Food Chemistry **42** (1991) 153–165

A Study of Factors Affecting Extraction of Peanut (*Arachis hypogaea* L.) Solids With Water

Ismail Y. S. Rustom,[a] M. H. López-Leiva[a]
& Baboo M. Nair[b]

[a] Department of Food Engineering, [b] Department of Applied Nutrition,
Chemical Center, University of Lund,
PO Box 124, 221 00 Lund, Sweden

(Received 24 January 1990; revised version received and
accepted 21 March 1990)

ABSTRACT

*The effects of pH, temperature, time, solids-to-water ratio, speed of agitation, partial hydrolysis of proteins, and soaking on the extraction of peanut (*Arachis hypogaea *L.) solids with water were studied using a fractional factorial set of experiments. Temperature, pH, solids-to-water ratio, hydrolysis, and soaking were significant for protein extraction. Temperature, speed of agitation, hydrolysis, and soaking were significant for fat extraction. Only hydrolysis and soaking were significant for carbohydrate extraction. Protein recovery was increased when hydrolyzed. Increased pH and solids-to-water ratio also increased protein recovery. Fat recovery was greatest at high agitation speed. Soaking highly increased carbohydrate contents of extracts, and decreased protein and fat. Hydrolysis was found to depend on temperature and time.*

INTRODUCTION

Recently, peanuts have attracted a great deal of interest as a source of low-cost protein to supplement human diets. In addition to the traditional food uses, peanut butter and roasted peanuts, they have been successfully utilized in supplemented foods such as bakery products, as extenders in meat product formulations, and in soups and desserts (Conkerton & Ory, 1988). Some workers incorporated peanuts in milk substitutes (Elahi & Ali, 1971;

Food Chemistry 0308-8146/91/$03·50

Schmidt & Bates, 1976), fermented peanut milk (Beuchat & Nail, 1978; Bucker *et al.*, 1979), and peanut beverage systems (Schmidt *et al.*, 1978; Rubico *et al.*, 1987).

The idea of making protein-rich beverages from peanuts has prompted workers to put much effort into this discipline. Peanuts are produced in surplus quantities (about 20 million tons per year) mainly in the developing countries where malnutrition exists (FAO, 1986). Peanuts are an excellent source of nutrients from a dietary viewpoint (Woodroof, 1969). It is reported that peanut proteins have suitable functional properties, such as solubility, viscosity and emulsifying capacity, most desirable in beverage systems (Sekul *et al.*, 1978; McWatters & Cherry, 1982). Many people in the developing countries can hardly drink mammalian milk because they are lactose-intolerant. Fortunately, peanut carbohydrates are lactose-free making them useful for the solution of this problem. Finally, many of these countries are located in temperate regions where sterilized food products with long storage life are economical.

Effective utilization of peanuts in beverage-type products is still in need of research and development. One of the primary aspects to be studied carefully is the extraction of peanut solids with water. There are several potential factors governing the extraction process of the peanut major constituents, protein and fat, with water. Among these factors are the pH of the suspension, extraction temperature and time, soaking of peanut kernels, solids-to-water ratio, speed of agitation, partial hydrolysis of peanut proteins with enzymes, and ionic strength of extraction medium (Elahi & Ali, 1971; Rhee *et al.*, 1972). The methods so far used to study the extraction of peanut solids with water adopted the one-factor-at-a-time approach. This approach is insufficient because it does not elucidate the effect of each individual factor, and the interactions between different factors cannot be followed (Box *et al.*, 1954). The purpose of the present work is to study the effect of some factors on the extraction of peanut total solids, protein, fat and carbohydrates by means of an experimental design approach so that the optimization of the process can be conceived.

MATERIALS AND METHODS

Peanuts

Peanuts (*Arachis hypogaea* L.), of unknown variety, imported from China, were purchased from the local market and used as the raw material for preparation of peanut milk throughout this work.

Papain

Papain (papainase; EC3.4.22.2, type III, Sigma Chemical Co., St. Louis, Mo) was used to hydrolyze peanut proteins.

Soaking

Peanut kernels were soaked in deionized water for 1 h at 20°C. A kernels-to-water ratio of 1:5 (w/v) was used. The soaked kernels were then taken from the water and stored at 5°C until used.

