

Functional properties of barinas nut flour (*Caryodendron orinocense* Karst., Euphorbiaceae) compared to those of soybean

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Protein solubility studies showed that the protein of *Caryodendron orinocense* flour was soluble at both acidic and basic pH, and NaCl (0.5 M) increased the solubility of all flours tested. The water absorption capacity was less, while the oil absorption was higher for *Caryodendron* flour than soybean flour. Both flours presented similar emulsion capacity and stability, while the foaming capacity was much smaller for *Caryodendron* flours than soybean flour; NaCl increased the foaming capacity of soybean flour; these results might be due to differences in protein concentration. NaCl concentrations greater than 0.25 M increased the emulsifying activity in *Caryodendron* flours, but did not influence the emulsion stability. The lowest gelation concentration and temperature of gelling were similar to that of soybean flour. These results suggest that *Caryodendron* flour might have some similar uses as soybean flour in the food industry. Copyright © 1996 Elsevier Science Ltd

INTRODUCTION

The study of non-conventional food sources is important owing to the need for new food and feed products for humans and animals, and to increase the economic development of countries through the exploitation of their resources.

Vegetable proteins are important in human nutrition, especially in the Third World countries where the ingestion of proteins is below the recommended allowance. The production of high protein foods from under-utilized food sources should contribute to improve this condition.

Proteins not only provide a nutritional component in foods but also perform other functions. Among the most important attributes of proteins are their various functional properties. The functional properties are characteristic of the proteins, but the method of extraction and the source have some effect on the functional quality of the product. The procedures used to obtain seed flours, concentrates and isolates cause denaturation and alter their functional properties. Carbohydrates, lipids, fibre, etc., may also influence the functional properties of proteins (McWatters & Cherry, 1977).

Caryodendron orinocense K. called in Venezuela 'nuez de Barinas', 'Nogal de Barquisimeto', 'nueza' and

'taque', which Reckin (1982) named 'orinoconut' owing to its natural distribution adjacent to the headwaters of the Orinoco river, belongs to the Euphorbiaceae and also grows wild at the base of The Andes in the Barinas, Lara and Apure States of Venezuela. It is a non-conventional food, consumed by the farmers of the region in diverse ways, and is sometimes used as a milk substitute in infant feeding (CRBS, 1984). The seeds have a high fat content (30% dwb), and after extraction the remaining cake could represent a source of protein for human and animal consumption. The use of vegetable proteins as functional ingredients in foods depends mainly on the benefits that they can produce (McWatters & Cherry, 1977). Taking into consideration actual developments in the food industry, the Barinas nut flour might represent a useful raw material. Its use will depend upon its functional properties; nevertheless as there have been no studies done until now, the objective of the present work is to determine these properties and compare them to those of soybean flour to find possible applications in food products.

MATERIALS AND METHODS

The seeds were obtained from the Calderas region (Barinas State) and the Yacambú Park (Lara State). Nestlé of Venezuela donated the soybean (*Glycine max*) flour.

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After extraction of fat with hexane, the broken, dehulled seeds were dried at 50°C under vacuum for 24 h, then ground and sifted through a 60-mesh sieve.

Proximate composition of the flour was carried out in triplicate using the methods described by the AOAC (1984).

Crude fibre was determined by using the Tecator model Fibertec M system (Sweden), following the procedure described by the manufacturer.

Protein solubility

To 0.5 g of the flour, 50 ml of water or a salt solution was added and the pH of the suspension adjusted to the desired value by the addition of 0.1 N NaOH or H₂SO₄. The suspension was then stirred for 20 min at room temperature (~25°C), and centrifuged at 10 000 g for 10 min. Aliquots of 10 ml were taken for nitrogen estimation by the Kjeldahl method. The percentage total flour nitrogen extracted was calculated. The salt solutions used were: NaCl at 0.05, 0.25 and 0.75 M.

Bulk density

The method described by Okaka & Potter (1979) was used using 25 g of the flour samples

Oil and water holding capacity

To 2 g of sample, 20 ml of water, or 12 ml of corn oil (Mazeite), was added, stirred in a vortex and stored at room temperature for 1 h. The procedure of Okaka & Potter (1979) was followed.

