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ANALYSIS OF THE ACID COMPOSITION OF PLANTS OF FAMILY *SCROPHULARIACEAE*

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Abstract. This article studies the amino and fatty acid composition of the aboveground parts of the plant *Verbascum*, of *Scrophulariaceae* family, collected in the East Kazakhstan region during the fruiting period in 2024. The relevance of the work is the identification of biologically active substances from plants growing in the region and the assessment of their potential for use in pharmaceutical production. During the study, the quantitative composition of various amino and fatty acids in the aboveground parts of the plant was determined, using paper and gas chromatography methods. As a result, amino acids (leucine, lysine, arginine) and unsaturated fatty acids (linoleic, oleic) important for the human body were found in the plant composition. The data obtained confirm the pharmacological potential of the plant *Verbascum* and indicate that it can be used in the production of medicinal products and food additives. The results of the study are important for future research in the field of phytochemistry, pharmaceuticals and the food industry.

The aboveground part of the plant is a source of many important compounds. Using paper chromatography, phenolic acids, flavonoids, and carbohydrates were identified from the plant. The results of the study showed that the plant species contains a sufficient amount of biologically active substances, which in the future will allow Kazakhstan to expand the range of domestically effective medicinal products available in medicine and agriculture of the republic.

Keywords: *Scrophulariaceae, Verbascum,* amino acids, fatty acids, gas-liquid chromatography, paper chromatography.

Introduction. Currently, the focus is on herbal medicines. Kazakhstan is rich in natural resources and has a significant potential for the production of medicinal plants. The diversity of geographical conditions in the eastern regions of our country determines the richness and diversity of the flora. The morphological and anatomical features of plants are closely related to their growth conditions and environment.

More than 6,000 species of plants grow in Kazakhstan, of which 515 are endemic to this territory [1]. Their roots, stems, and leaves are widely used in medicine to treat various diseases and injuries, as well as in psychiatry and to maintain the cardiovascular system. However, many plants of Kazakhstan are still waiting for their researchers [2].

Amino acids are the main building blocks of protein molecules. About 300 amino acids are found in nature, but only 20 of them are proteinogenic, that is, they participate in the construction of proteins [3]. Protein amino acids are α-amino acids that have a common structure with carboxyl and amino groups. In terms of the nutritional value of protein, amino acids are divided into non-essential and essential [4]. Essential amino acids, such as valine, leucine, methionine, phenylalanine, threonine, tryptophan, and lysine, play an important role in various processes. For example, threonine is involved in the synthesis of fatty acids, lipids, and carbohydrates, while methionine, cysteine, and cystine provide the body with organic sulfur. Tryptophan deficiency can negatively affect the function of the gonads [5].

Fatty acids are structural components of cell membrane lipoproteins and participate in important biochemical reactions. Fatty acids with two or more double bonds, such as linoleic, linolenic, and arachidonic acids, are especially valued. Enzymatic oxidation of arachidonic acid leads to the formation of important metabolites such as prostaglandins, thromboxanes, and

leukotrienes [6]. Prostaglandins, acting as intracellular bioregulators, affect the cardiovascular, respiratory, reproductive, and other systems and are used in the treatment of hypertension, asthma, thrombosis, peptic ulcer disease, and gynecological diseases. Thromboxanes are involved in the processes of thrombus formation and hematopoiesis, and leukotrienes affect allergic reactions. Unsaturated fatty acids prevent atherosclerosis, reduce blood clotting and the risk of blood clots, increase the body's defenses against infections, and protect against skin diseases. There is evidence of their protective effect against substances that cause tumor development [7].

The aim of the study is to study the chemical composition of the plant *Verbascum orientale* L., to obtain biologically active substances. To identify its main clusters, to conduct qualitative and quantitative analysis of biologically active substances using single- and double-system paper chromatography. To determine the amount of extractive substances using different percentage aqueous-alcoholic solutions, and to separate flavonoids from the plant composition. To consider the scheme - a variant of the separation of biologically active substances.

Materials and methods of study. The object of study is the above-ground part of the plant *Verbascum orientale*, belonging to the family *Scorchulariaceae*, growing in the territory of East Kazakhstan. It was ground in accordance with the requirements of GOST 24027. 1-80. Determination of the moisture content of the raw material and the yield of extractive substances was carried out in accordance with GOST 24027. 2-80. Studies of the quantitative composition of the main BAC of plants were carried out according to the methodological instructions of the XI State Pharmacopoeia.