Experimental design

Seven factors, each at two levels, were examined. The change in the levels of factors was made in the direction considered more likely to increase the extractability of peanut constituents. Factors with their corresponding chosen symbols and levels are shown in Table 1. Sixteen experiments were carried out under varied operating conditions according to a two-level fractional factorial arrangement shown in Table 2 (Natrella, 1963). Each experiment was carried out only once. Experiments were performed in a random order.

Peanut milk preparation

Dry peanut kernels (5·00% moisture, 22·0% protein, 48·23% fat, 2·62% ash and 22·1% carbohydrates) or soaked (14·36% moisture, 20·4% protein, 48·05% fat, 2·23% ash and 14·9% carbohydrates) were decorticated by hand and ground twice into a meal in a kitchen meat mincer. The peanut milk

TABLE 1
Factors and Their Corresponding Symbols and Levels

Factor	*Symbol*	*Level*	
		Lower	*Higher*
pH	A	6·95 ± 0·02	8·00 ± 0·02
Extraction temperature (°C)	B	20 ± 2	60 ± 2
Extraction time (min)	C	15	40
Solids-to-water (w/v)	D	1:5	1:9
Speed of agitation (r/min)	E	5000	10000
Partial hydrolysis	F	Unhydrolyzed	Hydrolyzed
Soaking	G	Dry	Soaked

TABLE 2
Experimental Design Arrangement

	Level of factor[a]						
Exp.	*A*	*B*	*C*	*D*	*E*	*F*	*G*
1	−	−	−	−	−	−	−
2	+	−	−	−	+	−	+
3	−	+	−	−	+	+	+
4	+	+	−	−	−	+	−
5	−	−	+	−	+	+	−
6	+	−	+	−	−	+	+
7	−	+	+	−	−	−	+
8	+	+	+	−	+	−	−
9	−	−	−	+	−	+	+
10	+	−	−	+	+	+	−
11	−	+	−	+	+	−	−
12	+	+	−	+	−	−	+
13	−	−	+	+	+	−	+
14	+	−	+	+	−	−	−
15	−	+	+	+	−	+	−
16	+	+	+	+	+	+	+

[a] + = Factor is at its higher level.
− = Factor is at its lower level.

samples were prepared by suspending 15 g of the meal in either 75 or 135 ml of distilled water. The suspension pH was adjusted by 1N sodium hydroxide solution. In experiments where peanut proteins were partially hydrolyzed, papain was added to the suspension in a concentration of 1:50 (mg papain/mg peanut protein). The extraction temperature was kept constant by means of a temperature-controlled water bath. The suspension was agitated by an Ultra-Turax homogenizer (type: TP 18/10, 12·7 mm rotor diam.). Agitation was carried out for 5 min followed by 5 min at rest throughout the total extraction time. Table 2 shows the set of operating conditions assigned for each experiment. The mixture was then filtered in a vacuum filter through Munktell's filter paper, No. 3. The residual cake was discarded and the filtrate, referred to as peanut milk, was stored at −10°C until analyzed.

Analysis of peanut milk

Peanut milk samples were analyzed for protein content using a Kjeltic Auto 1030 Analyzer. Total solids content and ash were evaluated according to the methods of the Association of Official Analytical Chemists (AOAC,

1984). Fat content was determined by the method of the International Dairy Federation (IDF, 1969). Carbohydrate content was calculated by difference.

Statistical analysis

Results were expressed as percent extracted solids, protein, fat and carbohydrates. They were statistically analyzed using Yates' method of analysis (Natrella, 1963). Three-and-higher-order interactions were assumed negligible. Their combined mean squares was used as an estimate of the experimental error for testing the significance of factors at 5% confidence level.

