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# PREPARATION AND PARTIAL CHARACTERIZATION OF SUNFLOWER (HELIANTHUS ANNUUS L.) SEED FLOUR

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To evaluate the potential of sunflower seed protein (SSF) as human food, grits were milled from dehulled sunflower seed and defatted, then milled into flour with a roller mill, and a hammer mill. Heat-treated, sunflower seed flour (SSF) was prepared by autoclaving for 30 min. at 121 C and "declumping" in a Laboratory Stein mill.

Fatty acid analysis indicated total oleic and linoleic acid of 89.61%, 79.70%, and 78.76%, respectively, in sunflower seed oil, flour and protein isolate.

Distribution of isolated flour proteins indicated that the major portion was globulin. The two major amino acids were glutamic and aspartic acids. Water-soluble protein contained the highest quality of lysine; salt soluble protein and sodium proteinate were low in lysine.

## Introduction

Sunflowers for agricultural purposes are on the increase annually in the United States. Oil from sunflowers is considered superior to soybean oil in Europe and the USSR but very little is known about the production of sunflower seed flour (SSF), the protein and of the sunflower seed oil (SSO) cake. We need improved ability to process protein from sunflowers and other raw agricultural materials into food and feed suitable for the demands of society. The major challenge is to better utilize available protein materials. The purpose of this study was the preparation and partial characterization of sunflower (Helianthus annuus L.) seed flour (SSF).

## Materials and Methods

### Preparation of Sunflower Seed Flour

Dehulled SS (Helianthus annuus L.) of large seed type were obtained from a commercial source. The seeds were cleaned, and coarse ground on the experimental roller mill of the Department of Grain Science and Industry, Kansas State University. The ground seeds were defatted by extraction with petroleum ether (Skellysolve F) with a Soxhlet extractor for 48 hr. The desolventized products were hammer milled to SSF according to procedures shown in Figure 1. The heated SSF was prepared by simply autoclafing the SSF in a Mason jar for 30 min at 121$^{o}$C (1), then regrind to flour by Laboratory Stein Mill (The Fred Stein Laboratories, Atchinson, Kansas). Both heated and unheated SSF were stored in airtight cans at 4$^{o}$C.

