Effect of Germination on Electrophoretic, Functional, and Bread-Baking Properties of Yellow Pea, Lentil, and Faba Bean Protein Isolates[1]

D. L. HSU,[2] H. K. LEUNG,[2] M. M. MORAD,[2] P. L. FINNEY,[3] and C. T. LEUNG[4]

Cereal Chem. 59(5):344-350

ABSTRACT

The physicochemical properties of protein isolates from ungerminated and germinated yellow peas, lentils, and faba beans were investigated. Polyacrylamide gel electrophoretic patterns revealed different degrees of protein modification among the three legumes after four days of germination. The effect of germination on nitrogen solubility, emulsifying capacity, foam capacity and stability, viscosity, gelation, and water sorption properties of the protein isolates from the three legumes also varied. Replacement of wheat flour with 5 or 8% legume protein isolates had deleterious effects on loaf volume and crumb grain of bread. Germination affected the baking properties of protein isolates from faba beans, but not those from peas or lentils.

The potential application of legume flours and legume protein concentrates or isolates has received much attention in recent years (D'Appolonia 1977, Gwiazda et al 1979, Jeffers et al 1978, McWatters 1977, McWatters and Cherry 1977, Sosulski et al 1978). According to Mizrahi et al (1967), isolated proteins often improve appearance and taste compared with the original meal and therefore can be better utilized as nutritional and functional ingredients in food products. Thompson (1977) found that, although mung bean protein isolates had some adverse effects on dough and bread quality, acceptable breads were produced that were supplemented at the 10% level using sodium stearoyl-2-lactylate as a dough conditioner.

Protein quality of certain legumes improves as a result of germination (Bau and Debry 1979, El-Hag et al 1978, Kakade and Evans 1966, Subbulakshmi et al 1976). Protein modification that occurs during germination has been reported for peas (Basha and Beevers 1975), chick-peas (Kumar and Venkataraman 1978), mung bean (Coffman and Garcia 1977), and cottonseed (Cunningham et al 1978). However, very few studies have been done on the effect of germination on the functional properties of legume proteins.

Pomeranz et al (1977) reported that bread supplemented with 10% flour from germinated soybeans resulted in highly satisfactory bread quality. Finney (1977) showed that replacing 7% wheat flour with wet, mashed, germinated soybeans produced breads with no objectionable taste or odor. Finney et al (1980) replaced 15% of wheat flour with flours from germinated and ungerminated, dehulled faba beans. They produced acceptable breads using both the sugar and the sugar-free formulas. Hsu et al (1980) reported that germination adversely affected the baking properties of yellow peas and lentils but not faba beans. The adverse effect of germination on baking properties of legume flours is partly due to changes in starch (Morad et al 1980). The present study was designed to investigate the effect of germination on the physicochemical properties and bread-making performances of protein isolates from yellow peas (*Pisum sativum*), lentils (*Lens culinaris*), and faba beans (*Vicia faba*).

MATERIALS AND METHODS

Sample Preparation

Dry yellow peas (Latah) and lentils (Chilean) were obtained from Dumas Seed Company, Moscow, ID. A composite of three varieties of faba beans (Diana, Ackerperle, and Herz Freya) were donated by the Department of Plant Science, University of Manitoba, Winnipeg, Canada. Legumes were germinated for four days for this study. The germination process and the legume flour preparation were the same as those described by Hsu et al (1980).

The laboratory procedure for preparing the protein isolates from yellow pea, lentil, and faba bean flours is based on the protein isolation method of Sosulski et al (1978) with minor modification. To prepare protein isolates, 10% (w/v) dispersions of legume flours were made in distilled water, adjusted to pH 8.5 with 1*N* NaOH, and mixed with a magnetic stirrer for 30 min at 40° C. To prevent excessive foaming during extraction, an antifoaming agent, Dow Corning A Compound, was added to the flour dispersion. The extract was then separated from the insoluble residue by centrifuging at 220 × *g* for 15 min at 10° C, using a Beckman model J-21C refrigerated centrifuge. The extraction procedure was repeated once to increase protein yield. The pH of the combined extracts was adjusted to 4.5 with 1*N* HCl to precipitate the major proteins. The mixture was centrifuged at 1,570 × *g* for 10 min at 10° C, and the supernatant was separated from the residue by decantation. The surface of the protein curd was washed with distilled water and the curd redispersed in distilled water, adjusted to pH 7.5 with 1*N* NaOH, and dialyzed against running tap water for 24 hr at 10° C. The dialyzed protein was lyophilized.

Chemical Analyses

The chemical composition of wheat flour and protein isolates from germinated and ungerminated legumes were determined by the AACC approved methods 44-15A and 46-20 (1976). Moisture content was determined by the air-oven method (130° C for 60 min). Protein was determined by the macro-Kjeldahl procedure (N × 6.25).

Polyacrylamide Gel Electrophoresis

Sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) was performed according to Laemmli (1970) with some modifications. Slab gels instead of tube gels were used. Dimension of the slab gel was 10 cm (length) × 15 cm (width) × 0.75 mm (thickness). A custom-built apparatus similar to the Hoffer SE 500 Vertical Slab Unit was used. About 200 μg of protein was used for each sample. Bovine serum albumin (mol wt 68,000), alcohol dehydrogenase (mol wt 37,000), and carbonic anhydrase (mol wt 29,000) were used as marker proteins (Weber and Osborn 1969). The current was set at 20 mA per two slabs during stacking and 30 mA per two slabs during separation. After electrophoresis, the gels were placed in fixing solution containing 45% methanol, 10% acetic acid, and 45% water for 1 hr, and stained with 0.075% Coomassie blue R-250 in water for 2 hr. The gels were destained overnight in a solution containing 10% methanol, 10% acetic acid, and 80% water. The gels were dried using a Bio-Rad model 224 Gel Slab Dryer. Densitometric scans were recorded at 570 nm on a Beckman CDS-100-F Computing Densitometer System.

