

FORTIFICATION OF SOME BAKERY PRODUCTS WITH PROTEIN CONCENTRATES

By

WAFAA KAMEL BAHGAAT

B.Sc. Agric. Sc. (Food Science), Ain Shams University, 1991

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Under the supervision of:

Prof. Dr. Nagwa Mosa Hassan Rasmy

Prof. of Food Science and Technology, Department of Food Science,
Faculty of Agriculture, Ain Shams University (Principal Supervisor)

Prof. Dr. Mohamed Abd El-Gelil Khorshid

Prof. of Dairy Science, Department of Dairy Science, Food Technology
& Nutrition Division, National Research Center

Dr. Gamal A.A. El-Shatanovi

Associate Prof. of Food Science and Technology, Department of Food
Science, Faculty of Agriculture, Ain Shams University

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1. INTRODUCTION

Cereals remain the dominant vegetable protein source in the human diet, although they have a protein content of only about 10-12%. In the developing world, about 80-90% or even more of the protein intake is represented by cereal proteins (**Hambraeus, 1980**). As the protein value of cereal is not very high, the addition of either legume products or whey protein concentrates is a way to improve the diet of low-income population (**Renz-Schauen and Renner, 1987**).

Kantha et al. (1986) reported that legumes are important sources of proteins, minerals and vitamins for millions of people in the world, particularly in the developing countries and they are the second largest plant sources after cereals, which could be used either for human food or animal feed. They are usually rich in protein (20% to over 40%).

Much of the world population relies on legumes as staple foods particularly in combination with cereals. Legumes are often advocated in western diets because of their beneficial nutritional effects and because they are a low cost source of protein. Use of legumes in the human diet might be increased in less developed regions of the world and also in western countries. Therefore, more information is needed about the potential nutritional implications of legume based diets (**Gustafsson and Sandberg, 1995**).

Utilization of legume as human food is below their potential, however, partly due to the presence of several antinutritional factors including trypsin, chymotrypsin and amylase inhibitors, hemagglutinins or phytates. In addition, raw legumes have low protein quality with deficiency in sulphur containing amino acids and with the resistance to proteolysis of proteins (**Kavas and EI, 1992**).

Whey as a by-product of cheese manufacture presents a major water pollution problem, as it contains half the solids of milk i.e., lactose, and protein. Therefore, recovering of these components and at the same time reducing the biological oxygen demand (BOD) of the whey before disposal of the effluent will share in solving the pollution problem (**Khorshid et al., 1994**).

Cheese whey contains approximately 93% water, 5.1% lactose, 0.9% protein and mineral salts. Various methods were reported for recovery of whey protein,

either by heating the whey under acidic or alkaline conditions in the range of 50.73% (**El-Sayed *et al.*, 1998**).

Different types of whey protein concentrates can be produced by ultrafiltration of whey, i.e., WPCs with a protein content ranging from 25% to 80% protein/total solids (**Ottosen, 1991**).

The nutritive value of the bread and the other cereal products depends upon the protein level in the flour and on the balance of various amino acids that make up the protein (**Howe *et al.*, 1965**). Normal cereal grain including wheat are low in some essential amino acids such as lysine, threonine, methionine, tryptophan and isoleucine (**Ibrahim, 1975**). Recognition of the beneficial nutritional attributes of legumes due to the complementarity of their essential amino acids with those of cereals, has led to world-wide attempts to fortify traditional bakery products, such as bread, biscuits (cookies) and also regionally popular bakery products (**Patel and Venkateswara-Raot, 1995**).

When dairy ingredients are added to non dairy foods to improve the nutritional quality, whey protein are primarily used. A combination of whey proteins with vegetable proteins results in a higher biological value of the mixture. The reason is the increased content of essential amino acids, mainly lysine (**Renz-Schauen and Renner, 1987**).

The baking industry is a major market for whey. Bakers began to utilize whey in bread many years ago. They found that whey was an economical source of milk solids when they switched from milk bread to white pan bread. More recently, they began to use whey-based products in cakes, biscuits and other bakery products (**Huginin, 1980** and **Schaap, 1992**).

Therefore, the objectives of the present investigation can be summarized as follows:

1. Evaluation of the studied legume flours and their protein concentrates (i.e., soybean, field pea and sweet lupine) for their chemical composition, functional properties and trypsin inhibitors.
2. The chemical and functional properties of sweet whey powder and ultrafiltration whey protein concentrates as a source of animal protein.

3. Studies the rheological properties of wheat flour dough as affected by supplementation with different levels of either legume products or whey protein products as concentrates.
4. Production high protein pan bread and biscuit by substituting of wheat flour with different levels of legumes products and whey protein concentrates.
5. Determination the chemical and sensory characteristics of the produced bread and biscuits as affected by supplementation.

2. REVIEW OF LITERATURE

2.1. Bakery products:

The first good primitive baking was perhaps done in Egypt, as depicted in scenes of tomb of Ti, 2600 B.C. Egyptians are also credited with the first use of leavening. They maintained a stock of sour doughs and mixed portions of this to fresh doughs. This method continues to be in use ever since and a number of bakeries in Egypt as also in India continue to rely on this method of fermentation. The art of baking then spread throughout the world. The availability of the enclosed baking utensils or the oven made the baking of thicker loaves and cakes possible. The bread forms multiplied and knowledge of fermentation made it feasible to make the loaf structure lighter and more digestible.

In Greece the art of using unique ingredients to dough was developed. In the beginning baked products were made of mixed seeds with predominance of barley. Later on it was seen that wheat flour responds best to puffing or leavening action of fermentation producing a light porous structure in baked products; thus wheat flour became a must for bakery products (**SBP, 1984**).

Wheat-based baked goods such as bread, cakes, biscuits and cookies are popular foods and provide an excellent means of improving the nutritional quality through incorporation of vegetable protein (**McWatters, 1978**).

Bakery industry needs a variety of raw materials; the raw material requirement differs with different bakery items. Even within the same product, the input composition varies considerably, and depends on the nutritional requirements of the end product, the consumer taste, and the pricing of the product. Whereas, the raw materials requirement for conventional bread and biscuits produced in mechanized units are well defined, a variety of raw materials in different "mixes" are used for a number of traditional bakery items and also items such as cakes, pastries etc., depending upon quality and taste requirements and local practices. Bakery industry has an important role to play in the economic development of the country, in fuller utilization of its wheat resources, and in building up the health of its people (**SBP, 1984**).

The leavened bread, inspite of all the varieties has a limited shelf life. Thus these breads are not suitable for long journeys by sea, trade or warfare and the demand for a bread form which had a much longer shelf life of more than a few weeks and even months, became very high. Thus biscuits were developed. 'Bis' means twice and 'cuit' means baked which suggests that the produce was intended originally to be twice baked. If properly prepared, biscuits were observed to be capable of being kept for a long time and hence these came to be used as a common form of bread at sea. After this came the development of several other products based on unfermented doughs both saltish and sweet. Bread and biscuits are good nutrition supplements. In recent times bread is usually fortified with vitamins and minerals such as thiamine, niacin and iron etc. The addition of 0.5% L-lysine, and 0.1 to 0.2% thiamine considerably improves the protein quality of the bread (**SBP, 1984**).

2.1.1. Bread

Man mastered the art of bread making thousands of years ago. Excavations of the oldest baker's oven in the world show that bread was known in Babylon 4000 BC. In the Old Kingdom of Egypt bread was baked in hot ashes or on heated stone slabs. At least as long ago as 2500 BC wedge-shaped bakers' ovens were known. Bread was baked on the inner surface of those ovens, and it still is in some parts of the Near East today.

From Egypt, bread making including fermentation, spread to other Mediterranean countries. Around the world, bread is the principal food and provides more nutrients than any other single food source. In over 50% of the countries bread supplies over half of the caloric intake; in almost 90% of the countries, over 30%. In most West European countries it is the source of half the carbohydrates, one-third of the proteins, over 50% of the B vitamins, and over 75% of vitamin E (**Pomeranz, 1987**).

Africa is a continent of great extremes in bread culture and bread consumption. In some parts of the continent bread has been known for about 6,000 years; in others it has been introduced fairly recently. Bread consumption is very high in Egypt, North Africa, West Africa and South Africa. In Egypt, cereal grains provide 75% of the calories and 90% of the proteins in the diet. In Kenya,

Uganda and the Congo, cereal grains constitute a relatively small amount of the total food consumed. The main food of the Near East and North Africa is wheat; barley and corn are important supplementary cereals in some of those countries. People in parts of tropical Africa eat sorghum; in some of the coastal areas of West Africa and in Madagascar, rice predominates; and corn is the staple food in the east and south of the African continent (**Pomeranz, 1987**).

There are about 100 types of bread baked today in Egypt. European-type breads (mainly French bread and sliced and wrapped pan bread) are popular in cities and comprise up to one-third of the bread consumed. In villages, flat bread is the most common, whereas in cities almost all the bread is made in commercial bakeries, in villages only 20% is made by professional bakers. The local flat balady bread with a diameter of 20 cm is the most widely accepted. It appears in two forms: the maui and mayar types. In addition, there is the Syrian bread that goes through a second baking stage at 200°C for 2-3 minutes. The Syrian bread is often made from Arabian balady dough. Bread is the important food of the Copt's, the Christian minority in Egypt. The bread is baked in bell-shaped ovens. It is called batauah in Upper Egypt and marahauh in Lower Egypt. The round loaves, up to 7 cm high are made from sour-fermented doughs. Thinner loaves are baked from rather stiff doughs that are sheeted to a diameter of up to 75 cm. Bread is mixture of wheat, flour, sugar, shortening, salt and water made into doughs, raised by the action of the added yeast, followed by fermentation and final baking. It is used as a staple food in many countries and varies in size, shape, texture, taste and composition from one country to another. The name of the variety therefore, denotes either the area of its origin or the presence of certain special ingredients e.g. French, Italian, Vienna, Crunch, Raisin, Rye, etc.... (**Pomeranz, 1987**).

2.1.2. Biscuit

Biscuits have become a traditional and significant food in many countries. Their variety in form and taste combined with long shelf life and convenience of use has perpetuated their popularity. It is generally recognized that biscuit products are cereals based and baked to a moisture content of less than 5%. The cereal component is variously enriched with two major ingredients, fat and sugar, but thereafter the variety is almost endless (**Manley, 1991**).

In biscuit making, the main ingredients are flour, water, sugar and salt. Varying the proportions of these ingredients may produce a variety of shapes and textures. The quality of biscuit is governed by the nature and quality of the ingredients used. At present the quality criteria of finished product changes, mainly due to the absence of a significant correlation between the characteristics of the raw materials (flour) and the quality of the product. Several authors have nevertheless attempted to describe the effect of ingredients in a dough and formula balance on the final structure of the product (**Gaines, 1982; Mizukoshi, 1985** and **Abboud et al., 1985**).

Biscuits, cookies and crackers differ from other baked cereal products such as bread and cakes by having low moisture content. The low moisture content ensures that biscuits are generally free from microbiological spoilage and confers a long shelf life on the products, provided of course that they are protected from uptake of moisture from a damp atmosphere or damp surroundings. Their low moisture content also gives biscuit a relatively high energy density compared with other baked goods (**Peter, 1988**).

Biscuits enriched with protein, usually from soya flour and caseinate, have been developed for special feeding programs, usually for children in developing countries. **Manley (1991)** has shown however, that in many cases malnutrition is due to not enough food, not only a lack of protein. Care should be taken about making nutritional claims, such as 'high protein', as there are usually statutory requirements to be observed. The main problems with soya-enriched biscuits are the strong and unattractive flavour that soya gives.

2.2. Legume as a source of plant protein

Legumes are the edible dicotyledons of plants in the family leguminose, the second largest family of seed plants. They are economical sources of protein and calories and are considered to be one of the cheapest and most convenient high-protein materials for offsetting the amino acid deficiency of cereal proteins (**Bahnassey et al., 1986** and **Duszkiewicz-Reinhard et al., 1988**).

Grain legumes are important sources of proteins, minerals and vitamins for millions of people in the world, particularly in the developing countries, which

could be used either for human food or animal feed (**Kantha *et al.*, 1986**). Legumes have low protein quality with deficiency in sulphur-containing amino acids and the resistance to proteolysis of proteins (**Chang and Mo, 1985**).

Fahmy *et al.* (1995) mentioned that legumes have a high protein, that is twice greater than cereals, ranging from 17% to 25% on a dry weight basis. Because legume was viewed as a good source of protein in vegetarian based diets, much effort had been spent investigating the protein quality of legumes and legume, cereal blends.

Cepeda *et al.* (1998) reported that the vegetable proteins are important in human nutrition due to population growth and widespread protein malnutrition, especially in third world countries. Production of high protein foods from non-conventional sources should improve this condition. In less developed countries, high protein mixes which are consumed in beverage form are used extensively, and in industrialized countries, the use of these mixes is ready to use dry milk products with soybeans as a major ingredient (protein beverages, high protein diet formulas, baby soups, etc.).

2.2.1. Chemical composition

The nutritional quality of food protein was mainly determined by the composition of the essential amino acids and by the digestibility of the protein. The sulfur-containing amino acids were the first-limiting amino acid in legume protein (**Chang and Sotterlee, 1981**).

The general analysis of 8 legumes was determined by **Hegazy (1981)**. He found that the protein content varied widely from 44% in soybean and lupine to 21% in lentil and chickpea. The oil content also ranged from 23% for soybean to 9.0% for field pea. On the other hand, variations in crude fiber and ash were relatively small. Glutamic acid, however come in the first order of the amino acid followed by aspartic acid in all legume samples.

The dry seeds of legumes generally had a similar chemical composition, with the exception of *Arachis* (peanuts) and *Glycine* (soybean), which had high fat and comparatively low carbohydrate contents (**FAO, 1958**). Component analysis of legumes included protein (15-38%), fat (1-2%), moisture, fiber (4-6%), ash (3-4%), minerals, vitamins and carbohydrates (**Sathe *et al.*, 1984**).

The stored proteins of legume seeds, comprising about 80% of the total protein, served to supply amino acids and a pool of nitrogenous compounds to the young seedlings. These proteins were located primarily in protein bodies, the protein content of which was approximately 75%. The remainder of the protein body was composed of phytic acid and mineral elements (**Stanley and Aguilera, 1985**). Although carbohydrate was a major constituent of legumes, detailed knowledge of the nature and properties was limited. Two obvious reasons for this component were first, the broad heterogeneity of material, ranging from simple sugars to complex heteropolysaccharides and secondly, the practice of analyzing for them by difference, viz. deducting the sum of all other constituents (moisture, protein, lipid, fiber, and ash) from 100%.

Soybean (*Glycine max* (L.) Merrill) is a legume increasingly consumed for economical and nutritional reasons (**Garcia et al., 1997**). In fact, soybean products are an important low-cost source of proteins, minerals, phosphorus and vitamins. Furthermore, soybean products play an important role in health (**Messina, 1995** and **Sirtori et al., 1995**). The intake of soybean is not only suitable for people with allergenic reactions caused by animal milk, but it is also recommended to prevent heart disease, obesity hypercholesterolemia, cancer, diabetes, kidney disease and osteoporosis. These reasons have promoted the recent appearance of numerous products derived from soybean such as soybean flour, textured soybean, soybean dairy-like products, meat, bakery products prepared with soybean etc., in order to facilitate its consumption and to improve its flavour (**Ishii and Yamaguchi, 1992** and **Ladodo and Borovik, 1992**).

The soybean is different from the other oil seeds in high content of oil and protein. Also, a high proportion of polyunsaturated lipids and lack of cholesterol are additional nutrition characteristics of soybean (**Lee and Chang, 1993**).

Soybean are limiting in sulphur-containing amino acids for most animal species, including humans, but contain sufficient lysine to help overcome the lysine deficiency of cereals. The amount of protein in soybean, 38-44%, is larger than the protein content of other legumes, 20-30% and larger than, 8-15%, for cereals. This larger quantity of protein in soybean along with excellent quality

increases their value as feed stuff and is one of the reasons for economic advantage that soybeans have over other oil seeds (**Snyder and Kwon, 1987**).

Soybean proteins are the components, which form about 40% of the total solids and play the most important role in food processing. About 90% of the proteins are extracted by water. Approximately 90% of the resultant proteins are precipitated at pH 4.5-4.8 and called acid perceptible protein or soybean globulins. These proteins are storage proteins and therefore do not possess any biological activities. The proteins contained in the supernatant are called whey proteins. They are composed of trypsin inhibitors, hemagglutinins, lipoxygenases, β -glucosidases, β -amylases, phosphatases, cytochrome C and the like, which are biologically active proteins. These proteins occur in very small quantities, but some of them such as lipoxygenases and β -glucosidases are very important in food processing (**Fukushima, 1991**).

Maciejewska et al. (1993) determined the changes in dry matter (DM), total sugars, soluble sugars, proteins, and fiber in Polan variety soybeans and mungbeans during germination. DM contents of soybeans and beans decreased by 12.7% for soybeans and 14.4% for beans. Total sugars decreased by 20.5% and soluble sugars decreased by 86.8% during germination of soybean seeds. Total sugars in bean seeds decreased by 21% while soluble sugars increased by approximately 70% protein contents decreased by 16.7% for soybeans and 19% for beans. Nutritional quality of the legumes was improved by germination.

Chemical composition of soybean can vary depending on the variety and growing conditions, but reasonable average figures are 40% protein, 20% lipid, 35% carbohydrate and 5% ash on a dry weight. The moisture content at harvest is an important factor and has an influence on the handling characteristics and keeping quality of the beans. Ideally moisture should be about 13% at harvest (**Tanteeratarm, 1993**).

Peas (*Pisum sativum*) are used extensively as human food and in some areas such as eastern Europe and Russia, for livestock but there appears to be little information available on the nutrient composition and protein quality of ripe peas (**Bell and Youngs, 1970**).

Dry peas contain 20-30% lysine-rich protein. Air classification and alkaline solubilization with isoelectric precipitation separate legume storage globulins and albumins into concentrates and isolates (**Swanson, 1990**).

Lupine seeds (*Lupinus angustifolius*) are similar to soybeans as sources of protein and in some species, also of oil. The limitation of a wider use of lupines has been their content of quinolizidine-alkaloids. This has been gradually overcome by establishment of new commercial low-alkaloid varieties developed by breeding programs (**Hill, 1977**). Breeding programs have produced "Sweet" varieties with as low as 0.002% alkaloid content which makes them safe for human consumption. The seeds must be defiltered from some varieties and after cooking, they may be used directly for human consumption as snacks, in soups, stews or mixed salads. Dried and milled grains may be also used as an ingredient for hot dishes and bakery products (**Gross, 1982**).