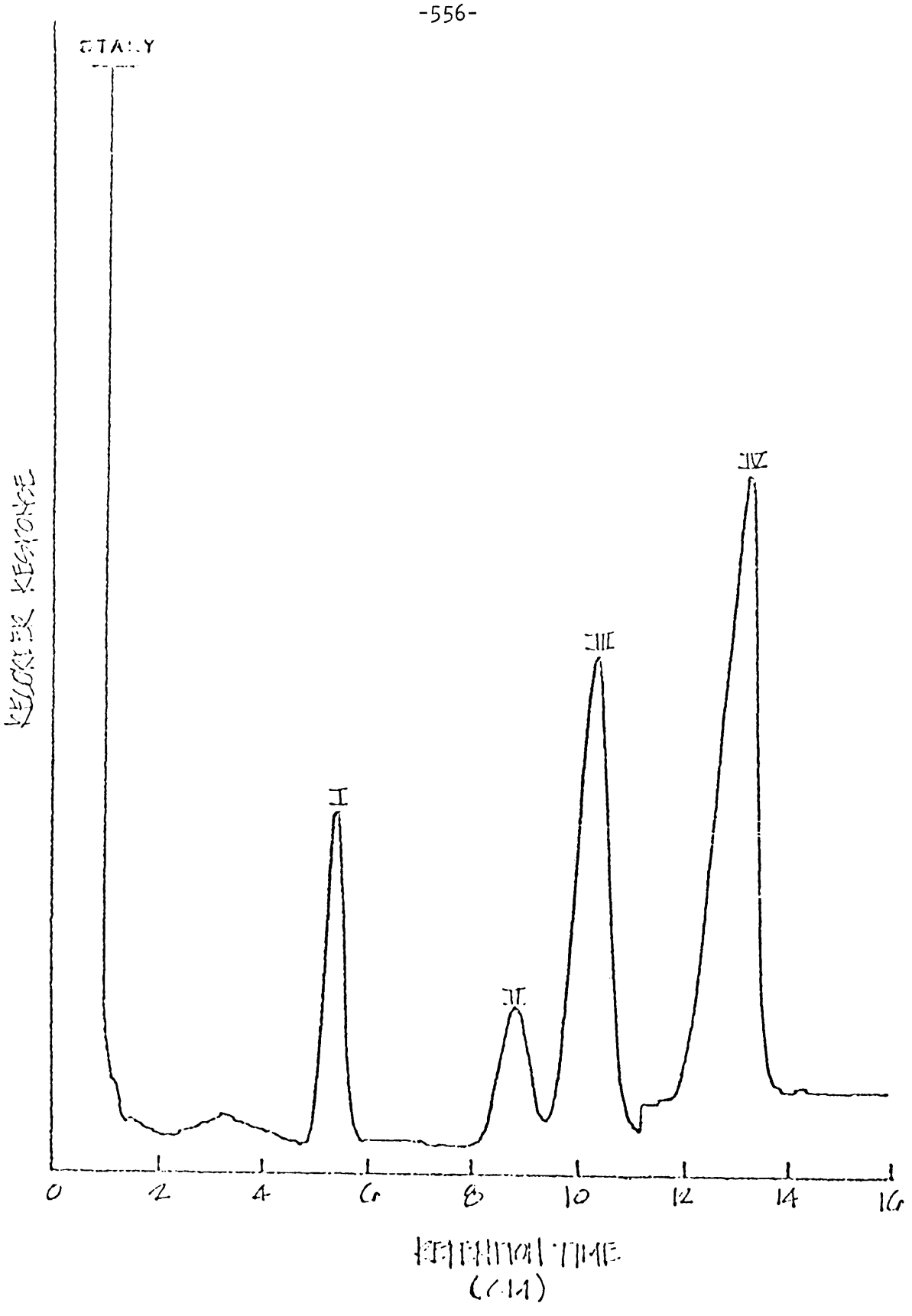


FIGURE 3. Gas-liquid chromatogram of fatty acid methyl esters from sunflower seed flour (I. Palmitate; II, Stearate; III. Oleate; IV, Linoleate).

## Preparation of Isolated Proteins From Sunflower Seed Flour (SSF)

The procedure of Sosulski and Bakal (2) was used to prepare water and salt soluble protein from SSF. A colorless protein isolate from SSF could be produced with the procedure of Gheyasuddin et al (3). Sunflower seed sodium proteinate was prepared using the flowsheet developed by Wolf (4) with some modification as shown in Figure 2.

## Proximate Analyses of SSF and Its Isolated Protein

Moisture analysis was carried out according to American Association of Cereal Chemists (AACC) Laboratory Method 44-40 (9). Ash analysis was performed using AACC Laboratory Method 08-02 and crude fiber analysis by AACC Laboratory Method 32-15 (9). American Official Agricultural Chemists (AOAC) Method 22.033 (5) was followed for crude fat determination and crude protein was analyzed according to AOAC Micro-Kjeldahl Methods 38.012-38.014 (5) with the factor 5.30 being used to convert percent nitrogen to percent protein.

## Fatty Acid

The fatty acid composition of SSF, SSP isolate, SSO was determined by extracting crude lipid from these samples according to the procedure of Lima et al (10) and Zanden (11) by preparing and recovering the fatty acid methyl ester. These methyl esters were analyzed using a Barber-Coleman gas-liquid chromotograph equipped with hydrogen flame ionization detector using a glass column (6 ft. x 1/8 in.) packed with 7.5% diethyleneglycol succinate (DEGS) on chromosorb G (80-100 mesh) with nitrogen as the carrier gas at a flow rate of 40 ml/min; oven temperature 190°C; injection port temperature, 250°C. Standards of fatty acid methyl ester were also analyzed.

