# Influence of pH and Salt Concentration on Protein Solubility, Emulsifying and Foaming Properties of Sesame Protein Concentrate

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**ABSTRACT:** The effects of pH and NaCl concentration on protein solubility, emulsification, and foamability of sesame protein concentrate from dehulled seeds were investigated. The protein content of the concentrate was 70.7%. Protein solubility, emulsion, and foaming capacities varied with pH and ionic strength. Protein solubility, which was least at pH 4, (2.1%) ranged from 6.6% at pH 2 to 13.1% at pH 10. The solubility increased with increase in ionic strength, ranging from 9.8% at 0.0 M to 16.1% at 1.0 M concentration. The emulsion capacity ranged from 6.2 mL oil/g sample at pH 2 to 19.4 mL oil/g sample at pH 10. The emulsion capacity increased from 11.5 mL oil/g sample at 0.0 M to 20.9 mL oil/g sample at 1.0 M salt concentration. Stability of the emulsion increased with increase in NaCl concentration, ranging from 42% at 0.0 M concentration to 70% at 1.0 M concentration, but 0.5 M NaCl produced the most stable foam after 120 min of whipping while the least stable was at 1.0 M.
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**KEY WORDS:** Emulsification, foamability, NaCl concentration, pH, protein solubility, sesame protein concentrate.

Serious protein deficiencies in parts of the world and increasing costs of many food ingredients from animal sources have generated considerable interest in plant proteins as substitutes for milk and meat protein. The growing industry, based on functional properties of soy protein in food product formulation, has stimulated interest in the functional potential of other major oilseeds. Sesame (*Sesamum indicum* L.) is an important oilseed that is extensively cultivated in the tropical and subtropical areas of the world, including Nigeria. The oil-free meal, containing approximately 50% protein, is at present used primarily to feed animals (1). The protein in the meal, as reported by Nilo Rivas *et al.* (1), is composed of globulins (67.3%), albumins (8.6%), prolamine (1.4%), and glutelin (7%). Unlike many other oilseeds, sesame does not contain undesirable pigments or off-flavors and is apparently devoid of antitrypsin compounds (2). Compared with the FAO reference protein, sesame protein is low in lysine but contains a high level of methionine, which is normally deficient in most other plant proteins (1,3). Because of this unique property, sesame protein may become an important source of high-quality food protein for supplementing other plant protein, such as peanut, soybeans and other legumes, in tropical diets. However, for sesame proteins to be successfully exploited for use in food formulations, they should possess satisfactory functional properties.

If oil-free meal is eaten by human beings, dehulling is necessary because the hull contains undesirable levels of oxalic acid (2–3%), which could complex calcium and reduce its availability (2). According to Johnson *et al.* (3), dehulling improves nutritional and flavor characteristics of sesame meal and leads to the production of a glossy white product. Also, it leads to an increase in protein content, reduction in fiber content, and improvement in the functional characteristics of the food ingredient (3).

The contribution of sesame protein concentrate, prepared from dehulled seeds, to human nutrition seems promising in view of its unique characteristics. Emulsification, foamability, and protein solubility are among the important functional properties that will influence its application in food formulation. Solubility of the protein is required if desired functional properties are to be achieved (4). Kinsella (5) listed many factors that have significant influence on the functional properties of proteins. Included in the list are pH and ionic strength of the medium.

Information concerning the functional properties of sesame protein concentrate and the influence of various processing factors and conditions is limited. This study was, therefore, designed to provide basic information on the effect of pH and salt concentration on emulsification, foaming, and protein solubility of sesame protein concentrate.

## MATERIALS AND METHODS

Sesame seeds were purchased from three different markets in Makurdi, the capital of Benue State, Nigeria, West Africa. Seeds were thoroughly mixed to obtain a composite sample that was processed into sasame protein concentrate.

*Seed dehulling.* Seeds were dehulled as described by Abou El-Khier *et al.* (6). Seeds were soaked in a lye mixture (0.04% NaOH and 5% $Na_2CO_3$) for 40–45 min at room temperature (31 ± 2°C) at a seed-to-lye ratio of 1:3 (wt/vol). Seeds were

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washed with tap water, and while being washed, they were rubbed by hand to decorticate them and to remove lye. Seeds, together with hulls, were sun-dried (33 ± 2°C), and hulls were removed by winnowing and screening.

*Concentrate preparation.* Dehulled seeds were flaked and extracted with food-grade *n*-hexane for 8 h in a Soxhlet apparatus. The meal was dried under vacuum at 60°C and milled to pass through a 75-mesh screen. The acid wash process, as described by Campbell *et al.* (7), was followed for production of the concentrate. The defatted flour was dispersed in distilled water at a flour-to-water ratio of 1:6 (wt/vol), and the pH was adjusted with 0.1 M HCl to pH 4.5 at room temperature to make the protein and polysaccharide insoluble, while the soluble sugars remained in solution. The dispersion was then centrifuged at 4000 × g for 30 min after 30 min of extraction. The wet protein concentrate was dried under vacuum at 60°C for 24 h. The dried concentrate was used for various determinations.