The genus *Lupinus* typically contains 36-52% protein, 5-20% oil and 30-40% fiber (**Gross et al., 1988** and **Petterson and Mackintosh, 1994**). The variation in composition is due to genetic and environmental differences (**Hill, 1986**).

The main lupine species are *Lupinus albus*, *L. luteus*, *L. angustifolius* and *L. mutabilis*. However, *L. mutabilis* is the species often most used for animal feed or human food. All species of lupine contain alkaloids (quinaolizidines) to a greater or lesser extent (range 0-4% of the seed, w/w), **Blaicher et al. (1981)**. They give (bitter) or sweet qualities to a particular variety.

Lupines, especially *Lupinus angustifolius* were becoming crop of increasing importance as a source of high protein food for human consumption (**Brooke et al., 1996**). It contained 28-30% crude protein, 5-7% ether extract lipid, 37-46% nitrogen free extract and 13-17% crude fiber as mentioned by **Summerfield and Roberts (1985)**.

Mohamed and Rayas-Duarte (1995) reported that the proximate analysis showed that *L. albus* L. 2043N was higher in protein (38%) and lower in starch (3%) than other common legumes. Ash content (4%) was similar to other lupine

species and similar to soybeans. Oil content (10%) was lower than in soybeans. The total carotenoid content of the whole grain was 36 ppm.

Sweet white lupine proteins are used as a complement in bread pastry and meat products and in dietetic products (**Morad et al., 1980**). The lupine proteins used for human food need to be treated to improved their physicochemical properties and nutritive value. Lupine proteins are very heat-sensitive (**Karara, 1989**). Their solubilities decrease with increasing temperature and duration of heating.

2.2.2. Nutritional quality improvement

Legume seeds are important sources of energy and protein in many parts of the world, both for animal and human nutrition. However, their nutritional value may be limited in part by the presence of undesirable components known as antinutritional factors. These factors include protease inhibitors, lectins, phenolic compounds, phytates and indigestible carbohydrates of the raffinose family (**Deshpande et al., 1984**). The content of these components may vary for different legumes, and this difference may be reflected in the efficiency of nutrient utilization.

Della-Gatta et al. (1988) reported that among the antinutritional factors, serious consideration should be given to trypsin inhibitors (TI) which were present in all legume seeds. These toxic factors combined with trypsin to form an inactive complex, thereby reducing protein digestion. Thus, the content and type of trypsin inhibitors could be used as an important parameter in evaluating the quality of legumes.

With the recognition of the presence of a trypsin inhibitor in soybean, it was tempting to conclude that the growth inhibition which it evoked in animals was simply due to an inhibited digestion of dietary protein by proteolytic enzymes present in the intestinal tract. The most destructive blow to this theory was the observation that preparations of trypsin inhibitor were capable of inhibiting growth even when it was incorporated into diets containing predigested protein of free amino acids. Such experiments obviously ruled out an inhibition of proteolysis as the sole factor responsible for growth inhibition, and thus served to focus attention

on some alternative mode of action of the trypsin inhibitor (**Liener, 1981** and **Hudson, 1983**).

The amounts of trypsin inhibitors levels obtained from the different legumes were 15 (TIU/mg sample) chickpea, 2.6 yellow lupine, 2.4 garden pea, 2.1 red lentil, 2.0 giant lentil, 3.9 and 5.1 in two varieties of faba bean, 66.0 soybean, 10.0 cowpea, 20.0 and 16.0 in two varieties of white bean and 32.0 runner bean (**Della-Gatta et al., 1988**).

There are many different processing methods used to eliminate the antinutritional factors present in legumes. The applied treatments included soaking, boiling, germination, fermentation, autoclaving and microwave heating and irradiation (**Lilian and Maria, 1985; Chang and Harrold, 1988; Sattar et al., 1990; Abou-Arab and El-Shatanovi, 1993** and **Idris, 1997**).

Of the several processing methods used for legume seeds processing, germination is a relatively simple method, does not require intensive energy input, and also yield natural product. Germination of legume seeds is accompanied by the metabolism of the reserve protein stored in proteins of bodies in the cotyledons (**Reddy et al., 1982**).

Germination improved the nutritive value of legumes by inducing the formation of enzymes which eliminated or reduced the antinutritional and indigestible factors in legumes. In addition, germination caused changes in protein and starch digestibility which probably also resulted from enzyme action (**Nnanna and Phillips, 1988 & 1990**). However, germination often caused undesirable effects, such as lipid degradation, modification of amino acid composition of proteins and microbial contamination (**Bates et al., 1977**).

Mostafa et al. (1987) described that one night soaking and 6-days germination depressed TIA by 32% for Calland variety soybean. They also, reported that germination process resulted in a marked increase in the relative contents of both essential and non-essential amino acids. The rate of relative increase in essential amino acids was 8.9% after 3 days of germination, 22.4% after 6 days of germination. The corresponding relative increases in non-essential amino acids were 17.6 and 17.5% after 3 and 6 days of germination, respectively.

The reduction in indigestible dietary fibers caused by germination was very important, as a diet with high content of indigestible dietary fibers affects the digestion and the intestinal absorption of nutrients induced by a great foecal loss of energy and in most instances of nitrogen and fat (**Weinstock and Levine, 1988**).

Germination degrades trypsin inhibitor slowly in the beginning. The content of the trypsin inhibitor in ungerminated soybeans was 20.4 ± 6 mg/g dry seeds, but decreased by 25.5% after 7 days of germination as was detected by **Roozen and DeGroot (1989)**.

Dagnia et al. (1992) compared the chemical composition of kernels from *Lupinus angustifolius* (Lupine) seeds with those for sprouts after 6 days germination. Germination resulted in an apparent increase in protein content from 395 to 435 g/kg dry matter (DM). Fat and carbohydrate contents decreased. Oligosaccharide content of the sprouted lupine fell to a negligible level, while the phytate and alkaloid concentration fell from 4.7 to 1.6 g/kg and from 0.72 to 0.16 g/kg, respectively.

Abou-Arab and El-Shatanovi (1993) studied the effect of dehulling and germination on the chemical characteristics of some legumes. They reported that germination caused a significant increase in protein content and decreased fat and carbohydrate in all germinated legume meals. They also found that removal of seed coats increased significantly the protein and fat contents.

Bau et al. (1997) found that both the total protein content and the non-protein nitrogen soybean increased after 5 days of germination of (*Glycine max.*). On the other hand, there was a gradual decrease in the available lysine level and lipid content as germination progressed. They also reported that germination was beneficial in reducing a number of antinutritional factors and increasing the biological availability of minerals and certain vitamins of seeds. It appeared possible to improve the biological value, flavor and nutritional qualities of seeds by this process. They also reported that phytic acid in the seeds was degraded by the phytase activated during germination, thus increasing the availability of minerals present in the germinated seed. Germination could degrade the trypsin

inhibitor and the degradation was enhanced if germination process lasted more than 4 days.

Hamza (1997) demonstrated that trypsin inhibitor content decreased after germination from 29.1, 21.6 and 22.5 to 14.4, 10.9 and 11.9 (mg/100 gm) in soybean, chickpea and mungbeans, respectively.

2.3. Legume flours and their protein concentrate

2.3.1. Preparation and chemical composition

Fernandez-Quintela *et al.* (1993) reported that legume flour and their protein concentrates can be used as ingredients in different foods or in a variety technological processes taking advantage of their functional properties.

The use of plant proteins in foods is expected to increase substantially in the future as a means of meeting the world wide demand for proteins. Therefore, there is a growing interest in the utilization of flours and fractions from different types of legumes (**McWatters, 1980** and **Chau and Cheung, 1998**).

Preparation of legume flours depended basically on, cleaning the seed, separating the hull and grinding the dehulled kernels then sifting (**El-Dash and Sgarbieri, 1980**).

Defatted meal is the principal source of soy flours and grits, although whole, dehulled or partially defatted soybeans can be used. The flour is ground until it passes through 100-mesh screen. For full-fat soy flour, whole soybeans are steamed or boiled, dried to 5% moisture, cracked, dehulled and ground. It contains 18% fat. Commercially, most flours are made from defatted meal. Defatted soy flour is made from defatted flakes and contains less than 1% fat. The nutritional quality and the functional properties of the flour depend on the heat treatment given to the protein during processing. Defatted soy flour contained, 59% protein, 1% oil, 6% ash and 3% fiber (**Singh *et al.*, 1987**).

Hung and Nithianandan (1993) produced full fat sweet lupine flour and full fat chickpea flour by cleaning the seeds from foreign materials then washing with water, draining and air drying at 35-38°C for 72 hours. The prepared seeds were ground and passed through a 250 µm sieve. Grinding was repeated until little, unpassable residue (250 µm) was left and discarded.

Lupine flour was obtained by debittering the seeds by soaking for 12 hours in water then discarding the water (**Abd El-Lateef, 1995**). The seeds were soaked in boiling water for 30 minutes, then the seeds were soaked in water for 48 hours at room temperature to remove residual bitterness. The water was changed every 4 hours. The debittered seeds were dried in an air oven at 50°C, ground and sifted on 60 mesh sieve.

Defatted soybean flours had the following chemical composition as mentioned by **Sarhan et al. (1986)** had 8.93% moisture, 5.93% ash, 1.59 ether extract, 3.98 crude fiber and 49.8% crude protein. However, **Hafez (1996)** found that defatted soy flour contained high amount of protein and ash being 49.7 and 7.75%, respectively.

Faheid and Hegazy (1991) found that defatted soybean flour contained 8.93% moisture content, 56.36% protein, 1.03% fat, 6.84% ash and 4.16% fiber (on dry basis). On the other hand, lupine flour had 11.52 moisture, 26.30% protein, 11.90% fat, 2.05 ash and 2.21% fiber.

The use of protein concentrates or isolates, alone or in combination with other processes which usually involve thermal treatment, has become an important choice in these strategies and has been applied to several legume seeds, e.g. soybean, pea, faba bean and mung bean (**Aremu, 1990**). These treatments usually extensively modify protein structure, leading to important alterations in their nutritive value as well as the amount of antinutritional factor(s) present.

Hassan (1980) extracted protein from legume by using salt solution (Na_2CO_3 , NaCl and Na_2SO_4), sodium hydroxide solution and enzymatic method (α -amylase and glucoamylase). He compared the three methods and found that the enzymatic method was the best but very expensive and needed to special condition, and the extraction with sodium hydroxide was better than the extraction with salt solution.

Soya protein concentrates are manufactured by extraction of the water-soluble carbohydrates, minerals and other minor constituents and inactivation of off-flavour producing enzymes and antinutritional factors. These products possess a low flavor level compared to the flavor associated with some soya proteins.

These concentrates vary in color, flavor, particle size water and fat absorption, and all characteristics important to bakery food manufacture. Concentrates are used mostly for water and fat absorption and where protein levels higher than those in soya flours are required for nutritional purposes (**Dubois and Hoover, 1981**).

Bahanassey et al. (1986) obtained protein concentrates from legumes (navy bean, pinto bean and lentil) by acid precipitation from dilute alkali solution. They found that the means of the chemical composition data of legume protein concentrates were 77.37%, 5.10%, 5.07% and 0.54% for protein, fat, ash and non-dietary fiber, respectively.

Clark and Proctor (1994) reported that soy protein concentrate is obtained by removing soluble carbohydrate either by acid extraction (pH 4.5), hot water extraction or washing with 60-80% ethanol to increase the protein content from 40 to 70%.

Soy protein concentrates from central soya Aarhus offer a number of benefits to consumers and food processors, desirable in a great variety of food applications. Whether in ground meat systems, whole muscle meat, emulsified meat, poultry, seafood or vegetarian products, textured and functional soy protein concentrates are designed to withstand stresses to the food system created by multiple cooking, microwave cooking, freeze/thaw, sterilization and extended holding or storage time. This functional stability combined with the healthy food image of the soy protein concentrate makes it a unique food ingredient. Therefore, **Pedersen and Taisbak (1995)** produced soy protein concentrates by extracting the soluble sugars as well as flavour component and antinutritional factors from the defatted white flakes by a mixture of ethanol and water at neutral pH. The traditional or standard range of soy protein concentrate was produced either in a coarse grits form or finely milled and provides water holding capacity and viscosity. Soy protein concentrate can be extruded to form a texture soy protein concentrate. Textured concentrate provides water holding capacity and texture improvement as well as some capacity to entrap fat particles. Textured concentrates absorb three to four times its own weight of water and can be incorporated into foods even at high doses without affecting the flavour of the final

product. By applying further potential processing steps primarily consisting of a high temperature, high shear treatment of an aqueous solution followed by spray drying, it is possible to obtain a soy protein concentrate with a high protein solubility, this product type known as "functional soy protein concentrate", ensures dispensability, solubility, viscosity, water absorption and excellent emulsifying properties.

2.3.2. Functional properties

The utilization of soy flours, concentrates and isolates in prepared foods has increased rapidly and exceeds that of other concentrated seed proteins. The functional and physical properties of these proteins have defined their role in baked goods, meat products and soy-beverage (**Johnson, 1970**).

Briskey (1970) and **Kinsella (1976)** mentioned that the most important functional properties in food applications were sensory properties (e.g. color, flavour, taste and texture), hydrophilic properties (e.g., wettability, water absorption, swelling, gelling, water holding capacity, foaming and protein solubility), hydrophilic-hydrophobic properties (e.g., fat-binding and emulsification), texture properties (e.g., softness, elasticity, viscosity, adhesion, hardness) and rheological (e.g. aggregation dough formation, stickiness and fiber formation).

Thus systematic determination of functional properties should be made when developing new sources of proteins, protein concentrates and isolates. These are required to evaluate and possibly help to predict how new proteins may behave in specific systems, as well as demonstrate whether or not such proteins can be used to simulate or replace conventional proteins. The functional properties of proteins denote any physicochemical property which effects the processing and behaviour of protein in food systems as judged by the quality attributes of the final product. These reflect complex interaction between the composition, structure, conformation physicochemical properties of the proteins other food components and the nature of the environment in which these are associated or measured (**Kinsella, 1976**).

Sosulski (1977) defined functionality as the physical, chemical and organoleptic properties of the colloidal suspend protein which affect the

structure, texture, flavor and color of the formulated food product. He also added that the number of desirable functional properties associated with behavior of proteins in aqueous colloidal system, and in the presence of carbohydrates, fats, minerals and other food ingredients, can be extremely large.

Most functional properties are determined by the balance between forces underlying protein-protein and protein-solvent interactions. This balance is affected by changes in the pH value, concentration, nature of solvent and presence of other components. The conditions that favor protein-solvent interactions increase the solubility. The major force favoring protein-protein interactions in aqueous solution was the hydrophobic interaction between the nonpolar surface on the protein. Crude correlation have been found between functional properties and protein solubility index. Thus, nitrogen solubility index determinations were often used as a quality control tests in preparing products for certain functional uses (**Ahmed, 1994**).

Solubility characteristics under various conditions are very useful in selecting the optimum conditions for extracting proteins from natural sources (**Betschart and Kinsella, 1973**). Its behavior provides a good index of the potential and limitation applications of proteins. Protein solubility also gives an information which is useful in the optimization of processing procedure and in determining the effect of heat treatments which affect actual and potential applications (**Hermansson, 1973**).

Protein solubility is very complex and can be affected by many variables such as electrostatic interactions, hydrophobic interactions and hydrogen bonding. The level of these three major forces contributed to protein solubility by favoring protein-protein interactions, which was indicated by lower protein solubility or by favoring protein-solvent interactions, which was indicated by higher protein solubility (**Kinsella et al., 1985**).

Fan and Sosulski (1974) determined the solubility characteristics of protein in nine legumes species and demonstrated wide differences in nitrogen extraction and precipitation curve. They found that the alkali extracted proteins had lower

solubility at pH values of 2-3. Greater dispensability was found at higher pH values.

King et al. (1985) prepared lupine protein isolates by alkaline extraction at different pH values and investigated their functional properties. They found that lupine protein isolates showed better solubility than soybean isolate. Generally, it is possible to consider lupine protein as a potential substitute for soybean proteins in food application.

Sousa et al. (1996) determined the solubility of lupine protein extracted from *Lupinus luteus* seeds. They found that the relationship between solubility and pH for the lupine protein isolate was similar to that reported in the literature for soy isolates.

The capacity of plant proteins to interact with and bind water or lipid materials is important in food formation and processing. Also, the rate of hydration is an important characteristic when water is being incorporated with dry ingredients such as baking or the preparation of extended meat products. Organoleptic characteristics associated with the degrees of hydration include dryness, juiciness and mouth feel. The same functional properties of proteins, which determine the total water absorption may control water retention after baking or shrinkage during cooking. Fat absorption has been equated with fat emulsification properties but there is no supporting evidence to confirm that these characteristics are related (**Sosulski, 1977**).

Globulin proteins are generally more hydrophilic than prolamine and gluten because they contain more polar side chains. Therefore, proteins such as soybean (70-85% globulins) will absorb relatively high levels of water and retain it in the finished product (**Wolf and Cowan, 1975**).

Fleming et al. (1974) determined water absorption of sunflower and soybean flour, concentrates and isolates. Among the soybean products, isolates had the highest water absorption followed by concentrates which were higher than flours. One shortcoming of this test was, that, if a protein was completely soluble, it would show no apparent water absorption, but if it was incorporated in a food

system, it might be insolubilized or gelled by heating and show excellent water absorption characteristics.

Soybean and sunflower products with good oil emulsifying properties tended to be low in fat absorption. The soybean proteins, which had oil absorption value of 84.4-154.5% were less lipophilic than sunflower products which absorbed 207.8-256.7% oil (Lin *et al.*, 1974 and Sosulski, 1977).

Hutton and Compbell (1981) and Kinsella *et al.* (1985) stated that the amount of lipid bound was markedly affected by the method used, the protein content, the surface area, the hydrophobicity and liquidity of the oil. It was conceivable that the binding capacity was enhanced by destroying hydrophobic domains, denaturation might reduce fat binding. However, it was probably that most of the oil hold by protein was actually physically entrapped and therefore the amount bound was influenced mainly by the surface area and bulk density of protein preparation.

Deshpande *et al.* (1982) studied the effect of dehulling on functional properties of dry beans (*Phaseolus vulgars* L.) flours. They found that dehulling improve the water and oil absorption capacities of bean flours by 3-39% and 10-44%, respectively.

Abou-Arab and El-Shatanovi (1993) showed that dehulling and germination improved oil and water absorption and emulsion capacity of some legumes. Oil absorption % of chickpea seeds improved by dehulling from 141.3% to 168.6%, also water absorption % improved from 214.1% to 242.9%. Generally, dehulling and/or germination process of legumes could be considered as effective means of improving their functional properties and therefore, increased their utilization in different formulated foods.