Retention times for all significant peaks were measured for all chromatograms. The area under each peak with the concentration of each fatty acid ester was measured and reported as percent of total. Identification of the peaks was accomplished according to the chart and method reported by Hofstetter et al (12).

Amino acid composition of SSF and its derived products was determined by the method of Speckman et al (6) using a Beckman Model 120B amino acid analyzer.

The study on the distribution of proteins from SSF accoridng to the solubility classification was conducted by a modified Lund and Sandstrom procedure (13).

## Results and Discussion

The hammer milled unheated sunflower seed flour (SSF) obtained had a nearly white appearance, but the heated SSF was light brown in color. This was in agreement with the work of Rooney et al (14).

The composition of unheated and heated SSF is shown in Table 1. The heated SSF was lower in protein content when compared to that of unheated SSF. There was no significant difference in crude fiber and ash content between those two flours.

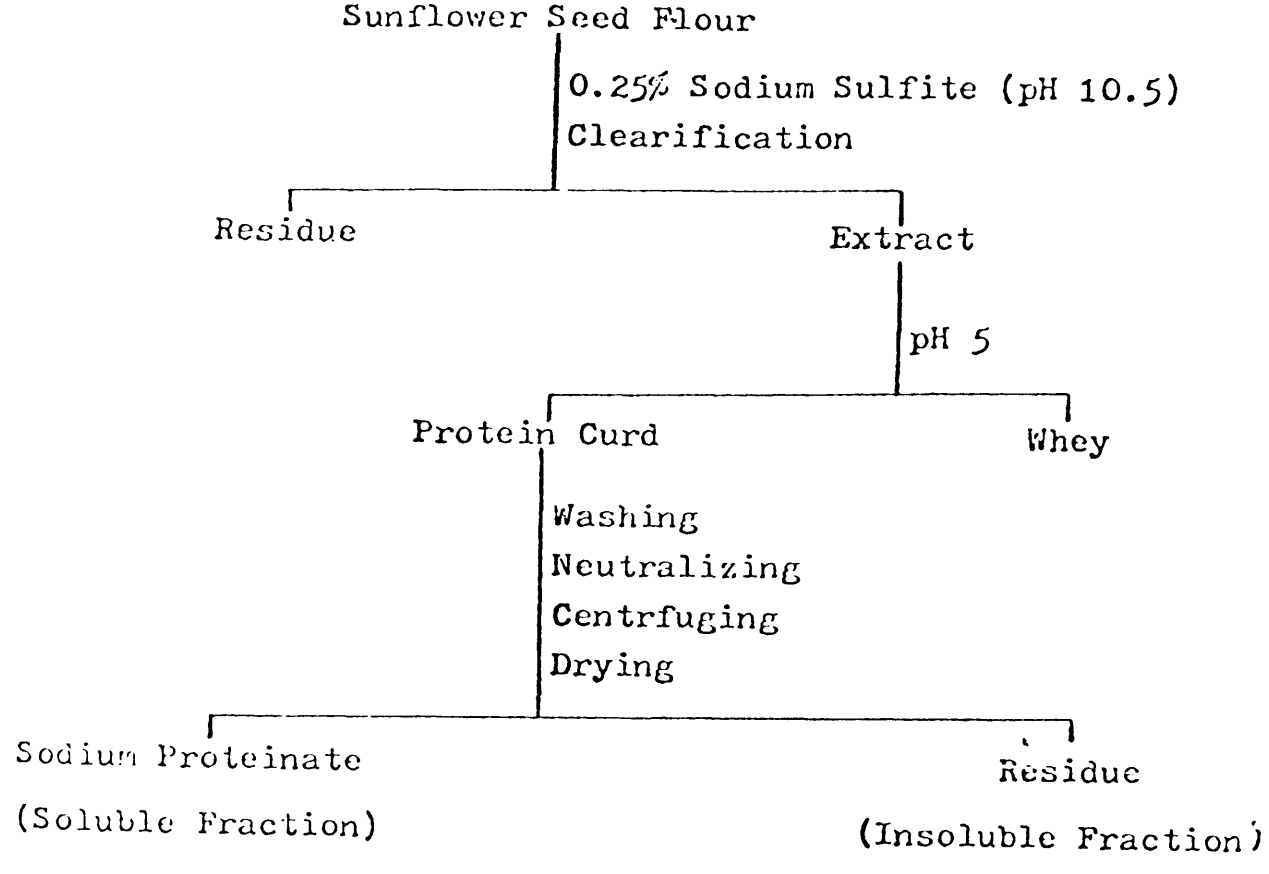


FIGURE 2. Flowsheet for isolation of sodium proteinate from sunflower seed flour.

TABLE 1. Chemical composition of heated and unheated sunflower seed flour.

| Description | Unheated Sunflower Seed Flour | Heated Sunflower Seed Flour |
|---|---|---|
| Moisture, % | 6.80 | 8.57 |
| Protein, % | 45.50 | 42.37 |
| Crude Fat, % | 4.40 | 9.20 |
| Crude Fiber, % | 2.00 | 1.90 |
| Ash, % | 8.10 | 7.70 |