[1] Scientific Paper 6059, College of Agriculture Research Center, Washington State University, Pullman 99164.

Mention of a trademark or proprietary product does not constitute a guarantee or warranty of the product by the USDA and does not imply its approval to the exclusion of other products that may also be suitable.

[2] Department of Food Science and Technology, Washington State University, Pullman 99164.

[3] USDA, Agricultural Research Service, Western Wheat Quality Laboratory, Pullman, WA 99164.

[4] Department of Animal Science, Washington State University, Pullman 99164.

Nitrogen Solubility

The protein isolates from germinated and ungerminated legumes were weighed in duplicate (0.25 g, db) directly into 50-ml centrifuge tubes. Twenty-five milliliters of distilled water was added to each sample, and the mixture was magnetically stirred for 5 min to disperse the protein. The pH was adjusted to the desired values (pH 2, 3, 4, 5, 6, 8, 10, or 12) by adding either 0.1*N* or 1.0*N* HCl or NaOH. The centrifuge tubes containing the protein dispersions were supported by a rack and immersed in a water bath. The samples were mixed for 25 min at 30° C using stirring bars. The protein dispersions were centrifuged using a Beckman model J-21C centrifuge at 12,100 × *g* for 15 min, and the supernatants were decanted through Whatman no. 1 filter paper. Ten-milliliter aliquots of the filtrates were used for Kjeldahl nitrogen determinations with a Kjeltec System II nitrogen-analyzer unit. Nitrogen-solubility index was calculated as percentage water-soluble nitrogen divided by total nitrogen in the sample.

Emulsifying Capacity

Emulsifying capacity of protein isolates prepared from ungerminated and germinated legume were determined in triplicate according to the electrical resistance method described by Webb et al (1970) as modified by Eisele (1980). Emulsifying capacity was expressed as milliliters of oil emulsified per milligram of protein at 22° C.

Foam Capacity and Stability

To determine the foam capacity and stability of protein isolates from germinated and ungerminated yellow peas, lentils, and faba beans, 30-ml aqueous suspensions of 3% (w/v) protein isolates were gently dispersed in an 800-ml beaker. The suspensions were whipped at high speed for 3.5 min using a Braun AG type M140 portable electric mixer. The mixer was mounted on a ring stand and connected to a timer so that beater position and whipping time were constant. The resulting mixtures were transferred into 100-ml graduated cylinders, and the total volumes were recorded 30 sec after whipping. The percent increase in volume was calculated based on the volume before and after whipping (Lawhon et al 1972).

Foam samples were allowed to stand in the cylinders at room temperature (22° C) for 1 hr, and the volumes of the liquid were recorded after 30, 60, and 120 min. Percent syneresis, which is inversely related to foam stability, was calculated according to Satterlee et al (1975).

Egg white and a commercial soy protein isolate, promine D (Central Soya Co., Chicago, IL), were used as references. The soy protein was treated the same way as all legume samples throughout the whipping test. Egg white at room temperature (22° C) was beaten slightly to blend the thin and thick albumins before a 30-ml volume was transferred to the 800-ml beaker for whipping. It was whipped at high speed for 30 sec to produce a soft peak foam for the evaluation of foaming properties. All whipping and foam stability tests were conducted in duplicate.

Viscosity and Gelation

Viscosity and gelation characteristics of slurries of protein isolates from ungerminated and germinated yellow peas, lentils, and faba beans were evaluated by comparison with a soy protein isolate (promine D) as control. Duplicate samples of protein slurries were prepared by blending 10% (w/v) legume protein isolates in distilled water for 10 min in a Sorvall Omni-Mixer at 8,000 rpm. The slurries were centrifuged at 3,000 × *g* for 2 min at room temperature to eliminate the foam. The viscosities of these unheated slurries were determined at 22° C with a Brookfield RVT viscometer with a number 2 spindle at 50 rpm. Three viscosity readings were taken for each sample.

Gelation experiments were conducted by heating and stirring the protein slurries in covered beakers to 90° C for 10 min in a water bath. The heated slurries were cooled to 25° C and stored overnight at 4° C. After equilibrating to 22° C, the viscosity of the heated samples was determined with the Brookfield Helipath Stand and the T-spindles operating at 2.5 or 5 rpm. Gelation characteristics of the legume protein dispersions were also evaluated visually.

Water Adsorption

Water adsorption of the protein isolates from ungerminated and germinated legumes was determined by exposing the dry protein samples to 85% relative humidity (rh) (Hagenmaier 1972). The protein isolates were dried under vacuum at 50° C for 48 hr. Duplicate samples of about 1 g each were weighed in small petri dishes and placed in a desiccator over saturated KCl slurry (Troller 1977). The desiccator was evacuated for 15 min to facilitate equilibration. The samples were equilibrated at room temperature (22° C) for two weeks. The weight gain was used to determine the equilibrium moisture content of the sample.

Baking Studies

The wheat flour used throughout the study was an unmalted, commercial, straight grade baker's flour with a medium mixing time of 4 min and good loaf volume potential (Centennial Mills, Spokane, WA). The 10-g mixograph was used to estimate physical dough properties including mixing time, mixing tolerance, and water absorption of legume protein-wheat flour blends according to Finney and Shogren (1972).

The microbake method developed by Shogren et al (1969) and the short-time sugar-free system of Magoffin et al (1977) were combined in this study. The method employed optimum mixing time, water absorption, potassium bromate (2.5–3.0 ppm), and a formula that included 10-g legume protein-wheat flour blends (14% moisture content), 0.76 g of fresh baker's yeast, 0.15 g of NaCl, 0.3 g of vegetable shortening, water extract from 0.06-g Ross malted barley (50.0 DU/g, 20° C), and 75 ppm ascorbic acid. Nonfat dry milk (0.4 g) was included only in the wheat flour control formula. The legume proteins were formulated on a replacement basis at 3, 5, or 8%. Doughs were fermented for 70 min and proofed for 55 min at 30° C. Doughs were punched after 40 and 60 min, and immediately before panning. Breads were baked at 232° C for 14 min and weighed. Loaf volume was measured by rapeseed displacement. Crumb grain, crust color, and overall appearance of breads were subjectively evaluated by members of the SEA-USDA Western Wheat Quality Laboratory. All analyses were conducted in duplicate.