El-Adawy (1996) reported that the oil absorption % of mung bean protein concentrate was 145.2% and for protein isolate was 98.3%.

The emulsifying property is an important functional property of a protein. Two main approaches had been used this property might be expressed as emulsifying capacity (EC) or emulsion stability (ES). The former measured the maximum oil addition until phase separation occurred, whereas the latter measured the

tendency for the emulsion to remain unchanged. Emulsion or emulsifying capacity is usually defined as the volume of oil (ml) that can be emulsified by gram protein before phase inversion or collapse of emulsion occurs. Emulsion stability referred to the ability of a protein to form an emulsion that remained unchanged for a particular duration and under specific conditions (**Kinsella, 1976**).

DeKanterewicz et al. (1987) showed that the emulsifying capacity of proteins depended on the suitable balance between the hydrophilic and lipophilic characteristics rather than merely on the high value for each one. The calculation of the water oil absorption index (WOAI), as a measure of the relative simultaneous of the emulsifying capacity of proteins. Maximum emulsion capacity was achieved when the WOAI was nearly two, that is when the protein absorbed twice as much water as oil. However, it was observed that an optimum WOAI (corresponding to proteins with the highest emulsifying capacity) did not ensure maximum stability properties.

Cheftel et al. (1985) reported that many factors influence the characteristics of emulsion and the results of emulsion test: equipment type and geometry, intensity of energy input, rate of oil addition, oil phase volume, temperature, pH, ionic strength, presence of sugars, presence of low molecular weight components, exposure to oxygen, kind of oil (melting point), concentration of soluble protein and emulsifying properties of the proteins.

Use of plant proteins in emulsified food systems could promote fat binding to reduce cooking losses, improve EC and maintain stability of the emulsion system (**McWatters and Cherry, 1977** and **Abe, 1989**). High protein concentration could increase the stability of an emulsion with less fat and water separation (**Grenwelge et al., 1974**). The stability effect of proteins in emulsions is related to high electrical charge and more hydrophilic-lipophilic groups within protein structures that increase the protein-lipid and protein-water interactions (**Jones, 1984** and **Li-Chan et al., 1984**). These interactions were the major factors of emulsion formation and affected the appearance, color, texture and yield of finished products. The pH of the medium indirectly affected EC of proteins by influencing protein solubility. The EC increased when the pH of the system

diverged from the isoelectric point of protein (**Pearson *et al.*, 1965** and **Grenwelge *et al.*, 1974**).

Yatsumatsu *et al.* (1972) found that emulsification capacity of soybean products correlated positively with protein content and negatively with fiber content.

Lin *et al.* (1974) demonstrated that wheat, soybean and sunflower flours had relatively good oil emulsification properties when compared to concentrates and isolates. The oil emulsification capacity were unrelated to water or fat absorption characteristics but high protein solubility index was associated with the percentage of oil emulsified in a model system.

Franzen and Kinsella (1976) reported that the pH and ionic strength of the aqueous markedly affected the emulsifying properties of soybean protein. Emulsifying activity followed the typical pH solubility profile. **McWatters and Cherry (1977)** added that the components other than proteins possibly carbohydrates might contribute to emulsification properties of protein containing products.

Deshpande *et al.* (1982) found that emulsion capacity of dry bean flour increased by 70.3-75.1% as a result of dehulling. Also, they found that emulsions of dehulled bean flours were however, less stable than those of whole bean flours.

Foda *et al.* (1984a) showed that low fat soy flour, variety "Clark" and its protein isolate had lower emulsion capacity at pH values. Emulsions obtained from low fat soy flours were highly stable, on the other hand, emulsions formed by using protein isolates prepared from different soybean varieties were less in their stability as compared to the corresponding low fat flour.

Both emulsifying capacity (EC) and emulsion stability (ES) increased with increasing concentrations from 0.4% to 0.81% of soy flour (SF), soy concentrate (SC), soy isolate (SI) and corn germ protein flour (CGPF) when studied by response surface methodology. EC and ES increased as pH increased from 6 to 8 in all samples. Increasing incubation temperatures of protein solutions from 20-70°C or from 4-20°C did not effect EC or ES, respectively, SF had the highest EC followed by SI, SC and CGPE (**Wang and Zayas, 1992**).

2.4. Fortification of bakery products with legume flours and their protein concentrates

Legumes are an economical source of protein in developing countries and can be a nutritional source of carbohydrates and particularly fiber. Recent nutritional studies indicated that dietary changes might protect against diabetes, hypertension, cardiovascular diseases (CHD) and obesity. Fiber rich foods have important effects on reducing serum cholesterol (**Anderson and Gustafson, 1988**). Complex carbohydrates from grain legumes could be an energy replacement for saturated fat in western diets (**Hetzel, 1983**).

The total yield of bread grains in Egypt not satisfy the needs of the country. The total production of wheat grains cover only about 25% of the total needs. The way to overcome this problem is to search for the native cereal sources which could be supplementation of wheat flour for bread making (**Foda et al., 1987**).

In Egypt, cost plays a large part in the kind of food consumed and animal protein is beyond the economic means of many people. Hence, it is important to develop protein mixtures that use local unexpensive sources, such as cereals and legumes. Legumes are considered important sources of different nutrients specially protein and minerals. Baked products (i.e. bread, biscuit, cake, muffins, cookies, etc.) are consumed on a large scale all over the world. Therefore, fortification of baked products with high protein legume flours could provide a good opportunity to improve the nutritional quality of protein consumed by many people (**Rooney et al., 1972** and **Hoover, 1979**).

Legume seeds have been employed by **Fernandez-Quintela et al. (1993)** is human nutrition, but its sulphur amino acid imbalance and the presence of some antinutritional factors have hampered a wider utilization. The application of number technologies allows to obtain different products with high protein levels, in which a great part of the undesirable components are discarded. These isolates and concentrates can be used as ingredients in different foods or in a variety technological processes taking advantage of their functional properties.

Many researchers reported that the use of plant proteins in food is expected to increase substantially in the future as a means of meeting the world wide

demand for protein. The extent to which plant derived proteins are successfully used will largely depend upon understanding of the physical and functional quality they impart to foods and of their acceptability to consumers (**Anonymous, 1974; Jeffers et al., 1978; Okaka and Potter, 1977 & 1979** and **Sosulski and Fleming, 1979**).

Efforts to increase the availability of protein in man's diet have encouraged use of high-protein plant materials as ingredients in a variety of foods. Such wheat based baked goods as breads, cakes and cookies are popular foods and provide an excellent means of improving nutritional quality through incorporation of vegetable proteins (**McWatters, 1978**).

The quality of protein for utilization as food and feed depends on four major elements, the composition, mainly the essential amino acid content, the occurrence and content of antinutritional factors (ANF) such as trypsin inhibitors and lectins, the amino availability and finally for food uses their functional properties like viscosity, solubility, emulsifying properties, water and fat binding capacity etc., (**Wiege et al., 1993**).

2.4.1. Chemical composition and nutritional quality

Onymi and Lorenz (1978) reported that addition of up to 5% soy concentrate did not adversely affect white bread quality. Generally, unless higher protein levels are demanded, soy flour has the functionality and economics to be used in bakery products in the place of concentrates.

EI-Dash et al. (1980) studied the effects of addition of sweet flour to bread. They reported that the incorporation of lupine flour at 10% level resulted in a satisfactory bread quality with a PER value of 1.28 (PER value for the control bread was 0.81).

Mabesa et al. (1983) replaced a certain ratio of wheat flour with flour of germinated navy bean (*Phaseolus vulgaris*), mung bean (*Vigna radiate*), cowpea (*Vigna sinensis*), soybean (*Glycine max.*) and rice bean (*Phaseolus calcarotus*) during making some wheat products such as biscuits, kopeck, vegetable loaf and noodles. Relative nutritive value was estimated. The fortified products were nutritionally superior in many respects to the equivalent products made with only wheat flour. Results indicated that the flour of germinated legume seeds can

replace a part of wheat flour in some products to improve both quality and nutritive value.

Foda *et al.* (1984b) used low fat soy flour as a partial replacement for wheat flour in biscuit production at levels of 0, 10, 20 and 30%. They observed that supplementation with soy flour led to significant increase in moisture, crude protein and minerals contents of biscuit, which had a favourable effect on increasing nutritive value of the product.

Faheid and Hegazy (1991) utilized soybean flour (SF), chickpeas flour (CF) and lupines flour (LF) to replace 0, 5, 10 and 15% of wheat flour in cookies. Results indicated that protein, ash and fiber contents as well as moisture content of supplemented cookies were higher than the control. Total protein content increased by about 1.5, 1.0 and 0.7% with each increment of SF, LF and CF, respectively. Mineral contents and amino acid score of the supplemented cookies increased as compared with the unsupplemented ones, due to the improvement in lysine and other essential amino acids except sulphur-containing amino acids. Consequently, both PER and BV of supplemented cookies improved with unsupplemented ones.

Lorimer *et al.* (1991) found that replacement of wheat flour with protein, high-lysine ingredients such as legume flours, protein concentrates and isolates improves the amino acid balance and increases the protein content of products baked from the blended flours.

Niola *et al.* (1992) analyzed fifteen retail samples of soy flour containing biscuits for moisture, ash, crude protein, lipids, starch, sugar, cellulose and acidity. IR spectra were determined to assess freshness and shelf-life. Soy contents ranged from 3 to 25% with most samples being in the range 10-15%. Composition of the biscuits varied widely, the samples with the highest soy content tended to have the highest crude protein content.

2.4.2. Rheological properties

Campos and El-Dash (1978) found that addition of sweet lupine flour to wheat flour increased the water absorption, dough development time and the mixing tolerance index, while it reduced dough stability. They also found that

dough extension and maximum resistance to extension showed a proportional reduction as the level of sweet lupine flour increased.

Onymi and Lorenz (1978) showed that addition of 5% soy concentrate or isolate to wheat flour did not significantly change Farinograph absorption, mixing time and proofing time of bread. Soy concentrate and isolate produced good quality bread especially when they were used at levels not exceeding 5%.

Hsu et al. (1980) studied the bread baking properties of wheat flour and dry peas, lentil and faba beans. The legume flour was formulated on a replacement basis of levels of 5, 10, 15 and 20%. The mixograms which reflect the dough properties of the commercial straight grade control flour and the legume-wheat flour blends are shown in either germinated legume wheat flour blends. The overall mixographs properties of yellow peas, lentils and faba beans were similar to those of control, except than for germinated lentil, prolonged mixing time and germinated faba bean reduced mixing tolerance. In all cases, water absorption decreased with increasing level of supplementation. These observations were in agreement with the results reported by **Jeffers et al. (1978)** when wheat flour fortified with raw pea flour.

Domah (1983) studied the effect of adding lupine on the physical properties of dough and baking quality of bread. He found that the rheological characteristics of dough was improved with increasing levels of lupine flour up to 10%. His studies indicated that the use of 10% lupine flour improved both protein content and baking quality of the produced bread.

Foda et al. (1987) found that defatted soy flour improved resistance to extension and proportional number and lowered dough extensibility and energy. **Campos and El-Dash (1978)** found that addition of sweet lupine flour to wheat flour increased the water absorption, dough development time and the tolerance index while reduced dough stability. They also found that dough extension and maximum resistance to extension showed a proportional reduction as the level of sweet lupine flour increased.

Levels of 5, 10 and 15% of legume flour, i.e. soybean, lupine and chickpeas were used to supplement cookies by **Hegazy and Fahied (1991)**. The effect of this supplementation on the rheological of the resulting dough was investigated

using the farinograph and extensograph as objective methods for quality assessment of the final product. It was found that, there was an increase in water absorption capacity, dough stability, arrival time, dough development time and mixing tolerance index as a result of supplementation of cookies with legume flour.

Hafez (1996) studied the effect of addition of soy flour to two types of wheat flours (72% and 82% extraction) at levels 5, 10 and 15%. He found that water absorption was lower at 72% extraction flour than 82%. Adding defatted soy flour increased dough stability and water absorption at 72% and 82%, but weakening was decreased. Addition of defatted soy flour decreased extensibility and increased resistance to extension.

Much researches were done concerning substituting various proportions (2.5-20%) of soy flour into cakes, cookies, muffins or biscuits. Studies indicated that 10-15% substitution of soy flour could be added to wheat flour without affecting its pasta making properties (**Hannigan, 1979**).

2.4.3. Organoleptic properties

Levinson and Lemancik (1974) predicated that soy protein in baked products serve the following functions: improve eating quality, lessen moisture loss during baking, make doughs more pliable and easier to handle, increase rate of browning and provide a better crust color, increase shelf-life and improve texture of baked products.

Fleming and Sosulski (1977) studied the fortification of wheat flour with vital gluten and sufficient soy flour, sunflower concentrate, faba bean concentrate and field pea concentrate to produce breads. They found that field pea bread were given "excellent" protein ratings of more than 40, but soy bread was given a "good" rating of 37.5 due to the higher moisture content and therefore lower protein content on a fresh weight basis. Sunflower bread also received a "good" rating while wheat bread had a rating of less than 20.

Onymi and Lorenz (1978) noticed that the specific loaf volume of bread was generally depressed by increasing the amount of soy protein.

Abdel-Rahman and Youssef (1978) found that wheat flour fortified with defatted soy flour yielded loaves of bread with a slightly smaller volume than the control sample (no soy flour).

Baking and organoleptic qualities of baking powder biscuits made by replacing milk protein with cowpea (*Vigna unguiculate*) and field pea (*Pisum sativum*) protein were investigated by **McWatters (1980)**. Flour from two varieties of cowpea (G143 and Dixie cream) prepared by a dry milling process and from a flour and a protein concentrate prepared from field peas by pin-milling and air classification were included. Biscuits containing unheated and steamed (100°C, 30 min) pea products were compared to reference biscuits made with whole milk. Doughs containing the pea protein products were slightly less sticky than reference dough. Sensory scores revealed that pea products in biscuits adversely influenced aroma and flavor qualities more than appearance, color and textural attributes. Steam heating of the pea products improved some biscuit quality attributes but not to the level of acceptability of the reference biscuits. Biscuits containing pea products browned less during baking and had lower weight/volume ratios than did the reference biscuits. The crust color of reference biscuits had lower L (Lightness) and higher b (yellowness) Gardner values than did biscuits containing the pea products. The crumb color of reference biscuits and of those made from cowpea flours was lighter and less yellow than that of biscuits made from field pea products.

Fortifying wheat flour with 10% soy flour increased specific volume of biscuit and had no significant effect on organoleptic evaluation such as appearance, tenderness, flakiness, color and flavor. Negative effect were also obtained when mixing was carried out by using 20 or 30% soy flour (**Foda et al., 1984b**).

Cookies enriched with 0, 5, 10, 15, 20 and 25% full-fat sweet lupine flour (FFSL) were evaluated by a sensory panel using the rank of preference and paired comparison tests by **Wittige De Penna et al. (1987)**. Cookies with 0, 5 and 10% FFSL were preferred while those containing 20 and 25% FFSL were rejected ($P \leq 0.01$). Studied conducted with school children showed similar acceptability for 0 and 10% FFSL-containing cookies which was different ($P = 0.05$) from those

containing 20% FFSL. Fortification of the basic formula with 10% FFSL was recommended on the basis of acceptability.

Grover and Gurmukh (1994) studied the effect of incorporation of 5 commercial defatted soy flour samples into wheat flour on physical (thickness, diameter and spread ratio) and sensory (top grain, texture, flavor and overall acceptability) characteristics of cookies. Wheat flour was supplemented with 5-25% defatted soy flour samples. Increasing levels of defatted soy flour reduced diameter and increased thickness of cookies resulting in significantly reduced spread ratio. Incorporation of all types of defatted soy flours increasing amounts, affected sensory properties and significantly decreased overall acceptability. The various type of flours tested differed significantly in the level of reduction of spread ratio. However, overall acceptability values of cookies made with these flours did not differ appreciably. They concluded that replacement of wheat flour by up to 15% soy flour was possible without adversely affecting sensory characteristics of cookies.

Hafez (1996) mentioned that the adding of 5% or 10% defatted soy flour to wheat flour improved produced loaves quality and 5% was better, but addition of 15% defatted soy flour produced unsatisfactory bread.

Ranjana et al. (1996) mentioned that sweet biscuits prepared from wheat flour with 0-50% replacement by defatted soybean flour (DSF) were evaluated for physical, chemical and sensory properties. Thickness biscuits increased, whereas diam. spread ratio and spread factor decreased as DSF level increased. Sensory properties (appearance, color, texture, flavor, overall acceptability) indicated that up to 20% DSF could be used in biscuit formulation without substantial adverse effects on overall quality.

2.5. Whey products as a source of animal protein

Whey protein may hold the key to innovative product development, particularly in the growing sports nutrition marketplace. Also, whey protein can be manufactured to be stable and not precipitate when heated, even at low pH, so it works well in acidic systems. Other interesting application possibilities of whey protein include meal replacers, central and medical nutritional products, lactose-

free and fat-free formulations and systems in which lactase is functionally undesirable because of concerns about the Maillard browning reaction or lactase intolerance (**Huffman, 1996**).

Since the emergence of the ultra-filtration technique during the early 1970s and its subsequent improvements, a large variety of whey protein concentrates (WPC) with protein, total solids ratio in the range of 35-85% has been produced by the dairy industry. These WPC are used in baby food formulations fermented sausages and protein-enriched bakery products and beverages due to their functional properties and their nutritional value (**Marshall and Harper, 1988**).

It will be possible to select a WPC ingredient with not only the desired protein content but also the desired functional attributes. Obtaining the desired function properties can be achieved by blending with other ingredients, from processing variables employed during the WPC manufacture, or from post-processing steps such as protein and lactose hydrolysis. As the variety of WPCs increases, it will become more important to evaluate them based on functionality information obtained from simple test systems, selected model foods or the actual target application (**Jacobson, 1997**).

2.5.1. Chemical composition and nutritional quality

Whey is the fluid protein of milk obtained after coagulation and removal of casein during the manufacture of cheese or casein. Whey contains approximately 50% of the milk solids, i.e. most of the lactase, between 20 and 24% of the protein and almost all of vitamins and minerals. Basically, there are two major types of whey arising from cheddar, Swiss and other rennet cheeses and acid whey from cottage and similar cheeses and from acid casein manufacture. The typical composition of raw cheddar and cottage cheese wheys is shown in Table (1):

Table (1): The composition of cheese whey.

Chemical composition	Cheddar ¹	Cottage ²
Protein	0.62	0.70
Non protein nitrogen	0.19	-
Fat	0.04	-
Lactose	4.60	4.50
Ash	0.56	0.60
Total solids	6.10	6.50

1 from **Delaney et al. (1973)**.

