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# STUDY OF THE AMOUNT OF AMINO ACIDS IN THE DRY EXTRACT "GLABRUS" OBTAINED FROM THE PLANT RHUS GLABRA

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**Abstract**

This article presents the results of research conducted jointly with scientists from the Tashkent Pharmaceutical Institute and the O.S. Sadykov Institute of Bioorganic Chemistry to determine the total amino acid content in the dry

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extract based on the conditional name "GLABRUS," obtained from the local medicinal plant raw material Rhus glabra L.

Based on the results of the conducted research, the content of proteinogenic and non-proteinogenic amino acids in the dry extract was determined by the HPLC method.

**Keywords:** Totum (sumah), Rhus glabra L., dry extract, amino acids, extraction, standard sample, HPLC, proteinogenic and non-proteinogenic, Kochen method.

## Introduction

According to the World Health Organization, as a result of bad climatic conditions, improper nutrition, stress, and serious illnesses, the amount of proteins in the human body significantly decreases. Failure to prevent this in a timely manner leads to a disruption of protein levels in the body and disruption of metabolic processes.

This article summarizes information on amino acids synthesized by plants, their metabolic functions, and the pharmacological activity of some of them, especially in the formation of protective factors against Parkinson's disease, the second most common neurodegenerative disease in geriatric practice.

The literature contains numerous scientific studies dedicated to the Totum plant, where the plant's phytochemical composition has been thoroughly studied and it has been determined to contain tannins, polyphenols, flavonoids, organic acids, and essential oils [9]. Hundreds of biochemical processes occur continuously in the human body, in which macro- and microelements directly participate. According to the general calculation of macroelements, the dry extract "glabrus," obtained from the plant Rhus glabra, contains potassium -76%, magnesium - 9%, calcium - 4%, phosphorus

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- 4.5-5%, sodium - 3%, sulfur - 1-2%, and microelements B-55% and Si-27% [10].

It is known that when standardizing the qualitative and quantitative composition of amino acids in plant raw materials, as well as medicinal plants, it is necessary to standardize the overall composition of amino acids.

About 300 types of amino acids are found in the body, but it has been established that there should be 20 essential amino acids (8 essential, 10 essential, and 2 partially essential), without which protein synthesis in the body is impossible [7].

Tyrosine and cysteine are conditionally substituted amino acids, since their synthesis requires the essential amino acids phenylalanine and methionine, respectively. Medicinal plant preparations contain various biologically active substances (BAS), including amino acids. Amino acids also give other biologically active substances an easily digestible and harmless form, while simultaneously enhancing their pharmacological effect [8].

Plants have the ability to synthesize all the nitrogen compounds they need. In the process of amino acid synthesis, ammonium nitrogen is converted into organic compounds.

**Purpose of the Research:**

Analysis of the amino acid number in the dry extract obtained from the leaves of "R. glabra" using 80% ethyl alcohol by a modern method.

**Materials and Research Methods**

Method for determining free amino acids.

1 g (precise amount) was weighed from the samples on analytical scales and placed in a flat-bottomed flask. 10 ml of distilled water was poured into the flasks, and the extraction was carried out in an ultrasonic bath of the brand

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8510 BRANSON for 8 hours with stirring every hour for 10 minutes. Then the mixture in the flask was centrifuged for 20 minutes at a rate of 6000 rpm, the supernatant was separated, and the precipitate was discarded. To eliminate the proteins and peptides in the extract, 1 ml of 10% trichloroacetic acid was added to 1 ml of supernatant. After 10 minutes, the resulting precipitate was centrifuged for 15 minutes at a rate of 6000 rpm, the supernatant was separated, and the precipitate was discarded. To determine the content of free amino acids, 250 µl of the supernatant was dried in a lyophilic dryer.

