ISOLATION OF PROTEIN FROM TOMATO SEED MEAL

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The results are given of a study of the extraction and precipitation of tomato seed protein. Some physicochemical properties and the amino acid composition of tomato seed protein has been studied. Under laboratory conditions 11% of protein on the weight of the initial seeds was obtained.

Interest in the problem of obtaining protein substances from the seeds of oil plants and the meals obtained from them is due to the search for methods of quantitatively increasing protein resources and eliminating the qualitative shortage of proteins in food products.

One of the promising sources of protein is represented by tomato seeds. Tomatoes amount to 1/5 of all vegetable crops in the USSR and occupy first place in the variety of vegetable preserves, the wastes from the production of which amount to 95 thousand tonnes per year [1]. The seeds contain not only a high-quality oil but also 25-35% of nitrogenous substances [2-4]. We have investigated the process of isolating protein from the defatted seeds (meal) and have studied some of its physicochemical properties. For the experiments we used seeds of tomatoes of the 1978 harvest collected in the Dzhambul province, KazSSR. The ground tomato seeds were defatted with gasoline. The oil was extracted by the steeping method at room temperature. The yield of oil from the ground seeds was 25% of the initial weight of the seeds. The solvent was eliminated from the defatted seeds (meal) in the air, and the air-dry meal was used for the experiments. Aqueous solutions of NaCl and NH_4Cl and weak solutions of NaOH were used as extractants for isolating the protein from the tomato meal. The protein was extracted by steeping at room temperature for 30 min with a 10-fold amount of extractant, which was then separated from the insoluble residue by vacuum filtration. The extract was then treated with a 5% solution of HCl and the precipitated protein was separated off by centrifuging. The crude protein was dehydrated with acetone.

The experiments showed that solutions of salts (with concentrations of 5-10%) extracted an insignificant proportion of the protein and the yield in the best case amounted to 7%, while when a 0.2% solution of NaOH was used the yield was a maximum (11.2%).

To study the influence of the time of extraction on the yield of protein, 100-g portions of meal were extracted with 0.2% NaOH solution (1000 ml), with a variation in the time of steeping. The bulk of the soluble fraction of protein passed into the extract in 30 min.

In an investigation of the influence of the temperature on the protein extraction process, 100-g portions of meal were extracted under identical conditions with a variation in the temperature of extraction. The protein extract was separated from the insoluble part of the meal and was cooled to room temperature, and the protein was precipitated. With a rise in the temperature of extraction to 60°C, the yield of protein rose to 12.5%, but at the same time it became darker in color.

To determine the isoelectric point of the protein the extracts obtained from 500 g of meal were separated into five equal parts, and the protein was precipitated by the addition of a 5% solution of HCl to predetermined values of the pH of the solution. The isoelectric point of the alkali-soluble fraction of the protein of tomato seeds is pH 5.5.

To study some properties of the protein we used samples obtained by extracting the meal under the optimum conditions found. Below we give some indices of the protein and its amino acid composition:

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Index of the Protein	Amount, % on Weight of the Protein
Total protein (N_{tot} × 6.25)	100.4
Soluble protein	99.57
Total ash	3.58
Cellulose	0.67

Amino Acid Composition of the Protein	Amount, %
Aspartic acid	11.0
Threonine	7.2
Serine	5.75
Glutamic acid	24.7
Proline	7.3
Glycine	6.35
Alanine	6.0
Cysteine	—
Valine	7.9
Methionine	1.95
Isoleucine	4.75
Leucine	4.75
Tyrosine	2.55
Phenylalanine	4.28
Histidine	1.9
Lysine	3.7
Arginine	5.55

In its total amino acid content, this protein can be assigned to the moderately balanced plant isolates.

EXPERIMENTAL

Defatting of the Seeds. A 30-kg batch of tomato seeds was passed through a roller mill (gap about 0.5 mm) and was charged into a 100-liter extractor. Defatting was carried out with gasoline (ratio to the raw material 1:2.5). The yield of oil from five extracts (50 liters each) was 7.5 kg.

Extraction of the Protein. The protein was extracted from 22.5 kg of defatted meal in a reactor (V = 0.63 m^3) with a working stirrer by 250 liters of 0.2% NaOH solution at room temperature for 30 min. The suspension was fed to a vacuum filter. Filtration was carried out by using "Bel'ting" filtration fabric. This gave 220 liters of decolorized extract.

Precipitation of the Protein. The clarified extract was fed to a precipitator (tank with a stirrer, V = 300 liters) and, with the stirrer working (60 rpm), 5% HCl solution was added in a thin stream until the protein had been precipitated completely (pH 5.5). The protein suspension was separated from the serum in a SGO-100 centrifuge.

Drying of the Protein. The protein precipitate (paste) in an amount of 12 kg was mixed with 15 liters of acetone in a glass apparatus with a stirrer. The protein was separated by filtration, and the operation was repeated three times. The dehydrated protein was ground and dried in the air. The yield of protein was 3.2 kg, i.e., 11% of the weight of the dry seeds.

Determination of the Composition of the Protein. The total nitrogen was determined by an accelerated Kjeldahl method [5] with subsequent recalculation to the crude protein content. The amount of soluble protein was determined in accordance with GOST 13979.3-68 and the total ash according to GOST 13879.6-69. The amount of cellulose was found by the method of Kürschner and Hanak [6]. The amino acid composition of the protein was determined on a LKB-4101 amino acid analyzer using a standard method [7] for hydrolysis.

SUMMARY

It has been established that when defatted tomato seeds are extracted with a 0.2% solution of NaOH and the protein is precipitated by acidification to pH 5.5 the yield of protein amounts to 11% on the weight of the initial raw material.

In terms of quality, the tomato protein belongs to the moderately balanced plant isolates.

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IDENTIFICATION AND FRACTIONATION OF THE TOTAL HISTONE OF THE COTTON PLANT

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In order to isolate individual fractions of the histones of the cotton plant of variety 108-F and to investigate them further, the total histone has been chromatographed on Acrylex P-60 and Bio-Gel P-30. It has been shown that the most complete fractionation is achieved on a Acrylex P-60. By electrophoresis of the fractions in 15% PAAG in the presence of 6.25 M urea and on the basis of amino acid compositions, the following order of elution of the histones has been established: H1, H2B + H2A, H3, and H4. In a comparison of the amino acid composition of the H3 and H4 histones of the cotton plant with histones enriched with arginine from other species, a somewhat higher lysine : arginine ratio in these fractions has been found. On comparing the electrophoretic mobilities of the histone fractions from the cotton plant with the histones of calf thymus, some difference is observed which is apparently connected with a difference in their molecular weights.

Histones, basic proteins of the cell nucleus, are assigned a leading role in the structural organization and functioning of the chromosomes, which is responsible for the great attention devoted to their investigation.

At the present time, a considerable amount of material has been accumulated on histones of animal origin [1-4]. Information relating to plant histones is less detailed, although interest in their isolation and study is increasing.

The majority of methods for isolating histones do not always enable homogeneous fractions to be obtained because of cross-contamination [5-7].

We have previously [8] fractionated cotton-plant histones by a modification of Johns' method. This method enabled us to obtain the individual histones H1, H2B, and H4. Further purification was required for the other fractions. However, the chromatography of several plant histones on Bio-Gels enables individual fractions to be isolated without additional purification [9-12].

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