2. from **Merson (1971)**.

Aruna (1994) reported that the only certainty with whey protein products is that their functionality will vary from batch to batch and manufacture to manufacture. Therefore, an understanding of the types of products and their manufacture may help in rationalizing the observed variabilities in functionalities of whey products. Typical composition of wheys are given in Table (2):

Table (2): Composition % of different types of whey from cow's milk (**Fevrier and Bourdin, 1977 and Morr, 1984**).

Composition %	Rennet	Lactic	Mixed	Sweet	Acid
Dry matter	7.08	6.58	7.05	7.00	6.50
Lipids	0.51	0.09	0.34	0.20	0.04
Lactase	5.18	4.53	5.05	4.90	4.40
Total nitrogen	0.15	0.12	0.15	0.13	0.11
Lactic acid and citric acid	0.16	0.78	0.32	0.20	0.05
Ash	0.53	0.07	0.47	0.50	0.80

In all considerations of the nutritive value of a protein, an important factor is the limiting amino acids. In almost every food the limiting amino acids is either lysine or methionine plus cystine. The whey proteins are unusual in having a relative surplus for the majority of the essential amino acid including lysine and the combined sulphur-containing amino acids. The high content of essential amino acids in WPC suggests that it could be utilized to supplement low quality protein foods. For example, cereal grains are deficient in several amino acids and up to 60% of their potential nutritive value as protein is not utilized in the absence of proper supplementation (**Smith, 1976**).

When whey is dried the moisture content of sweet whey powder approaches 4.6% while that of acid whey is 3.9. Because of their low nitrogen contents, whey powders are not regarded as rich sources of functional proteins (**Morr, 1984**).

Products containing more than 35% protein on dry basis are called whey protein concentrates. A number of different techniques have been developed to concentrate the proteins. **Morr (1986)** Classified these techniques as laboratory and commercial processes. Laboratory processes include those based on different solubility (e.g. polyphosphate, carboxy-methyl cellulose complexes), polyethylene glycol precipitation, pH-temperature precipitation, demineralization

and chromatographic techniques (ion exchange and molecular size exclusion). Laboratory processes were either cost prohibitive for commercial scale up or produced nonfunctional products. Commercial products with 35, 50 or 80% protein on a dry basis are routinely available. Commercial processes rely on ultrafiltration and diafiltration. Composition of such products is provided in the following table. **Mangino (1992)** observed that as the protein content of the products increases so do manufacturing costs. **Morr (1986)** reported that concentration of whey by ultrafiltration to 4% protein in the retentate followed by evaporation and spray drying results in a 35% whey protein concentrate. If the retentate containing 4% protein is diafiltered to 16% protein and then evaporated and spray-dried a 50-75% protein, whey protein concentrate results.

Huffman (1996) mentioned that whey powder is dried whey with 10% ash, 1% fat, 76% lactose and 13% protein. The 35% whey protein concentrate (WPC) has 34-35% protein, 53% lactose and typically 4% fat and 8% ash. This composition is similar to that of nonfat dry milk. The 50% WPC contains about 53% protein, 35% lactose, 5% fat and 7% ash. In 80% WPC, the protein concentration increases to 80% the lactose content decreases to about 7% and the fat and ash range between 4-7% as shown in Table (3).

Table (3): Composition of WPC powders.

Constituent	Whey protein concentrates protein (%)			
	35	50	65	80
Moisture	4.6	4.3	4.2	4.0
Crude protein	36.2	52.1	63.0	81.0
True protein	29.7	40.9	59.4	75.0
Lactose	46.5	30.9	21.1	3.5
Fat	2.1	3.7	5.6	7.2
Ash	7.8	6.4	3.9	3.1

An important aspect that whey processors will face in commercialization of WPC is the development of application of the product into different food systems: price, functional properties, and competitive proteins. The extent of processing and the extent of quality control of the product will determine the cost and, therefore the market potential of the product. This is shown graphically in Figure

(1). As the product is upgraded by increased protein concentration, the cost per unit of protein increases and the market potential decrease. For the food processor to utilize an expensive WPC, the product must exhibited a certain functionality that is worth the price (**Melachouris, 1984**).

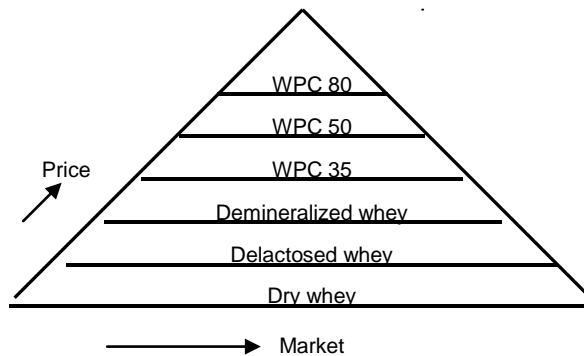


Fig. (1): Price and market potential of whey and modified whey products of increasing protein content. The WPC 35, 50 and 80 refer to whey protein concentrates (WPC) with 35, 50 and 80% protein content.

2.5.2. Functional properties

The term functionality refers to the functional demands made on food products, with regard to their desired properties such as aeration, fat-binding, water binding and structure-forming capacity. These functional demands are simply related to similar physio-chemical protein properties in aqueous solution. This implies that the functional requirements of food products are frequently solved by trial and error through additions of arbitrarily selected protein before food processing. For every new product this empirical procedure has to start afresh, without any possible help from systematic background information (**De Wit, 1988**).

Morr (1979) observed that for any food protein ingredient to be useful, it must be free from toxic and antinutritional factors, free of off-flavors and off-colors, compatible with other ingredients and process, readily available at an affordable price and serve a function in the product.

Proteins are highly functional ingredients that yield important benefits in foods such as cost reduction, nutrition or function. Functional benefits can include flavor,

enhancement, texture or storage improvement and stabilization of emulsions and foams. Not only do proteins influence final properties of the product, they also can affect processing parameters (**Jacobson, 1997**).

Solubility is often heralded as the first necessity in functional protein. The protein must be soluble so that it can subsequently interact with other ingredients, including water and contribute to texture formation in foods. Soluble proteins can have low water-binding properties, which allow addition of high levels of protein without causing viscosity increases (**Jacobson, 1997**).

Macromolecules are not truly soluble in the same manner as low molecular weight solutes. However, amino acids in protein chains interact with water and proteins can be suspended in water. This property is often used as an indicator of whey protein denaturation. Protein solubility is often affected by temperature, pH and the presence of other solutes and salts, and the values for solubility obtained are particular least soluble in the pH range close to their isoelectric point, but when proteins are soluble at these pH values. The wide range of pH values over which whey proteins are soluble make them ideal for use in a variety of products (**Kilara, 1994**).

Whey protein is highly soluble in water compared with most other proteins. The solubility of WPC is high at all pH values whereas the solubility of other proteins such as caseinate and soy protein is highly dependent on pH values. Furthermore, the solubility of WPC is hardly influenced by low molecular weight solutes such as salt and sugar. And as most food systems have pH values in the range of 3-7, it follows that the solubility of WPC in practically all liquid or moist foods will be excellent (**Ottosen, 1991**).

Whey proteins are sensitive to heat and this is exploited in preparing lactalbumin. Whey proteins prepared by heat treatments are insoluble, gritty and have very poor functional properties (**Robinson et al., 1976**). Thus, much research is being focussed on developing practical methods for the isolation of undenatured whey proteins with good solubility which should increase their uses in various food products (**Jelen, 1979** and **Marshall, 1982**). **Morr et al. (1973)**

studied the solubility of several whey proteins prepared by a variety of methods, solubility ranged from 6 to 10 mg/ml at neutral pH.

Whey proteins that have not been heat-denatured demonstrate excellent solubility over a wide pH range. However, heating to temperature above 70°C can cause partial loss of solubility between pH 3-5 because some of the whey proteins aggregate and precipitate at their isoelectric points (pH 4.5-5.3). Even with a heat treatment of 90°C for 5 minutes of an aqueous solution of WPC, more than 80% of the whey protein remains in solution. The solubility of whey protein in heated products can be increased by the addition of sugar, which improves the heat stability of whey proteins. The good acid solubility of whey proteins is especially important in applications such as acid beverage and salad dressings (**Huffman, 1996**).

Morr and Foegeding (1990) analyzed solubilities of several commercial whey protein concentrates and observed that solubilities at pH 3, 4.5 and 7 were good and that whey protein isolates were more soluble at any given pH than whey protein concentrates.

Whey protein is a good emulsifier. It contains both hydrophilic and lipophilic groups and therefore has the ability to reduce the surface tension between oil and water or, in other words to form oil-in-water and water-in-oil emulsions. The emulsifying properties of WPC are highly dependent on the solubility of the protein and will diminished with decreasing solubility on the other hand as WPC is highly soluble under acid conditions, it will act as an emulsifier even at low pH values where most other proteins are insoluble. Furthermore, in some food systems, the combined emulsifying and heat gelling properties of WPC are of special interest, e.g., in the production of minced meat products and salad dressings (**Ottosen, 1991**).

Behaviour of proteins at the oil/water interface are of interest of foods. Emulsions can be liquids, semi-solids or solids and standardized methods to study emulsion properties do not exist. **Kilara (1994)** observed that as whey protein concentration increased from 0.5 to 5.0% and dispersed phase volume was kept constant at 25%, droplet size of the emulsion decreased. After more

than 15 passes in a piston homogenizer droplet size decreased was dramatic. This could also be due to the slower rate of adsorption of protein at the interfaces. Factors affecting whey protein emulsions include pH and ionic strength. Around their isoelectric point, whey proteins form poor, unstable emulsions. Also, there is no adverse effects of pasteurization of milk or whey on emulsification, but pasteurization of the retentate greatly reduced the emulsion capacity. Denaturation of proteins caused by the heat treatment was speculated to result in the observed loss of emulsion properties. The emulsion capacity and stability are important attributes in many food products.

Whey proteins are thought to form interfacial membranes around oil or water globules that prevent creaming, coalescence and oiling off. After adsorption at the fat/water interface, the protein partially unfolds to stabilize the globules. Because whey protein maintain their solubility under acidic conditions, they perform well in such applications as salad dressing. In addition, WPC can provide emulsion stability in heated foods, such as sauces, via their thickening and gelling properties. Increased viscosity reduces fat globule mobility and minimizes coalescing. Gelation can provide total entrapment of the fat emulsion within the gel network (**Huffman, 1996**).

2.6. Fortification of bakery products with whey protein concentrate (WPC)

Cereals remain the dominant vegetable protein source in the human diet, although they have a protein content of only about 10-12%. In the developing world, about 80-90% or even more of the protein intake is represented by cereal proteins. As the protein value of cereal foods is not very high, the addition of whey protein concentrate (WPC) is one way to improve the diet of low-income population groups. Also, in institutional feeding there is often a need to improve the nutritive value of traditional products at reasonable costs. Bread and pasta products belong to the group of foods whose nutritive value should be increased (**Renz-Schauen and Renner, 1987**).

Whey is one of the least expensive potential ingredients in a baker's formulation. It is at parity with flour, less expensive than dextrose or sucrose and

for less expensive than shortening and non-fat dry milk. Most important, whey can maintain or improve the final product (**Huginin, 1980**).

Vetter (1984) mentioned that dried whey can partially replace dried skim milk in bakery products. Also, modification of the functional properties of whey products used in bakery products such as whey protein concentrate improved the quality of these products.

Because of their excellent nutritional and functional properties protein concentrates obtained by ultrafiltration whey are considered to be valuable ingredients of a large range of food products, e.g. cereal products. In former investigations, the effect of whey protein concentrates (WPC) with varying protein content and denaturation degree, which had been added in varying concentration to wheat flour, on the quality of French-type bread and noodles (macaroni) has been examined (**Sanchez et al., 1989**).

Sanchez et al. (1988) mentioned that the whey protein concentrates (WPC) were produced with protein contents of 35, 45 and 60% each WPC was also manufactured with 3 different degrees of heat denaturation (low, medium and high).

2.6.1. Chemical composition and nutritional quality

Whey proteins are an excellent source of all the essential amino acids and are easily digested. Some foods lack adequate amounts of certain amino acids (e.g., wheat flour and rice are both low in lysine and soy is low in methionine). Foods consumed together can balance each other by balancing the deficits and surpluses of essential amino acids supplementing and fortifying foods with complementary proteins increases the overall nutritional value of the available protein. Whey proteins also contain high levels of the branched chain amino acids-leucine, isoleucine and valine. These amino acids are considered useful in sports drinks. Whey proteins can also used in nutritional applications such as infant and enteral formulas, weight-gain and weight-reduction diet foods, protein fortified fruit juices and other healthy foods and drinks (**Huffman, 1996**).

Whey dairy ingredients are added to nondairy foods to improve the nutritional quality, whey proteins are primarily used. A combination of whey proteins with

vegetable proteins results in a higher biological value of the mixture. The reason is the increased content of essential amino acids, mainly lysine, which is usually the limiting amino acid in cereal proteins. For instance a 50:50 cereal/whey mixture (on a protein basis) has a protein efficiency ratio (PER) 215% of that of wheat flour (**Hernandez et al., 1981**). The PER values of such mixtures vary between 2.78 and 3.87, compared to 0.45 for unsupplemented wheat. Whey protein concentrate (WPC) is considered to be the most efficient wheat protein supplement. Adding only a relatively small amount of whey protein considerably improves the protein quality (**Forsum, 1979**).

Whey protein concentrates (WPC) are by-products of cheese processing industries and are underutilized as human food. Whey proteins have a high protein efficiency ratio (PER). The availability in whey and different processes, the lack of knowledge about the interactions of whey proteins with other components such as carbohydrates during extrusion, as well as their influence on texture formation, have limited their utilization (**Martinez-Serma and Villota, 1992**).

The protein value of the bread was improved by addition of WPC because of the higher biological value of the whey proteins 104 versus 54 of wheat protein. This is shown by the chemical score relating the concentration of essential amino acids in individual proteins to the Food and Agricultural Organization reference protein. Adding WPC to wheat flour increased the chemical score of the bread protein from 36 up to 66, again depending on the amount and the protein content of the added WPC. This can be explained by an increase in the concentration of all essential amino acids. There was a remarkable increase of about 42% in the lysine content (**Renner, 1983**).

Renz-Schauen and Renner (1987) reported that WPCs were added in amounts of 2, 4 and 6% wheat flour, while the hydration was kept between 54 and 60% to get the same dough consistency in each blend. By adding the WPCs to wheat flour, the protein content of the bread was increased from 12.8% up to 15.9%, depending on the amount of the protein content of the added WPC. Addition of WPC to cereals will also lead to increased calcium content in the fortified products.

Addition of 4% whey proteins to corn, wheat or rice would significantly improve protein efficiency ratio. Further, whey proteins compared with many other proteins are less likely to mask added flavours. The fortification of cereals with WPC (average calcium content 500-700 mg/100 g) substantially increased the calcium content (**Gupta and Thapa, 1991**).

As WPC have a high protein content, the protein content of the crackers can be significantly increased when wheat flour is partly substituted by WPC. The protein content of WPC as well as the amount of WPC added result in increased protein values of the resulting crackers. While the control samples contain 11.2 protein on average, the protein value can be increased up to about 18% with the highest protein content and at the highest substitution level (**Sanchez et al., 1989**).

Voronetskene and Mikalauskaite (1991) showed that biscuits enriched with dried skim milk or dried butter milk tended to contain more lysine (2.67-3.11 g/100 g) than those enriched with soy flour (1.43-2.33 g/100 g) and had improved amino acid (AA) balance as indicated by the lower coefficient of variation for AA score (31-41 versus 47-52). Comparison of another brand of biscuits enriched with whey concentrate (11.7 g/100 g) or malted barley (5% flour replacement) showed that enrichment with malted barley (obtained as a by-product from the brewing industry) increased lysine content from 1.18 to 1.58 g/100 g and essential AA:N ratio from 1.39 to 1.49.

2.6.2. Rheological properties

Smith (1976) recommended use of whey product to improve the water absorption capacity and handling characteristics of yeast-fermented dough.

Zadow (1981) studied that examination of traces indicated that the presence of WPC in dough resulted in increased relaxation heights and an increased number of steps in the trace that corresponded to gross loss of CO₂ from the system. These results are interpreted in terms of WPC addition resulting in weaker, less elastic dough that ruptures more readily on gas expansion during baking. This conclusion is supported by microscopic examination of the structure of proofed doughs.

Holsinger (1983) reported that dough absorption was increased slightly with the addition of whey products. Non fat dry milk also increased absorption slightly. Addition of whey products to the flour increased arrival time in all cases and in general, minor increases in peak time were also observed. Most products had little effect on dough stability.

Sanchez et al. (1984) studied the effect of fortification of French-type bread with WPC on the rheological properties of the fortified doughs. Results showed that the greater was the degree of denaturation, the more elastic were the doughs obtained. WPC with a medium degree of denaturation gave an elasticity of the dough which was similar to that of the control sample. WPC with a low denaturation degree reduced the farinographic water absorption.

Dairy ingredients such as non-fat dry milk (NFDM), whey and casein are widely used in the preparation of bakery products. The nutritional, organoleptic and some functional properties of bread enriched by dairy products are improved. Increased water absorption, reduced staling rate and increased crust color are some of the advantages of dairy ingredients in bread baking (**Dubois and Dreese, 1984**). On the other hand, dough slackening and volume-depressing effects with non heated dairy fractions have been reported frequently. The performance of dairy ingredients in baking has been the subject of many publications and almost every milk fraction has been described as loaf volume-depressing. Such fractions include whey proteins (Powders or concentrates), casein and lactose (**Zadow, 1981** and **Harper and Zadow, 1984**).

The ability to produce soluble, high water binding, heat-denatured whey proteins may have applications in bakery products where high-heat treated non fat dry milk (NFDM) has long produced a more functional ingredient in dough than low-heat NFDM. The limitation of WPC in dough due to low water absorption previously observed (**Melachouris, 1984**) may no longer apply to all WPCs.

Zadow and Marston (1984) studies the effect of the addition of undenatured whey protein concentrates on the rheological behaviour of proofed bread dough, as assessed with an Instron Universal Tester. The behaviour of proofed doughs free from and containing undenatured WPC were compared. The results showed that after application of an initial pressure, the observed force decreased as a

result of ongoing fermentation. The presence of whey protein concentrates in dough resulted in greater relaxation heights and an increased number of steps in the trace that corresponded to gross loss of carbon dioxide from the system. These results indicate that addition of whey protein concentrates results in a weaker, less elastic dough that ruptures more readily on gas expansion during proofing and baking, compared to the control doughs free from WPC. This conclusion was supported by microscopic examination of the structure of proofed dough. On baking, the doughs containing undenatured WPC yielded loaves of poor volume.

