

Food Chemistry 82 (2003) 361–366

Food Chemistry

[www.elsevier.com/locate/foodchem](http://www.elsevier.com/locate/foodchem/a4.3d)

Solubility and functional properties of sesame seed proteins as influenced by pH and/or salt concentration

E.K. Khalid^a, E.E. Babiker^{b,*}, A.H. EL Tinay^b

^aNational Research Institute, Khartoum, Sudan

^bUniversity of Khartoum, Faculty of Agriculture, Department of Food Science and Technology, Shambat, Sudan

Received 5 August 2002; received in revised form 18 November 2002; accepted 18 November 2002

Abstract

The protein content, solubility and functional properties of a total protein isolate prepared from sesame seeds (Kenana 1 cultivar) as a function of pH and/or NaCl concentration were investigated. The protein content of the seed was found to be 47.70%. The minimum protein solubility was at pH 5 and the maximum was at pH 3. The emulsifying capacity, activity and emulsion stability as well as foaming capacity and foam stability were greatly affected by pH levels and salt concentrations. Lower values were observed at acidic pH and high salt concentration. The protein isolate was highly viscous and dispersable at pH 9 with water holding capacity of 2.10 ml H_2O/g protein, oil holding capacity of 1.50 ml oil/g protein and bulk density of 0.71 gm/ml. \odot 2003 Elsevier Science Ltd. All rights reserved.

Keywords: Sesame; Kenana; Protein isolate; Functional properties; Sudan

1. Introduction

Plant proteins play significant roles in human nutrition, particularly in developing countries where average protein intake is less than that required. Because of inadequate supplies of food proteins, there has been a constant search for unconventional legumes, as new protein sources, for use as both functional food ingredients and nutritional supplements ([Onweluzo, Obanu,](#page-4-0) [& Onuoha, 1994](#page-4-0)). Plant protein products are gaining increased interest as ingredients in food systems throughout many parts of the world; the final success of utilizing plant proteins as additives depends greatly upon the favourable characteristics that they impart to foods. Therefore, the interrelationships of protein quality and processing parameters, that affect the functional performance of protein products, is worthy of an extensive investigation ([Jane, Rivas, & John, 1981\)](#page-4-0). For plant proteins to be useful and successful in food application, they should ideally possess several desirable characteristics, referred to as functional properties, as well as providing essential amino acids ([Wang & Kinsella, 1976\)](#page-5-0).

These properties are intrinsic physicochemical characteristics, which affect the behaviour of proteins in food systems during processing, manufacturing, storage and preparation ([Kinsella, 1979\)](#page-4-0). Proteins have unique surface properties due to their large molecular size and their amphiphilic properties. However, the industrial applications of food proteins are limited, because proteins are generally unstable to heating, organic solvents and proteolytic attack [\(Sakamoto, Kumazawa, &](#page-5-0) [Motoki, 1994](#page-5-0)). Therefore, if proteins could be converted into stable forms, their applications would be greatly broadened. Attempts have been carried out to modify plants proteins to improve their physical functionality, i.e. gelation, viscosity, emulsification and foaming [\(Saka](#page-5-0)[moto et al., 1994\)](#page-5-0). Several molecular parameters, such as mass, conformation, flexibility, net charge and hydrophobicity, as well as interaction with other food components, have already been shown to play an important part, in both their emulsifying and foaming properties [\(Nakai &](#page-4-0) [Voutsinas, 1983](#page-4-0)). However, most chemical modifications are not applicable to the food industry. Therefore, in this study, we investigate the effects of pH and salt concentration on solubility and functional properties of sesame (Kenana 1) protein isolate and predict its compatibility, prior to modification, in different food systems.

Corresponding author. E-mail address: elfadilbabiker@hotmail.com (E.E. Babiker).

2. Materials and methods

2.1. Materials

Sesame cultivar (Kenana 1) was grown at the demonstration Farm, Faculty of Agriculture, Shambat, Sudan during the 1992–1993 seasons. The seeds were sundried, carefully cleaned and ground to a powder (0.4 mm screen). Groundnut and corn oils were obtained from Bittar Co. Ltd., Khartoum, Sudan. Unless otherwise stated, all reagents used in this study were of reagent grade.

2.2. Methods

2.2.1. Preparation of protein isolate

Sesame protein isolate from defatted flour was prepared according to the [Parakash and Nandi method](#page-5-0) [\(1978\)](#page-5-0).