A mixture was prepared in a clean test tube at a ratio of water: acetonitrile: triethylamine (1:7:1) and phenylthiocarbamoyl (FTK) derivatives of amino acids were obtained according to the Cohen method. 100 µl of a mixture (water: acetonitrile: triethylamine (1:7:1)) was added to each sample. Then it was placed in a drying bottle again for 1 hour, taken from the drying bottle and placed in a thermostat at 37℃ for half an hour in a ratio (100 (1:7:1):1) (water: acetonitrile: triethylamine): phenylylisotiocyanate (100:1). It was removed from the thermostat, placed in the refrigerator for 15-20 minutes, and again placed on a lyophilic dryer for 1-1.5 hours. Then, after double washing with acetonitrile, a HSS analysis of the synthesis of phenylthiocarbamoyl (FTK) derivatives of amino acids in the hydrolysates was carried out.

A set of amino acids from the company "SERVA" was used as a standard. For this, a mixture of standard amino acids with a concentration of 0.01 mg/ml was prepared. FTK derivatives of these amino acids were synthesized by the Kochen method.

When identifying amino acids in the samples, the timing of the amino acids in the standard sample exiting the column was used, and for quantitative analysis, the surface of the peaks of each amino acid on the chromatogram was used.

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Conditions for HPLC: Chromatograph: Agilent Technologies 1200 (with DAD detector); column: Discovery HS C18 (75×4.6 mm); mobile phases: solution A: 0.14 M sodium acetate + 0.05% triacetylamine, pH=6.4; Solution B: acetonitrile (CH3CN); flow rate:1.2 ml/min; detection wavelength:269 nm gradient (solution B, %/min):1-6%/0-2.5 min; 6-30%/2.51-40 minutes; 30-60%/40.1-45 minutes; 60-60%/45.1-50 minutes; 60-0%/50.1-55 minutes.

**Discussion of the obtained results:**

The results of the study of the content of amino acids in the dry extract "Glabrus," obtained from the plant Rhus glabra, are presented in Table 1.

Table 1 Dry extract obtained from the leaves of "Rhus glabra" amino acid count

| № | Name of amino acids | Amount of amino acids in the dry extract of Rhus glabra, mg/g |
|---|---|---|
| 1. | Aspartic acid | 4.287 |
| 2. | Glutamic acid | 0.641 |
| 3. | Serine | 0.901 |
| 4. | Glycine | 2.981 |
| 5. | Asparagine | 2.964 |
| 6. | Glutamine | 0.565 |
| 7. | Cysteine | 0.909 |
| 8. | Histidine | 1.019 |
| 9. | Argenin | 0.577 |
| 10. | Alanine | 1.471 |
| 11. | Proline | 2.081 |
| 12. | Tyrosine | 0.846 |
| 13. | Valine* | 2.710 |
| 14. | Methionine* | 1.373 |
| 15. | Threonine | 1.786 |
| 16. | Isoleucine* | 1.963 |
| 17. | Leucine* | 1.712 |
| 18. | Tryptophan* | 1.004 |
| 19. | Phenylalanine* | 1.862 |
| 20. | Lysine* | 0.447 |
| In total | | **32.099** |

