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GENERAL CHARACTERISTICS OF THE PROTEINS OF TOMATO SEED FLOUR AND TOMATO SKIN FLOUR

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The chemical compositions of tomato seeds and skin have been investigated. The total protein contents (TCA-precipitable) of the seeds and the skin amounted to 30.5% and 16.5%, respectively. The pH dependence of the extraction of protein from the seeds and skin in the presence and absence of oil has been established in the pH interval from 2.0 to 10.0. The protein spectra of the seeds and skin have been determined.

The wastes from tomato processing can be used as a raw material for the production of food protein and oil. The seeds and skin are rich sources of minerals [1]. A number of publications have demonstrated the possibility of using dry tomato pressing residues as a fodder for animals and poultry [2-4]. It has been shown that this nontraditional source of food protein contains no antifeedant substances [3, 5, 6].

In the Republic of Uzbekistan, tomatoes occupy one of the leading positions in the variety of vegetable crops, and the residues remaining after the processing of the tomatoes are practically unutilized. As has been shown previously [4], in the processing of tomato products 5-15% of wastes are formed. The wastes contain 40-48% of seeds. There is no information in the literature [1-6] on the fractional composition of the proteins of the seeds and of the skin of the tomato or the structures of the main protein components and their functional properties.

We have investigated the wastes from local tomato varieties. The chemical compositions of the tomato seeds and skin are given in Table 1.

The amounts of protein extractable from the seeds and skin are shown in Table 2. As can be seen from Table 2, the total yields of extractable protein from the flour of tomato seeds and that of tomato skin were 30.5% and 16.5%, or 83.6% and 76.0% of their amounts in the flour, respectively. The yields of protein from extracts on isoelectric precipitation amounted to 12.8% and 9.3%, respectively, for the seeds and the skin, which represent 42.0% and 56.5% of the extractable protein.

We have studied the extractability of protein from the seed flour and the skin flour under various conditions (pH of the medium and the presence or absence of oil, Fig. 1). In the presence of oil (undefatted flour, Fig. 1, curves *2* and *4*) more protein passed into the solution, this being particularly characteristic for the seeds.

It must be mentioned that the extraction of protein began only at pH > 7. In the pH range of 8-9 the yield of protein rose by 4.4-9.0%. In the case of the skin, the yield of protein in the presence of oil likewise rose by 1.5%, and on curve *4* in the pH 7-9 region a small plateau was observed, after which, with a rise in the pH, the yield continued to increase and reached 10% at pH 12. The pH maxima for the precipitation of the seed and skin proteins were determined by turbidimetric titration; they were 4.7-5.2 and 2.8-4.8, respectively.

The separation of the seed and skin proteins into fractions was achieved by a scheme adopted in biochemical practice [8] by extracting the proteins successively with water and with salt, aqueous–alcoholic, and alkaline solutions. Table 3 gives the yields of the fractions obtained from the seeds and the skin. As the experiment showed, the tomato seeds contained a high level of the globulin fraction (14.0%), including not only salt-soluble proteins but also proteins extractable by water and precipitating on dialysis (readily soluble globulins). Then, in order of amounts present, followed a glutelin fraction (4.0%), albumins (2.6%), and prolamines (1.2%).

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TABLE 1. Chemical Compositions of Tomato Seeds and Skin, g/100 g of Flour

Sample	Moisture	Ash	Cellulose	Protein (N × 5.7)	Lipids
Seeds	8.1—8.2	4.3—4.9	24.2—26.8	36.5	20.4—20.9
Skin	6.7—6.8	3.7—3.9	49.0—50.1	21.7	3.3—5.8

TABLE 2. Yields of Protein from Tomato Seed Flour and Tomato Skin Flour

Sample	Yields of protein (%) on repeated extraction				Total yield of protein, %	Precipitation at the IEP, %
	1	2	3	4		
Seeds	21.5	5.6	2.1	1.3	30.5	12.8
Skin	10.3	4.1	2.1	—	16.5	9.3