RESULTS AND DISCUSSION

Composition of the peanut milk samples is shown in Table 3. The corresponding calculated responses: percent extracted solids, protein, fat

TABLE 3
Peanut Milk Composition

Exp.	*TSC*[a,b]	*Peanut milk composition (g/100 g)*			
		Protein[a,c]	*Fat*[a]	*Ash*[a]	*Carbohydrates*
1	10·82 ± 0·00	3·47 ± 0·00	4·27 ± 0·32	0·32 ± 0·00	2·76
2	10·59 ± 0·00	3·08 ± 0·00	4·41 ± 0·00	0·29 ± 0·02	2·81
3	8·19 ± 0·03	3·07 ± 0·02	3·16 ± 0·60	0·26 ± 0·03	1·70
4	10·01 ± 0·07	3·71 ± 0·03	4·32 ± 0·00	0·41 ± 0·01	1·57
5	10·78 ± 0·00	3·19 ± 0·02	5·44 ± 0·36	0·29 ± 0·00	1·86
6	8·30 ± 0·01	2·83 ± 0·00	3·59 ± 0·16	0·28 ± 0·00	1·60
7	8·86 ± 0·03	2·91 ± 0·01	2·72 ± 0·26	0·30 ± 0·00	2·93
8	13·29 ± 0·03	3·91 ± 0·00	6·03 ± 1·33	0·35 ± 0·03	3·00
9	4·70 ± 0·03	1·68 ± 0·00	1·97 ± 0·21	0·16 ± 0·04	0·89
10	7·18 ± 0·09	2·26 ± 0·00	3·52 ± 0·00	0·25 ± 0·00	1·15
11	7·55 ± 0·01	2·31 ± 0·05	3·84 ± 0·33	0·17 ± 0·02	1·23
12	5·02 ± 0·02	1·82 ± 0·01	1·42 ± 0·02	0·18 ± 0·00	1·60
13	6·71 ± 0·01	1·93 ± 0·00	3·03 ± 0·12	0·16 ± 0·02	1·59
14	5·12 ± 0·03	2·18 ± 0·00	1·80 ± 0·00	0·22 ± 0·03	0·92
15	5·60 ± 0·06	2·18 ± 0·01	2·26 ± 0·02	0·23 ± 0·01	0·93
16	5·73 ± 0·02	1·90 ± 0·00	2·75 ± 0·01	0·21 ± 0·02	0·87

[a] Values are means ± SD of three independent determinations.
[b] TSC = Total solids content.
[c] Kjeldahl nitrogen × 5·46 (Woodroof, 1969).

TABLE 4
Percent Extracted Solids, Protein, Fat and Carbohydrates

Exp.	*Per cent extracted*			
	Solids	*Protein*	*Fat*	*Carbohydrates*
1	56·64	78·3	44·03	62·1
2	63·23	77·1	46·93	96·2
3	45·37	71·3	31·20	54·0
4	52·87	84·5	44·94	35·6
5	68·80	87·8	68·39	51·0
6	52·96	75·7	40·82	58·6
7	47·05	64·8	25·75	89·3
8	57·13	72·5	51·06	55·4
9	52·19	78·2	38·99	56·9
10	67·19	91·2	64·88	46·2
11	65·32	86·2	65·44	45·7
12	53·20	80·9	26·82	97·3
13	68·12	82·1	54·82	92·6
14	48·66	89·3	33·70	37·6
15	53·27	89·4	42·35	38·0
16	58·67	81·6	50·18	51·1

and carbohydrates are shown in Table 4. A summary of statistical analysis giving the mean effect of each factor with respect to different responses is shown in Table 4. A negative mean effect of a factor indicates that changing this factor from its lower to higher level causes a decrease in the measured response.

TABLE 5
Summary of Statistical Analysis[a]

Factor	*Mean effect with respect to extracted:*			
	Solids	*Protein*	*Fat*	*Carbohydrates*
pH	−0·36 ns	1·82 s	−1·46 ns	−1·45 ns
Extraction temp.	−5·61 s	−3·59 s	−6·85 s	−4·35 ns
Extraction time	−0·17 ns	−0·56 ns	0·48 ns	−2·57 ns
Solids-to-water	2·82 s	8·37 s	3·01 ns	−4·60 ns
Speed of agitation	9·62 s	1·08 ns	16·94 s	2·14 ns
Partial hydrolysis	−1·00 s	3·56 s	4·15 ns	−23·08 s
Soaking	−3·64 s	−8·42 s	−12·41 s	28·04 s

[a] s = Significant at 5% level.
ns = Non-significant at 5% level.