*Proximate analysis.* Moisture, fat, protein, fiber, and ash were determined according to AOAC (8). Carbohydrate was calculated by difference (9).

*Protein solubility.* Sesame protein concentrate (2% suspension, wt/vol) was dispersed in either distilled water with pH adjustments over a range of 2–10 (by adding either 1 N HCl or 1 N NaOH) or in 0.0, 0.1, 0.5, and 1.0 M sodium chloride solutions with the pH adjusted to pH 7. The dispersions were stirred for 30 min and centrifuged at 13000 × g for 10 min, and the protein content of each of the supernatants was determined by the Kjeldahl procedure (8).

*Emulsion capacity (EC) and emulsion stability (ES).* The method of Sathe and Salunkhe (10) was used. The sample (2 g) was blended at high speed in a Waring blender, either with 100 mL distilled water for 30 s with pH adjustments over a range of 2–10 or in 100 mL of 0.0, 0.1, 0.5, and 1.0 M sodium chloride solutions. To each of the suspensions, refined golden soy oil was added in 5-mL portions during blending. The drop in consistency (break point), as judged by a decreased resistance to blending (subjectively), was considered to be the point of discontinuation of oil addition. The quantity of oil added up to this point was noted, and EC was expressed as the quantity (mL) of oil emulsified by 1 g of product.

The emulsion was allowed to stand in a 100-mL graduated measuring cylinder, and volumes of water separated at 30-min time intervals were noted to determine ES. All experiments were conducted at room temperature (31 ± 2°C).

*Foam capacity (FC) and foam stability (FS).* The method of Okezie and Bello (11) was used. FC was expressed as percentage volume increase and calculated as shown below:

$$FC = \frac{\text{vol after whipping} - \text{vol before whipping}}{\text{vol before whipping}} \times \frac{100}{1} \qquad [1]$$

FS was determined by measuring the foam height at 30-min time intervals up to 120 min. Both FC and FS were determined over a pH range of 2 to 10 and also in salt concentrations of 0.0, 0.1, 0.5, and 1.0 M. All determinations were done at room temperature (31 ± 2°C).

## RESULTS AND DISCUSSION

*Proximate composition.* Proximate compositions of the dehulled sesame seed, defatted flour, and protein concentrate are shown in Table 1. Because sesame is an oilseed, extraction of oil from dehulled seed meal led to increases in other constituents in the flour. The major increase was in protein content, which ranged from 24.1% in the meal to 59.7% in the extracted flour. Processing of the flour to protein concentrate resulted in a further increase in protein content from 59.7% in the flour to 70.7% in the protein concentrate.

*Protein solubility.* Protein solubility of the concentrate, as influenced by pH (within pH range of 2–10) and sodium concentration (0.0 M–1.0 M), is illustrated in Figure 1. As the graph indicates, protein solubility was pH-dependent. The least protein solubility (2.2%) occurred at pH 4. At pH above or below 4, solubility increased and averaged 6.6% at pH 2 and 13.1% at pH 10. According to Okezie and Bello (11), protein solubility at various pH values may serve as a useful indicator of how well protein materials will perform when incorporated into food systems. Similar variations in protein solubility with pH were reported by McWatter and Holmes (12), Dench (13), Abbey and Ibeh (14), and Inyang and Nwadimkpa (15) for peanut flour, wing bean flour, cowpea flour, and sesame seed flour, respectively.

Protein solubility of the concentrate was also affected by ionic strength of the dispersing medium. Thus, increasing ionic strength from 0 to 1.0 M NaCl at pH 7 progressively increases protein solubility of the concentrate (Fig. 1). The increase was from 9.8% at 0.0 M concentration to 16.5% at 1.0 M concentration. Similar increases in protein solubility with increase in ionic strength have been reported (5,12,1). According to Nilo Rivas *et al.* (1), the major protein fraction in sesame flour is salt-soluble globulins. Increase in protein solubility of the concentrate with ionic strength could therefore be attributed to the globulins. Kinsella (5), however, stated that the increased solubility could be due to the greater activity and binding capacity of the chloride ions to the positively charged protein groups, thereby enhancing its solubility.