2.2.2. Protein content

Nitrogen contents of defatted samples were determined by micro-Kjeldahl technique, following the method of the AOAC (1980). Protein content of each sample was calculated by multiplying the nitrogen content by a factor of 6.25.

2.2.3. Nitrogen solubility

Nitrogen solubilities of the proteins at 2% (w/v) were determined by the method of [Beuchat, Cherry, and](#page-4-0) [Quinn \(1975\)](#page-4-0) over a pH range from 2 to 10. The dispersions were stirred at different pHs at 24 $^{\circ}$ C for 45 min and then centrifuged at 3000 g for 30 min. Nitrogen content of the supernatants was determined following the method of the AOAC (1980). Nitrogen solubility was expressed as percent of the nitrogen content of the sample.

2.2.4. Water- and oil-holding capacity

The method of [Carcea Benecini \(1986\)](#page-4-0) was used with a slight modification. One gramme of protein samples was stirred in 10 ml of distilled water or corn oil and then centrifuged at 2200 g for 30 min. The volume of the supernatant was measured. The water-holding capacity was expressed as the number of g of water held by 1.0 g of protein sample. The oil-holding capacity was expressed as the number of g of oil held by 1.0 g of protein sample. Density of the oil was found to be 0.92 g/ml.

2.2.5. Apparent viscosity

Apparent viscosity of a 20% of a total protein isolate (w/v) , at different pH levels, was determined by the method of [Quinn and Beuchat \(1975\).](#page-5-0) The samples were heated at 25 or 70 \degree C and then cooled to room temperature. The apparent viscosity was determined at 24 \degree C with a Brookfield (Model RVT) viscometer equipped with a No. 1 spindle. Apparent viscosity in cetipoises (cps) was reported as the average of three readings.

2.2.6. Emulsion measurements

Emulsification capacity was determined according to the procedure of [Beuchat et al. \(1975\)](#page-4-0) and is expressed as millilitres of oil emulsified per g of protein. Emulsifying activity and emulsion stability were determined according to the method of [Pearce and Kinsella \(1978\)](#page-4-0).

2.2.7. Foam measurements

Foam capacity and stability at different pH levels or NaCl concentrations were determined according to the [Aruna and Prakash method \(1993\).](#page-4-0) One hundred millilitres of distilled water at different pH or NaCl concentrations were separately added to 3 g of defatted sesame protein isolate and the mixture homogenized at 300 rpm for 5 min in a Virtis homogenizer at 27 C and transferred to a measuring cylinder. The volume of foam at 30 s was calculated, and the volume increase is expressed as percent foam capacity. The foam stability was determined by measuring the decrease in volume of foam as a function of time up to a period of 90 min.

2.2.8. Bulk density

The bulk density was determined according to [Wang](#page-5-0) [and Kinsella \(1976\)](#page-5-0) using samples of 20 g and a 50 ml graduated cylinder. Bulk density was calculated as g/ml.

2.2.9. Dispersibility

The dispersibilities of sesame protein isolates at different pH levels was measured according to the method of [Karuna, Kulcarni, and Ingle \(1991\).](#page-4-0)

3. Results and discussion

3.1. Protein content and nitrogen solubility

The protein content of sesame cultivar (Kenana 1) was found to be 47.70%, with moisture content of 3.61%. [Fig. 1](#page-2-0) shows the variations in nitrogen solubility at different pH levels of a total protein isolate (TPI). The minimum nitrogen solubility of TPI was 12% at pH 5 and also observed at pH 4 and 6. On either side of pH 4 and 6, there was a sharp increase in the solubility for the total protein isolate. At pH 3, about 90% of the nitrogen was soluble, and about 72% of the nitrogen was soluble at pH 10. Total protein isolate studied showed good solubility in both acid and alkaline pH regions, which is an important characteristic for food formulations [\(Idouraine, Yensen, & Weber,](#page-4-0) [1991\)](#page-4-0). [Prakash and Narasinga \(1986\)](#page-4-0) reported similar observations.

Fig. 1. Effect of pH on solubility of a total protein isolated from sesame flour.