The results concerning moisture, protein content, as well as the yield of the isolated proteins from SSF are shown in Table 2. Generally speaking, those products had very high protein content. Sunflower seed protein (SSP) isolate contained the highest level of protein, but the yield was quite low. All products were tan in color except for sodium proteinate and protein isolate, which were brown to nearly white in appearance. Color of SSF and protein isolate as a function of pH is shown in Table 3. Sodium proteinate had the advantage of being water dispersible, while the protein isolate (isoelectric precipitation) was insoluble in water. Solubility is an important factor governing food applications of protein products, and soluble ingredients are always easier to formulate in food.

TABLE 2. Moisture, protein content, and yield of isolated proteins from sunflower seed flour.

| Products | % Moisture | % Proteins | % Yield |
|---|---|---|---|
| Water soluble protein | 6.80 | 66.07 | 10.07 |
| Residue from water soluble protein preparation | 7.47 | 59.97 | - |
| Salt soluble protein | 6.33 | 79.62 | 23.00 |
| Sodium proteinate | 2.20 | 84.38 | 18.27 |
| Precipitation fraction from sodium proteinate preparation | 3.30 | 84.13 | 13.03 |
| Protein isolate | 1.40 | 86.26 | 31.26 |

Data obtained in this study (Table 4 and Figure 3) indicated that 89.61% of the total fatty acids in SSO was oleic (15.45%), linoleic acid (74.16%). This result was in agreement with the work of Earle et al (7) and Trotter et al (8). Similarly, 79.70% and 78.76% of total fatty acids being oleic and linoleic acids were found in SSF and SSP isolate respectively. An interesting aspect concerning fatty acid composition of SSO, SSF, and SSP isolate is that the high amount of essential fatty acid (linoleic acid) make them desirable for food products in terms of fatty acid composition and content.