Analysis of amino acids [8]. 1 g of raw material was hydrolyzed in 5 ml of 6N HCl at 105^0C for 24 hours in an ampoule sealed with argon. The resulting hydrolysate is evaporated to dryness three times at 40°C in a rotary vacuum evaporator, then the precipitate obtained is dissolved in 5 ml of 5% sulfosalicylic acid after centrifugation at 2500 rpm. Over the next 15 minutes, the supernatant is passed through a Dausk 504-8 ion exchange column with a rotation speed of 200-4000 rpm at a rate of 1 drop per second. First, the resin is washed with 1-2 ml of D. I . water and 0.5 N acetic acid, and then again with distilled water until a neutral pH is reached. To elute the amino acids, 3 ml of 6N solvent is dripped through the column at a rate of 2 drops per minute. The eluate is collected in a round-bottom flask together with the distilled water used to wash the column until the pH is neutral. The eluate in the flask should be dried in a rotary evaporator at 1 atmosphere and a temperature of $50\text{-}60^0$C.

As for the next step, 1 drop of fresh 2,2-dimethoxypropane and 1-2 ml of saturated HCl propanol are added to the flask and heated to 110^0C for 20 min and evaporated to dryness again in a rotary vacuum. In the next step, 1 ml of fresh oxidizing reagent (1 volume of acetic oxide, 2 volumes of triethylamine, 5 volumes of acetone) is added and heated to dryness at 600^0C for 1.5-2 min, 2 ml of ethyl acetate and 1 ml of saturated NaCl solvent are added. The flask should be mixed thoroughly until two liquid layers are formed. The upper layer (containing ethyl acetate) should be removed for gas chromatographic analysis, which was carried out on a "CARLO-ERBA-420" gas chromatograph. Since the column temperature reached 2500C, it was necessary to hold until all amino acids were released. The analysis data are shown in Table 1.

Analysis of fatty acids [8]. The extracted and shaken aerial parts of the plant raw material are extracted with a chloroform-methanol mixture (2:1) for 5 minutes. The obtained extract is filtered through a paper filter and concentrated to dryness. After that, 10 ml of methanol and 2-3 drops of acetyl chloride are added to the extract, and methylation is carried out in a special system at a temperature of $60\text{-}70^0$C for the last 30 minutes. Methanol is removed using a rotary evaporator and the samples are extracted with 5 ml of hexane, which is analyzed for 1 hour on a "CARLO-ERBA-420" gas chromatograph. As a result, chromatograms of fatty acid methyl esters were obtained. 8 fatty acids were identified by column elution time compared with reliable samples [9]. The results are shown in Table 2.

To determine the content of components, the internal normalization method is used, and the concentration of components is calculated using the following formula [10]:

$$c_i = \frac{Si}{\sum_{n=1}^{n} Si} \cdot 100$$

Research results.

The collected plant material was dried according to the state standard. The dried plant material was ground in a special grinder, 100 g of plant material was taken, poured with 30% and 70% aqueous alcohol in a ratio of 1:8, and left for 72 hours.

Figure 1 − Aqueous-alcoholic extract

By two-system paper chromatography method; n – butanol: acetic acid: water (40:12.5:29) and 6% acetic acid systems, flavonoid aglycones, phenolic acids and flavonoid glycosides were found in the extracts. The results of the GC showed that the biologically active substances were particularly pronounced in the 70% alcoholic extract of the quality composition of the plant Verbassum orientale, so the work was carried out with this percentage of the extract [11].

Table 1 − Amount of amino acids in the composition of *Verbascum*

№	Amino acids	Amount, mg/100g
1	Alanine	**1253**
2	Glycine	482
3	Valine	256
4	Leucine	315
5	Isoleucine	286
6	Threonine	244
7	Serine	715
8	Proline	956
9	Methionine	74
10	Aspartic acid	**1246**
11	Cystine	24
12	Oxyproline	4
13	Phenylalanine	318
14	Glutamic acid	**3892**

15	Ornithine	5
16	Tyrosine	285
17	Histidine	302
18	Arginine	448
19	Lysine	310
20	Tryptophan	182

Table 2 − Amount of fatty acids in the composition of *Verbascum*

№	**Fatty acids**	**Percentage**
1	Myristic acid (C14:0)	0,6
2	Pentadecyl acid (C15:0)	0,8
3	Palmitic acid (C16:0)	**11,2**
4	Stearic acid (C18:0)	1,1
5	Palmitoleic acid (C16:1)	3,2
6	Oleic acid (C18:1)	**50,2**
7	Linoleic acid (C18:2)	**32,5**
8	Linolenic acid (C18:3)	0,4