* Essential amino acids

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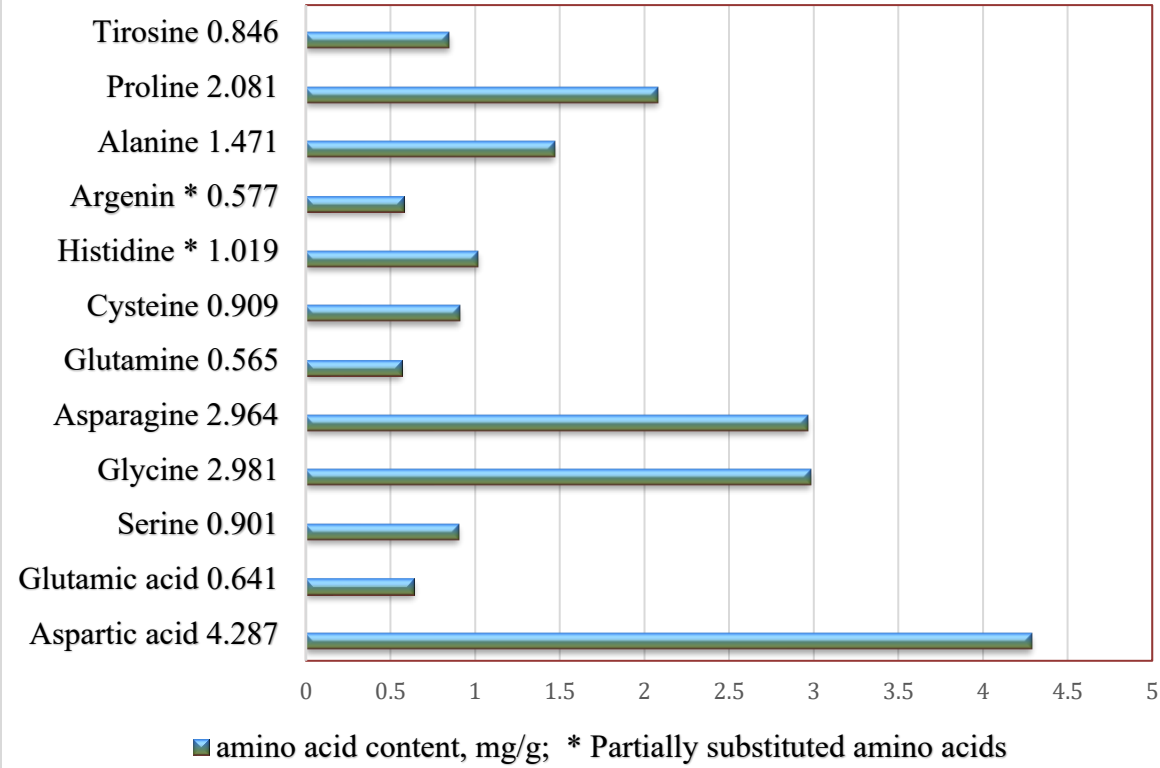


**Figure 1. Results of the analysis of the content of metabolizable amino acids in the dry extract**

As can be seen from Figure 1, the content of aspartic acid and glycine in the dry extract is higher than in other amino acids. In the human body, aspartic acid is present not only in peptides but also in free form, but its concentration is especially high in brain tissue, as it is the second most important amino acid with neurotransmitter properties [1]. Glycine is the simplest amino acid in terms of structure and plays an important role in the central nervous system

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(CNS). It acts as a fast-acting primary inhibitory neurotransmitter and as an excitation neurotransmitter modulating factor [6].