RESULTS AND DISCUSSION

Chemical Analyses

Moisture and protein content of the wheat flour was 10.1 and 11.7%, respectively. Moisture, protein content, and percent protein recovery of the protein isolates from ungerminated and germinated yellow pea, lentil, and faba bean flours are given in Table I. The freeze-dried protein isolates contained 86.3–92.5% protein. Approximately 70% of the total protein in the legume flours was recovered after the extraction. The protein recoveries from lentil flour were slightly lower than those from faba bean and yellow pea flours. These data were comparable to those reported by Fan and Sosulski (1974), who used a similar extracting procedure to prepare

TABLE I
Moisture, Protein Content, and Protein Recovery of Legume Protein Isolates

Protein Isolates	Moisture (%)	Protein[a] (%)	Protein Recovery (%)
Yellow pea			
Ungerminated	5.8	87.3	73.6
Germinated	6.2	87.2	73.6
Faba bean			
Ungerminated	5.0	92.3	73.7
Germinated	6.4	92.5	74.0
Lentil			
Ungerminated	6.0	86.3	68.5
Germinated	5.2	87.6	70.1

[a] N × 6.25, dry-weight basis.

protein isolates from various legume flours. The protein contents of isolates from ungerminated and germinated legumes showed little difference.

Gel Electrophoresis

The SDS-PAGE patterns of faba bean proteins indicated seven subunits with molecular weights of 30,000–70,000 (Fig. 1), whereas pea and lentil proteins contain about twice as many subunits. The protein modification in the three legumes during germination showed most change for lentils but least change for faba beans. For yellow pea and lentil proteins, small subunits generally increase as a result of four-day germination. Using SDS-PAGE to study changes in chick-pea proteins, Kumar and Venkataraman (1978) observed a progressive decrease in large protein subunits and

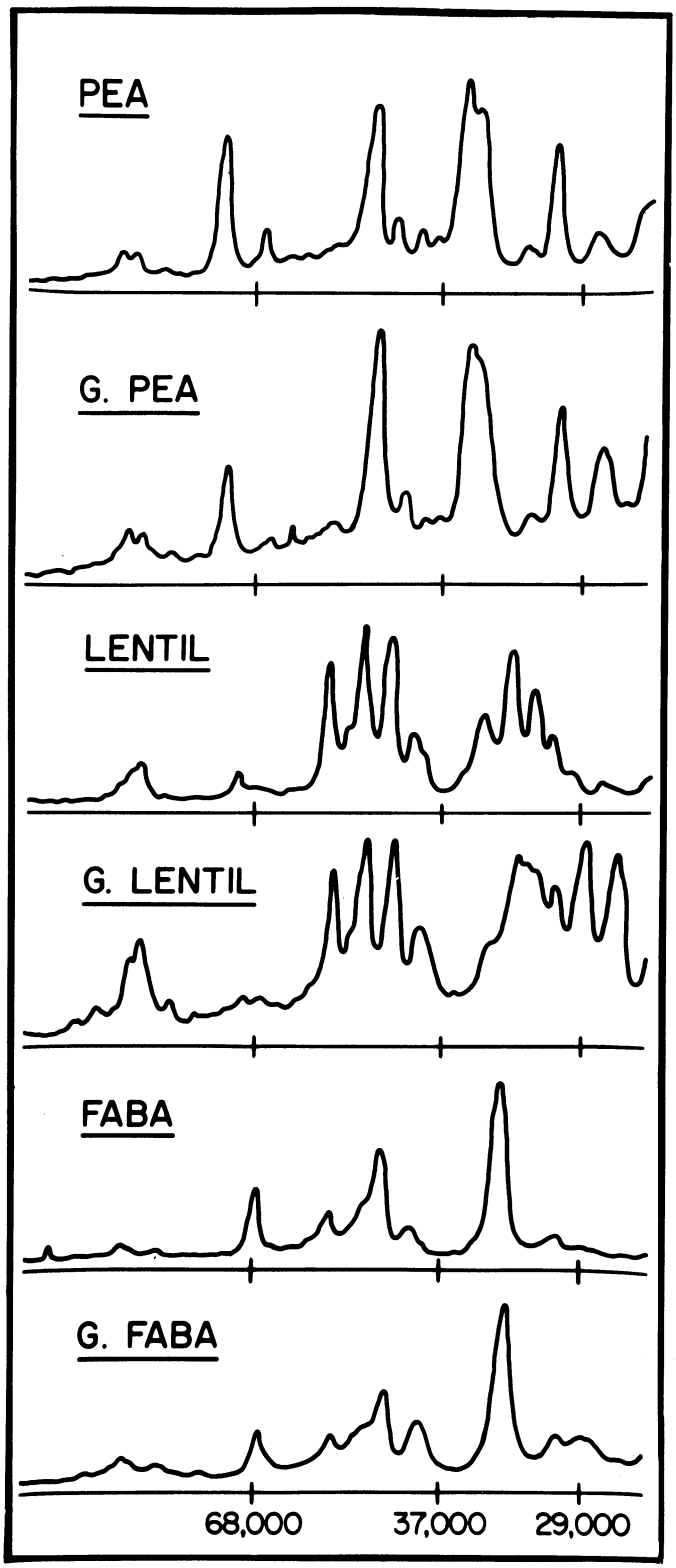

Fig. 1. Densitometric scans of sodium dodecyl sulfate-polyacrylamide gel electrophoresis patterns of protein isolates from germinated (G) and ungerminated yellow peas, lentils, and faba beans. Molecular weights of the marker proteins are indicated at the bottom of the figure.