Emulsifying activity and emulsion stability

Emulsifying activity (EA) was determined by the turbidimetric method of Pierce & Kinsella (1978) with slight modification. A suspension of the flour was prepared to a protein concentration of 0.50% (w/v) in an aqueous solvent, and the pH adjusted to 6.4, in a 100 ml volumetric flask. The solvents used were water, and NaCl in 0.05, 0.25, 0.50, and 0.75 M solutions. The suspensions were stirred for 1 min at 750 rpm. To 20 ml of the suspension 20 ml of oil were added and homogenized in a T-line homogenizer for 1 min at 2500 rpm. Immediately after homogenization, aliquots of 30 µl of each emulsion were diluted to 10 ml with a 0.1% sodium dodecyl sulfate (SDS) solution and its absorbance was determined at 500 nm with a Shimadzu spectrophotometer, using the SDS solution in the reference cell. A plot of absorbance vs sodium chloride concentration is used to determine the emulsifying activity. The emulsion stability (ES) was determined by reading the volume of emulsion remaining after 24 h at room temperature.

Foam capacity and stability

These properties were assessed by the Srinivas & Rao Narasinga (1986) method, where 3 g of flour were dilu-

Table 1. Proximate analyses of the *Caryodendron* and soybean flours*

Component	Lara (%)	Barinas (%)	Soybean (%)
Moisture	1.58 ± 0.02	2.57 ± 1.53	8 (max)
Crude fat	15.08 ± 0.32	14.09 ± 0.11	2 (max)
Protein (N x 6.25)	16.6 ± 0.30	17.94 ± 0.19	50 (min)
Ash	3.17 ± 0.23	3.50 ± 0.05	6.5 (max)
Crude fibre	4.45 ± 0.02	6.60 ± 0.41	17 ¹ (max)
Carbohydrates	59.09	55.30	16.5

*Mean of $n=3$ and expressed on dry weight.

¹Expressed as dietary fibres.

ted to 100 ml with an aqueous solvent. The pH was adjusted to the desired value with 0.1 N NaCl or H₂SO₄ solution. The suspensions were stirred in a blender for 5 min at 1600 rpm and then poured into a 250 ml measuring cylinder, reading the volume of foam after 30 s. The solvents used were: water, and NaCl at 0.05, 0.25, 0.50 and 0.75 M solutions. The foam stability (FS) was determined by measuring the volume of foam at 5, 30, 60 and 120 min after pouring the suspension into the measuring cylinder and is expressed as % volume.

Gel formation

The gelation was analysed by the method of Coffman & Garcia (1977) with slight modification as suggested by Deshpande *et al.* (1982). Water dispersions of the samples at concentrations in the range of 2–20% (w/v) with increments of 2% were prepared. Aliquots of 5 ml were transferred to each of three test tubes for each concentration, and heated for 1 h in a boiling water bath, followed by rapid cooling under running tap water and then stored at 4°C for 2 h. The least gelation concentration was recorded as the concentration when the sample did not fall or slip from the inverted test tube.

The time needed to form a gel was measured by the Jongasma & van Pijkeren (1985) method. Suspensions of the samples were prepared at the previously determined minimum flour concentration, placed in a water bath at 80°C, and observations made at 2 min intervals. The following indices are used to describe the observed situation:

Gelling		Gel structure
–	no gelling	liquid
+ –	some flocules	pourable
+ + –	almost homogeneous gel	gel remains fixed on turning the tube upside down
+ + +	complete gelation	gel remains fixed on shaking the tube upside down

Statistical analysis

To compare mean differences, the Student's *t*-test at 95% confidence interval and $P < 0.05$ was used.

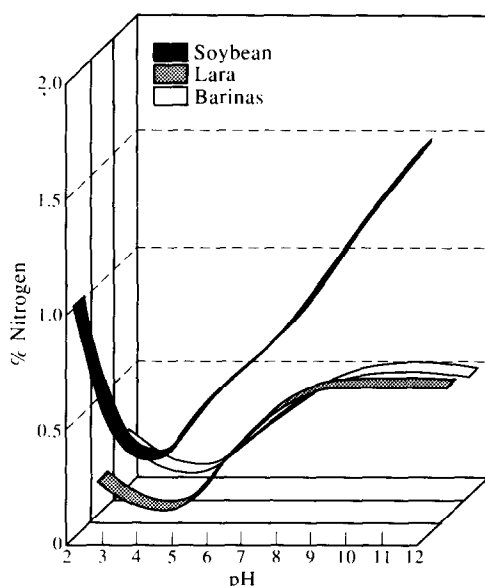


Fig. 1. pH effect on the solubility profile of *Caryodendron* and soybean flours.