pH

Peanut proteins are 90% globulins and 10% albumins with an isoelectric point between pH 3·00 and 4·00 depending on the type of extraction medium used (McWatters *et al.*, 1976). When the extraction medium does not contain ionic salts, the solubility of peanut globulins in water can be increased by setting the pH of the medium in the alkaline region (or acidic region, pH between 1·00 and 2·00). In our experiments, changing the pH of the suspension from its initial value 6·95 $\pm$ 0·02 (which was almost the same for all samples) to 8·00, caused significant increase in the extraction of the proteins. The fat recovery was decreased in that alkaline medium. This is indicated by the negative mean effect of the pH produced on the extraction of fat, and can be attributed to the fact that oil recovery from raw peanuts in aqueous systems was found to be maximum at pH 4·00 (Rhee *et al.*, 1972).

Extraction temperature

Table 5 indicates that the temperature is significant for the extraction of protein and fat. The higher temperature used (60°C) seems to be too high giving negative mean effects with respect to all responses. This may be attributed to the denaturation of the major peanut storage protein, arachin, due to exposure to heat at that temperature. Heat has been proposed to dehydrate protein molecules leading to peptide linkages between free amino and carboxyl groups. Conversely, cleavage of peptide bonds by heat leads to rearrangements of the peptide chains which can then react with each other or with macromolecules such as fats and carbohydrates giving rise to larger molecules (Neucere, 1972). Upon filtration, these large molecules were liable to be retained by the filter paper causing decreased recovery of protein, fat and carbohydrates.

Extraction time

Changing the time from 15 to 40 min made the time factor nonsignificant for the extraction of all peanut constituents. Practically, the time taken to adjust the pH of the suspension and to conduct filtration interferred with that assigned for a given experiment. This extended the lower time level to about 30 min. In the experiments where peanut proteins were partially hydrolyzed with papain, the time factor became a significant one for the protein extraction as concluded from the time–hydrolysis interaction shown in Table 6.

TABLE 6
Analysis of Time–Hydrolysis Interaction Based on the Percent of Protein Extracted[a]

Hydrolysis	*Protein extracted (%)*		
	15 min		*40 min*
Unhydrolyzed	78·3		64·8
	77·1		72·5
	86·2		82·1
	80·9		89·3
	322	Total	309
Hydrolyzed	71·3		87·8
	84·5		75·7
	78·2		89·4
	91·2		81·6
	325	Total	334

[a] Observations are taken from Table 4 in accordance with the design arrangement shown in Table 2.

Solids-to-water ratio

This ratio was found to be nonsignificant for fat and carbohydrates extraction. On the other hand, it was found to be very significant for the protein extraction with the second highest mean effect. Protein extraction could be enhanced if this ratio was increased as far as 1:20 (Rhee *et al.*, 1972), but this will be at the expense of suitable peanut milk composition. However, determination of a compromise ratio is a matter of optimization.

Speed of agitation

Increasing the speed of agitation produced the highest mean effect for total solids and fat recovery. This is because of the particle size reduction of fat globules due to increased shear rate, making many of them able to pass through the filter paper. Protein and carbohydrates were less affected by the change in the speed of agitation.

Partial hydrolysis

Partial hydrolysis of peanut proteins with papain significantly increased their extraction. Enzymatic hydrolysis of peanut proteins was reported to

increase their solubility in water due to degradation of large-molecular weight globulins, and to improve some of their functional properties most desired in beverage systems (Sekul & Ory, 1977; Sekul *et al.*, 1978). Enzymatic hydrolysis should be preferred to acid or base since hydrolysis by either acid or base might require that the final product be neutralized, thereby increasing salt concentration which is undesirable when the final product is destined to serve as a drink.