3.2. Functional properties of sesame protein isolate (SPI)

3.2.1. Water- and oil-holding capacity

TPI had a water-holding capacity of 2.10 ml H_2O/g protein, similar to that reported by [Prakash and Nar](#page-4-0)[asinga \(1986\)](#page-4-0) and within the range of the commercial values of protein concentrates (1.90–2.20), as reported by [Lin and Zayas \(1987\)](#page-4-0). It has been reported that the protein concentrate exhibits poor water-binding capacity compared to that of the isolate. This is likely due to the fact that the TPI has great ability to swell, dissociate and unfold, exposing additional binding sites, whereas the carbohydrate and other components of the protein concentrate may impair it [\(Kinsella, 1979\)](#page-4-0). The oilholding capacity of SPI was 1.50 ml oil/g protein. Sesame proteins showed a lower oil-holding capacity than soybean flour [\(Marina, 1986\)](#page-4-0) but had a higher value than chickpea flour ([Marina, 1986](#page-4-0)). [Kinsella](#page-4-0) [\(1979\)](#page-4-0) explained the mechanism of fat absorption as a physical entrapment of oil and several authors have related the oil absorption capacity to the nonpolar side chains of the protein as well as to the different conformational features of the proteins. Our results suggested that SPI had both good water-holding and good oil-holding capacity.

3.2.2. Viscosity and dispersibility

The results of viscosity and dispersibility measurements of TPI at different pHs are presented in Table 1.

Table 1

Effect of pH on viscosity at different temperature and dispersibility of total protein isolated from sesame flour

PН	Viscosity (cps)		Dispersibility $(\%)$	
	25 °C	70 °C		
5.0	44.70 (± 0.86)	134.20 (± 1.10)	71.0 (± 2.10)	
7.0	53.10 (± 1.20)	178.90 (± 0.96)	$85.0 (\pm 1.21)$	
9.0	67.10 (± 2.30)	223.70 (± 2.15)	91.0 (± 0.86)	

Values are means of triplicate samples $(\pm S.D.)$.

At room temperature (25 \degree C), it was clear that alkaline pH resulted in a higher viscosity value (67.10%) than that obtained at acidic pH. Heating TPI at 70° C for 15 min resulted in an appreciable increase in viscosity, especially at alkaline pH. It has been reported that heating of aqueous proteins activated the protein solution to the progel state, which is characterized by a marked increase in apparent viscosity. Moreover, further increase in apparent viscosity was observed after cooling the progel [\(Rivero & Pauda, 1983\)](#page-5-0). Results of the present study indicate that the apparent viscosity of the protein depends on the pH, which is likely to affect the conformational characteristics of the protein since viscosity is conformation-dependent [\(Idouraine et al.,](#page-4-0) [1991\)](#page-4-0). The reconstitution property of TPI, in terms of dispersibility, was significantly higher at alkaline pH than acidic (Table 1). It was reported that higher dispersibility enhances the emulsifying and faoming properties of proteins, which was observed during making of bread, macroni and cookies [\(Kinsella, 1979\)](#page-4-0).

3.3. Emulsifying properties

The effects of pH and NaCl concentration on emulsion capacity of total protein isolate (TPI) are shown in Figs. 2 and 3, respectively. It was found that SPI had a minimum capacity (70 ml oil/g protein) at pH 5 (Fig. 2) with an increase on either side of pH 5, and it was observed to be 150 ml oil/g protein at pH 2. Results revealed that emulsion capacity was pH-dependent and alkaline pH improved the emulsion capacity more than did the acidic pH. Dependence of emulsion capacity on pH was expected, as it is known that emulsion capacity of a total protein depends upon the hydrophilic–lipophilic balance, which is affected by pH ([Sathe, Desh](#page-5-0)[pande, & Salunkhe, 1982](#page-5-0)). Addition of NaCl at concentrations up to 1.0 M at pH 5 (isoelectric pH) increased the emulsification capacity of the protein [\(Fig. 3](#page-3-0)), due to the fact that addition of NaCl improved solubility of the protein at pH 5 and, accordingly, the emulsifying capacity. Beyond this salt concentration,

Fig. 2. Effect of pH on emulsifying capacity of a total protein isolated from sesame flour.