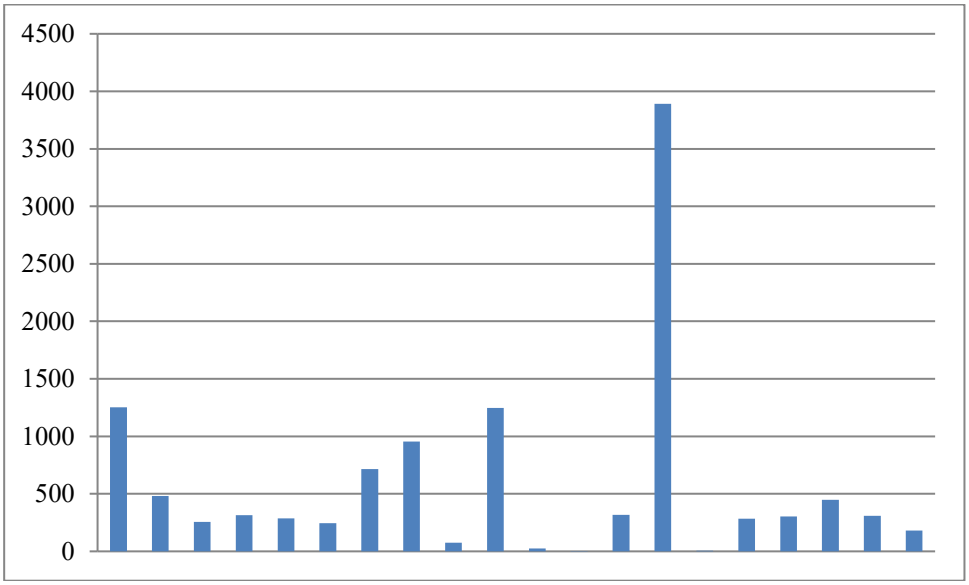

Diagram 1 - Amount of amino acids in the composition of the *Verbascum*

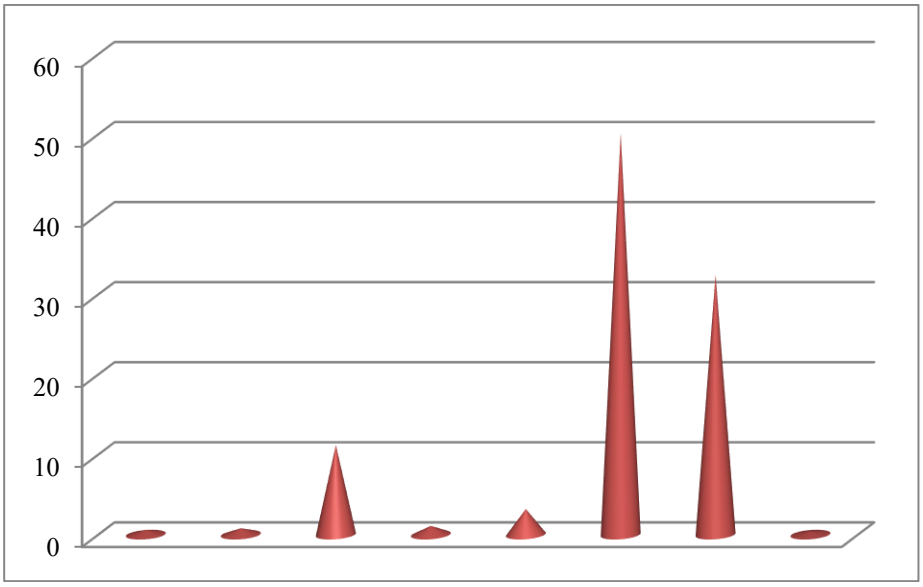

Diagram 2 - Amount of fatty acids in the composition of *Verbascum*

The quantitative content of 20 amino acids in the plant *Verbascum orientale* was determined. The highest amino acids in 100 g of raw material were glutamate 2600 mg, aspartate 1096 mg and proline 898 mg.