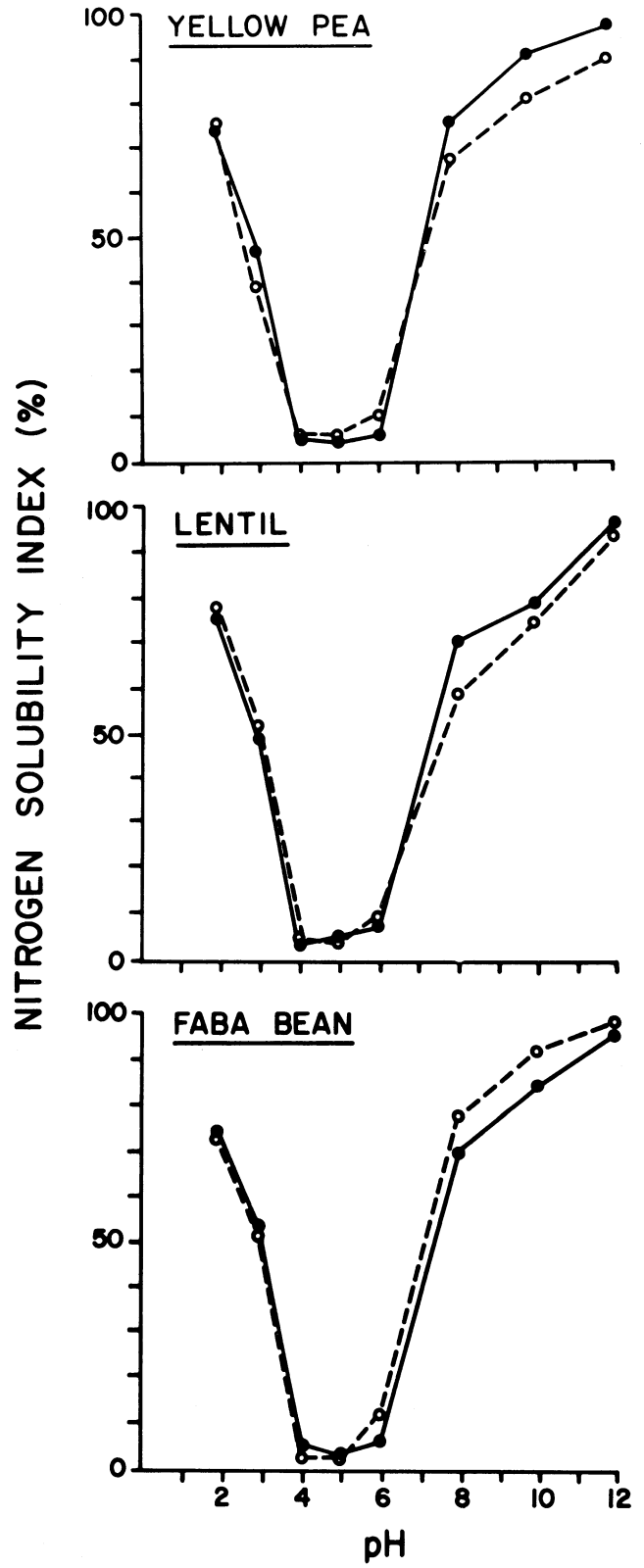

Fig. 2. Nitrogen-solubility index of protein isolates from germinated and ungerminated yellow peas, lentils, and faba beans as a function of pH. --o-- = germinated, —●— = ungerminated.

formation of small subunits as germination progressed. Findings by Basha and Beevers (1975) using SDS-PAGE also indicated that pea seed globulins undergo modifications before eventual hydrolysis during germination.

Nitrogen Solubility

The pH-solubility profiles were similar among the three legume protein isolates and had only minor differences (Fig. 2). They all exhibited a relatively broad apparent isoelectric pH range between 4 and 6. These results were in general agreement with earlier reports of Fan and Sosulski (1974) on nitrogen extraction of various legume flours. Similar findings were also reported for mung bean protein isolates (Coffman and Garcia 1977, Thompson 1977).

The effect of germination on the nitrogen solubility was small for all three legume protein isolates. Protein isolates from germinated faba beans showed an increase in nitrogen solubility above pH 6. Some decreases in solubility in the alkaline range were observed in protein isolates from germinated peas and lentils. Germination seemed to have little effect on the isoelectric point of the protein isolates.

Emulsifying Capacity

The emulsifying capacities of legume proteins were relatively poor at pH 6.5 but were improved at pH 7.5 (Table II). This is probably due to increased protein solubility as the pH was raised above the apparent isoelectric range of the legume proteins. In general, the emulsifying capacities of faba bean proteins were larger than those of yellow pea and lentil proteins. The emulsifying capacities of legume protein isolates increased after germination. The emulsifying capacity of protein isolate from germinated faba beans was the greatest among the six legume samples. Emulsifying properties apparently were influenced by the type of seed and protein modification as a result of germination.

Foam Capacity and Stability

The foaming properties of the legume proteins, soy protein isolate, and egg white are given in Table III. Soybean protein isolate produced smooth egg-whitelike foam. Although its foam capacity, measured by percentage volume increase, was less than that of egg white foam, the soy protein foam was more stable upon standing than the egg-white foam. Compared to soy protein isolate, the protein isolates from the ungerminated legumes had coarser air cells, smaller percentage volume increases after whipping, and slightly more syneresis, indicating weaker foam structure. Protein isolates from germinated legumes showed increased foam expansion and decreased foam stability compared to the ungerminated samples. These changes were most noticeable in lentils and were relatively minor in faba beans (Table III).