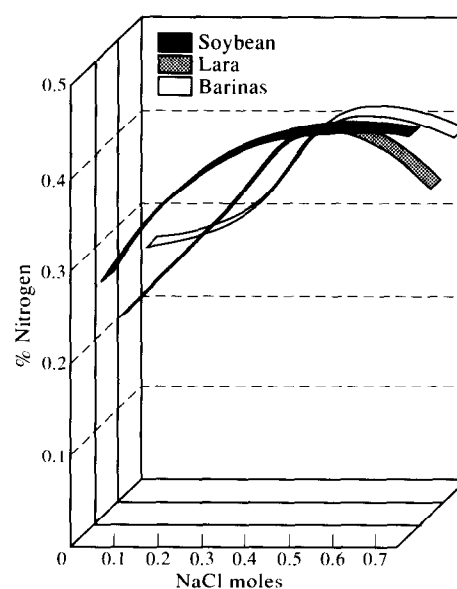


Fig. 2. NaCl concentration effect on the solubility profile of *Caryodendron* and soybean flours at pH 6.4.

RESULTS AND DISCUSSION

Proximate analysis

The results show differences in some of the components (Table 1).

Protein solubility

The differences in protein solubility as affected by pH are shown in Fig. 1. All flours at pH 4 are less soluble; this might represent the isoelectric point region. Solubility increased both above and below pH 4; this behaviour is similar to many other seed flours, as has been reported by Akobundu *et al.* (1982); the only difference is that the solubility of *Caryodendron* flours reaches a plateau at pH 8, while soybean solubility continues to increase.

These results suggest that, at alkaline pH there is a greater extraction of the soluble proteins as has been indicated by Ma & Harwalkar (1984), or there is a denaturation which increases solubility.

Figure 2 shows the effect of NaCl concentration on the solubility of the proteins. The solubility increases with NaCl concentration up to 0.5 M for all samples, showing a small decrease afterwards for *Caryodendron* flours and remaining constant for soybean flour. This behaviour is due to the low ionic strength of the NaCl ($\mu = 0.1$), which allows its dissociation and consequent interaction with the proteins, increasing their solubility ('salting in' effect). With higher concentrations, NaCl produces a dehydrating effect over the proteins which tend to aggregate, and in this way their solubility decreases, ('salting out' effect).

These results corroborate what has been reported by Kinsella (1979) and Shen (1981) that the solubility is influenced by factors such as source of the protein, processing conditions, pH, ionic strength and presence of other ingredients.

Table 2. Bulk density of *Caryodendron orinocense* and soybean flours*

Flour samples	Density (g/ml)
Barinas	0.2988 ± 0.0004 ^a
Lara	0.3061 ± 0.0022 ^a
Soybean	0.5598 ± 0.0665 ^b

*Mean of $n=3$. Values with different letters are statistically different ($P < 0.05$).

Table 3. Comparison of water and oil absorption capacity of *Caryodendron orinocense* and soybean flours*

Flour samples	Water %	Oil %
Barinas	34.05 ± 0.6578 ^a	35.70 ± 1.2456 ^a
Lara	43.57 ± 1.9719 ^b	35.08 ± 2.0325 ^a
Soybean	112.43 ± 2.3097 ^c	29.59 ± 0.4406 ^b

*Mean of $n=3$. Values with different letters are statistically different ($P < 0.05$).

Density

Table 2 shows the values for bulk density of the different samples. There are significant differences between soybean and *Caryodendron* flours; this may be due to differences in particle size as has been indicated by Dench *et al.* (1981).