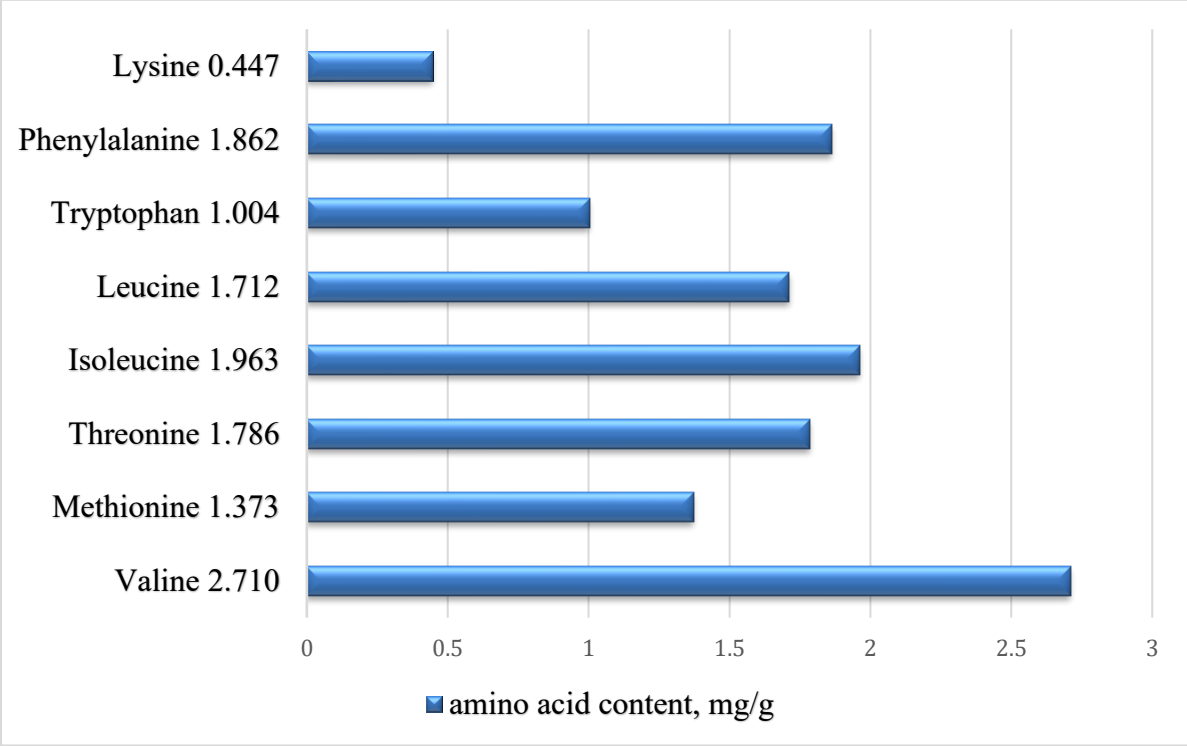


Figure 2. Results of the analysis of the content of essential amino acids in the dry extract.

Amino acids play many roles in the central nervous system, acting as neurotransmitters, neuromodulators, as well as regulators of energy metabolism [2].

According to scientific sources provided by Chinese scientists, a high level of phenylalanine is associated with a lower risk of Parkinson's disease ("safety factor"). That is, according to genetic data, if there is more phenylalanine, the

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probability of PD development decreases. A 2024 Mendelian randomization analysis found that individuals with a genetic profile with higher levels of phenylalanine were less likely to develop Parkinson's disease. This can be understood as the effect of the "safety factor," that is, the presence of sufficient phenylalanine in the body positively affects dopamine synthesis and the balance of neurotransmitters [3].

Arginin has high therapeutic potential not only for the treatment of Alzheimer's and PolyQ diseases, but also for a wide range of neurodegenerative diseases associated with protein malformation and aggregation [4].

Branched-chain amino acids (BCAA) - i.e., leucine (LEU), isoleucine (ILE), and valine (VAL) - are among the most common essential amino acids in the human body, and the results of the conducted research show that the branched-chain amino acids leucine-1.712, isoleucine-1.963, and valine-2.710 have high biological value in the dry extract of the conditional name Glabrus. Branched-chain amino acids pass through the blood-brain barrier via large neutral amino acid carriers. These carriers also carry tryptophan (TRP) and tyrosine (TYR), which are precursors of serotonin (5-HT) and dopamine (DA), respectively. This competitive mechanism in the transport process means that an increase in the amount of BCAA in the body can reduce the penetration of TRP and TYR into brain tissue, which increases the likelihood of disruption of the synthesis and function of monoamine neurotransmitters [5].

## Conclusion

The amino acid composition of the alcohol-water extract of Rhus glabra consists of 20 components and is characterized by a high content of aspartic acid, glycine, threonine, proline, valine, isoleucine, phenylalanine, a low

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content of histidine, alanine, methionine, leucine, and tryptophan, and a very low content of glutamic acid, glytamine, arginine and lysine.

It should also be noted that the neuroprotective effect of R. glabra, along with the main biologically active substances, increases the flow of Na+ and Ca+2 ions, which has a beneficial effect on the heart muscle in neurodegenerative diseases and is associated with a high level of asporbic acid. The results of the conducted research will serve as the basis for our further research.

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