Viscosity and Gelation

Results of apparent Brookfield viscosity measurements are presented in Table IV. All of the unheated slurries except soy protein isolate had low viscosity. Protein isolates from germinated

TABLE II
Emulsifying Capacity of Legume Protein Isolates

Protein Isolates	Emulsifying Capacity[a] pH 6.5	pH 7.5
Yellow pea		
Ungerminated	1.17 ± 0.05 a[b]	1.30 ± 0.03 a
Germinated	1.25 ± 0.02 a	1.40 ± 0.01 b
Faba bean		
Ungerminated	1.21 ± 0.02 a	1.41 ± 0.02 b
Germinated	1.32 ± 0.03 b	1.55 ± 0.04 c
Lentil		
Ungerminated	1.06 ± 0.03 c	1.17 ± 0.03 d
Germinated	1.24 ± 0.03 a	1.35 ± 0.04 ab

[a] Milliliters of oil/milligrams of protein.
[b] Expressed as means of triplicates ± standard deviations. Values in a column followed by the same letter are not significantly different at $P = 0.05$.

TABLE III
Foam Capacity and Stability of Legume Protein Isolates

Protein Isolates	Volume Increase (%)	Syneresis (%) 30 min	60 min	120 min
Egg White	330	30.0	53.0	64.9
Soybean (promine D)	230	29.5	49.0	55.3
Yellow pea				
Ungerminated	116	33.0	54.0	62.2
Germinated	212	75.5	80.0	79.9
Faba Bean				
Ungerminated	100	50.0	54.0	58.4
Germinated	134	57.0	64.5	71.3
Lentil				
Ungerminated	125	45.5	54.5	63.6
Germinated	297	61.5	68.5	85.9

TABLE IV
Apparent Viscosity of Legume Protein Slurries Before and After Heating and Cooling

Protein Isolates	Brookfield Viscosity[a] (centipoise) Unheated[b]	Heated and Cooled	Appearances of Slurries After Heating and Cooling Gelation	Syneresis
Soybean (promine D)	314.4 ± 2.0	116.5M ± 3.1M[c] (T-B)[d]	Smooth, soft gel	No
Yellow pea				
Ungerminated	23.1 ± 0.8	34.4M ± 3.0M (T-B)	Smooth paste	Slight
Germinated	16.0 ± 0	24.0M ± 1.4M (T-B)	Granular curd	Severe
Faba bean				
Ungerminated	29.7 ± 0.6	30.4M ± 1.8M (T-B)	Smooth slurry	Little
Germinated	24.0 ± 2.4	377.0M ± 4.6M (T-D)	Soft curd, custardlike	Little
Lentil				
Ungerminated	28.1 ± 0.3	440.0M ± 10M (T-E)	Medium-firm curd	Little
Germinated	21.2 ± 0.8	43.2M ± 3.2M (T-B)	Coarse paste	Severe

[a] Values are averages of three readings on duplicate samples ± standard deviations.
[b] Spindle no. 2.
[c] $M = 1{,}000$.
[d] Spindle type.

faba beans, yellow peas, and lentils consistently exhibited lower apparent viscosities than their ungerminated counterparts. Viscosity is a useful index of structural changes in proteins and, subsequently, of hydrodynamic properties of modified food proteins (Kinsella 1976). The differences in viscosity and gelation characteristics observed between protein isolates from ungerminated and germinated legumes are indications of enzymatic modification of proteins during germination.

The viscosity of soy protein isolate was about 12 times greater than that of faba bean, yellow pea, and lentil protein isolates. Obviously, the protein conformations in these legume isolates do not facilitate hydration as readily as those of soybean isolate. Soy isolate promine D has been reported to contain spherical aggregates in which the majority of the hydrophilic groups are on the surface and easily undergo hydration (Gwiazda et al 1979).

The viscosity of all slurries increased markedly after heating and subsequent cooling. The apparent Brookfield viscosity and appearances of the legume slurries varied greatly after the heating and cooling treatment. The soy protein isolate slurry formed a stable soft gel with a pectin gel consistency. This gel had a relatively high viscosity. The lentil protein slurry formed a medium-firm curd resembling a custard with slight syneresis. The viscosity of the lentil protein gel was much greater than that of soy isolate. Neither faba bean nor yellow pea protein isolates formed gels. These heated slurries were pastelike after cooling. These observations are different from those of Fleming et al (1975), who found that faba bean and pea protein isolates formed weak gels when heated to 90° C for 45 min. The differences could be caused by variations in extraction and gelation procedures.

Germination had varying effects on the gelation characteristics of legume proteins. Protein isolate from germinated faba beans formed a soft, smooth curd with a 12-fold increase in viscosity over its ungerminated counterpart. Heated protein isolate slurries of germinated yellow peas and lentils exhibited severe syneresis.

TABLE V
Equilibrium Moisture Contents of Legume Protein Isolates at 85% Relative Humidity

Protein Isolates	Water Adsorbed[a]
Soybean (promine D)	0.24
Yellow pea	
Ungerminated	0.20
Germinated	0.19
Faba bean	
Ungerminated	0.19
Germinated	0.20
Lentil	
Ungerminated	0.20
Germinated	0.16

[a] Grams of water/grams of solid.

Water Adsorption

Equilibrium moisture content is defined as the water vapor adsorbed by a dry substance after equilibration at a known relative humidity. The mean values for water adsorbed by the protein isolates at 85% rh are presented in Table V. The equilibrium moisture contents of the legume protein isolates were approximately 20% (db) except for germinated lentil protein (16% db). Soy protein adsorbed more water (0.24 g/g of solid) than did the other legume proteins. These data are consistent with the viscosity and foaming properties discussed earlier. Soy protein isolate is more hygroscopic than the legume protein isolates. Thus, it was readily hydrated in water, resulting in aqueous dispersion with greater viscosity than the other legume protein samples. The more viscous protein dispersion of soy protein isolate yielded greater foam stability than the legume protein isolates. On the other hand, protein isolate from germinated lentils, which absorbed less water than other protein isolates, generally had lower viscosity, poorer gelation property, and less foam stability.