Soaking

Soaking is a physical process characterized by water uptake by the seed. It is the early step in seed germination. It is accompanied by activation of endogenous enzymes which act on seed storage materials giving rise to different vital changes in the seed contents. In this experiment, the extractions of protein and fat were depressed, and that of the carbohydrates was highly increased as a result of soaking (see Table 5). A lot of fibrous residue was obtained in the filter cake when peanut milk was prepared from soaked kernels. The effect of soaking time on the total solids content of peanut milk was examined in a separate experiment. Peanuts were soaked in deionized water (1:5 (w/v) kernels-to-water) at 20°C. Peanut milk samples

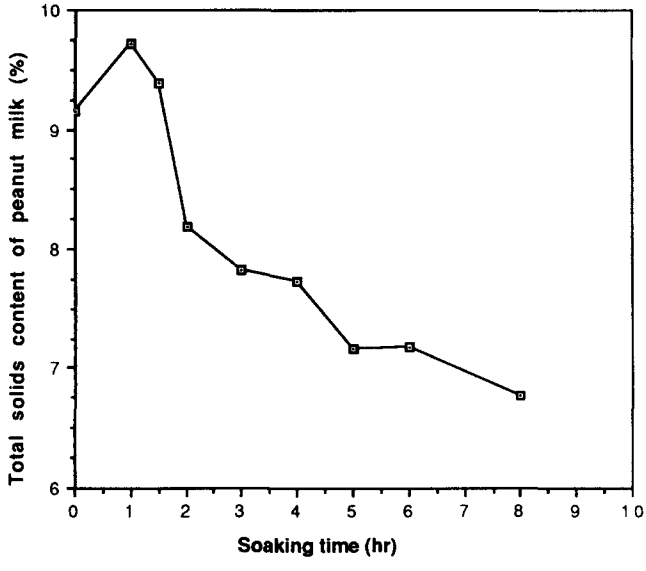

Fig. 1. Effect of soaking time on the total solids content of peanut milk. Peanut kernels were soaked in deionized water in a ratio of 1:5 (w/v) at 20°C. Peanut milk was prepared from soaked kernels using solids-to-water ratio = 1:6 (w/v), pH = 7·00, temperature = 20°C ± 2°C and extraction time = 30 min.

were then prepared as described before (1:6 solids-to-water, pH = 7·00, temperature = 20°C ± 2°C and extraction time = 30 min). The result is shown in Fig. 1, which indicates that a greater extraction yield might have been achieved if peanuts were soaked (in experiments 2, 3, 6, 7, 9, 12, 13 & 16) for less than 1 h.

Interactions

Using a fractional factorial design to reduce the number of experiments made the effects of two-factor interactions confounded with each other in the statistical analysis. Another way to help understand the nature of these interactions is by the construction of two-way tables. For example, Table 7 shows that the temperature–time interaction is significant for protein extraction. Increasing the temperature from 20°C to 60°C at 15 min decreased the protein extractability by 1·99 units, whereas it was decreased by 26·71 units when the time was 40 min. The hydrolysis of peanut proteins was found to depend on time and temperature as can be concluded from Tables 6 and 8, respectively. It was less affected by the change in the pH value (see Table 9).

TABLE 7
Analysis of Temperature–Time Interaction Based on the Percent of Protein Extracted[a]

Time (min)	*Protein extracted (%)*		
	20°C		*60°C*
15	78·3		71·3
	77·1		84·5
	78·2		86·2
	91·2		80·9
	325	Total	323
40	87·8		64·8
	75·7		72·5
	82·1		89·4
	89·3		81·6
	335	Total	308

[a] Observations are taken from Table 4 in accordance with the design arrangement shown in Table 2.

TABLE 8
Analysis of Temperature–Hydrolysis Interaction Based on the Percent of Protein Extracted[a]

Hydrolysis	*Protein extracted (%)*		
	20°C		*60°C*
Unhydrolyzed	78·3		64·8
	77·1		72·5
	82·1		86·1
	89·3		80·9
	327	Total	304
Hydrolyzed	78·2		71·3
	87·8		84·5
	75·7		89·4
	91·2		81·6
	333	Total	327

[a] Observations are taken from Table 4 in accordance with the design arrangement shown in Table 2.

TABLE 9
Analysis of pH–Hydrolysis Interaction Based on the Percent of Protein Extracted[a]

Hydrolysis	*Protein extracted (%)*		
	pH = 6·95		*pH = 8·00*
Unhydrolyzed	78·3		77·1
	64·8		72·5
	86·2		80·9
	82·1		89·3
	311	Total	320
Hydrolyzed	71·3		84·5
	87·8		75·7
	78·2		91·2
	89·4		81·6
	327	Total	333

[a] Observations are taken from Table 4 in accordance with the design arrangement shown in Table 2.