Oil and water holding capacity

Table 3 shows the oil and water absorption capacity. Soybean flour presents the highest water holding (112.43%) and the lowest oil absorption capacities (29.59%), and there are significant differences among

Table 4. Emulsifying activity and emulsion stability of *Caryodendron orinocense* and soybean flours*

Flour samples	Emulsifying activity ¹	Emulsion stability ²
Barinas	0.64 ± 0.0938 ^a	61.79 ± 3.0215 ^{ab}
Lara	1.08 ± 0.1058 ^b	59.46 ± 0.0000 ^b
Soybean	1.28 ± 0.2504 ^c	61.75 ± 0.6749 ^a

*Mean of $n=3$. Values with different letters are statistically different ($P<0.05$).

¹Absorbance at 500 nm.

the samples. These results suggest that soybean proteins are more hydrophilic due to a higher number of carboxyl and amino groups, or soybean flour contains more carbohydrates (polysaccharides) which entrap more water than proteins. Conversely, *Caryodendron* samples are more lipophilic and this may be owing to a higher number of non-polar amino acids in their composition, because fat absorption is attributed mainly to the combination of fats to the non-polar groups of proteins (Kinsella, 1976) or the availability of lipophilic groups (Sumner *et al.*, 1981). These properties have an influence in texture and mouthfeel of foods, particularly, comminuted meats, extenders or analogues, and baked doughs (Cheftel *et al.*, 1985; Okezie & Bello, 1988).

Emulsifying activity (EA) and emulsion stability (ES)

Emulsion characteristics of proteins contribute much to their functionality in foods. Soluble proteins are surface active and known to promote oil-in-water emulsions (Subba Rau & Srinivasan, 1988). Table 4 indicates that there are significant differences among the samples at $P<0.05$ for the EA determined at pH 6.4 without the addition of NaCl. At this pH, soybean flour presents the highest solubility. For ES there are significant differences between *Caryodendron* and soybean flours, but not between *Caryodendron* flours.

Figure 3 shows the EA by relating absorbance to NaCl concentration; here the most important thing to be noted is that, even though soybean flour starts with a higher EA, it has an inverse relationship with the NaCl concentration. However, *Caryodendron* flours reached a minimum EA at 0.25 M NaCl to increase thereafter, finishing with higher or similar EA at 0.75 M NaCl. These results may be related to protein concentration differences (soybean 50%, *Caryodendron* 17%) as reported by Holm & Eriksen (1980).

The results shown in Fig. 4 suggest that the NaCl concentrations do not have an influence on the ES of any samples tested.

Foam capacity and stability

Figure 5 shows the foam capacity (FC) vs pH profile of *Caryodendron* and soybean flours. The foam volume increased as pH increased for all samples tested; nevertheless, soybean flour, probably due to its higher content of protein, showed a higher increase than

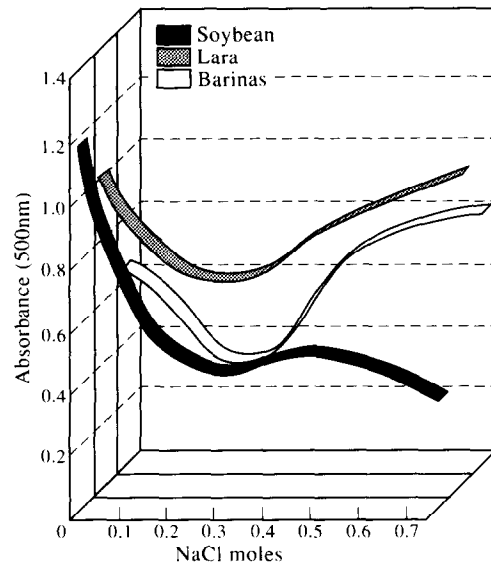


Fig. 3. NaCl concentration effect on the emulsifying activity of *Caryodendron* and soybean flours at pH 6.4.

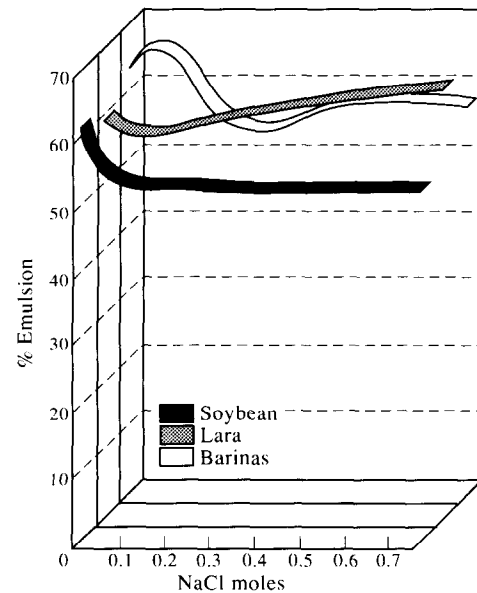


Fig. 4. NaCl concentration effect on the emulsion stability of *Caryodendron* and soybean flours at pH 6.4.