Conclusion. As a result of quantitative and qualitative analysis of the plant **Verbascum orientale**, belonging to the family *Scorchulariaceae*, growing in the East Kazakhstan, 30% and 70% ethanol solvents were used to separate the biologically active complex from its composition, and 70% was found to be effective. Flavonoids, carbohydrates, phenols, phenolic acids, and amino acids were studied in plant raw materials using paper chromatography. The amino and fatty acid composition of the raw materials was determined using gas-liquid chromatography, and it was found that alanine, aspartic acid, and glutamic acid were more abundant in the amino acids, while palmitic acid, oleic acid, and linoleic acid were more abundant in the fatty acids.

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АНАЛИЗ КИСЛОТНОГО СОСТАВА РАСТЕНИЙ СЕМЕЙСТВА *SCROPHULARIACEAE*

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Аннотация. В данной статье изучен аминокислотный и жирнокислотный состав надземных частей растения *Verbascum*, относящегося к семейству *Scrophulariaceae*, собранного в Восточно-Казахстанской области в период плодоношения в 2024 году. Актуальность работы заключается в выявлении биологически активных веществ из растений, произрастающих в регионе, и оценке их потенциала для использования в фармацевтическом производстве. В ходе исследования был определен количественный состав различных аминокислот и жирных кислот в надземных частях растений, с использованием методов бумажной и газовой хроматографии. В результате в составе растения обнаружены важные для организма человека аминокислоты (лейцин, лизин, аргинин) и ненасыщенные жирные кислоты (линолевая, олеиновая). Полученные данные подтверждают фармакологический потенциал растения *Verbascum* и свидетельствуют о возможности его использования в производстве лекарственных средств и пищевых добавок. Результаты исследования важны для будущих исследований в области фитохимии, фармацевтики и пищевой промышленности.

Надземная часть растения является источником многих важных соединений. С помощью бумажной хроматографии из растений были идентифицированы фенольные кислоты, флавоноиды и углеводы. Результаты исследования показывают, что вид растения обладает достаточным количеством биологически активных веществ, что в перспективе позволит Казахстану расширить ассортимент отечественных эффективных лекарственных средств, доступных в медицине и сельском хозяйстве республики.

Ключевые слова: *Scrophulariaceae, Verbascum*, аминокислоты, жирные кислоты, газожидкостная хроматография, бумажная хроматография.

SCROPHULARIACEAE ТҰҚЫМДАСЫНА ЖАТАТЫН ӨСІМДІКТЕРДІҢ ҚЫШҚЫЛДЫҚ ҚҰРАМЫН ТАЛДАУ

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Аңдатпа. Бұл мақалада Шығыс Қазақстан облысында 2024 жылы жеміс беру кезеңінде жиналған *Scrophulariaceae* тұқымдасына жататын *Verbascum* өсімдігінің жер үсті бөліктерінің амин және май қышқылдық құрамы зерттелген. Жұмыстың өзектілігі – өңірде өсетін өсімдіктерден биологиялық белсенді заттарды анықтау және оларды фармацевтикалық өндірісте қолдану мүмкіндігін бағалау. Зерттеу барысында өсімдіктерінің жер үсті бөлігіндегі әртүрлі амин және май қышқылдарының сандық құрамы анықталып, қағазды және газды хроматографиялық әдістер қолданылды. Нәтижесінде, өсімдік құрамынан адам ағзасы үшін маңызды амин қышқылдары (*лейцин, лизин, аргинин*) және қанықпаған май қышқылдары (*линол, олеин*) табылды. Алынған деректер *Verbascum* өсімдігінің фармакологиялық әлеуетін растап, оны дәрілік препараттар мен тағамдық қоспалар өндіруде пайдалануға болатынын көрсетеді. Зерттеу нәтижелері фитохимия, фармацевтика және тағам өнеркәсібі саласындағы болашақ зерттеулер үшін маңызды болып табылады.

Өсімдіктің жер үсті бөлігі көптеген маңызды қосылыстардың көзі болып табылады. Қағаз хроматографиясының көмегімен өсімдіктерден фенол қышқылдары, флавоноидтар және көмірсулар анықталды. Зерттеу нәтижелері көрсеткендей, өсімдік түрінің биологиялық белсенді заттардың жеткілікті мөлшері бар екендігі, болашақта Қазақстан республиканың медицинасы мен ауыл шаруашылығында қолжетімді отандық тиімді дәрілік препараттардың ауқымын кеңейте алады.

Түйін сөздер: *Scrophulariaceae, Verbascum*, аминқышқылдары, май қышқылдары, газды-сұйық хроматография, қағаз хроматографиясы.