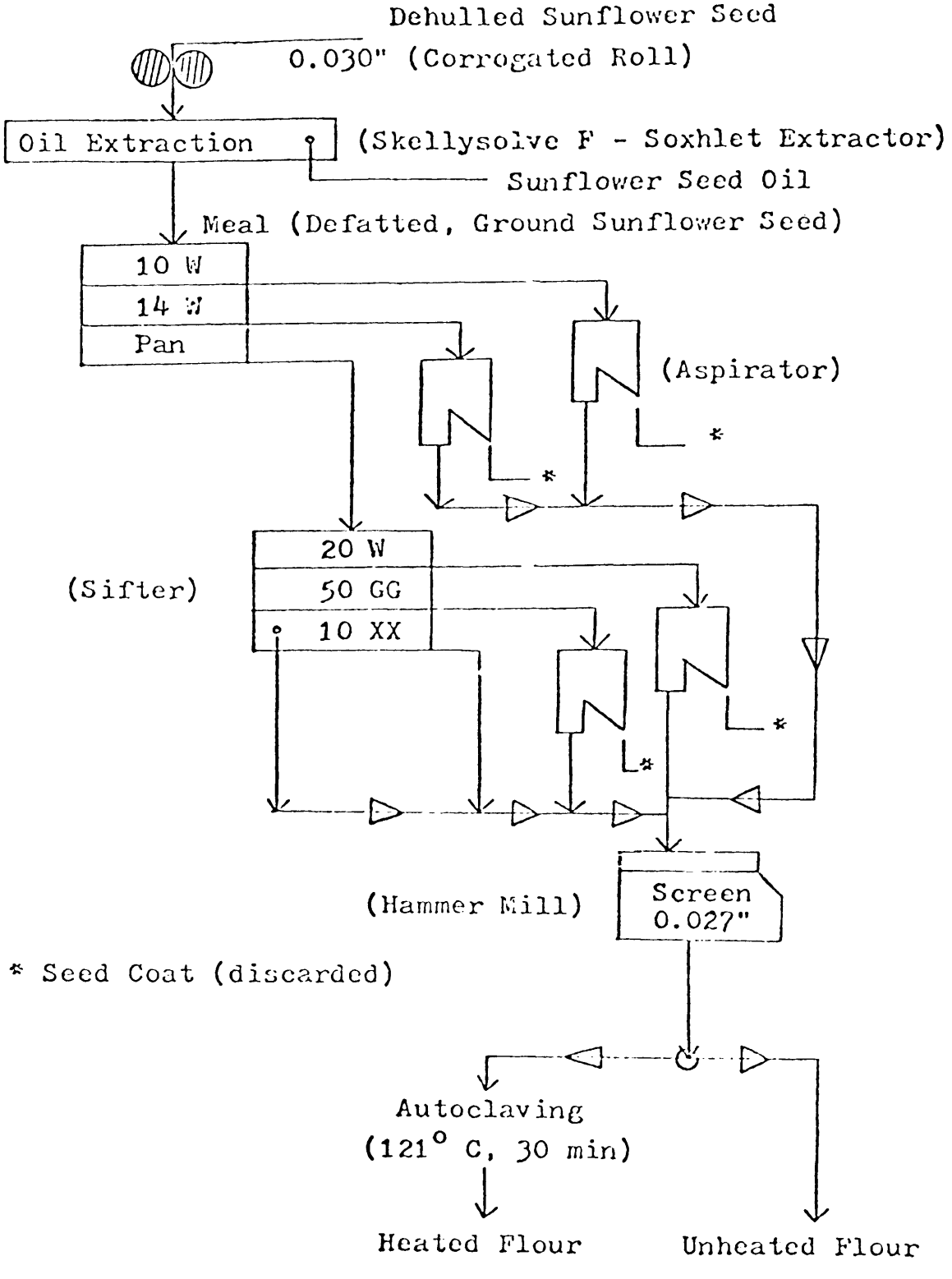


FIGURE 1. Scheme used to prepare unheated and heated sunflower seed flour.