Vasin (1986) found that whey increased the dough resistance to mechanical processing and controlled rheological properties of dough.

Erdogdu-Arnoczky et al. (1996) determined the effects of 4% dairy ingredients on dough absorption and mixing time, parameters of fermentation, loaf volume and bread characteristics. Dairy ingredients generally increased water absorption and decreased mixing time. The decrease in mixing time was to some extent reversed by heat treatment (at 80 or 95°C) of non fat dry milk (NFDM), casein or whey.

Jacobson (1997) examined the addition of dairy proteins to dough for faster dough development during mixing and for added strength in the final product. Native whey protein tends to have the reverse effect by lengthening the time of dough development and decreasing the loaf volume. Newer WPCs with heat-denatured, soluble protein may be better ingredients in a fresh dough. However, the same native whey protein that interfered with gluten development in fresh dough may confer a protective effect on the gluten network during freeze thaw cycles in frozen dough. Whey proteins which allow longer expansion during baking and yield softer and more fragile cakes, may be a choice ingredient in lean cakes, where tougher textures and reduced volume can occur.

Matthey and Hanna (1997) reported that the addition of WPC reduced the expansion and water absorption index under some conditions. Wheat extender addition of milk protein to cereal starch affected the extruded products quality. The decrease in protein solubility and the simultaneous increase of protein content in starch suggested formation of a protein-starch complex.

In bakery-products, WPC-35 can replace whole milk powder or skim milk powder and thereby reduce the cost of production. Because of its hydrophilic and lipophilic properties, WPC will give a good fat distribution and thus a good structure in the bread. Furthermore, because of the surface denaturation of whey protein, its water binding ability will be built up during the baking process. This means that such bakery products will keep fresh for a long time. WPC-80 at least that produced from sweet cheese whey-contains components which are gluten relaxing in wheat doughs. This is an advantage in the production of hard biscuits and crackers because it helps obtain nicely shaped products and secures a good oven rise (**Ottosen, 1991**).

Srivastava et al. (1996) studied the use of whey solids as a substitute for non-fat dried milk (NFDM) in biscuit mix at 3-15%, dough was analyzed for rheological properties and the products for quality and storage stability. Inclusion of whey solids at 3 and 10% lowered the farinograph dough stability value from 5.5 minutes (control) to 4.0 and 3.0 minutes, respectively and resistance to extension from 775 BU (control) to 715 and 680 BU, respectively.

2.6.3. Organoleptic properties

General improvement in the baking properties of bread by the addition of whey has attributed to its lactic acid and riboflavin content. Improved flavor, appearance and reduction in fermentation time was accomplished by the addition of 10-20% whey (in terms of weight of flour) to the dough in the manufacture of bread and other bakery products (**Smith, 1976**).

Specifically, whey and whey based products have generally been found to improve the flavor, aroma, color, texture and in some cases the shelf life of bakery products. Slight increases in whey levels can generally be effected with very few changes in formulation and process. If the economic of functional potential of whey products are to be maximized, some experimentation and cooperation between whey processors and bakers will be necessary (**Huginin, 1980**).

Zadow (1981) observed that the addition of WPC to bread dough resulted in only a slight reduction in the height of the loaf after final proofing. The major reason for the reduced loaf volume was the small increase in height on baking (oven spring) of these samples compared to the controls. Typically, when baking by the no-time system, an average increase in height of approximately 1.3 cm was found in the controls, whereas an increase of only 0.1 cm was observed in

the WPC containing samples. This difference in oven spring was reflected in the average loaf volumes of the controls and WPC samples, respectively 205 and 150 ml. It was also shown that the addition of WPC to the dough had no significant effect on the rate of fermentation in the system. It was clear therefore, that the WPC was influencing the physical structure of the dough resulting in either a very stiff structure that resisted expansion or an open weak structure that ruptured under the stress of the expanding gases.

Renner *et al.* (1982) found that the specific volume of bread is reduced compared with the control samples by adding whey protein concentrates. The same tendency can be observed as to the sensoric evaluation using a point-scheme. Fairly good results are obtained by using a concentrate with a medium denaturation degree and a medium protein content.

Mizyakin (1983) reported that concentrated whey and the hydrolysate concentrated whey could be used in the manufacture of bread. The sensory properties of the products were very good. The crumb and crust were higher with a better keeping quality.

In a sensory view, it could not be seen a significant change in acceptance, when WPC were added in a concentration of up to 6% by **Sanchez *et al.*, 1984**. Another report described that WPC addition in bread resulted in loaf volume reduction, increased protein and amino acid contents and that the bread was acceptable (**Sanchez *et al.*, 1986**).

Renz-Schauen and Renner (1987) showed a significant difference between the fortified bread samples and the control samples particularly when 6% of low denaturated WPC was used. However, the test panel members very often said that in spite of the different taste, the taste of the fortified bread samples was accepted very well.

Srivastava *et al.* (1996) mentioned that replacement of non-fat-dry-milk (NFDM) by whey solids at the 3% level improved color, texture and taste of biscuits. However, increasing the level of whey solids to 10% in order to improve nutritional value, resulted in biscuits with a harder texture, as indicated by the increase in breaking strength (from 2.23 to 3.03 kg) and a slightly sour taste. This was overcome by use of an emulsifier such as glyceryl monostearate or stearyl lactylate and a strong flavouring such as cardamom. Biscuits had a shelf-life of 6

months when packed in metallized polyester/polyethylene laminate under ambient conditions of 65% relative humidity and 27°C.

3. MATERIALS AND METHODS

3.1. Materials

3.1.1. Legume seeds

Three varieties of legume seeds, i.e. soybean (*Glycine max.* L.) variety Giza 21, field pea (*Pisum sativum*) variety Little Marvel and Sweet lupine (*Lupinus angustifolius*) were used in this investigation. Soybean and field pea were obtained from Food Legumes Research Department, Field Crops Research Institute, Agricultural Research Center, Giza, Egypt during the season of 1996 whereas sweet lupine was obtained from local market (imported 1996 from Australia). Different sample of legume seeds were stored under cooling at 4-6°C until used.

3.1.2. Whey powder and whey protein concentrate

Spray dried sweet whey powder (SW) extra grade Dutch origin, fresh production, safe for human consumption was obtained from Taly Establishment (Holland). Standards of Esprion 580 ultrafiltrated-whey protein concentrate (WPC) was obtained from DMV international veghel-the Netherlands.

3.1.3. Wheat flour

Hard wheat flour (72% extraction) was obtained from North Cairo Flour Mills Company, El-Hoda Mill, Shoubra El-Kheima, Egypt.

3.1.4. Trypsin

Trypsin was obtained from Sigma Chemical Company.

3.1.5. Corn oil

Commercially available corn oil was obtained from Crystal, Arma Food Industries, Egypt.

3.1.6. Vegetable shortening

Vegetable samna (Alnakhletein) was obtained from Misr Gulf Oil Processing Company, Suez, Egypt.

3.2. Methods

3.2.1. Technological treatments and processing

3.2.1.1. Preparation of legumes flours

Dry legume seeds were cleaned and sieved by hand to be free from sand, stones and any other foreign materials and then washed by water. The washed seeds were dried in an air and divided into two groups. The first group was ground to obtain the seed meal which was used to prepare the ungerminated seed flour (UGF) by separating hulls and extracting the oil with petroleum ether. The second group was germinated according to the method described by **Abou-Arab and El-Shatanovi (1993)**, The cleaned legume seeds were soaked over night in distilled water (1:5 w/v) at room temperature ($25\pm 5^{\circ}\text{C}$). The soaked seeds were washed with water for 10 minutes, drained and spreaded evenly in a single layer on top of wet paper towels, covered with another paper towel and wetted by spraying water twice daily. Germination process was carried out at room temperature up to 4 days. After germination the seeds were manually decoated to separate the hulls and then dried in an air drying oven at 50°C . The oil was extracted with petroleum ether and the residue was ground in Wiley Mill to produce the germinated seed flour (GF). Both legume flours (UGF and GE) were passed through a U.S. No. 60 sieve 250 μm .

3.2.1.2. Preparation of legume protein concentrates

Legume protein concentrates were prepared from the previous obtained legume flours according to the method described by **Baker et al. (1979)**. The soluble constituents of the flours (250 g) were eliminated with six, 20 min extraction, the slurry was filtered under vacuum through Whatman No. 2 filter paper followed by washing with one volume of ethanol 70%. The obtained protein concentrates were dried under vacuum at 50°C over night, ground, sifted and stored at -15°C till used.

3.2.1.3. Processing of bread and pan biscuit

Legume flours and their protein concentrates from soybean, field pea and sweet lupine, (as a source of plant protein) and whey protein, medium whey protein and high WPC (as a source of animal protein) were used as partial

replacement for wheat flour in pan bread and biscuit production at levels of 0, 5, 10 and 15%.

3.2.1.3.1. Pan bread processing

The straight dough process was carried out according to the method outlined by **Kent-Jones and Amos (1967)** as follows:

One hundred grams of flour, together with 25 ml of freshly prepared yeast suspension (12 gm fresh compressed yeast suspended in 100 ml water) and 25 ml of 4% sodium chloride solution were placed in a mixing bowl and the ingredients were mixed for using the rest of water obtained from farinograph at a temperature that bring the mixture about 27°C. The dough was removed from the bowl and rounded manually by folding for 20 minutes. Fermentation was carried out for 20 minutes through three consecutive stage at 30°C and 85% relative humidity. The first punch was after 105 minutes. The second was after 590 minutes and moulding was after 25 minutes. 120 gm from the fermented dough was placed in baking pans (5 x 9 x 8) and tightly greased to prevent the loaves from sticking to the tins. This was followed by proofing for 55 minutes in a cabinet at 30°C and 85% relative humidity. After proofing the pans were baked in Monline oven at 230±2°C for 25 minutes. Pan bread was cooled and packed in polyethylene bags until analysis.

3.2.1.3.2. Biscuit formulation and preparation

The formula used for the control biscuit contained 500 g wheat flour 72% extraction, 162 g sugar, 65 g vegetable shortening, 0.8 g skim milk powder, 0.4 g vanelin, 2.8 g sodium bicarbonate, 8.6 g ammonium carbonate, 0.2 g sodium bisulfate and 325 ml water (**Foda et al., 1984b**).

Fat and sugar were creamed, using the first speed in the mixing machine. The NaHCO₃ and NH₄ HCO₃ were dissolved in water and added to the creamed sugar and fat. Creaming was continued until it became light and fluffy. The other different ingredients has been added and were stirred well together. The dough was shaped handly using a biscuit cutting form. Baking was carried out in the laboratory oven at 180±2°C for 30 minutes, cooled and packed in polyethylene bags until analysis. The fortified biscuits were prepared by the same formula.

3.2.2. Chemical analysis

Different materials used for pan bread and biscuits production were subjected to chemical analysis. All determinations were carried out in triplicates.

3.2.2.1. Moisture content

Moisture content was determined as described in the **A.O.A.C. (1990)**.

3.2.2.2. Total nitrogen

Total nitrogen was determined using micro-kjeldahl method as recommended by **A.O.A.C. (1990)**. Protein content was calculated by multiplying total nitrogen percentage by the factors 5.7 for the wheat flour, 6.25 for legumes and 6.38 for whey products. For wheat flour and legumes blends, a combined conversion factor was interpreted taking into consideration the wheat flour : legume production in the blend.

3.2.2.3. Crude fat

Lipids were extracted from the samples in Soxhlet apparatus using petroleum ether and calculated as percentage according to **Less (1975)**.

3.2.2.4. Crude fiber

Crude fiber was determined according to **A.O.A.C. (1990)**.

3.2.2.5. Ash content

Ashing was carried out using a muffle furnace at 550°C until constant weight was obtained according to the method described in the **A.O.A.C. (1990)**.

3.2.2.6. Trypsin inhibitor activity (TIA)

The method obtained by **Roy and Reo (1971)** was employed for determining the trypsin inhibitor activity for different legume samples.

Extraction of Trypsin inhibitor: One gram of fine ground samples of the dry legumes were treated with 10 ml of 0.05 M sodium phosphate buffer adjusted at pH 7.0. The mixture was shaken for 3 hours at room temperature, kept over night at refrigerator and centrifuged at 2000 rpm for 30 minutes at 15°C. The supernatants were filtered through Whatman No. 1 to get clear solutions of which 1.0 ml was diluted to 10 ml using distilled water.

Determination of TIA: A 2% casein solution in phosphate buffer (0.1 M, pH 7.6) was used as substrate, while the enzyme used was trypsin (5 mg/ml HCl 0.001 M), Sigma Co. The incubation mixture consisted of 0.5 ml of trypsin

solution, 2.0 ml of 2% casein, 1.0 ml of phosphate buffer, 0.4 ml of hydrochloric acid solution (0.001 M) and 0.1 ml sample extract. The mixture was incubated at 37°C for 20 min. after which 6.0 ml of 5% trichloroacetic acid (TCA) was added to stop the reaction. The absorbance of the mixture was measured using ultra violet spectrophotometer model Shimadzu UV-2401 PC/2501 PC. At 280 nm, against a blank consisting of 0.5 ml HCl solution (0.001 M) and 0.1 ml phosphate instead of the sample. One trypsin unit (TU) is arbitrarily defined as an increase of 0.01 absorbance unit for 10 ml of the reaction mixture. The trypsin inhibitor activity was calculated as the number of trypsin units inhibited by milligram of dry sample.

3.2.3. Physical measurement

The percentage of water absorption of 100 g of legume seeds were followed during soaking and germination by weighing the seeds before and after soaking and germination. Hydration ratio was computed by dividing the weight of hydrated sample by the initial weight. Volume of soaked and germinated seeds was determined by using seed displacement method.

Wet and dry glutes were separated from different blends and determined according to the method outlined in **A.A.C.C. (1983)** and calculated the hydration ratio as follows:

$$\frac{\text{Wet gluten} - \text{dry gluten}}{\text{dry gluten}} \times 100$$

Baking quality was measured by weighing bread loaves (g) after their removal from the pan within one hour of baking. The volume (cm³) was measured by clover seed replacement method. The specific volume was obtained by dividing the loaf volume by its weight (**A.A.C.C., 1983**).

3.2.4. Functional properties

Functional properties were carried out on flours and protein concentrates obtained from ungerminated and germinated legume seeds.

3.2.4.1. Water and oil absorption

Water and oil absorption were determined according to **Benchat (1977)** as follows: One gram of each sample was mixed with 10 ml either distilled water or corn oil for 1 min. in a 25 ml centrifuge tube. The samples were then allowed to stand at room temperature for 30 minutes, centrifuged at 1500 rpm for 30 minutes

and the volume of supernatant was noted in 10 ml graduated cylinder. The results were calculated as g water or oil (density of oil = 0.9198 g/ml) by 100 g dry sample.

3.2.4.2. Emulsifying properties

Oil emulsifying capacity (EC) was evaluated in 100 ml of 1% (w/v) aqueous dispersion of each sample at pH values 3, 4.5, 6, 7.5 and 9 by titration with corn oil. To the break point of emulsion using warning blender at low speed (**Marshall et al., 1975**). Emulsifying capacity was expressed as ml oil emulsified by 1 g sample. The emulsion was transferred to 250 ml graduated cylinder and emulsion stability (ES) was recorded in terms of the percent aqueous phase separated at time intervals of 0.25, 0.50, 2.00, 3.00 and 48.00 hrs (**Dipak and Kumar, 1986**).

3.2.4.3. Nitrogen solubility index (NSI)

Nitrogen solubility index was determined according to the method described by **Thompson and Cho (1984)**. The 1% aqueous suspension (w/v) of each protein product adjusted to pH values of 3, 4.5, 6, 7.5 and 9. Each suspension was stirred for 30 minutes then centrifuged for 30 minutes at 3500 rpm. The supernatant was then decanted, filtered through Whatman filter paper No. 40 and analyzed for nitrogen content by the micro-kjeldahl procedure (3.2.2.2.). The results are calculated as percentage of soluble nitrogen based on nitrogen of the sample.

3.2.5. Rheological properties

Farinograph tests

Farinograph tests were carried out on the wheat flours along with their mixtures with different ratios of legume-flours and protein concentrates, used in the processing of bakery products. According to the farinograph schedule, an amount of 49.43 gm flour (13% moisture) was used for farinograph test (**A.A.C.C. 1983**). The temperature was kept at $30 \pm 2^\circ\text{C}$. When the mixing curve level was high than 50, Brabender unit (B.U.) more water was added and the bowl was covered with a glass plate to prevent evaporation. The first titration attempted rarely produced a curve which was maximum resistance centered on 500 B.U. line, therefore, in a subsequent titration the absorption was adjusted up or down

until this was achieved to within 20 B.U. for final titration. All water was added within 25 seconds after opening the buret stopcock. The following constants were determined:

- A) **Water absorption:** was calculated by means of the following equation:

$$\text{Absorption \%} = 2 [X + (Y-50)]$$

Where : X = ml water required to produce a curve with maximum consistency on 500 B.U. line.

Y = grams of flour used, equivalent to 50 grams at 14% moisture basis.

- B) **Arrival time:** Minutes required for the curve to reach the 500 B.U. line after the mixer had been started through addition of water.
- C) **Dough development time:** the time in minutes from first consistency or minimum mobility, till leaving the curve 500 B.U. line.
- D) **Dough stability:** Differences in time, to the nearest minute, between the time when the curve first intercepted the 500 B.U. line (arrival time) and the time when the curve leave the 500 B.U. line.
- E) **Mixing tolerance index:** Difference in Brabender units from the top of curve at the peak to the top of curve measured 5 minutes after the break.
- F) **Time to break down:** the time in minutes from beginning of mixing till center of the curve at 470 B.U. after leaving the curve 500 B.U. line.
- G) **Weakening degree:** Difference in Brabender units from 500 B.U. line to center of the curve measured after 12 minutes leaving the 500 B.U. line.

3.2.6. Organoleptic evaluation

Fresh loaf samples baked under the previous experimental treatments were organoleptically evaluated by eleven members semi trained preference taste panel from the staff of the Food Sci. and Dairying Dept., National Research Center. The external and internal characteristics were scored using the report sheet according to **Kramer and Twigg (1962)**.

Table (4): Taste panel scores for pan bread samples (**Kramer and Twigg, 1962**).

Characteristics	Score	Samples					
		1	2	3	4	5
Appearance	20						
Crumb texture	20						
Crumb grain	20						
Crust color	10						
Taste	20						
Odor	10						
Total score	100						

Appearance, color, odor, taste, mouth-feel, texture and crispiness of baked biscuit were evaluated organoleptically as described by **Saleh (1998)** as follows:

Table (5): Panel scores for biscuit samples (**Saleh, 1998**).