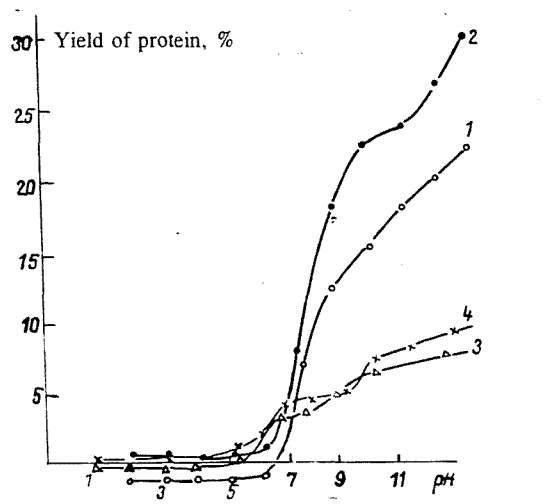

Fig. 1. Extractability of protein from tomato seed flour and tomato skin flour: 1) defatted seed flour; 2) inital seed flour; 3) defatted skin flour; 4) initial skin flour.

In the skin, the largest fraction found was that of the readily-soluble globulins (3.8%), while a 10% NaCl solution extracted in total (supernatant solution and precipitate) only 0.8% of proteins. The glutelin content was somewhat lower than in the seeds (3.3%). The yield of the albumin fraction was only 1.5%, and that of prolines 0.8%. The yields of the protein fractions were determined gravimetrically in relation to the initial flour and from the total nitrogen content. The total yields of the protein fractions corresponded to the amount of protein on one-stage extraction.

An electrophoretic investigation of the protein fractions under various conditions showed that they were inhomogeneous and consisted of complex mixtures. Electrophoresis was conducted in blocks of 7% and 15% PAAG in the presence and in the absence of NaDDS. Figure 2a, b, gives the results of the investigation of the seed proteins in 7% PAAG.

As can be seen from Fig. 2a, the albumin fraction (1) was separated in PAAG into five main bands, while the patterns of the globulin fraction and of the readily soluble globulins coincided. In the presence of 0.02% NaDDS (Fig. 2b) the albumin fraction (1) was represented by a single component with R_f 0.56. Fractions 2, 3, and 4 were close in composition. The globulin fraction (4) consisted of three main, in terms of intensity, bands.

Figure 3a, b, gives the results of an investigation of the skin proteins in 7% and 15% PAAG in buffer and in 0.02% NaDDS, respectively.

The albumin fraction (4) was represented by a broad band. Fraction (5) of readily soluble globulins (the main fraction of mixture quantitatively) apparently consisted of a multiplicity of bands since there was no clear separation. Fraction (6)