Physical Dough Tests

Supplementing wheat flour with legume protein isolate did not influence the water absorption of the flour blend at the 3 and 6% levels but caused a slight decrease at the 8% level, with the exception of ungerminated faba bean protein (Table VI and Fig. 3). All legume protein-wheat flour blends had longer mixing times

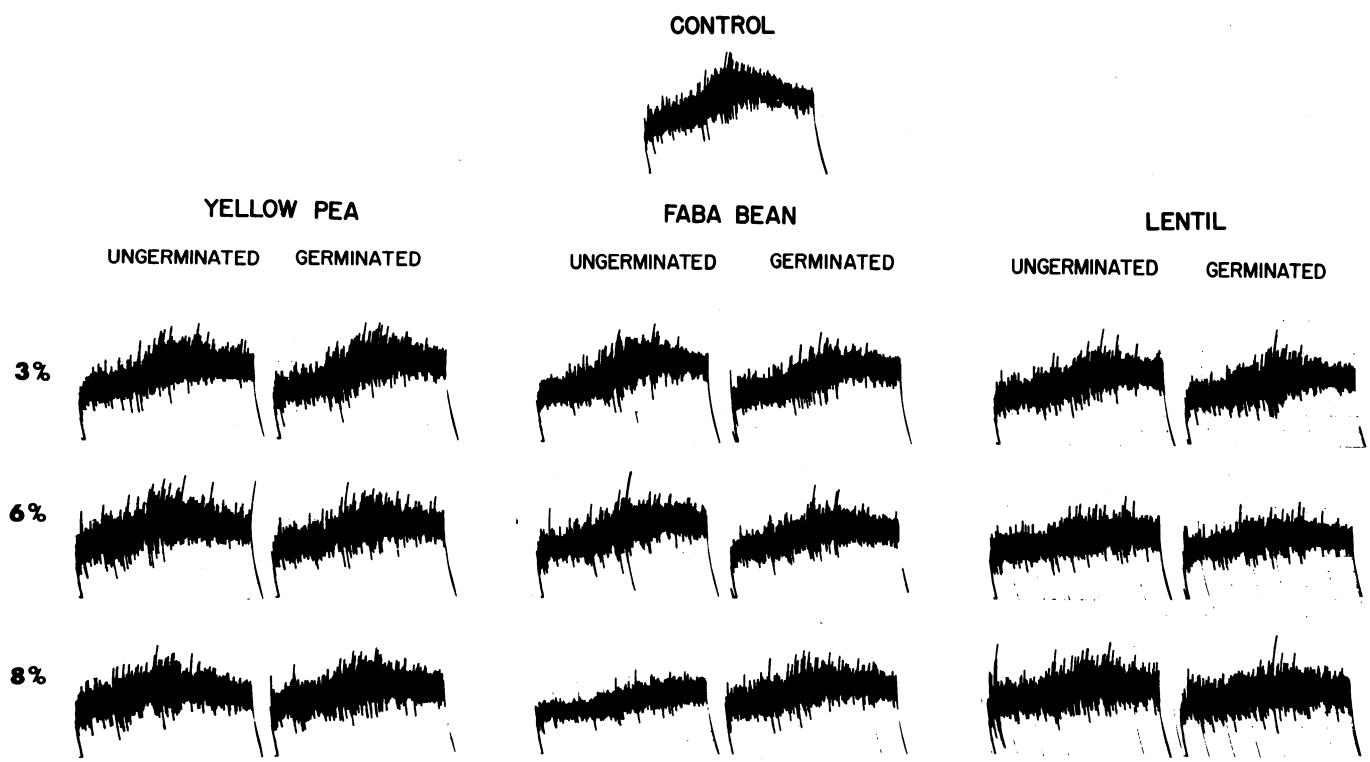

Fig. 3. Mixograms (10-g) for wheat flour-protein isolates from germinated and ungerminated legumes at 3, 6, and 8% replacement concentrations.

than that of the wheat flour control. Replacing wheat flour with legume protein isolates affected dough strength as indicated by the decreased mixogram peak heights. Some differences in mixing time were found between germinated and ungerminated samples.

Baking Studies

The data obtained from comparative baking studies on wheat flour supplemented with protein isolates from germinated and ungerminated legumes are shown in Table VII. The standard deviation for loaf volume based on duplicate samples was 2.1 cc for all flour blends. A difference of 5 cc or more in loaf volume is significant at $P = 0.05$. Slightly deleterious effects on loaf volume and crumb grain were observed in breads made from flour blends supplemented with 3% legume protein isolates. Replacement of wheat flour with 8% legume proteins reduced the baking absorption markedly and adversely affected the bread-making performance of the flour blends, resulting in decreased loaf volume and unsatisfactory crumb grain (Fig. 4 and Table VII). The bread supplemented with legume protein showed decreased crust browning with increased supplementation. At 8% supplementation, the breads had a noticeably pale crust color. Replacing part of the wheat flour with legume protein in the fortified blends decreased the reducing sugars necessary for Maillard browning. Unlike the wheat flour control, the legume protein-wheat flour blends did not contain nonfat dry milk. Therefore, lactose was not available to help produce a brown crust.

In general, baking properties of protein isolates from germinated and ungerminated faba beans were better than those from peas or lentils. Germination had an adverse effect on the baking properties of protein isolates from faba bean at 5 and 8% replacement concentrations but had little effect on pea or lentil protein isolates. The clear difference in baking properties between protein isolates from germinated and ungerminated faba beans is surprising because SDS-PAGE patterns of faba bean protein isolates showed little changes after germination. Kumar and Venkataraman (1978) noted that changes in electrophoretic patterns of seed proteins as a result of germination may be difficult to interpret because different protein components may have similar electrophoretic mobilities. Some changes in faba bean proteins during germination may not have been detected by the SDS-PAGE used in this study. Changes in minor components other than protein in the isolates may also account for some of the changes in functional properties.

