



## Amino acid composition of some *Verbascum* species

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### ABSTRACT

Amino acids are elementary structural units for the formation of proteins and some of them cannot be synthesized within the human body but should enter from outside. When the indispensable amino acids are lacking or some of them are completely absent in food the synthesis of full-blooded proteins becomes impossible with the following disturbances in the work of a number of systems and the appearance of various diseases. We have studied the amino acid composition of the aboveground and underground organs of two species of *Verbascum* widely growing on the territory of the Republic of Kazakhstan. 16 amino acids have been identified in the studied objects, 8 of which being indispensable, their quantitative composition has been established.

**Key words:** amino acids, *Verbascum*, chromatography.

### INTRODUCTION

*Verbascum* species used for different purposes in folk medicine of various nations; therefore, scientists have tested them for different types of biological activities. *Verbascum* species contain biologically active compounds, such as flavonoids, phenylethanoid and neolignan glycosides, saponins, iridoid and monoterpene glycosides. Traditional medicinal use of mullein flower connected to catarrh of the upper respiratory tract, cough and colds documented in handbooks and scientific literature [1-3].

Mullein (*Verbascum*) flowers are highly valued herbal drugs around the world used in the treatment of inflammation, asthma, spasmodic coughs and other respiratory tract diseases.

Amino acids take a special place in the modern medicine. Many of them due to their effects are relate to centralneurotransmitters as stimulates and inhibitory neurotransmission in synapses in the central nervous system, which determine their pharmacological direction [4]. They also contribute to a more rapid absorption and potentiation of trace elements and other biologically active compounds.

However, current knowledge about the content of amino acids in *Verbascum* is limited.

The aim of our study was to investigate amino acid composition of the aboveground and underground parts of *Verbascum Thapsus* and *Verbascumsongaricum* by gas chromatography.

### EXPERIMENTAL SECTION

Object of current study is the aboveground and underground parts of *Verbascum Thapsus* and *Verbascumsongaricum*.

To determine the amino acid in 1 g of substance, it hydrolyzed in 5 ml of six normal (N) hydrochloric acid at 105°C for 24 hours in vials sealed under a stream of argon. The hydrolyzate evaporated to dryness three times on a rotary evaporator at a temperature of 40-50°C and a pressure of one atmosphere. The resulting precipitate dissolved in 5 ml of sulfosalicylic acid. After centrifugation (1500 rev / min) for 5 minutes, the supernatant passed through a column of ion exchange resin Dowex 50, H-8, and 200-400 mesh, at a rate of 1 drop per second. Thereafter, the resin washed with 1-2 mL of deionized water and 2 ml of 0.5 N acetic acid; Resin washed to neutral pH deionized water.

Amino elution from the column passed therethrough 3 ml six N NH<sub>4</sub>OH solution at 2 drops per second. The eluate collected in a round bottom flask with deionized water used for washing the column to neutral pH. The flask contents evaporated to dryness on a rotary evaporator under a pressure of one atm. and a temperature of 40-50°C.

After addition to the flask of 1 drop of a freshly prepared 1.5% solution of SnCl<sub>2</sub>, 1 drop of 2,2-dimethoxypropane and 1-2 ml of saturated hydrochloric acid-propanol, heated to 1100S, maintaining this temperature for 20 minutes and then the contents of the flask was evaporated again on a rotary evaporator.

In the next step in the flask, 1 ml of freshly prepared acylating reagent (1 volume of acetic anhydride, triethylamine, 2 volumes and 5 volumes of acetone) and heated at 600 ° C for 1.5-2 min. The sample re-evaporated to dryness on a rotary evaporator and added to the flask 2 mL of ethyl acetate and 1 ml of a saturated solution of NaCl. The flask contents mixed thoroughly and as two layers of liquids clearly formed - take the upper (ethyl acetate) for gas chromatography analysis, which conducted on gas-liquid chromatography "Carlo-Erba-4200" (Italy-USA).

The conditions of chromatography:

- Temperature of the flame ionization detector - 300°C
- Evaporator temperature - 250°C
- The initial temperature of the column - 110°C
- The final column temperature - 250°C
- Column temperature programming speed: from 110°C to 185°C - 60°C in the minutes; from 185°C to 250°C - 32°C min. When the temperature reaches 250°C column it should remain until the full release of all amino acids.

For the separation of amino acids a stainless steel column, size 400 mm 3 filled with a polar mixture of 0.31% Carbowax 20 m 0.28 5% Silar CP and 0.06% on the Lexan hromasorbe WA-W-120-140 mesh were used . Counting carried out on the chromatogram external standard of company Altex [5].

## RESULTS AND DISCUSSION

Results of research presented in Table 1. 16 amino acids have been identified in the studied objects, 8 of which being indispensable (Leucine, isoleucine, histidine, tyrosine, glycine, lysine, valine, methionine).

**Table 1: The content of free amino acids in some *Verbascum* species**

№	Aminoacids	Content, %			
		<i>Verbascumsongaricum</i>		<i>Verbascumthapsus</i>	
		herb	roots	herb	roots
1	Threonine	0,400	0,412	0,426	0,390
2	Valine	0,358	0,375	0,400	0,362
3	Methionine	0,174	0,190	0,198	0,181
4	Isoleucine	0,365	0,389	0,412	0,374
5	Leucine	0,590	0,604	0,628	0,572
6	Phenylalanine	0,388	0,404	0,420	0,390
7	Histidine	0,300	0,325	0,310	0,310
8	Lysine	0,290	0,303	0,303	0,292
9	Arginine	0,632	0,644	0,658	0,633
10	Aspartic acid	1,020	1,148	1,155	0,996
11	Glutamic acid	2,540	2,590	2,650	2,530
12	Serine	0,482	0,495	0,520	0,490
13	Proline	0,862	0,884	0,900	0,855
14	Glycine	0,268	0,282	0,302	0,260
15	Alanine	0,720	0,796	0,900	0,762
16	Tyrosine	0,504	0,526	0,530	0,505

In all the objects, high content of aspartic acid and glutamic acid detected. The maximum accumulation of alanine observed in the *Verbascum Thapsus* herb. The results based for a deeper study of plants as a kind of source natural biologically active compounds.

## REFERENCES

- [1] P. Bradley, *British Herbal Compendium*, British Herbal Medicine Assoc., Bournemouth, **2006**.
- [2] W. Blaschek, S. Ebel, E. Hackenthal, U. Holzgrabe, K. Keller, J. Reichling, V. Schulz, Hager ROM, **2006**.
- [3] *Herbal Drugs and Phytopharmaceuticals*, 3rd Edition, Edited by Max Wichtl (Universität Marburg), CRC Press, Boca Raton, **2004**, 704.
- [4] Копытько Я.Ф., Костеникова З.П., Тимохина Е.А. *Фармация*, **1997**, 6, 31-34.
- [5] Adams R. J., *Chromatography*, **1974**, 95 (2), 188-212.