## FRACTIONATION AND AMINO ACID COMPOSITON OF PROTEINS OF THE WOODY VERDURE OF SEA BUCKTHORN

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The woody verdure of sea buckthorn is rich in protein substances. Among the soluble proteins, water- and alkali-soluble fractions predominate. The presence of 17 amino acids in the water-soluble proteins has been established. The sum of the essential amino acids amounts to 42.12-49.51%. The water-soluble proteins of the woody verdure of sea buckthorm possess a high biological value.

Sea buckthorn wastes may be an additional source of plant protein. Thus, in the collection of the berries a large amount of woody verdure of sea buckthorn accumulates: leaves, bark, unlignified twigs with a diameter up to 10 mm, and a small amount of berries. The sea buckthorn woody verdure is rich in protein substances (Table 1). In June it contains about 20% of total protein, which accumulates as the leaves grow and the fruit ripens. Its maximum amount is present in August-September (26.5%). In October after the ripening of the fruit and the yellowing of the leaves the total protein content falls to 19.6%.

In all cases, water-and alkali-soluble fractions predominate in the plant proteins of sea buckthorn woody verdure. They make up more than 90% of the total protein. The amounts of water-soluble proteins are practically the same in June and July (8.76 and 8.35%), but by September, with the ripening of the fruit, their amount falls to 1.38%.

The alkali-soluble proteins have a different nature. In July their amount is higher than in June (9.94% as compared with 9.29%) and the minimum amount is observed in September (5.62%). In October, the amount of alkali-soluble proteins almost doubles in comparison with September.

The salt- and alcohol soluble fractions of the proteins amount to a small percentage of the total soluble proteins. The difference in the amounts of salt-soluble fractions is slight: they range from 0.58 to 0.82%. The maximum amount of alcohol-soluble proteins is found in June (0.94%), while in August their amount halves, reaching a minimum.

The largest amount of soluble proteins is present in June, (97.36% of the total protein). On comparing the sum of the soluble proteins with the total amount of protein it can be seen that from June to September the proportion of soluble proteins falls by a factor of more than two, while in October there is almost twice as much of them as in September.

The water-soluble fraction of the proteins of sea buckthorn woody verdure has been studied in the most detail. Among the water-soluble proteins 17 amino acids have been identified and determined quantitatively. The amino acid composition of the water-soluble proteins depends on the time of collecting the sample. As can be seen from Table 2, the amount of essential amino acids ranges from 42.12 to 49.51%. This exceeds the amount in sea buckthorn juice, where their amount ranges from 22.60 to 31.1% [1].

Predominant among the essential amino acids are leucine and lysine, which are deficient in the majority of plant feedstuffs. The limiting amino acids are methionine and cystine, their proportion in the total amount of acids being less than 1%. Of the other amino acids we must mention the high levels of asparagine, glutamic acid, and alanine.

From the results of amino acid analysis we calculated the indices of the biological value of the water-soluble proteins (the BVP index) and the amino acid scores for such

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TABLE 1. Fractional Composition of the Proteins of Sea Buckthorn Woody Verdure, % on Absolutely Dry Substance

D. et al.	Time of collecting the sample						
Protein fraction	June	July	August	September	October		
Water-soluble Salt-soluble Alcohol-soluble Alkali-soluble Total soluble proteins Total protein	8,76 0,58 0,94 9,29 19,57 20,10	8,35 0,70 0,66 9,94 19,65 25,10	6,65 0,73 0,41 7,23 15,03 26,0	1,38 0,79 0,59 5,62 8,38 26,20	1,64 0,82 0,75 11,10 13,31 19,60		

TABLE 2. Amino Acid Composition of the Water-soluble Proteins of Sea Buckthorn Woody Verdure