TABLE VI
Mixograph Characteristics of Legume Protein-Wheat Flour Blends

Flour Blends	Mixing Time (min)	Peak Height (unit)	Water Absorption (%)
Control (commercial standard)	3.5	5.5	68.0
Ungerminated			
Yellow pea (%)			
3	4.5	5.0	68.0
6	4.5	4.8	68.0
8	4.2	4.5	67.0
Faba bean (%)			
3	4.0	5.2	68.0
6	5.0	4.5	68.0
8	5.5	4.2	68.0
Lentil (%)			
3	5.0	4.8	68.0
6	5.5	4.5	68.0
8	5.0	4.2	67.0
Germinated			
Yellow pea (%)			
3	5.0	5.2	68.0
6	5.0	5.0	68.0
8	4.8	4.4	67.0
Faba bean (%)			
3	4.2	5.0	68.0
6	4.2	5.0	68.0
8	4.5	4.5	67.0
Lentil (%)			
3	4.0	4.5	68.0
6	4.5	4.0	68.0
8	4.5	4.0	67.0

TABLE VII
Baking Data of Legume Protein-Wheat Flour Blends

Flour Blends	Baking Absorption[a] (%)	Loaf Volume (cc)	Crumb Grain[b]	Crust Color[c]
Control				
(commercial standard)	65.7	95	S	5.0
Ungerminated				
Yellow pea (%)				
3	64.7	84	S	3.5
5	65.7	72	Q-S	3.0
8	61.7	51	U	2.5
Faba bean (%)				
3	63.7	87	S	3.5
5	65.7	83	S	3.0
8	61.7	76	Q-S	3.0
Lentil (%)				
3	64.7	86	Q-S	3.5
5	65.7	70	Q-S	3.0
8	61.7	50	U	2.0
Germinated				
Yellow pea (%)				
3	63.7	87	Q-S	3.5
5	65.7	69	Q	3.0
8	60.7	52	U	2.5
Faba bean (%)				
3	63.7	91	S	3.5
5	65.7	76	S	2.5
8	60.7	64	Q-U	2.5
Lentil (%)				
3	64.7	85	Q	3.0
5	65.7	67	U	2.0
8	61.7	50	U	1.5

[a] 14% moisture basis.
[b] S = satisfactory, Q = questionable, U = unsatisfactory.
[c] Crust color rated subjectively on a 1–10 scale with increasing darkness.

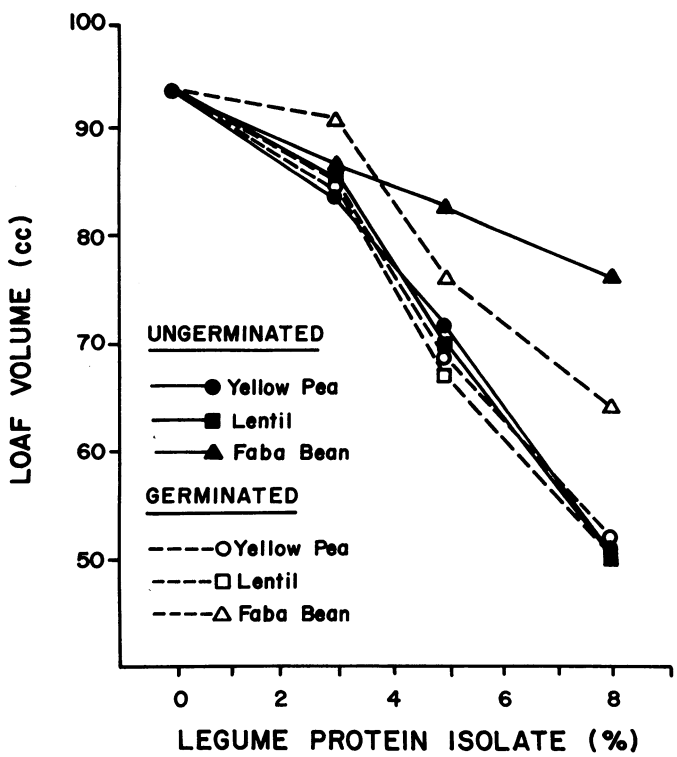

Fig. 4. Effect of legume protein supplementation on loaf volume of bread.

CONCLUSION

PAGE patterns of the legume protein isolates with SDS revealed definite changes after four days of germination for pea and lentil, but only slight modification for faba bean. Germination had small effects on emulsifying capacity and nitrogen solubility of the legume protein isolates. Foam capacity of the legume protein isolates increased substantially, but foam stability decreased as a result of seed germination. Viscosity, gelation, and water sorption properties of the protein isolates varied among the germinated and ungerminated legumes. Bread-baking properties of protein isolates from germinated and ungerminated faba beans generally were better than those from peas or lentils. Replacement of wheat flour by legume protein isolates at 5 and 8% resulted in deleterious effects on bread quality. Germination had an adverse effect on the baking properties of protein isolates from faba beans but not on those from peas or lentils. The marked changes in some functional properties of protein isolates from germinated and ungerminated faba beans cannot be explained by their SDS-PAGE patterns, which showed little protein modification after germination.

LITERATURE CITED

AMERICAN ASSOCIATION OF CEREAL CHEMISTS. 1976. Approved Methods of the AACC. Methods 44-15A and 46-20, approved October 1976; and Method 46-10, approved April 1961. The Association, St. Paul, MN.

BASHA, S. M. M., and BEEVERS, L. 1975. The development of proteolytic activity and protein degradation during the germination of *Pisum sativum* L. Planta 124:77.

BAU, H. M., and DEBRY, G. 1979. Germinated soybean protein products: Chemical and nutritional evaluation. J. Am. Oil Chem. Soc. 56:160.

COFFMAN, C. W., and GARCIA, V. V. 1977. Functional properties and amino acid content of a protein isolate from mung bean flour. J. Food Technol. 12:473.