*Effect of pH on emulsification capacity and stability.* The EC and ES were affected by pH within the range of 2–10 as shown in Table 2. The lowest EC of 5.5 mL oil/g sample was observed at pH 4. Above or below pH 4, an increase in EC was observed. Alkaline pH improved EC more than did acid

**TABLE 1**
**Proximate Composition of Dehulled Sesame Seed, Defatted Flour, and Protein Concentrate**

| Parameters (%) | Dehulled seed | Defatted flour | Protein concentrate |
|---|---|---|---|
| Moisture content | 4.1 ± 0.1 | 5.1 ± 0.0 | 4.9 ± 0.2 |
| Crude fat | 62.5 ± 0.2 | 0.6 ± 0.2 | 0.3 ± 0.0 |
| Crude protein | 24.1 ± 0.1 | 59.7 ± 0.3 | 70.7 ± 0.1 |
| Ash | 3.2 ± 0.2 | 6.0 ± 0.1 | 6.9 ± 0.0 |
| Fiber | 3.3 ± 0.2 | 1.0 ± 0.1 | 1.0 ± 0.1 |
| Carbohydrate | 2.8 ± 0.3 | 28.2 ± 0.2 | 16.1 ± 0.1 |

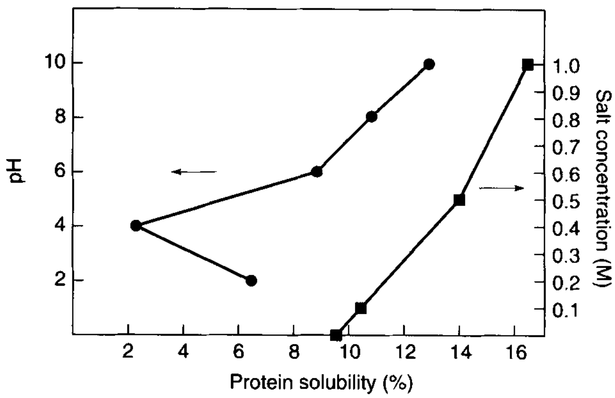


**FIG. 1.** Effects of pH (●) and salt concentration (■) on protein solubility of sesame protein concentrate.

**TABLE 2**
**Effect of pH on Emulsion Capacity and Stability of Sesame Protein Concentrate**

| pH | Emulsion capacity (mL/g) | Emulsion stability [mL $H_2O$ separated after time (min)] 30 | 60 | 90 | 120 | 150 | 180 |
|---|---|---|---|---|---|---|---|
| 2 | 6.2 ± 0.2 | 19.0 | 22.0 | 22.0 | 22.0 | 22.0 | 22.0 |
| 4 | 5.5 ± 0.2 | 18.0 | 20.0 | 20.0 | 20.0 | 20.0 | 20.0 |
| 6 | 9.4 ± 0.3 | 25.0 | 28.0 | 28.0 | 28.0 | 28.0 | 28.0 |
| 8 | 18.4 ± 0.3 | 23.0 | 24.5 | 24.5 | 25.0 | 25.0 | 25.0 |
| 10 | 19.4 ± 0.2 | 8.0 | 10.5 | 10.5 | 11.0 | 11.0 | 11.0 |

pH. This observation is in agreement with the findings of Franzen and Kinsella (16) who concluded that, as the net charge was near minimum in the isoelectric region, the protein might have aggregated and destabilized the interfacial membrane. The highest EC of 19.4 mL oil/g sample was observed at pH 10, whereas at pH 2, the value was 6.2 mL oil/g sample. The stability of the emulsion was also highest at pH 10 and least at pH 6. A similar result was reported by Ihekoronye (17) for groundnut protein concentrate.

*Effect of sodium chloride concentration.* The emulsion properties of sesame protein concentrate were also affected by the ionic strength of the dispersing medium at pH 7 as shown in Table 3. Increasing the concentration of sodium chloride from 0 to 1.0 M resulted in an increased emulsion capacity of the concentrate. The increase was from 11.5 mL oil/g sample at 0.0 M concentration to 20.9 mL oil/g sample at 1.0 M concentration. This increase in EC could be attributed to increased solubilization of the protein as the ionic strength was increased. Kinsella *et al.* (18) had stated earlier that, in dilute emulsion, solubility of proteins is an important prerequisite for their emulsifying properties. They concluded that, because the ionic strength of the dispersing medium affects the solubility and association–dissociation of soy protein subunits, the enhanced emulsifying activity observed with increased salt concentration may reflect the formation of a more cohesive protein film. The stability of the emulsion increased with increased salt concentration. A similar observation was reported by Kinsella (5). McWatter and Holmes (12) stated that salt may stabilize emulsions by reducing coulumbic interaction between neighboring droplets. The highest ES was observed at 1.0 M salt concentration (Table 3). This result suggests that the sesame protein concentrate could be used in comminuted meat products.