Fig. 3. Effect of NaCl concentration on emulsifying capacity of a total protein isolated from sesame flour.

the emulsification capacity gradually decreased due to the salting effect of NaCl. [Chobert, Bertrand, Nicolas,](#page-4-0) [Gaertner, and Puigserver \(1987\)](#page-4-0) have reported similar results. The effects of pH and NaCl concentration on the emulsifying activity (EA) and emulsion stability (ES) are shown in Table 2. Sesame protein isolate had a minimum emulsifying activity at pH 5 with emulsifying activities increasing on either side of pH 5 (Table 2). However, addition of NaCl (1.0 M) at pH 5 greatly improved the EA of the protein due to solubility improvement. Differences observed might account for the variations of the hydrophilic–lipophilic balance of the protein along the pH gradient from 2 to 10. Similar observations on the pH dependence of EA have been reported [\(Sathe et al., 1982](#page-5-0)). Moreover, the relationship between EA and pH for sesame protein was similar to that between nitrogen solubility and pH. This was in agreement with the general correlation between EA and nitrogen solubility found in previous studies ([Cren](#page-4-0)[welge, Dill, Tybor, & Landmann, 1974; Hung & Zayas,](#page-4-0) [1991\)](#page-4-0). Like the results of EA (Table 2), ES was pHdependent. [Hung and Zayas \(1991\)](#page-4-0) suggested that various factors, including pH, droplet size, net charge, interfacial tension, viscosity and protein conformation, could affect the values of ES.

3.3.1. Foaming properties

The foam capacity (FC) of sesame protein isolate (Fig. 4) was pH-dependent and was found to be lowest at pH 5 (2%). The lowest FC was attributed to the protein behaviour at its isoelectric point. Beyond pH 5, FC significantly increased, especially at pH 9 and 10. The higher FC at the above two pHs was likely due to the increased net charges on the protein, which weakened the

Fig. 4. Effect of pH on foaming capacity of a total protein isolated from sesame flour.

hydrophobic interactions but increased the flexibility of the protein. This allowed the protein to diffuse more rapidly to the air–water interface to encapsulate air particles and then enhance the foam formation [\(Aluko](#page-4-0) [& Yada, 1995\)](#page-4-0). The profile of FC against pH for the protein isolate was more or less similar to that of its nitrogen solubility against pH. Addition of NaCl at a concentration up to 1.0 M at pH 5 (Fig. 5) gradually improved FC of the protein and a higher increment was observed at this concentration. On either side of this concentration, there was a sharp decrease of FC. This may be attributed to the fact that addition of NaCl, at a concentration up to 1.0 M, enhances the protein solubility by weakening the hydrophobic interaction of the protein while high salt concentration had an adverse effect on FC due to the salting effect of NaCl. The effects of time (min), pH and 1.0 M NaCl on foam stability (FS) of sesame protein isolate (SPI) are shown in [Fig. 6.](#page-4-0) FS of the protein significantly decreased with

Fig. 5. Effect of NaCl concentration on foaming capacity of a total protein isolated from sesame flour.

Table 2

Effect of pH or NaCl concentration on the emulsifying activity and emulsion stability of total protein isolated from sesame flour

Functional properties	pH	1.0 M NaCl			
				10	
Emulsifying activity $(\%)$ Emulsion stability $(\%)$	87.00 (± 0.40) 75.20 (± 0.30)	41.00 (± 0.80) 37.80 (± 0.22)	70.00 (± 0.24) 70.02 (\pm 0.02)	75.00 (± 0.40) 62.00 (± 0.30)	52.00 (± 0.40) 42.80 (± 0.32)

Values are means of triplicate samples $(\pm S.D.)$.

Fig. 6. Effect of pH and NaCl concentration on foam stability of sesame protein isolated from sesame flour.

time at pH 2, 7 and 10, but slightly decreased at 1.0 M NaCl. When the foam stood for 90 min, FS was found to be 10, 11, 18 and 67% at pH 2, 7 and 10 and 1.0 M NaCl, respectively (Fig. 6). Addition of salt (1.0 M NaCl) greatly improved FS of the protein, due to increased solubility and surface activity of the soluble protein. Results revealed that foaming properties of the protein were pH-dependent.

In conclusion, a total protein isolate from sesame seed was found to be highly soluble at acidic and alkaline pH. Therefore, its emulsifying and foaming properties were higher than other proteins. Moreover, its waterholding, and fat-holding capacities, bulk density and other properties are good. Therefore, it can be used in food formulation systems.