Quality attributes	Score	Samples					
		1	2	3	4	5
Appearance	10						
Color	20						
Odor	10						
Taste	20						
Mouth feel	10						
Texture	15						
Crispiness	15						
Total score	100						

The mean values for each of the parameters in the organoleptic analysis were subjected to statistical analysis using analysis of variance and Duncan's Multiple Range Test.

3.2.7. Statistical analysis

The experimental design of all studies was a completely randomized with five replications. Analysis of variance (ANOVA) of the data was performed with the MSTAT-C Statistical Package (A Microcomputer Program for The Design, Management and Analysis of Agronomic Research Experiments, Michigan State Univ., USA) as outlined by **Gomez and Gomez (1984)**. Duncan's multiple range test and/or least significant difference (LSD) were used to compare treatment means as suggested by the method of **Duncan (1955)**.

4. RESULTS AND DISCUSSION

Part I. Legume as a source of plant protein

4.1. Hydration ratio of some germinated legume seeds:

Changes in weight and volume values of legume seeds were followed during soaking and germination of the seeds and the data are given in Table (6). Results showed that soaking of 100 g legume seeds overnight in the water caused a considerable increase % in their weight ranged between 228.00-251.33 g. However, volume of legume seeds raised from 85.00-87.33 cm³ to about 213.53-236.03 cm³ after soaking. On the other hand, specific weight slightly reduced after soaking of different seeds overnight.

Hydration ratio, a good parameter for measuring the swelling of a seed, was calculated as the ratio of the weight of swollen seeds to the weight of the dry seeds. Lupine seeds showed the lower hydration ratio compared with the other tested legumes. Same findings were observed by **El-Shatanovi (1992)**.

The weight and volume values of soaked-legume seeds were gradually increased during 4 days of germination. Soybean seeds recorded higher weight value, being (302 g) followed by field pea (294.33 g) and lupine (277 g). Similarly the maximum volume value was noticed also for soybean seeds (317.53 cm³) and the minimum value was obtained for lupine seeds (276.67 cm³). On the other hand, specific weight was reduced slightly during germination of different legume seeds. On contrary, hydration ratio was increased gradually during germination process. Statistical analysis of the obtained data showed that the weight and volume values of the seeds were increased significantly during soaking and germination except after the first day of germination. However, the rate of increase in weight and volume values were more pronounced during soaking than during germination. On the other hand, soybean seeds showed significantly higher weight and volume values in comparison with the other two legume seeds as a result of soaking and germination.

No significant differences were obtained for the specific weight of different legume samples as a result of soaking and germination. With respect to the hydration ratio results showed that soybean and field pea seeds recorded significantly higher hydration ratio values than sweet lupine.

Table (6): Changes in weight (g) and volume (cm³) of legume seeds as a result of soaking and germination process of 100 g seeds.

	Soybean	Field pea	Sweet lupine	Total mean*
Before soaking				
Weight (g)	100	100	100	100 ^e
Volume (cm ³)	87.33	85.00	85.33	85.89 ^e
Specific weight	1.15	1.18	1.17	1.17 ^a
After soaking				
Weight (g)	247.00	251.33	228.00	242.11 ^d
Volume (cm ³)	231.33	236.03	213.53	226.97 ^d
Specific weight	1.07	1.06	1.07	1.07 ^b
Hydration ratio	2.5	2.5	2.3	2.41 ^d
Germination period (day)				
(1 day)				
Weight (g)	252.67	252.00	225.00	243.22 ^d
Volume (cm ³)	236.80	244.43	218.63	233.29 ^d
Specific weight	1.07	1.03	1.03	1.04 ^{bc}
Hydration ratio	2.5	2.5	2.3	2.43 ^d
(2 days)				
Weight (g)	265.67	260.67	237.67	254.67 ^c
Volume (cm ³)	265.33	251.90	229.67	248.97 ^c
Specific weight	1.00	1.03	1.03	1.02 ^c
Hydration ratio	2.7	2.6	2.4	2.55 ^c
(3 days)				
Weight (g)	297.67	287.33	261.67	282.22 ^b
Volume (cm ³)	312.20	287.67	262.00	287.29 ^b
Specific weight	0.95	0.99	0.99	0.98 ^d
Hydration ratio	3.00	2.90	2.60	2.82 ^b
(4 days)				
Weight (g)	302.00	294.33	277.00	291.11 ^a
Volume (cm ³)	317.53	295.00	276.67	296.40 ^a
Specific weight	0.95	0.99	1.00	0.98 ^d
Hydration ratio	3.02	2.90	2.80	2.91 ^a
Mean of seed**				
Weight (g)	244.17 ^A	240.94 ^A	221.56 ^B	
Volume (cm ³)	241.76 ^A	233.34 ^B	214.31 ^C	
Specific weight	1.03 ^A	1.05 ^A	1.05 ^A	
Hydration ratio	2.74 ^A	2.68 ^A	2.48 ^B	

* Mean within the column followed by the same small letter(s) are not significantly different at $P \leq 0.05\%$.

** Mean within the same column followed by the same capital letter(s) are not significantly different at $P \leq 0.05\%$.

Same results were obtained by **Griswold (1962)**, **Hsu et al. (1980)** and **El-Shatanovi (1992)**.

4.2. Legume flours and protein concentrates:

4.2.1. Chemical composition:

Means of the data obtained for the major chemical constituents of whole legume seeds (soybean, field pea and sweet lupine), legume flours and their protein concentrates as affected by germination process were analyzed by analysis of variance and the differences among the means were compared by Duncan's multiple range test, (Table 7). Data showed that chemical analysis values differed significantly among the three ungerminated whole legume seeds used in this study. Lupine seeds contained significantly high amounts of protein content (43.05%) and crude fiber (12.71%) than the other two legume seeds. However, soybean seed had significantly higher values of fat (24.88%), ash (5.73%) and energy of value (438.62). The total carbohydrates content of field pea was significantly higher (49.63%) than the other two legumes.

Same findings were indicated by **Mohamed and Rayas-Duarte (1995)**. They showed that protein and total carbohydrates contents of lupine seeds were higher than that of soybeans. However, the oil content of lupine seeds was lower than that of soybean. **Danangelo et al. (1995)** found that soybean contained high ash content and low dietary fiber than lupine seeds. The soybean seeds had higher protein content and fat values and lower carbohydrate content than pea seeds (**Fernandez-Quintela et al., 1998**). Whole soybean seeds contained 40% protein, 21.0% oil, 4.9% ash and 34% carbohydrates (**Singh et al., 1987**).

Chemical composition of ungerminated legume flour prepared from dehulled seeds showed that different legume flours contained significantly more protein and fat contents and less crude fiber than whole legume seeds and these could be attributed to the removal of legume hulls. Dehulled samples showed similar ash content like hulled ones except for lupine.

Dehulling improved protein content by 7.24, 9.94 and 10.41% for soybean, field pea and sweet lupine flours respectively, Fig. (2).

Table (7): Chemical composition* of whole legume seeds, flours and their protein concentrates as affected by germination process.

Sample	Moisture %	Chemical composition calculated on dry weight basis					Total Carbo hydrate %**	Value of energy cal/100 g ***
		Protein %	Fat %	Ash %	Fiber %			
Whole seed								
Soy bean	7.45 h	39.39 k	24.88 c	5.73 d	6.98 c	23.02 i	438.61 c	
Field pea	8.99 cde	33.50 m	4.42 g	3.29 j	9.15 b	49.63 b	355.23 kl	
Sweet lupine	7.95 gh	43.05 j	11.66 de	3.59 hi	12.71 a	28.99 h	364.96 gh	
Ungerminated (UG)								
Dehulled flour								
Soy bean	6.54 i	42.24 j	28.50 a	5.55 d	1.38 j	22.32 i	475.98 a	
Field pea	8.51 d-g	36.83 l	6.72 f	3.23 j	3.55 ef	49.67 b	386.23 f	
Sweet lupine	8.79 c-f	47.53 i	12.43 d	3.20 j	1.83 hij	35.01 f	411.43 d	
Defatted flour								
Soy bean	9.15 cd	59.77 f	0.57 i	8.24 b	1.72 ij	29.70 gh	333.09 p	
Field pea	8.48 d-g	37.64 l	0.80 i	3.91 g	2.71 f-i	54.94 a	360.93 hij	
Sweet lupine	8.69 c-g	56.18 g	1.94 h	4.30 f	1.41 j	36.17 f	358.38 ijk	
Protein concentrate								
Soy bean	9.23 cd	74.26 b	0.50 i	7.01 c	3.56 ef	14.67 k	321.58 q	
Field pea	10.35 b	47.59 i	0.65 i	1.80 l	2.63 f-i	47.33 c	363.19 ghi	
Sweet lupine	9.41 c	70.45 c	1.95 h	3.22 j	1.86 hij	22.52 i	352.39 lm	
Germinated (G)								
Dehulled flour								
Soy bean	5.78 j	47.18 i	27.19 b	6.73 c	3.47 ef	15.42 k	454.08 b	
Field pea	10.19 b	42.73 j	5.07 g	3.77 gh	5.01 d	43.42 d	367.41 g	
Sweet lupine	5.10 j	51.50 h	10.98 e	4.04 fg	2.24 g-j	31.23 g	397.73 e	
Defatted flour								
Soy bean	11.11 a	65.13 d	0.37 i	9.21 a	2.99 fgh	22.30 i	319.87 q	
Field pea	10.31 b	43.56 j	1.28 hi	4.20 f	3.78 ef	47.18 c	356.93 jkl	
Sweet lupine	8.04 fgh	61.33 e	1.91 h	4.65 e	3.17 fg	28.93 h	346.56 no	
Protein concentrate								
Soy bean	10.42 ab	78.66 a	0.36 i	6.76 c	4.65 de	9.57 l	314.93 r	
Field pea	10.30 b	51.20 h	0.52 i	2.77 k	4.41 de	41.11 e	349.29 mn	
Sweet lupine	8.23 e-h	73.94 b	1.82 h	3.47 ij	3.50 ef	17.27 j	342.09 o	

* Means within the same column followed by the same letter(s) are not significantly different at 0.05 level.

** Calculated by difference.

*** (Protein x 3.47 + Fat x 8.37 + carbohydrate x 4.07).

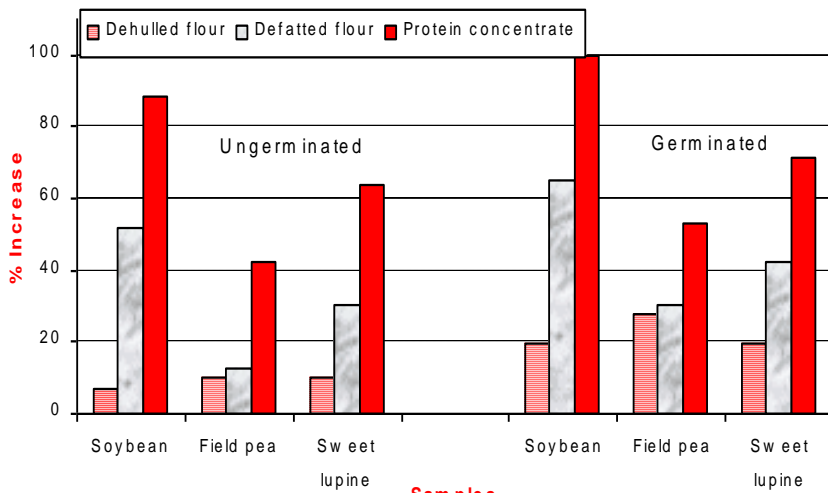


Fig. (2): Percentage of protein increase of legume flours and their protein concentrates as affected by germination (compared to that of whole seeds).

These results are in agreement with those obtained by **Deshpande et al. (1982)**, **Abou Arab and El-Shatanovi (1993)** and **Hassan (1998)**.

Defatting process significantly reduced fat content and increased protein content of the resultant defatted flours. Protein content improved by 51.74, 12.36 and 37.91% for defatted soybean, field pea and lupine flours, respectively, (Fig. 2). Defatting process significantly increased total carbohydrates and reduced the energy values of all legume flours. **Smith and Circle (1972)** reported that full-fat soybean flour contained 47% protein, 22% oil, 5% ash and 2% fiber. However, defatted soybean flour showed 59% protein, 1% oil, 6% ash and 3% fiber. Similar results are obtained by **Bressani (1981)**.

Results presented in the same table showed that legume protein concentrates prepared from ungerminated legume seeds contained much higher protein content than that of legume flours. Protein content differed significantly among all the three legume-protein concentrates used in this study and it reached 74.26, 47.59 and 70.45% for soybean, field pea and lupine protein concentrates, respectively. It was clearly noticed that ungerminated soybean protein concentrates characterized by highest protein, ash and crude fiber and lowest total carbohydrates. Same findings were reported by **Garcia et al. (1998)** and **Hassan (1998)**. However, **Meyer (1967)** and **Bressani (1981)** found that soybean protein concentrate contained high protein content (70%), with small amount of oil (0.3-1%), ash (5-6%), fiber (3.7-4%) and total carbohydrates (20%).

In Egypt, it is common to germinate some legume seeds which are rich in protein (20-50%) before direct eating, cooking or use in a salad dressing. Germination improves the nutritional value of the proteins which are hydrolyzed into easily assimilable polypeptide and essential amino acids, and decreases trypsin inhibitors (**Ahmed et al., 1995**).

Therefore, the effect of germination of legume seeds on the chemical composition of the legume flours and their protein concentrates were determined and the results are given in Table (7). It is clearly appeared that protein content increased significantly after four days of germination at room temperature for all the samples. The percentages of increase reached 19.78, 27.56 and 19.63% for soybean, field pea and lupine flours, respectively as a result of dehulling and

germination (compared to whole seeds) as could be seen in Fig. (2). These increases might be due to a synthesis of enzyme proteins or a compositional change following degradation of other constituents (**Bau et al., 1997**).

Total carbohydrates were reduced significantly for all legume flours as a result of germination and it could be attributed to the degradation of polysaccharide under the effect of amylase and phosphorylase enzymes in respiratory metabolism. Total carbohydrates were decreased from 22.32, 49.67 and 35.01% for dehulled soybean, field pea and lupine flours to 15.42, 43.42 and 31.23% for the former germinated samples, respectively.

Germinated dehulled flours showed higher ash and crude fiber than ungerminated flours. These results are in accordance with those reported by **Kavas and El (1992)** and **Hassan (1998)**. On the other hand, germination slightly reduced the fat content of legume flours with comparison with ungerminated samples. **Bau et al., (1997)** reported that lipid content of seeds gradually diminishes as germination progresses.

Results also revealed that flours and protein concentrates of germinated defatted legume significantly contained higher protein content than the corresponding ungerminated samples as can be seen in Fig. (2).

For example defatted flours of germinated soybean, field pea and lupine samples contained 65.13, 43.56 and 61.33% respectively. The corresponding values for the aforementioned protein concentrate samples were 78.66, 51.20 and 73.94% respectively. On the other hand, germinated legume products had higher crude fiber than ungerminated ones.

It was clearly observed from the previous results that soybean protein concentrates characterized by highest protein, ash and crude fiber and lowest total carbohydrates.

4.2.2. Trypsin inhibitor:

Trypsin inhibitor activity (TIA) of the whole legume seeds, ungerminated and germinated defatted legume flours and their protein concentrates are presented in Table (8). Whole soybean seeds had significantly higher TIA (46.35

Table (8): Trypsin inhibitor (TIA)* of legume products as affected by germination*.

Sample	TIA (TIU/mg sample)	TIA (TIU/mg sample)
Whole seeds		
Soybean	46.35 ^a	
Field pea	7.65 ^e	
Sweet lupine	7.47 ^{ef}	
Ungerminated		
	Defatted flour	Protein concentrate
Soybean	45.65 ^a	38.88 ^b
Field pea	6.91 ^{efg}	5.88 ^{gh}
Sweet lupine	6.20 ^{fgh}	6.00 ^{gh}
Germinated		
	Defatted flour	Protein concentrate
Soybean	30.12 ^c	26.23 ^d
Field pea	3.55 ^j	3.24 ^j
Sweet lupine	5.30 ^{hi}	4.10 ^{ij}

* Means in the same column followed by the same letter(s) are not significantly different at $P \leq 0.05$.

TIU/mg sample) than field pea (7.65 TIU/mg) and lupine (7.47 TIU/mg). Dehulling and defatting process reduced slightly TIA by 1.51, 9.02 and 17.00% for defatted flour of ungerminated soybean, field pea and lupine, respectively.

With respect to the effect of germination, results revealed that TIA reduced significantly by 34.02, 48.99 and 14.52% for germinated soybean, field pea and lupine flours, respectively (in comparison with ungerminated samples).

On the other hand, legume protein concentrates showed lower TIA than the corresponding samples of legume flours. For example, ungerminated and germinated soybean flours contained 45.65 and 30.12 TIU/mg reduced to 38.88 and 26.23 TIU/mg for ungerminated and germinated soybean protein concentrates, respectively. These results are in agreement with those obtained by **Hsu *et al.* (1982)**, **Sarita *et al.* (1996)**, **Bessar and El-Sayed (1997)**, **Idris (1997)** and **Hassan (1998)**.

4.2.3. Functional properties:

Plants constitute an enormous source of proteins for human consumption. However, to be exploited successfully, these protein must be presented in forms that are attractive and possess the flavor, texture and quality desired by the consumer. The properties of plant proteins that determine their uses in foods are collectively called functional properties (**Abou-Arab, 1991**).

4.2.3.1. Water and oil absorption:

Water and oil absorption capacities (WAC and OAC) and water-oil absorption index values (WOAI) of different legume products are represented in Table (9). Defatted ungerminated soybean and lupine flours showed similar WAC, being 269.2 and 268.1 g water/100 g sample respectively. However, WAC of ungerminated pea flour was lower significantly (180.8 g water/100 g sample) than the other two legume flours. Water absorption variations among the tested samples may be related to the nature and type of proteins. Hydrophilic properties of proteins are related to such polar groups as carbonyl, hydroxyl, amino, carboxyl and sulfhydryl. Water-binding capacity varies with the number and type of polar groups (**Kuntz, 1971**). Moreover, the increased water absorption of the defatted products may have been due to exposure of water-binding sites on side chains of proteins previously blocked in a lipophilic

Table (9): Water and oil absorption capacities* (WAC and AOC) and water-oil absorption index (WOAI) of some legume products.