*Effect of pH on FC and FS.* Data in Table 4 show the effect of pH on FC and FS of the sesame protein concentrate. The lowest FC of 9.0% occurred at pH 4. At pH above or below 4, the FC increased with the highest (97.0%) being at pH 10. At pH 2, the value was 22.0%. The poor FC at pH 4 could be due to low solubility in this pH range (18).

Stability of the foam decreased with increase in pH above pH 4. Maximum stability of the foam occurred at pH 2, while the least was at pH 10 (Table 4). Stability of the foam in the acid pH range could be due to the formation of stable molecular layers in the air–water interface, which impart stability and elasticity to the foams (5). A similar dependence of foaming properties on pH was reported by Dench *et al.* (19), Abbey and Ibeh (14) and Inyang and Nwadimkpa (15) for sesame isolates, cowpea flour, and sesame seed flour, respectively.

*Effect of NaCl concentration on foaming properties.* The influence of NaCl concentration on the FC and FS of sesame protein concentrate is shown in Table 5. The FC increased with increasing NaCl concentration. The increase was from 42.0%, when dispersed in distilled water (0.0 M concentration), to 70% at 1.0 M concentration. This increase is in agreement with observations reported by Kinsella (5) for soy

**TABLE 3**
**Effect of NaCl Concentration on Emulsion Capacity and Stability of Sesame Protein Concentrate**

| Salt concentration (M) | Emulsion capacity (mL oil/g sample) | Emulsion stability [mL $H_2O$ separated after time (min)] 30 | 60 | 90 | 120 | 150 | 180 |
|---|---|---|---|---|---|---|---|
| 0.0[a] | 11.5 ± 0.1 | 10.1 | 12.0 | 12.0 | 12.0 | 12.0 | 12.0 |
| 0.1 | 15.1 ± 0.0 | 11.5 | 12.5 | 13.0 | 13.0 | 13.0 | 13.0 |
| 0.5 | 17.3 ± 0.2 | 8.0 | 9.5 | 10.0 | 10.0 | 10.0 | 10.0 |
| 1.0 | 20.9 ± 0.1 | 9.1 | 9.5 | 10.0 | 10.0 | 10.0 | 10.0 |

[a]Distilled water.

**TABLE 4**
**Effect of pH on Foam Capacity and Stability of Sesame Protein Concentrate**

| Dispersion pH | Volume before whipping (mL) | Volume after whipping (mL) | Foam capacity (%) | Foam stability [vol (mL) at room temperature (31 ± 2°C) after time (min)] 30 | 60 | 90 | 120 |
|---|---|---|---|---|---|---|---|
| 2 | 50 | 61.0 ± 0.2 | 22.0 | 60.5 | 59.5 | 59.5 | 59.5 |
| 4 | 50 | 54.5 ± 0.3 | 9.0 | 50.5 | 50.5 | 50.5 | 50.5 |
| 6 | 50 | 71.5 ± 0.1 | 43.0 | 66.5 | 64.0 | 59.5 | 58.0 |
| 8 | 50 | 85.0 ± 0.1 | 70.0 | 60.0 | 57.0 | 54.5 | 54.5 |
| 10 | 50 | 98.5 ± 0.2 | 97.0 | 62.0 | 60.0 | 50.0 | 50.0 |

**TABLE 5**
**Effect of NaCl Concentration on Foam Capacity and Stability**

| NaCl concentration (M) | Volume before whipping (mL) | Volume after whipping (mL) | Foam capacity (%) | Foam stability [vol (mL) at room temperature (31 ± 2°C) after time (min)] 30 | 60 | 90 | 120 |
|---|---|---|---|---|---|---|---|
| 0.0[a] | 50 | 75.0 ± 0.1 | 42.0 | 57.5 | 53.5 | 51.5 | 51.0 |
| 0.1 | 50 | 77.5 ± 0.3 | 55.0 | 62.0 | 56.0 | 53.0 | 53.0 |
| 0.5 | 50 | 81.0 ± 0.2 | 62.0 | 73.0 | 65.5 | 62.0 | 62.0 |
| 1.0 | 50 | 85.5 ± 0.1 | 70.0 | 62.5 | 57.0 | 55.0 | 53.0 |

[a]Distilled water.

protein. The increase could be attributed to increased solubilization of protein as ionic strength was increased (Fig. 1). Kinsella (5) and Dench *et al.* (19) have stated earlier that only soluble proteins in the aqueous phase can participate in foam formation.

Stability of the foam varied with the ionic strength of the dispersing medium: 0.5 M salt concentration produced the most stable foam after 120 min of whipping, while 1.0 M gave the least FS. Dench *et al.* (19) had earlier reported a decrease in FS with an increase in ionic strength of the dispersing medium.

In conclusion, protein solubility, emulsion, and foaming properties of the sesame protein concentrate vary with pH and NaCl concentration. Increase in ionic strength of the dispersing medium resulted in increases in these properties.

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