CONCLUSIONS

For the production of peanut milk and beverage, protein is the item of greatest interest. Moreover, its extraction was found in this work to be significantly dependent on five of the factors examined: pH, temperature, solids-to-water ratio, partial hydrolysis and soaking. The optimization of these factors can therefore be based on maximum protein extraction and suitable peanut milk composition.

REFERENCES

AOAC (1984). *Official Methods of Analysis.* (14th edn), Sect. 16032 and 16035. Association of Official Analytical Chemists, Washington, DC.

Beuchat, L. R. & Nail, B. J. (1978). Fermentation of peanut milk with *Lactobacillus bulgaricus* and *L. acidophilus. J. Food Sci.*, **43**(2), 1109–12.

Box, G. E. P., Connor, L. R., Cousins, W. R., Davies, O. L., Himsworth, F. R. & Sillitto, G. P. (1954). Factorial experiments: Elementary principles. In *Design and Analysis of Industrial Experiments.* (1st edn), ed. Owen L. Davies. Oliver & Boyd, London, pp. 247–89.

Bucker, E. R., Mitchell, J. H. & Johnson, M. G. (1979). Lactic fermentation of peanut milk. *J. Food Sci.*, **44**(2), 1534–8.

Conkerton, E. J. & Ory, R. L. (1988). Peanuts as food proteins. In *Developments in Food Proteins.* (5th edn), ed. J. F. Hudson. Elsevier Applied Science Publishers, London, pp. 1–29.

Elahi. M. M. & Ali, S. M. (1971). Studies on milk substitute from groundnut. *Pakistan J. Sci.*, **23**(3), 172–5.

FAO. (1986). FAO statistics series No. 76. *FAO Production Year Book*, **40**, 111–12.

IDF (1969). *International Standard, FIL-IDF 1A: 1969.* International Dairy Federation, Belgium.

McWatters, K. H. & Cherry, J. P. (1982). Potential food uses of peanut seed proteins. In *Peanut Science and Technology*, ed. H. E. Pattee & C. T. Young, American Peanut Research and Education, Inc. Yoakum, Texas, USA, pp. 689–736.

McWatters, K. H., Cherry, J. P. & Holmes, M. R. (1976). Influence of suspension medium and pH on functional and protein properties of defatted peanut meal. *J. Agric. Food Chem.*, **24**(3), 517–23.

Natrella, M. G. (1963). Factorial experiments. In *Experimental Statistics.* United States Department of Commerce, National Bureau of Standards, Handbook 91. Chapter 12.

Neucere, N. J. (1972). Effect of heat on peanut proteins. *J. Agric. Food Chem.*, **20**(2), 252–5.

Rhee, K. C., Cater, C. M. & Mattil, K. F. (1972). Simultaneous recovery of protein and oil from raw peanuts in an aqueous system. *J. Food Sci.*, **37**(1), 90–3.

Rubico, S. M., Resurreccion, A. V. A., Frank, J. F. & Beuchat, L. R. (1987). Suspension stability, texture, and color of high temperature treated peanut beverage. *J. Food Sci.*, **52**(6), 1676–9.

Schmidt, R. H. & Bates, R. P. (1976). Sensory acceptability of fruit flavored oilseed milk formulations. *Proc. Fla. State Hort. Soc.*, **89**, 217–19.

Schmidt, R. H., Surak, J. G. & Hausknecht, D. R. (1978). Particle size distribution in citrus flavored soybean and peanut milk beverages. *Proc. Fla. State Hort. Soc.*, **91**, 153–6.

Sekul, A. A. & Ory, R. L. (1977). Rapid enzymatic method for partial hydrolysis of oilseed proteins for food uses. *J. Am. Oil Chem. Soc.*, **54**, 32–5.

Sekul, A. A., Vinnett, C. H. & Ory, R. L. (1978). Some functional properties of peanut proteins partially hydrolyzed with papain. *J. Agric. Food Chem.*, **26**(4), 855–8.

Woodroof, J. G. (1969). Composition and use of peanuts in the diet. *World Review of Nutrition and Dietetics*, **11**, 142–69.