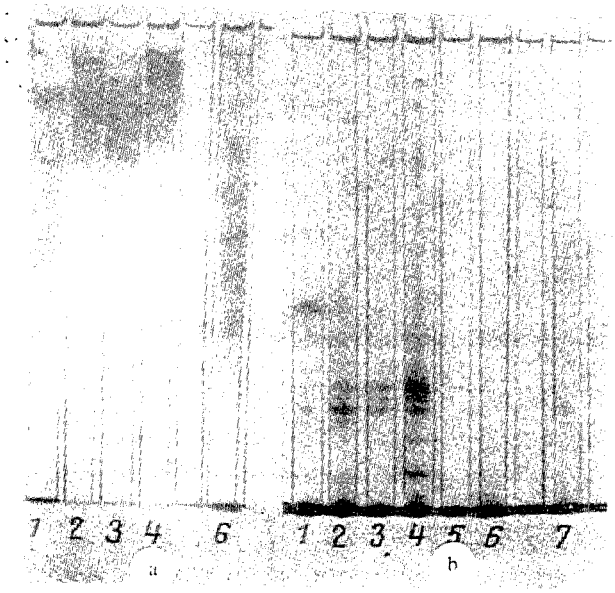

Fig. 2

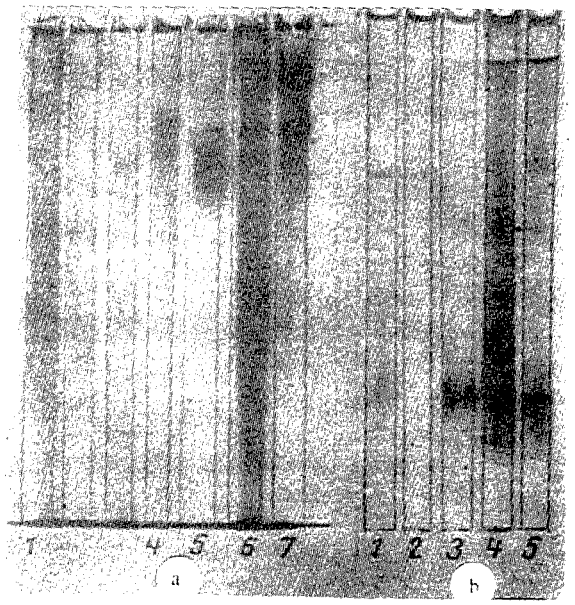

Fig. 3

Fig. 2. Electrophoregrams of tomato seed proteins in 7% PAAG in buffer (a) and in 7% PAAG in the presence of 0.02% NaDDS (b): 1) albumins (supernat. liquid); 2) albumins (precipitate); 3) globulins (supernat. liquid); 4) globulins (precipitate); 5) prolamines; 6) glutelins (supernat. liquid); glutelins (supernat. liquid); 7) glutelins (precipitate).

Fig. 3. Electrophoregrams of tomato skin proteins in 7% PAAG in buffer (a) and in 15% PAAG, 0.02% Na b): a) *1*) prolamines; *4*) albumins, supernat. liquid; *5*) albumins (precipitate); *6*) globulins (supernat. liquid); *7*) glutelins (supernat. liquid); b) *1*) albumins (supernat. liquid); *2*) albumins (precipitate); *3*) globulins (supernat. liquid); *4*) glutelins (supernat. liquid); *5*) globulins (precipitate).

consisted of a multiplicity of components, which were not separated under these conditions. In the presence of 0.02% NaDDS in 15% PAAG (Fig. 3b) the globulin fractions (3) and (5) were represented by a single component with R_f 0.74.

Thus, according to electrophoresis, quantitatively the main fractions in the seeds (globulins) and in the skin (readily soluble globulins) of the tomatoes were inhomogeneous and consisted of several components.

Proteins of plant origin usually contain deficient amounts of Lys, Met, Trp, and Thr. The protein isolated from tomato seeds and skin contained a large amount of Lys [9, 10] which, calculated to the ideal protein recommended by the FAO and WHO, amounted to 135.7% and 80.9%, respectively.

Table 4 gives the amino acid compositions of the protein fractions of the tomato seeds and skin. As can be seen from the amino acid compositions of the isolates and of the fractions obtained both from the seeds and from the skin, they contained large amounts of lysine, dicarboxylic acids, proline, valine, and leucine. A high level of lysine is characteristic for the albumin and the globulin fractions from the tomato skins (7.4 and 5.6%, respectively), although the isolate itself contained less lysine (3.4%), apparently through a negative contribution of the glutelin and prolamine fractions.

For the protein fractions of the seeds the pattern was somewhat different, namely: the level of lysine in the protein isolate was higher (5.7%) than in the globulin and albumin fractions (4.9 and 3.5%). The amount of proline in the protein fractions of the seeds was due to the albumin fraction (10.2%), and in the skin to the globulin fraction, although the yield of globulin fraction was smaller than that of the albumin fraction. A high glutamic acid content was characteristic both for the isolates (seeds and skin) and for the fractions composing them.

Thus, the chemical compositions of the proteins of the seeds and skin of local varieties of tomato have been investigated. The total levels of protein in the seeds and skin have been determined (30.5 and 16.5%, respectively). The pH dependence of the extraction of protein from the seeds and skin in the pH 2-10 interval has been determined.