References

- Aluko, R., & Yada, R. Y. (1995). Structure-function relationships of cowpea (Vigna unguiculata) globulin isolate: influence of pH and NaCl on physicochemical and functional properties. Food Chemistry, 53, 259–265.
- AOAC Official methods of analysis (13th ed.). Washington, DC: Association of Official Analytical Chemists.
- Aruna, V., & Parakash, V. (1993). Functional properties of the total proteins of sun flower (Helianthus annuus L.) seed. Effect of physical and chemical treatments. Journal of Agriculture and Food Chemistry, 41, 18–23.
- Beuchat, L. R., Cherry, J. B., & Quinn, M. R. (1975). Phsicochemical properties of peanut flour as affected by proteolysis. Journal of Agriculture and Food Chemistry, 23, 616–620.
- Carcea Bencini, M. (1986). Functional properties of drum dried chickpea (Cicer arietinum L.) flour. Journal of Food Science, 51, 1518– 1526.
- Chobert, J. M., Bertrand, H. C., Nicolas, M. G., Gaertner, H. F., &

Puigserver, A. J. (1987). Solubility and emulsifying properties of caseins chemically modified by covalent attachment of L-methionine and L-valine. Journal of Agriculture and Food Chemistry, 35, 639– 644.

- Crenwelge, D. D., Dill, C. W., Tybor, P. T., & Landmann, W. A. (1974). A comparison of the emulsification capacities of some protein concentrates. Journal of Food Science, 39, 175–177.
- Hung, S. C., & Zayas, J. F. (1991). Emulsifying capacity and emulsion stability of milk proteins and corn germ protein flour. Journal of Food Science, 56, 1216–1223.
- Idouraine, A., Yensen, S. B., & Weber, C. W. (1991). Tepary bean flour, albumin and globulin fractions functional properties compared with soy protein isolate. Journal of Food Science, 56, 1316-1318.
- Jane, E. D., Rivas, N. R., & John, C. (1981). Selected functional properties of sesame flour and two protein isolates. Journal of Science, Food and Agriculture, 32, 557–564.
- Karuna, D., Kulcarni, D. N., & Ingle, U. M. (1991). Sorghum maltbased weaning food formulations, preparation, functional properties and nutritive value. Food and Nutrition Bulletin, 13, 324–327.
- Kinsella, J. E. (1979). Functional properties of soy proteins. Journal of the American Oil Chemists' Society, 56, 242–249.
- Lin, C. S., & Zayas, J. F. (1987). Functionality of defatted corn germ proteins in a model system: fat binding and water retention. Journal of Food Science, 52, 1308–1311.
- Marina, C. B. (1986). Functional properties of drum-dried chickpea (Cicer arietinum L.) flours. Journal of Food Science, 51, 1518–1521.
- Nakai, S., & Voutsinas, L. P. (1983). A simple turbidimetic method for determining the fat binding capacity of proteins. Journal of Agriculture and Food Chemistry, 31, 58–63.
- Onweluzo, J. C., Obanu, Z. A., & Onuoha, K. C. (1994). Functional properties of some lesser known tropical legumes. Journal of Food Science and Technology, 31, 302-306.
- Pearce, K. N., & Kinsella, J. E. (1978). Emulsifying properties of proteins: Evaluation of a turbidimetric technique. Journal of Agriculture and Food Chemistry, 26, 716–723.
- Prakash, V., & Narasinga, M. S. (1986). Physico-chemical properties of oilseed proteins. CRC Critical Reviews in Biochemistry, 20, 265– 363.
- Prakash, V., & Nandi, P. K. (1978). Isolation and characterization of a-globulin of sesame seed. Journal of Agriculture and Food Chemistry, 26, 320–324.
- Quinn, M. R., & Beuchat, L. R. (1975). Functional property changes resulting from fungal fermentation of peanut flour. Journal of Food Science, 40, 475–479.
- Rivero, M., & Pauda, D. E. (1983). Some functional and utilization characteristics of sesame flour and proteins. Journal of Food Science, 48, 1145–1147.
- Sakamoto, H., Kumazawa, Y., & Motoki, M. (1994). Strength of protein gels prepared with microbial transglutminase as related to reaction conditions. Journal of Food Science, 59, 866–871.
- Sathe, S. K., Deshpande, S. S., & Salunkhe, D. K. (1982). Functional properties of lupin seed (Lupinus mutabilis) proteins and protein concentrates. Journal of Food Science, 47, 491–497.
- Wang, J. C., & Kinsella, J. E. (1976). Functional properties of novel proteins: alfalfa leaf protein. Journal of Food Science, 41, 286–289.