TABLE 3. Color of sunflower seed flour and protein isolate as a function of pH.

| pH | Color | |
|---|---|---|
| | Isolate | Flour |
| 2 | Cream white | Cream white |
| 3 | Cream white | Cream white |
| 4 | Cream white | Cream white |
| 5 | Cream white | Cream white |
| 6 | Cream white | Cream white |
| 7 | Cream white | Yellowish grey |
| 8 | Grey | Yellowish grey |
| 9 | Yellowish grey | Green |
| 10 | Light brown | Green |
| 10.5 | Dark brown | Green |

TABLE 4. Fatty acid compositions of sunflower seed oil, flour and protein isolate.

| Methyl Esters | Oil | Flour | Protein Isolate |
|---|---|---|---|
| I. Palmitate (16:1) | 6.62 | 12.04 | 12.52 |
| II. Stestrate (18:0) | 3.77 | 8.26 | 8.72 |
| III. Oleate (18:1) | 15.45 | 31.14 | 31.94 |
| IV. Linoleate (18:2) | 74.16 | 48.56 | 46.82 |

Values determined by comparison of the area under each peak to the sum of the areas under all peaks.

## Amino Acid Analysis

The amino acid analysis of SSF and isolated proteins is presented in Table 5. Glutamic and aspartic acids were the two major amino acids found in SSF, and they constitute about 35% of total amino acids of SSF protein; whereas they also constituted about 29 to 46% of the total amino acids in those other sunflower seed isolated products.

Water soluble protein from SSF had the greatest quantity of lysine while the salt soluble protein and sodium proteinate were lowest in lysine.

Results indicate that both residues from sodium proteinate preparation and protein isolate preparation from SSF contained more lysine than those of sodium proteinate and protein isolate themselves.

Autoclaving, as expected, caused some loss of lysine of SSF, and this is consistent with the findings of Rooney et al (14) and Liener (15). It also held true for arginine, histidine, and valine.

TABLE 5. Amino acid composition of sunflower seed flours (SSF), protein isolates.

| | Unheated SSF | Heated SSF | Protein isolate | Residue from prot. isolate prepara-tion | Water soluble protein | Residue from Water Soluble Protein | Salt Soluble Protein (5% NACL) | Na Protein-ate | Residue from Na-Prot. Prep. |
|---|---|---|---|---|---|---|---|---|---|
| Moisture | 6.80 | 8.57 | 1.40 | 5.76 | 6.80 | 7.47 | 6.30 | 2.20 | 3.30 |
| Protein[1] | 49.77 | 45.46 | 86.26 | 31.78 | 66.07 | 59.97 | 79.62 | 84.38 | 84.13 |
| Amino acids lysine | 3.394 | 3.3134 | 2.730 | 3.998 | 6.790 | 2.218 | 1.344 | 1.511 | 3.110 |
| Histidine | 2.408 | 1.983 | 3.461 | 2.677 | 2.176 | 1.925 | 1.031 | 1.558 | 1.695 |
| Arginine | 8.681 | 8.230 | 8.469 | 8.847 | 7.788 | 7.460 | 5.034 | 5.062 | 9.141 |
| Threonine | 3.443 | 3.944 | 3.658 | 3.760 | 3.181 | 3.258 | 3.291 | 3.639 | 6.417 |
| Glutamic acid | 25.480 | 24.935 | 24.555 | 27.788 | 21.696 | 31.807 | 30.263 | 27.796 | 24.592 |
| Valine | 4.982 | 4.765 | 5.205 | 4.846 | 2.973 | 4.475 | 4.757 | 5.047 | 4.898 |
| Methionine | 0.932 | 1.373 | 1.790 | 1.162 | 3.492 | 1.275 | 1.996 | 1.529 | 1.361 |
| Isolucine | 4.153 | 3.805 | 4.158 | 3.533 | 3.931 | 3.676 | 4.432 | 4.320 | 4.216 |
| Phenylalanine | 4.702 | 5.551 | 4 828 | 4.207 | 4.007 | 4.539 | 4.905 | 5.826 | 5.040 |

[1] % Nx5.30, moisture-free basis.

SSP contain adequate amounts of other essential amino acids although they are somewhat low in lysine and isoleucine. Therefore, the amino acid composition of SSP suggests that they may be used as a valuable ingredient in high-quality foods.

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