Sample	WAC (g water/100 g sample)	OAC (g oil/100 g sample)	WOAI (g water/ g oil)
Ungerminated			
Defatted flour			
Soybean	269.2 ^c	278.5 ^{ab}	0.96 ^{gh}
Field pea	180.8 ^d	225.9 ^e	0.80 ^{gh}
Sweet lupine	268.1 ^c	255.1 ^c	1.04 ^{efg}
Protein concentrate			
Soybean	341.9 ^b	153.6 ^h	2.23 ^a
Field pea	356.8 ^b	242.8 ^{cd}	1.47 ^{cd}
Sweet lupine	393.9 ^b	224.7 ^e	1.75 ^{bc}
Germinated			
Defatted flour			
Soybean	244.4 ^c	231.4 ^{de}	1.06 ^{efg}
Field pea	250.0 ^c	271.8 ^b	0.92 ^{gh}
Sweet lupine	244.8 ^c	215.4 ^e	1.14 ^{ef}
Protein concentrate			
Soybean	452.7 ^a	194.4 ^f	1.99 ^{ab}
Field pea	387.5 ^b	289.5 ^a	1.34 ^{de}
Sweet lupine	396.9 ^b	177.8 ^g	2.22 ^a

* Means in the same column followed by the same letter(s) are not significantly different at $P \leq 0.05$.

environment. Water absorption of legume protein concentrate was higher significantly than that of legume flours. WAC of ungerminated soybean, field pea and lupine protein concentrates were reached 341.9, 356.8 and 393.9 g water/100 g sample, respectively. No significant differences were obtained for WAC among the three samples of ungerminated legume protein concentrates. These results were in agreement with those obtained by **Sosulski and Fleming (1977)** for soybean flour and concentrates. Increased water absorption by legume products with increased protein contents was reported by **Fleming et al. (1974)**, **Al-Kahtani and Abou-Arab (1993)** and **Hassan (1998)**. However, water absorption of a particular sample need not parallel to its protein content. **Lin et al. (1974)** observed that all sunflower products had lower water absorption than those of soybean products, although their protein contents were similar. Also, **Tjahjadi et al. (1988)** showed that differences in carbohydrate content might also have affected water absorption.

Variations in the water absorption values were relatively small among the ungerminated and germinated legume products.

Germination process only improved significantly WAC of field pea flour from 180.8 to 250 g water/100 g sample and soybean protein concentrate from 341.9 to 452.7 g water/100 g sample. Same findings were obtained by **Hassan (1998)**. He found that germination process had variable effect on WAC of three legume products (chickpea, lupine and mung bean).

Oil absorption is mainly attributed to the physical entrapment of oil (**Kinsella, 1976**). It is also related to the number of nonpolar side chains on proteins that bind hydrocarbon chains on the fatty acid.

Results in Table (9) showed that ungerminated soybean flour had significantly higher oil absorption (278.5 g oil/100 g sample) followed by ungerminated lupine and field pea flours (255.1 and 225.7 g oil/100 g sample). On the other hand, ungerminated soybean and lupine protein concentrate had lower OAC than their flours. Oppositely, oil absorption of ungerminated pea flour was higher than their protein concentrates. **Al-Kahtani and Abou-Arab (1993)** found that defatted flour of soybean had higher oil absorption than their protein concentrate.

Germination process had variable effect on OAC of legume products as can be seen in the same table.

The water-oil absorption index (WOAI) of various legume samples were calculated and given in Table (9). This index is a measure of relative simultaneous attraction of a protein to water and oil. Ungerminated and germinated legume flours showed low WOAI (nearly one). However, most of the legume protein concentrates showed high WOAI (nearly two). Generally, soybean and lupine protein concentrates had higher WOAI than the field pea protein concentrates. Same results were obtained by **Al-Kahtani and Abou-Arab (1993)** and **Hassan (1998)**. This indicates that the protein molecule acted as a mediator in the formation of stable emulsion by binding both water and oil molecules to form thick barriers which prevented the oil particles from coalescing (**DeKanterewicz, et al., 1987**).

4.2.3.2. Emulsion capacity and stability

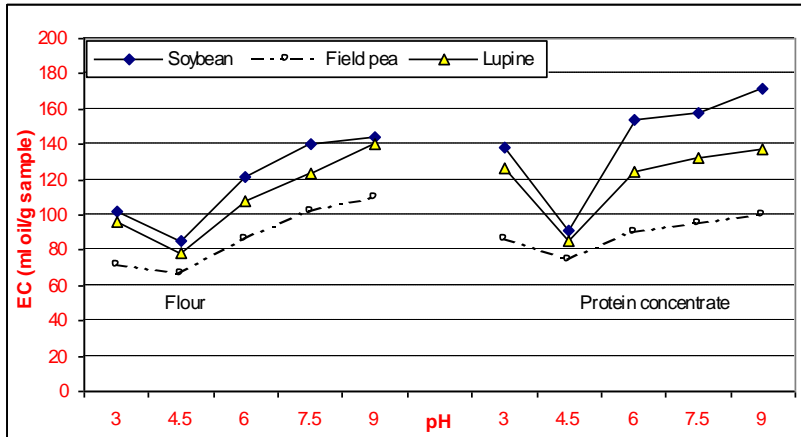
The ability of proteins to aid the formation and stabilization of emulsions was critical for many food applications. The emulsifying capacity of protein depended on the suitable balance between the hydrophilic and lipophilic characteristics rather than merely on the high values for each one (**De-Kanterewicz et al., 1987**). The stability of emulsions had also been related to the spreading coefficients of the internal phase liquid on the surface of a solution of the emulsifier in the continuous phase (**Petrowski, 1976**).

Emulsion capacity (EC) of flour and protein concentrate of ungerminated and germinated legume products are measured at different pH values and the results are given in Fig. (3).

Results revealed that minimal emulsifying capacities of different legume products were measured at pH 4.5, near the isoelectric point with the lower protein solubility and markedly increased below and above pH 4.5, reaching their maximum at pH 9.0.

These results are in accordance with those obtained by **Wang and Zayas (1992)**, **Al-Kahtani and Abou-Arab (1993)**, **El-Adawy and Khalil (1994)** and **Hassan (1998)**.

Ungerminated



Germinated

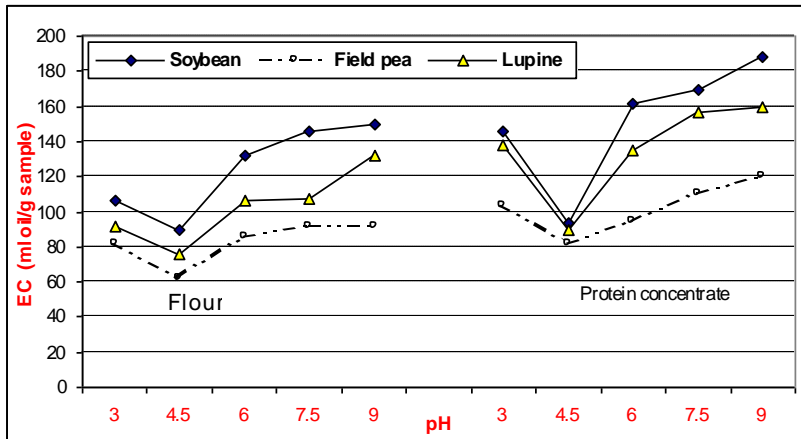


Fig. (3): Emulsion capacity (EC) of 1% dispersion of soybean, field pea and sweet lupine products.

The emulsion capacity values for soybean products were generally higher than those of the corresponding field pea and lupine products at all the pH values studied. At pH 4.5 (minimum solubility), the respective EC for defatted flour of ungerminated soybean, field pea and lupine seeds were 85, 67 and 78 ml oil/g sample respectively. With increasing the pH to 9.0, EC was increased to 144, 110 and 140 ml oil/g for the former samples.

Germination process had variable effect on EC, as it improved EC of soybean flours, it reduced EC of field pea and lupine flour. These results are similar to those obtained by **Abou-Arab and El-Shatanovi (1993)** and **Hassan (1998)**.

On the other hand, legume protein concentrates had significantly higher EC than the corresponding samples of legume flours. The maximum EC was recorded for soy protein concentrates followed by lupine and field pea protein concentrates, (Fig. 3).

The emulsion capacity versus pH profile of legume seed products closely resembled to protein solubility in shape, suggesting that emulsification was caused by the solubilized proteins. Similar observations on the relationship of pH and emulsifying capacity of proteins have been reported by several investigators (**Lin *et al.*, 1974**; **Abou-Arab and El-Shatanovi, 1993** and **Al-Kahtani and Abou-Arab, 1993**).

Emulsion stability (ES) is important because the success of an emulsion depends on its ability to maintain the emulsion in subsequent processing steps. Soybean flour and isolates are excellent emulsifiers and binders in high-fat foods, and this characteristics has been associated with their high water- and fat-absorption properties (**Porteous and Wood, 1983** and **Mittal and Usborne, 1985**).

Emulsion stability (ES) of 1% dispersion of flour and protein concentrate of ungerminated and germinated legume seeds were followed during 48 hr. and the results are given in Tables (10, 11 and 12).

The higher ES, i.e., the lower percentage of aqueous phase separated after 48 hrs for different legume samples, was noticed in most cases at higher pH value.

Table (10): Emulsion stability (ES) of 1% dispersion of soybean flour, and protein concentrate.

Sample	pH values	ES% Water separated after time, hr.					
		0.25	0.50	2.00	3.00	24.00	48.00
Ungerminated (UG)							
Soy bean flour (USF)	3.0	4.14	8.20	12.30	12.30	24.60	30.74
	4.5	31.53	33.78	33.78	33.78	36.04	42.79
	6.0	18.18	22.73	22.73	22.73	27.27	31.82
	7.5	0.00	0.00	4.17	4.17	12.50	12.93
	9.0	0.00	0.00	0.00	0.00	4.31	12.50
Protein concentrate (USPC)	3.0	0.00	8.13	12.19	16.26	20.33	20.33
	4.5	8.00	20.00	24.00	28.00	32.00	32.00
	6.0	0.00	12.77	14.89	17.02	21.28	21.28
	7.5	7.58	11.36	15.15	15.15	18.94	18.94
	9.0	1.89	3.79	11.36	11.36	15.15	15.15
Germinated (G)							
Soy bean flour (GSF)	3.0	0.00	2.03	4.07	6.10	10.16	10.16
	4.5	0.00	3.73	4.46	14.93	14.93	14.93
	6.0	0.00	2.38	4.76	4.76	14.29	14.29
	7.5	3.68	7.35	11.03	11.03	11.03	11.03
	9.0	0.00	0.00	0.00	0.00	6.25	6.25
Protein concentrate (GSPC)	3.0	0.00	0.00	5.36	19.53	23.44	27.34
	4.5	0.00	0.00	17.09	21.37	27.78	29.91
	6.0	17.36	20.83	24.31	24.31	27.78	27.78
	7.5	0.00	0.00	11.45	11.45	22.90	22.90
	9.0	0.00	0.00	10.71	14.29	17.86	21.31

Table (11): Emulsion stability (ES) of 1% dispersion of field pea flour, and protein concentrate.

Sample	pH values	ES% Water separated after time, hr.					
		0.25	0.50	2.00	3.00	24.00	48.00
Ungerminated (UG)							
Field pea flour (UPF)	3.0	4.95	9.90	14.85	17.33	24.75	34.65
	4.5	47.90	53.89	59.88	65.86	65.86	65.86
	6.0	0.00	0.00	23.25	34.88	46.51	46.51
	7.5	14.29	23.81	28.57	33.33	33.33	38.10
	9.0	5.38	5.38	10.75	10.75	10.75	16.13
Protein concentrate (UPPC)	3.0	10.53	15.79	21.05	26.32	31.58	31.58
	4.5	15.46	20.62	25.77	30.93	36.08	41.24
	6.0	21.51	26.88	26.88	26.88	32.26	32.26
	7.5	5.71	11.43	17.14	22.86	25.71	25.71
	9.0	0.00	0.00	15.00	15.00	20.00	20.00
Germinated (G)							
Field pea flour (GPF)	3.0	6.17	18.52	24.69	24.69	30.86	30.86
	4.5	10.75	16.13	21.51	21.51	26.88	32.26
	6.0	10.42	20.83	20.83	20.83	31.25	31.25
	7.5	5.21	10.42	15.63	15.63	26.04	31.25
	9.0	5.49	10.99	10.48	16.48	21.98	21.98
Protein concentrate (GPPC)	3.0	0.00	4.35	13.04	26.09	26.09	26.09
	4.5	0.00	24.79	28.93	33.06	37.19	37.19
	6.0	11.03	18.38	25.74	29.41	29.41	29.41
	7.5	0.00	0.00	14.02	21.03	28.04	28.04
	9.0	0.00	0.00	0.00	0.00	9.26	14.81

Table (12): Emulsion stability (ES) of 1% dispersion of lupine flour, and protein concentrate.

Sample	pH values	ES% Water separated after time, hr.					
		0.25	0.50	2.00	3.00	24.00	48.00
Ungerminated (UG)							
Lupine flour (ULF)	3.0	5.10	15.31	30.61	35.71	45.12	45.12
	4.5	5.68	11.36	22.73	28.41	51.14	51.14
	6.0	22.99	28.73	40.23	45.98	45.98	45.98
	7.5	4.67	14.02	23.36	32.71	42.06	42.06
	9.0	4.17	8.33	16.67	29.17	37.50	37.50
Protein concentrate (ULPC)	3.0	16.13	26.21	36.29	36.29	38.31	38.31
	4.5	40.00	47.50	50.00	52.50	52.50	52.50
	6.0	11.81	19.69	31.50	35.43	39.37	39.37
	7.5	19.38	23.26	32.26	27.13	29.07	31.00
	9.0	22.06	22.06	22.06	22.06	25.74	25.74
Germinated (G)							
Lupine flour (GLF)	3.0	0.00	0.00	0.00	0.00	32.26	32.26
	4.5	5.15	15.46	30.93	30.93	51.55	51.55
	6.0	0.00	0.00	14.85	24.75	29.70	34.65
	7.5	10.42	15.63	20.83	26.04	33.85	33.85
	9.0	5.56	11.11	16.67	22.22	27.78	27.78
Protein concentrate (GLPC)	3.0	8.40	16.81	21.01	25.21	25.21	25.21
	4.5	22.56	22.56	26.32	28.20	30.08	30.08
	6.0	15.75	19.69	19.69	23.62	27.56	27.56
	7.5	9.09	18.18	22.73	27.27	27.27	27.27
	9.0	7.69	15.38	23.08	23.08	26.92	26.92

The emulsion stability (ESs) formed by different legume samples were pH-dependent. At the pH 4.5, the ESs of these three legume products were found to be at a minimum state, high emulsion stability was observed at pH 3 and pH 9. Same results were obtained by **Chau and Cheung (1998)**. The relatively high ES observed at the extreme pHs could be possibly attributed to the higher levels of solubilized proteins, which influenced ES through film encapsulation and a balance of the attractive van der Waals and repulsive electrostatic forces.

Emulsion stability of soybean flour and their protein concentrate was generally higher than those of field pea and lupine products particularly at higher pH value.

From the above-mentioned results, it could be concluded that soybean products characteristics by high emulsion capacity and high emulsion stability.

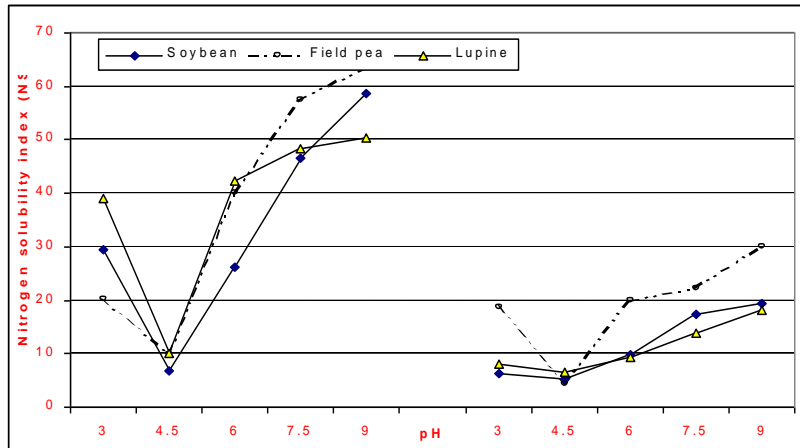
At pH 4.5 the emulsion stability of USF, UPF and ULF were 42.79, 65.86 and 51.14% respectively. At high pH value 9, the emulsion stability of the former samples reached 12.5, 16.13, 37.50%, respectively. On most cases, ungerminated legume protein concentrates had high emulsion stability than ungerminated legume flours. Germination had variable effect on ESs for all the studied legume samples.

Szuhaj and Sipos (1989) reported that protein aided formation of emulsions and helped to stabilize them during processing. Proteins form a charge layer around fat droplets causing natural repulsion, reducing interfacial tension and preventing coalescence.

4.2.3.3. Nitrogen solubility

Protein solubility is very complex and can be affected by many variables such as electrostatic interactions, hydrophobic interactions and hydrogen bonding. The levels of those three major forces contribute to protein solubility by favoring protein-protein interactions, which is indicated by lower protein solubility or by favoring protein-solvent interactions, which is indicated by higher protein solubility (**Kinsella *et al.*, 1985**).

Ungerminated



Germinated

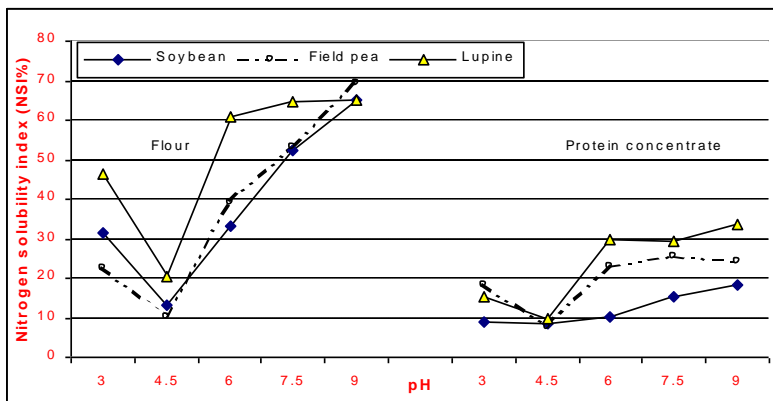


Fig. (4): Nitrogen solubility index (NSI) of legume flours and their protein concentrates.

Protein solubility profiles of soybean, field pea and lupine products are shown in Fig. (4). The minimum protein solubility for different legume products occurred at pH 4.5, near the isoelectric point, and as expected, it increased below and above this region reaching its maximum at pH 3.0 or pH 9.0. The ungerminated field pea flour was generally more soluble than the ungerminated soybean and lupine flour at pH 9.0.