Caryodendron flours. This increase is also the result of an increase in protein solubility as the pH increases, as reported by Satterlee *et al.* (1975) and Kinsella (1976). The FS also increased with pH and showed its maximum at the pH of highest solubility for each of the samples tested (Fig. 6). According to Badui (1981), for the formation of foam there must be a controlled denaturation process, therefore in the alkaline pH region there must be more denaturation, since there is higher foam formation.

The effect of NaCl on the FC of the samples in the range of 0.05–0.75 M NaCl concentration is shown in Fig. 7. In these measurements, the pH of the flour suspensions was 6.4. In contrast to soybean flour, which showed an increase in FC values only up to 0.5 M NaCl

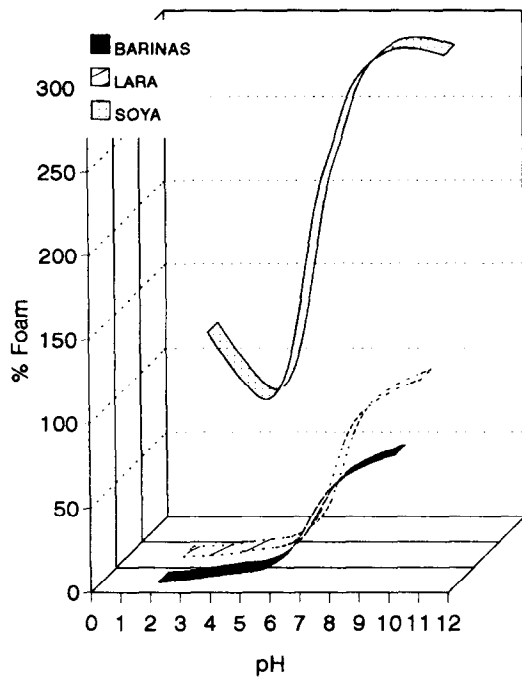


Fig. 5. pH effect on the foam capacity of *Caryodendron* and soybean flours.

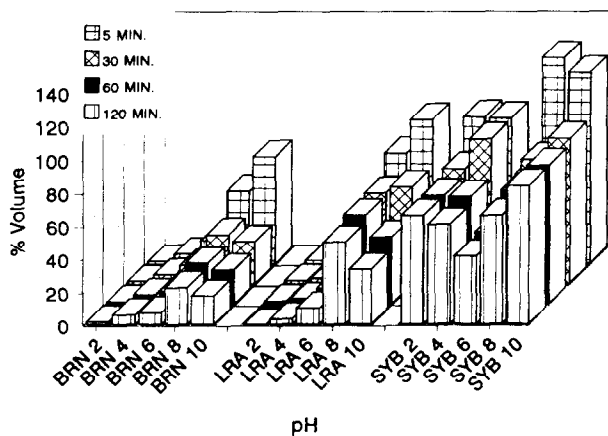


Fig. 6. pH effect on the foam stability of *Caryodendron*: BRN (Barinas), LRA (Lara) and SYB (soybean) flours.

concentration, *Caryodendron* flours showed higher FC values at 0.75 M NaCl. These results may be due to the increase in solubility produced by NaCl as also reported by Shanmugasundaran & Venkatamaran (1989). The differences among soybean and *Caryodendron* flours may also be explained by the differences in protein concentration.

The FS of *Caryodendron* flours was lower than that of soybean (Fig. 8). For *Caryodendron* flours (Barinas and Lara), the FS was higher at 0.25 and 0.05 M NaCl concentrations, respectively, while soybean flour showed its highest FS at 0.5 M NaCl concentration. These differences between soybean and *Caryodendron* flours may also be attributed to the differences in protein concentration. These two properties are important in foods that require a high FC and FS, as in the case of cake mixes and frostings (Ma & Harwalkar, 1984).