CUNNINGHAM, S. D., CATER, C. M., and MATTIL, K. F. 1978. Effects of germination on cottonseed proteins. J. Food Sci. 43:102.

D'APPOLONIA, L. 1977. Rheological and baking studies of legume-wheat flour blends. Cereal Chem. 54:53.

EISELE, T. A. 1980. Chemical, functional and nutritional properties of acylated beef heart myofibrillar protein. Ph.D. thesis. Washington State University, Pullman, WA.

El-HAG, N., HAARD, N. F., and MORSE, R. E. 1978. Influence of sprouting on the digestibility coefficient trypsin inhibitor and globulin proteins of red kidney bean. J. Food Sci. 43:1874.

FAN, T. Y., and SOSULSKI, F. W. 1974. Dispersibility and isolation of proteins from legume flours. Can. Inst. Food Sci. Technol. J. 7:256.

FINNEY, K. F., and SHOGREN, M. D. 1972. A ten-gram mixograph for determining and predicting functional properties of wheat flours. Bakers Dig. 46(2):32.

FINNEY, P. L. 1977. A review of older and some newer short-time bread baking studies. Bakers Dig. 51(5):81.

FINNEY, P. L., MORAD, M. M. and HUBBARD, J. D. 1980. Germinated and ungerminated faba bean (*Vicia faba*) in sugar and sugar-free conventional U.S. and Egyptian Balady breads. Cereal Chem. 57:267.

FLEMING, S. E., SOSULSKI, F. W., and HAMON, N. W. 1975. Gelation and thickening phenomena of vegetable protein products. J. Food Sci. 40:805.

GWIAZDA, S., RUTKOWSKI, A., and KOCON, J. 1979. Some functional properties of pea and soybean protein preparations. Nahrung 23:681.

HAGENMAIER, R. 1972. Water binding of some purified oilseed proteins. J. Food Sci. 37:965.

HSU, D., LEUNG, H. K., FINNEY, P. L., and MORAD, M. M. 1980. Effect of germination on nutritive value and baking properties of dry peas, lentils, and faba beans. J. Food Sci. 45:87.

JEFFERS, H. C., RUBENTHALER, G. L., FINNEY, P. L., ANDERSON, P.D., and BRUINSMA, B. L. 1978. Pea: A highly functional fortifier in wheat flour blends. Bakers Dig. 52(3):36.

KAKADE, M. L., and EVANS, R. J. 1966. Effect of soaking and germinating on the nutritive value of navy beans. J. Food Sci. 31:781.

KINSELLA, J. E. 1976. Functional properties of proteins in foods: A Survey. Crit. Rev. Food Sci. Nutr. 7:219.

KUMAR, K. G., and VENKATARAMAN, L. V. 1978. Chick-pea seed proteins: Modification during germination. Phytochem. 17:605.

LAEMMLI, U. K. 1970. Cleavage of structural proteins during the assembly of the head of Bacteriophase T4. Nature 227:680.

LAWHON, J. T., CATER, C. M., and MATTIL, K. F. 1972. A comparative study of the whipping potential of an extract from several oilseed flours. Cereal Sci. 17:240.

MAGOFFIN, C. D., FINNEY, P. L., and FINNEY, K. F. 1977. Short-time baking systems. II. A 70-min sugar-free formula for conventional and high-protein breads. Cereal Chem. 54:760.

McWATTERS, K. 1977. Performance of defatted peanut, soybean and field pea meals as extenders in ground beef patties. J. Food Sci. 42:1492.

McWATTERS, K., and CHERRY, J. 1977. Emulsification, foaming and protein solubility properties of defatted soybean, peanut and field pea and pecan flours. J. Food Sci. 42:1444.

MIZRAHI, S., ZIMMERMAN, G., BERK, Z., and COGAN, U. 1967. The use of isolated proteins in bread. Cereal Chem. 44:193.

MORAD, M. M., LEUNG, H. K., HSU, D. L., and FINNEY, P. L. 1980. Effect of germination on physicochemical and bread-baking properties of yellow pea, lentil, and faba bean flours and starches. Cereal Chem. 57:390.

POMERANZ, Y., SHOGREN, M. D., and FINNEY, K. F. 1977. Flour from germinated soybeans in high protein bread. J. Food Sci. 42:824.

SATTERLEE, L. D., BEMBERS, M., and KENDRICK, J. G. 1975. Functional properties of the Great Northern bean (*Phaseolus vulgaris*) protein isolates. J. Food Sci. 40:81.

SHOGREN, M. D., FINNEY, K. F., and HOSENEY, R. C. 1969. Functional (bread-making) and biochemical properties of wheat flour components. I. Solubilizing gluten and flour protein. Cereal Chem. 46:93.

SOSULSKI, F. W., CHAKRABORTY, P., and HUMBERT, E. S. 1978. Legume-based imitation and blended milk products. Can. Inst. Food Sci. Technol. J. 11:117.

SUBBULAKSHMI, G., GANESHKUMAR, K., and VENKATARAMAN, L. V. 1976. Effect of germination on the carbohydrates, proteins, trypsin inhibitor, amylase inhibitor and hemagglutinin in horsegram and mothbean. Nutr. Rep. Int. 13:19.

THOMPSON, L. U. 1977. Preparation and evaluation of mung bean protein isolates. J. Food Sci. 42:202.

TROLLER, J. A. 1977. Statistical analysis of A_w measurements obtained with the Sina scope. J. Food Sci. 42:86.

WEBB, N. B., IVEY, F. J., CRAIG, H. B., JONES, V. A., and MONROE, R. J. 1970. The measurement of emulsifying capacity by electrical resistance. J. Food Sci. 35:501.

WEBER, K., and OSBORN, M. 1969. The reliability of molecular weight determinations by dodecyl sulfate-polyacrylamide gel electrophoresis. J. Biol. Chem. 244:4406.

[Received November 2, 1981. Accepted February 22, 1982]