Amino acid	Amount, % of the total amino acids						
Alimio acid	June	June July		September	October		
Lysine Histidine Arginine Cystine Aspartic acid Threonine Serine Glutamic acid Proline Glycine Alanine Valine Methionine Isoleucine Leucine Tyrosine	8,40 1,90 6,64 0,34 11,68 4,05 5,60 15,88 3,39 6,30 10,54 4,93 0,38 2,70 9,81 3,93	8,47 2,49 7,28 0,36 13,91 3,89 5,48 15,80 2,34 5,83 9,57 4,44 1,05 2,24 9,87 3,59	7,00 2,35 6,18 0,78 18,84 4,16 5,11 14,08 5,28 8,63 3,85 0,31 2,04 8,87 3,53	5,55 1,73 6,41 2,43 18,37 3,99 6,24 17,33 2,25 6,93 6,76 4,68 0,52 2,60 8,49 2,43	9,40 3,30 7,53 0,50 13,08 4,30 4,79 17,00 3,11 5,04 7,47 4,86 0,31 1,43 10,27 3,44		
Phenylalanine Sum of essential amino acids	3,47 46,58	3,39 46.02	4,08 43,15	3,29 42,12	4,17 49,51		

essential amino acids as lysine, histidine + methionine, valine, isoleucine, threonine, and tyrosine + phenylalanine (Table 3). The ideal amino acid scale recommended by the FAO in 1973 was selected as the standard.

It can be seen from Table 3 that lysine, threonine, leucine, and tyrosine + phenylalanine have amino acid scores exceeding 100%. The BVP indices for all the water-soluble proteins are close to 100%. An exception is the woody verdure of sea buckthorn gathered in September: it has a low index of the amino acid score and BVP index. The indices of the biological value of the proteins and the amino acid score of the water-soluble proteins of sea buckthorn are superior to the analogous indices of such traditional crops as alfalfa and red clover. Thus, the protein of alfalfa, a valuable fodder crop, contains 43.5% of essential amino acids [2] and has a BVP of 91.64%, which is somewhat lower than the results that we have obtained.

## EXPERIMENTAL

The woody verdure of sea buckthorn growing in Altai Territory [1] was studied. Samples weighing 3-5 kg were gathered in June-October on the territory of the plantations of the Biisk Technical School of Forestry in the first ten days of each month from 20 shrubs during the times of the greatest vital activity of the plants, namely: in the periods of the incipient growth of shoots, the development of the leaves, the ripening of the fruit, and the fall of the leaves. They were ground in a laboratory mill and were separated into fractions according to degrees of solubility by the generally adopted procedure in the variant of S. M. Tyuterev and Z. V. Chmeleva [3].

The amount of nitrogen in each of the protein fractions was determined by an accelerated photometric method using a ninhydrin reagent in place of the Nessler reagent [4], which improved the accuracy and sensitivity of the method.

The purified protein fractions were subjected to acid hydrolysis with 6 N hydrochloric acid in sealed tubes at 105°C for 24 h [5]. The hydrolysates were freed from humins by filtration in vacuum at 40°C, and the dry residue was dissolved in an aliquot of buffer with pH 2.2. The amino acids were determined quantitatively on a model AAA-881 automatic amino acid analyzer using a standard mixture of amino acids as internal, and leucine as external, standard [6].

TABLE 3. Indices of the Biological Value of the Water-soluble Proteins of Sea Buckthorn Woody verdure

		Amino acid score, % of total							
Month	1ysine	cystine + methio- nine	threo- nine	val- ine	isoleu- cine	leu- cine	tyrosine + phenyl- alanine	BVP index	
June July August September October	152,7 154,0 127,3 10,9	2 6 40,3 31,3 84,3 23,1	101.2 97.2 1.4. 99.7 1.7.5	98.6 88.8 77.0 93,6 97,2	67.5 56.0 51,0 65,0 35,7	140,1 141,0 126,7 121,3 146,7	123,3 116,3 126,8 95,3 126,8	94,22 96,55 89,29 78 14 92,61	

## SUMMARY

- 1. The woody verdure of sea buckthorn is rich in protein substances (19.6-26.5% on the absolutely dry matter).
- 2. Among the soluble proteins the water-soluble and alkali-soluble fractions predominate (1.4-8.8% and 5.6-11.1%, respectively, on the absolutely dry matter).
- 3. In the water-soluble proteins 17 amino acids have been identified and determined quantitatively. The sum of the essential amino acids ranges from 42.12 to 49.51%.
- 4. The water-soluble proteins of sea buckthorn woody verdure possess a high biological value exceeding the biological value of the proteins of several fodder crops.

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