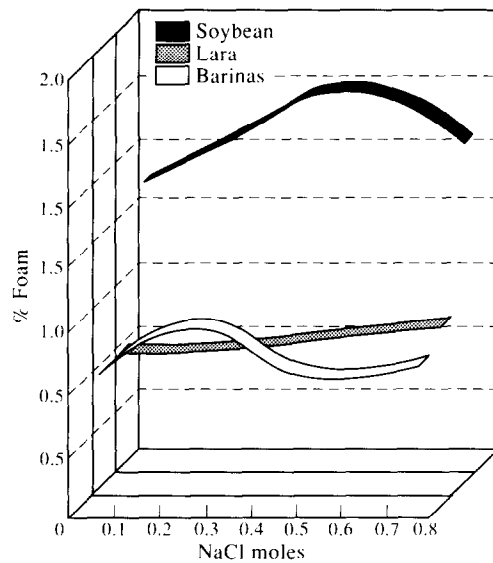


Fig. 7. Effect of NaCl concentration on the foam capacity of *Caryodendron* and soybean flours at pH 6.4.

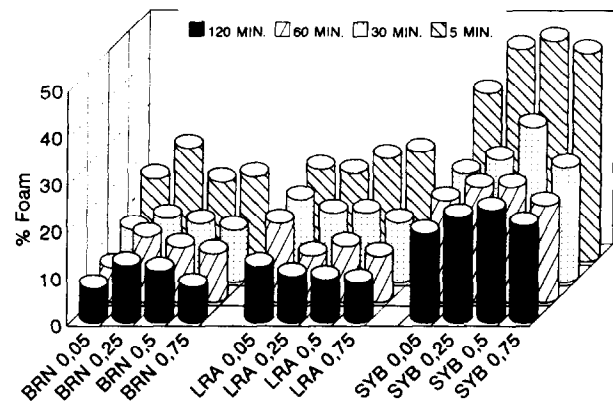


Fig. 8. NaCl concentration effect on the foam stability of *Caryodendron*: BRN (Barinas), LRA (Lara) and SYB (soybean) flours at pH 6.4.

Gel formation

The least gelation concentration (w/v) for *Caryodendron orinocense* seed flour is 14%, which is the same for Lupine seed (Sathe *et al.*, 1982) and *Adenopus breviflorus* (Oshodi, 1992) but higher than soybean flour (10%). It has been suggested (Sathe *et al.*, 1982) that variation in the gelling properties of different legume flours may be linked to relative ratios of different constituent proteins, carbohydrates, and lipids and that interactions between such components may affect functional properties. Gel strength improves with increasing protein concentration, but a minimum protein concentration is necessary for gelation; as the concentration is increased above this minimum, gelling time is reduced (Mulvihill & Kinsella, 1987). The time required for *Caryodendron* and soybean flours to gel was 12 and 10 min at 80°C. This gelling time is also dependent on temperature, with the time required for gelation decreasing as the heating temperature is increased (Hillier & Cheeseman, 1979). These results

show that the differences in protein concentration have a clear influence.

CONCLUSIONS

Some functional properties of *Caryodendron orinocense* flour are comparable to those of soybean and this flour therefore might have similar uses in the food industry.

