/г; як у вільній, так і зв'язаній формах), гліцину (1,02 мг/г) і серину (1,04 мг/г). Із незамінних амінокислот в екстракті незначно відрізняються кількості валіну (0,80 мг/г), фенілаланіну (0,67 мг/г), треоніну (0,66 мг/г), лейцину (0,63 мг/г) та ізолейцину (0,58 мг/г). Загальний вміст амінокислот в екстракті становить 3,2 %.
Висновки. Методом ВЕРХ вивчено амінокислотні профілі п'яти зразків стулок квасолі звичайної. Встановлено, що склад достатньо однорідний, а загальний вміст коливається в межах 0,7–1,1 %.
У сухому екстракті стулок квасолі визначено вміст 5 незамінних та 7 замінних амінокислот. Вміст вільних амінокислот в екстракті – 0,52 %, сумарний вміст вільних і зв'язаних амінокислот – 3,2 %.
При дослідженні стабільності і встановленні терміну придатності сухого екстракту стулок квасолі необхідно враховувати присутність вільних амінокислот і білкових речовин.

Ключові слова: стулки квасолі звичайної; сировина; екстракт; амінокислоти; ВЕРХ; кількісне визначення.

АМИНОКИСЛОТНЫЙ ПРОФИЛЬ СТВОРОК ФАСОЛИ ОБЫКНОВЕННОЙ И СУХОГО ЭКСТРАКТА НА ИХ ОСНОВЕ

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Цель работы. Изучение аминокислотного профиля створок фасоли обыкновенной и экстракта на их основе.

Материалы и методы. Пять образцов сырья створок фасоли обыкновенной (кустовые сорта с белыми семенами) были собраны в Тернопольской и Волынской областях, сухой экстракт створок фасоли был получен согласно разработанной ранее технологии. Изучение аминокислотного состава сырья створок фасоли и экстракта на их основе осуществляли методами тонкослойной хроматографии (ТСХ) и высокоэффективной жидкостной хроматографии (ВЭЖХ).

Результаты и обсуждение. Лучшее разделение аминокислот при ТСХ-исследовании сырья створок фасоли обыкновенной наблюдали в системе растворителей изопропанол – муравьиная кислота – вода (40 : 2 : 10). В результате исследования было идентифицировано аспарагиновую и глутаминовую кислоты, глицин, валин, тирозин и лейцин.

Аминокислотный профиль исследуемых образцов сырья достаточно однороден по составу: 7 незаменимых аминокислот – гистидин, треонин, валин, метионин, фенилаланин, изолейцин и лейцин и 8 заменимых – аспарагиновая и глутаминовая кислоты, аргинин, серин, глицин, аланин, тирозин и пролин; лизин был обнаружен среди связанных аминокислот в четвертом образце сырья. Среди свободных аминокислот во всех образцах сырья доминирует пролин. Среди связанных аминокислот наибольшее содержание глутаминовой кислоты, которая является продуктом гидролиза глутамина. Также высоким является содержание глицина и серина, аланина. Среди незаменимых аминокислот, в порядке убывания содержания, определено лейцин, фенилаланин, гистидин, треонин, изолейцин, валин.

Количественное определение аминокислот в экстракте створок фасоли показало, что содержание пролина является самым высоким – 12,47 мг / г, а также высокие содержание аспарагиновой (5,41 мг / г) и глутаминовой (3,41 мг / г) кислот, аргинина (5,10 мг / г) как в свободной так и связанной формах, глицина (1,02 мг / г) и серина (1,04 мг / г). Из незаменимых аминокислот в экстракте незначительно отличаются количества валина (0,80 мг / г), фенилаланина (0,67 мг / г), треонина (0,66 мг / г), лейцина (0,63 мг / г) и изолейцина (0,58 мг / г). Общее содержание аминокислот в экстракте составляет 3,2 %.

Выводы. Методом ВЭЖХ изучено аминокислотные профили пяти образцов створок фасоли обыкновенной. Установлено, что состав достаточно однороден, а общее содержание колеблется в пределах 0,7–1,1 %.

В сухом экстракте створок фасоли определено содержание 5 незаменимых и 7 заменимых аминокислот. Содержание свободных аминокислот в экстракте – 0,52 %, суммарное содержание свободных и связанных аминокислот – 3,2 %. При исследовании стабильности и установлении срока годности сухого экстракта створок фасоли необходимо учитывать присутствие свободных аминокислот и белковых веществ.

Ключевые слова: створки фасоли обыкновенной; сырье; экстракт; аминокислоты; ВЭЖХ; количественное определение.

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Надійшла до редакції / Received: 05.02.2019
Після доопрацювання / Revised: 11.02.2019
Прийнято до друку / Accepted: 14.02.2019

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