TABLE 3. Yields of Protein Fractions from the the Seeds and Skins of Tomatoes

Sample	Albumins		Globulins		Prolamines	Glutelins	
	supernat. soln.	ppte	ppte	supernat. soln.		supernat. soln.	ppte
Seeds	2.6	5.6	6.1	2.3	1.2	2.0	2.0
Skin	1.5	3.8	0.2	0.6	0.8	2.1	1.2

TABLE 4. Amino Acid Compositions* of the Proteins of Tomato Seeds and Skin, mole-%

Amino acid	Isolate		Albumins		Globulins	
	seeds	skin	seeds	skin	seeds	skin
Asp	8.7	10.6	7.9	10.4	5.5	10.2
Thr	2.9	4.3	2.7	5.6	1.8	3.4
Ser	4.1	5.3	5.1	5.8	3.7	5.8
Glu	19.7	20.1	21.4	17.5	19.9	20.6
Pro	9.7	9.3	10.2	7.0	7.5	9.0
Gly	3.6	3.0	4.1	18.0	4.4	5.3
Ala	5.1	10.4	5.3	—	5.1	8.7
Val	12.0	7.1	11.9	6.6	13.8	6.4
Met	1.5	1.1	3.4	1.8	1.4	3.1
Ileu	8.6	5.2	8.9	4.0	10.3	4.4
Leu	9.1	8.5	8.7	7.0	10.2	7.1
Tyr	2.0	3.8	1.6	3.1	1.9	3.3
Phe	4.8	6.0	3.3	4.2	7.6	3.4
His	2.1	—	1.8	2.4	1.9	3.8
Lys	5.7	3.4	3.5	7.4	4.9	5.6

*Excluding Arg and Trp, and Cys.

EXPERIMENTAL

The tomato processing wastes were obtained from the Tashkent cannery. The pressing residues (wastes) were dried in the air, and the seeds were separated from the skin manually. Grinding yielded light brown (from the seeds) and red-brown (from the skin) flours with a faint tomato odor. After the flours had been defatted with petroleum ether, they were dried, and the proteins were extracted under various conditions.

The moisture contents of the samples were determined in accordance with GOST [State Standard] 975–88 [11], and the amount of crude cellulose in accordance with GOST 1397910–69 [12].

Free lipids were determined by extraction with petroleum ether. A weighed sample of seeds (skin) that had been ground in a mill was covered with petroleum ether in a ratio of 1:20 (50) wt./vol. and the mixture was stirred with a magnetic stirrer for 1 h. Extraction was performed 5 times. The extracts were collected in a previously weighed flask, the solvent was distilled off in a rotary evaporator, and the lipid fraction was dried to constant weight. The yield was calculated on the initial flour.

The yields of protein (isolate) from the seed flour and the skin flour were determined at room temperature after three extractions with 0.2 N NaOH solution for 30 min at a ratio of flour to extractant of 1:20. The proteins were precipitated from the extract with 10% TCA, and the amounts of protein in the precipitates were determined by the micro biuret method [7].

Determination of the Yields of Protein at Various pH Values. With constant stirring by a magnetic stirrer, 1 N HCl or NaOH was added to a suspension of 1 g of seed (skin) flour in distilled water (flour:extractant ratio = 1:20) to bring the pH to a given value in the range from 2 to 12. Then the suspension was stirred for 30 min at the given pH, after which the extract was separated by centrifugation and the proteins were precipitated with 10% TCA. The amounts of protein in the precipitates were determined by the micro biuret method.

The proteins were separated into fractions as described in [8].

The electrophoretic investigation of the total proteins and their fractions was conducted in 7.5 and 15% PAAGs in a basic buffer (pH 8.9) and in the presence of 0.02% NaDDS in vertical plates. An AVGÉ-2 apparatus for vertical electrophoresis was used. Electrophoresis was conducted in Tris-glycine buffer, pH 8.3, for 2 h at I = 250 V and U = 33 mA

per plate. The plates were fixed in 10% TCA solution for 20 min and were stained for 1 h in a 0.2% solution of Coomassie Blue R-250. The excess of dye was washed out with 7% acetic acid solution.

The amino acid compositions of the protein factions were determined on a Czech mark T339 amino acid analyzer after hydrolysis of the samples with 5.7 N HCl at 110°C for 24 h.

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