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REFERENCES

- Akobundu, E. N. T., Cherry, J. P. & Simmons, J. G. (1982). Chemical, functional, and nutritional properties of egusi (*Colocynthis citrullus* L.) seed protein products. *J. Food Sci.*, **47**, 829–835.
- AOAC (1984). *Official Methods of Analysis*, 14 edn. Association of Official Analytical Chemists, Washington, DC.
- Badui, D. S. (1981). *Química de los Alimentos*. Edt. Alhambra Mexicana, SA, pp. 107–158, 343–372.
- Cheftel, J. C., Cuq, J. L. & Lorient, D. (1985). Amino acids, peptides and proteins. Ch.5. In *Food Chemistry*, 2nd edn., ed. O. R. Fennema. Marcel Dekker, New York, p. 299.
- Coffman, C. W. & Garcia, V. V. (1977). Functional properties & amino acid content of a protein isolate from mung bean flour. *J. Food Technol.*, **12**, 473–484.
- CRBS (1984). Comité de Recursos Botánicos Sub-utilizados. Boletín Informativo.
- Dench, J. E., Rivas, N. & Caygild, J. C. (1981). Selected functional properties of sesame (*Sesamum indicum* L.) flour and two protein isolates. *J. Sci. Food Agric.*, **32**, 557–664.
- Deshpande, S. S., Sathe, S. K., Cornforth, D. & Salunkhe, O. K. (1982). Effects of dehulling on functional properties of dry bean (*Phaseolus vulgaris* L.) flours. *Cereal Chem.*, **59**, 396–401.
- Hillier, R. M. & Cheeseman, G. C. (1979). Effect of proteose-peptone on the heat gelation of whey protein isolate. *J. Dairy Res.*, **46**, 113–120.
- Holm, F. & Eriksen, S. (1980). Emulsifying properties of undenatured potato protein concentrate. *J. Food Technol.*, **15**, 71–83.
- Jongsma, J. & van Pijkeren, H. (1985). The influence of different non-meat proteins on the heat gelling properties of various meat protein fractions. DMV International.
- Kinsella, J. E. (1976). Functional properties of proteins in food: a survey. *Crit. Rev. Fd. Sci. Nutr.*, **7**, 219–280.
- Kinsella, J. E. (1979). Functional properties of soy proteins. *J. Am. Chem. Soc.*, **56**, 242–258.
- Ma, C. T. & Harwalkar, V. R. (1984). Chemical characterization and functionality assessment of oat protein fractions. *J. Agric. Food Chem.*, **32**, 144–149.
- McWatters, K. H. & Cherry, J. P. (1977). Emulsification, foaming and protein solubility properties of defatted soybean, peanut, field pea and pecan flours. *J. Food Sci.*, **42**, 1444–1447.
- Mulvihill, D. H. & Kinsella, J. E. (1987). Gelation characteristics of whey proteins and β -lactoglobuline. *Food Technol.*, **41**, 102–111.
- Okaka, J. C. & Potter, N. N. (1979). Physico-chemical and functional properties of cowpea powders processed to reduce beany flavor. *J. Food Sci.*, **44**, 1233–1240.
- Okezie, B. O. & Bello, A. B. (1988). Physicochemical and functional properties of winged bean flour and isolate compared with soy isolate. *J. Food Sci.*, **53**, 450–454.
- Oshodi, A. A. (1992). Proximate composition, nutritionally valuable minerals and functional properties of *Adenopus breviflorus* Benth. seed flour and protein concentrate. *Food Chem.*, **45**, 79–83.
- Pierce, K. N. & Kinsella, J. E. (1978). Emulsifying properties of proteins: Evaluation of a turbidimetric technique. *J. Agric. Food Chem.*, **26**, 716–723.
- Reckin, J. (1982). The orinoconut. A promising tree crop for the tropics. *Int. Tree Crops J.*, **2**, 105–119.
- Sathe, S. K., Deshpande, S. S. & Salunkhe, D. K. (1982). Functional properties of Lupine seed (*Lupinus mutabilis*) proteins and protein concentrates. *J. Food Sci.*, **47**, 491–497.
- Satterlee, L. D., Bembers, M. & Kendrick, J. O. (1975). Functional properties of great northern bean (*Phaseolus vulgaris*) protein isolate. *J. Food Sci.*, **40**, 81–84.
- Shanmugasundaran, T. & Venkatamaran, L. V. (1989). Functional properties of defatted and detoxified madhuca (*Madhuca butyraceae*) seed flour. *J. Food Sci.*, **54**, 351–353.
- Shen, J. L. (1981). Solubility and viscosity. In *Protein Functionality in Foods*, ACS Symposium Series 147, ed. J. P. Cherry. American Chemical Society, Washington, DC, pp. 89–109.
- Srinivas, H. & Rao Narasinga, M. S. (1986). Functional properties of poppy seed meal. *J. Agric. Food Chem.*, **34**, 222–224.
- Subba Rau, B. H. & Srinivasan, K. S. (1988). Enzymatic modification of groundnut flour (by papain/protease) and its effect on functional properties. *Lebensm.-Wiss. u.-Technol.*, **21**, 126–130.
- Sumner, A. K., Nielsen, M. A. & Youngs, C. G. (1981). Production and evaluation of pea protein isolate. *J. Food Sci.*, **46**, 364–372.