However, germinated legume flours showed generally higher protein solubility at all the pH's studied compared with ungerminated legume flours. The protein of defatted legume flours were more soluble at alkaline pH than at acidic or neutral pH. These findings are comparable to those obtained by **Hsu *et al.* (1982)** and **Hassan (1998)**.

The same figure showed that nitrogen solubility index of both ungerminated and germinated legume protein concentrate was generally lower than those of legume flours at the all studied pH values.

These protein solubility curves are very similar to those of other plant protein flour and protein concentrates (**McWatters and Holmes, 1979; Dench, 1982; Narayana and Narasinga Rao, 1982; Sathe *et al.*, 1982a & b**).

4.3. Rheological and physical properties of wheat flour-legume products blends

4.3.1. Farinograph properties

Fortification of wheat flour with different levels of ungerminated or germinated legume flours or protein concentrates caused different effects on the farinograms (from 6 to 11) of the produced blends. Results in Table (13) showed that dough made from 100% wheat flour had 61.5% water absorption, supplementing wheat flour with legume flours or their protein concentrates caused an increase in water absorption of the blends except for those contained 5% of ungerminated or germinated soybean and field pea flours. However, blends contained different levels of legume protein concentrates characterized by high water absorption than those contained legume flours. Generally, blends fortified with different levels of lupine products absorbed more water than those supplemented with the same levels of soybean and field pea products.

Table (13): Farinogram parameters of wheat flour dough as affected by addition of different levels of ungerminated and germinated legume defatted flour and protein concentrates.

Sample	Replace- ment level (%)	Farinograph parameters					
		Water absorption (%)	Arrival time (min.)	Dough development (min.)	Dough stability time (min.)	Mixing tolerance index (B.U.)	Degree of softening (B.U.)
Control (100% wheat flour)	0.0	61.5	1.5	3.0	13.5	20	20
Ungerminated (UG)							
Flour							
Soy bean (USF)	5	60.0	3.0	10.0	17.0	15	20
	10	63.4	7.5	10.5	13.0	20	15
	15	68.0	12.0	14.5	9.0	20	15
Field pea (UPF)	5	61.0	2.0	6.5	20.5	15	20
	10	62.0	4.0	6.0	7.0	25	25
	15	61.6	4.5	6.0	4.5	50	60
Sweet lupine (ULF)	5	66.0	8.0	13.0	10.0	20	40
	10	67.0	6.0	7.0	3.0	30	75
	15	66.8	5.5	7.0	2.5	40	90
Protein concentrates							
Soy bean (USPC)	5	63.2	1.0	2.0	15.0	40	40
	10	69.4	2.0	9.5	19.0	20	35
	15	73.2	5.0	13.5	20.0	35	30
Field pea (UPPC)	5	63.4	1.5	2.0	11.5	40	50
	10	66.0	1.5	2.5	12.0	35	40
	15	68.6	1.5	2.0	17.0	20	10
Sweet lupine (ULPC)	5	74.0	1.0	2.0	12.0	70	20
	10	69.4	1.5	2.0	8.5	20	50
	15	75.0	2.0	6.0	7.5	65	70
Germinated (G)							
Flour							
Soy bean (GSF)	5	60.0	2.0	7.0	13.5	25	30
	10	64.0	7.0	9.5	10.0	25	35
	15	65.0	9.0	11.5	5.5	40	70
Field pea (GPF)	5	60.0	1.5	2.5	8.0	35	35
	10	63.0	4.0	7.0	6.5	40	65
	15	61.4	1.5	5.5	7.0	60	95
Sweet lupine (GLF)	5	64.2	4.0	6.5	13.0	20	15
	10	63.0	5.0	8.0	6.5	35	45
	15	65.0	6.5	8.0	5.0	40	50
Protein concentrates							
Soy bean (GSPC)	5	69.0	9.5	19.5	19.0	15	15
	10	74.0	11.0	17.0	14.5	25	30
	15	79.0	9.5	15.0	15.0	20	20
Field pea (GPCC)	5	64.2	1.0	1.5	17.0	20	25
	10	68.8	1.5	2.0	14.5	20	30
	15	74.8	1.5	11.0	21.0	15	20
Sweet lupine (GLPC)	5	69.0	1.5	12.0	27.5	15	10
	10	77.0	5.5	10.0	12.0	15	20
	15	83.0	5.5	9.0	11.5	15	30

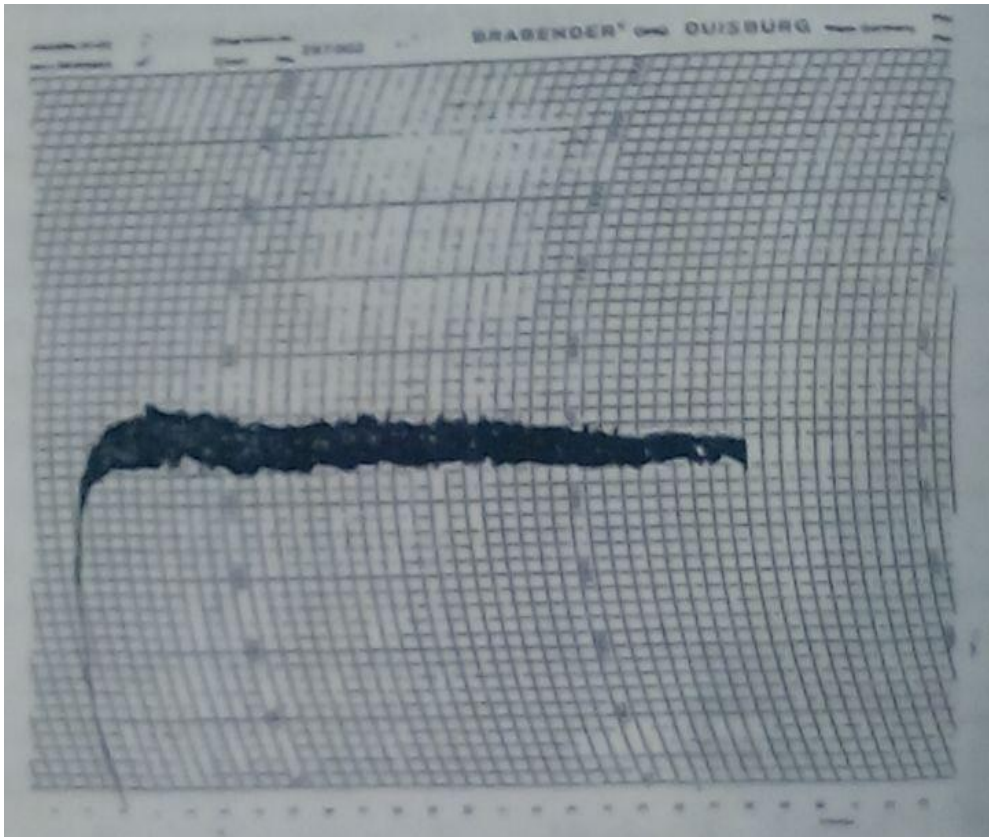


Fig. (5): Farinogram of 100% wheat flour (control).

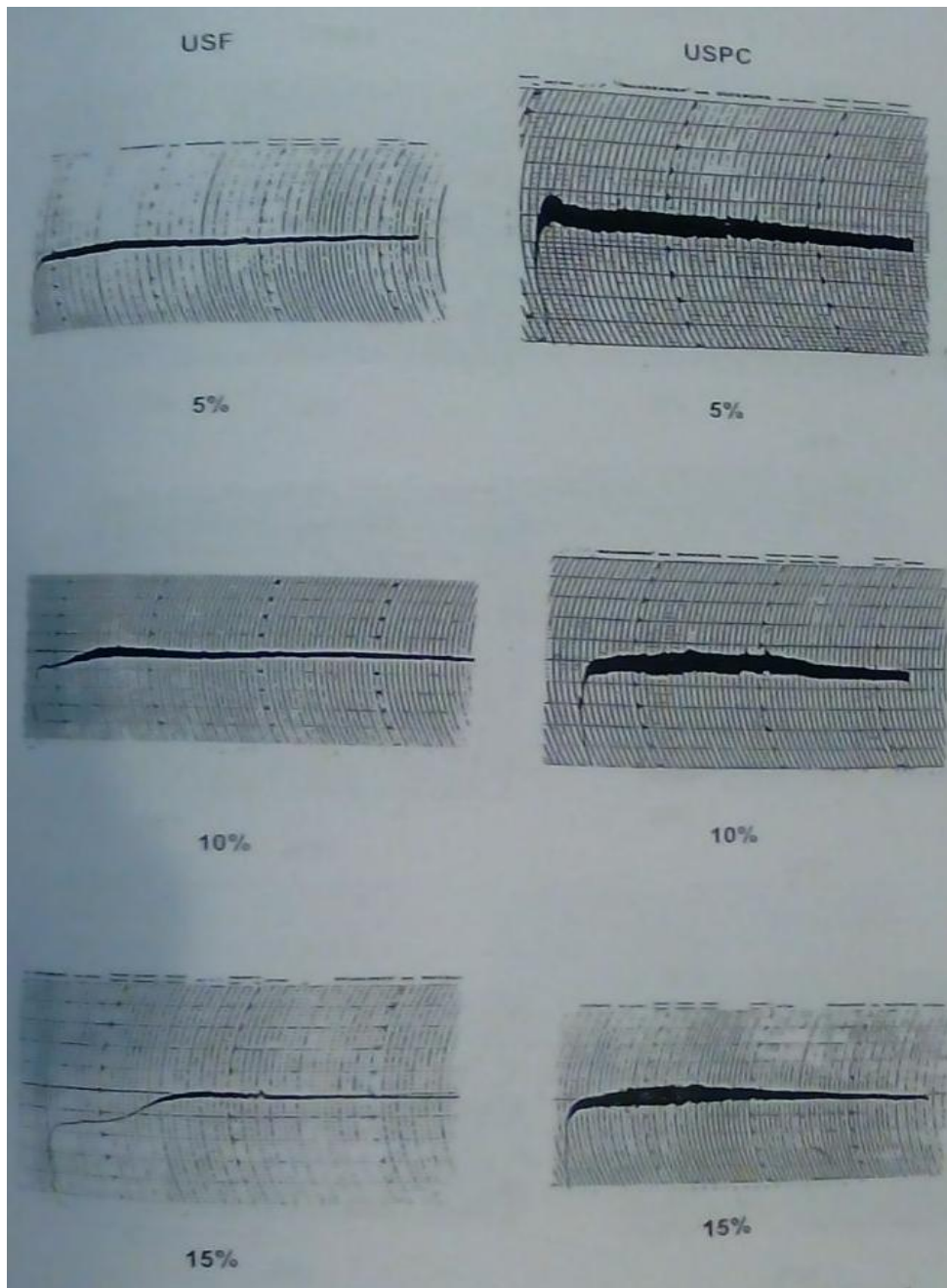


Fig. (6): Farinogram of blends containing wheat flour and different levels of ungerminated soybean flours (USF) and protein concentrate (USPC).

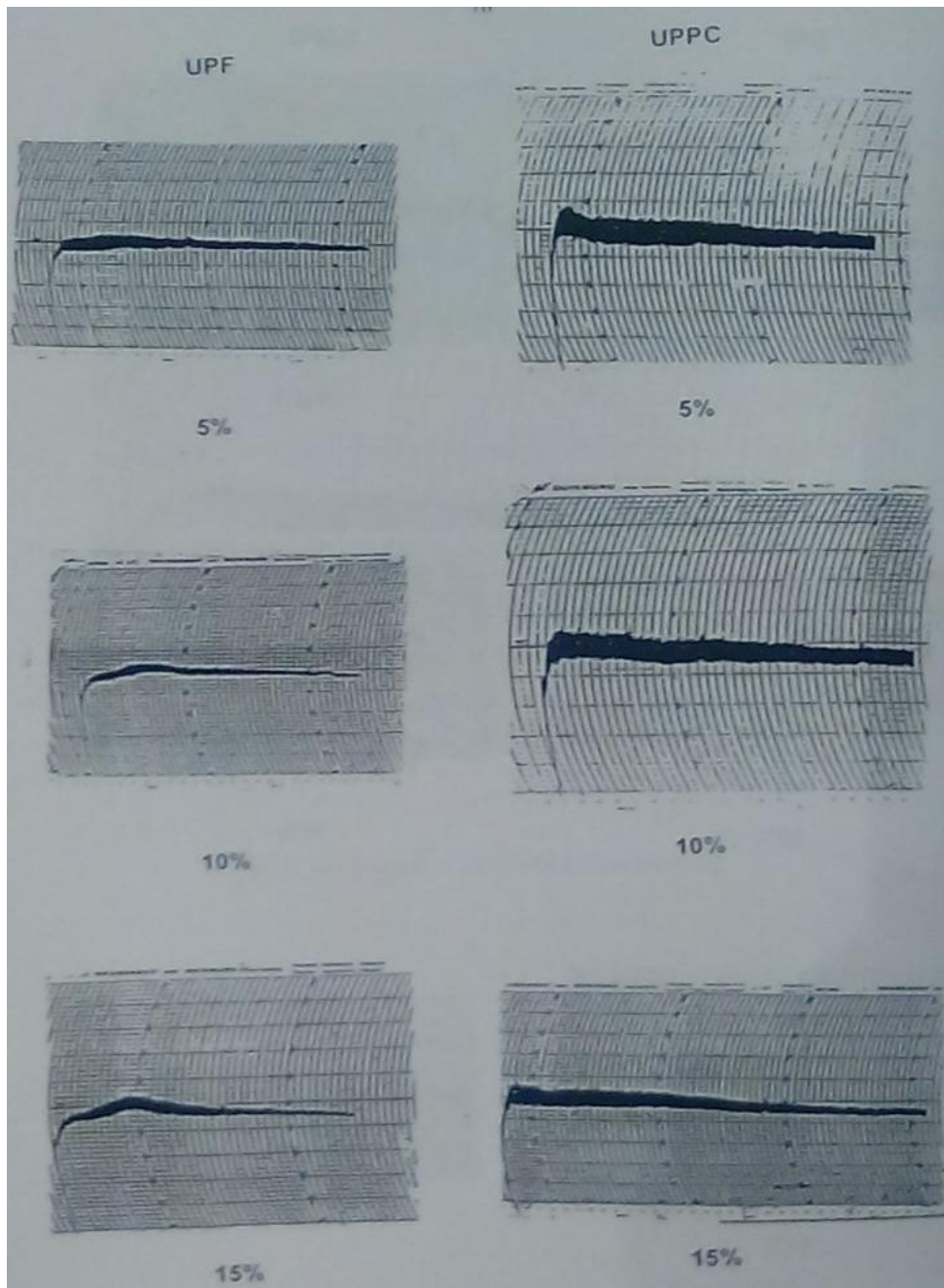


Fig. (7): Farinogram of blends containing wheat flour and different levels of ungerminated field pea flours (UPF) and protein concentrate (UPPC).

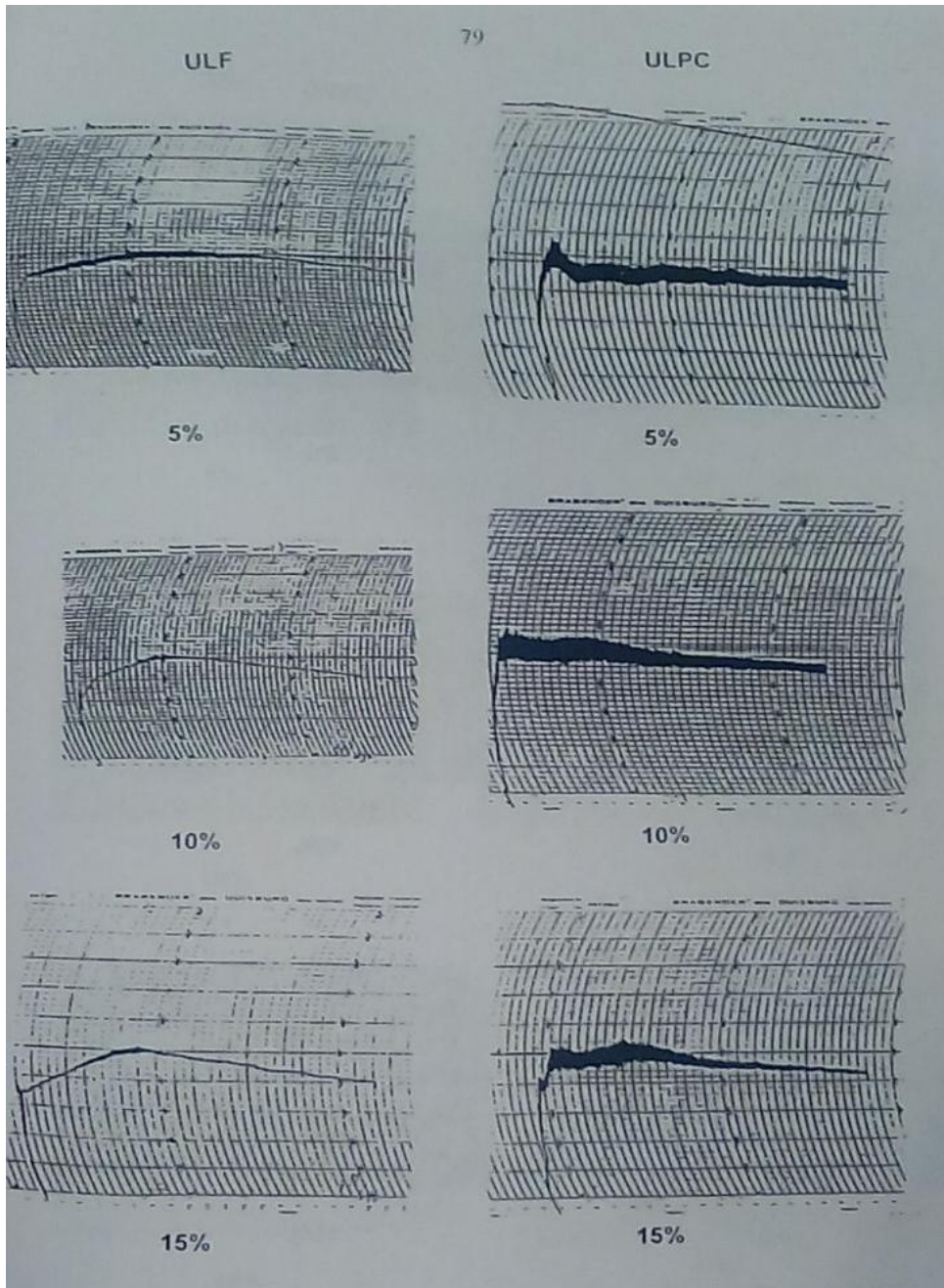


Fig. (8): Farinogram of blends containing wheat flour and different levels of ungerminated lupine flours (ULF) and protein concentrate (ULPC).

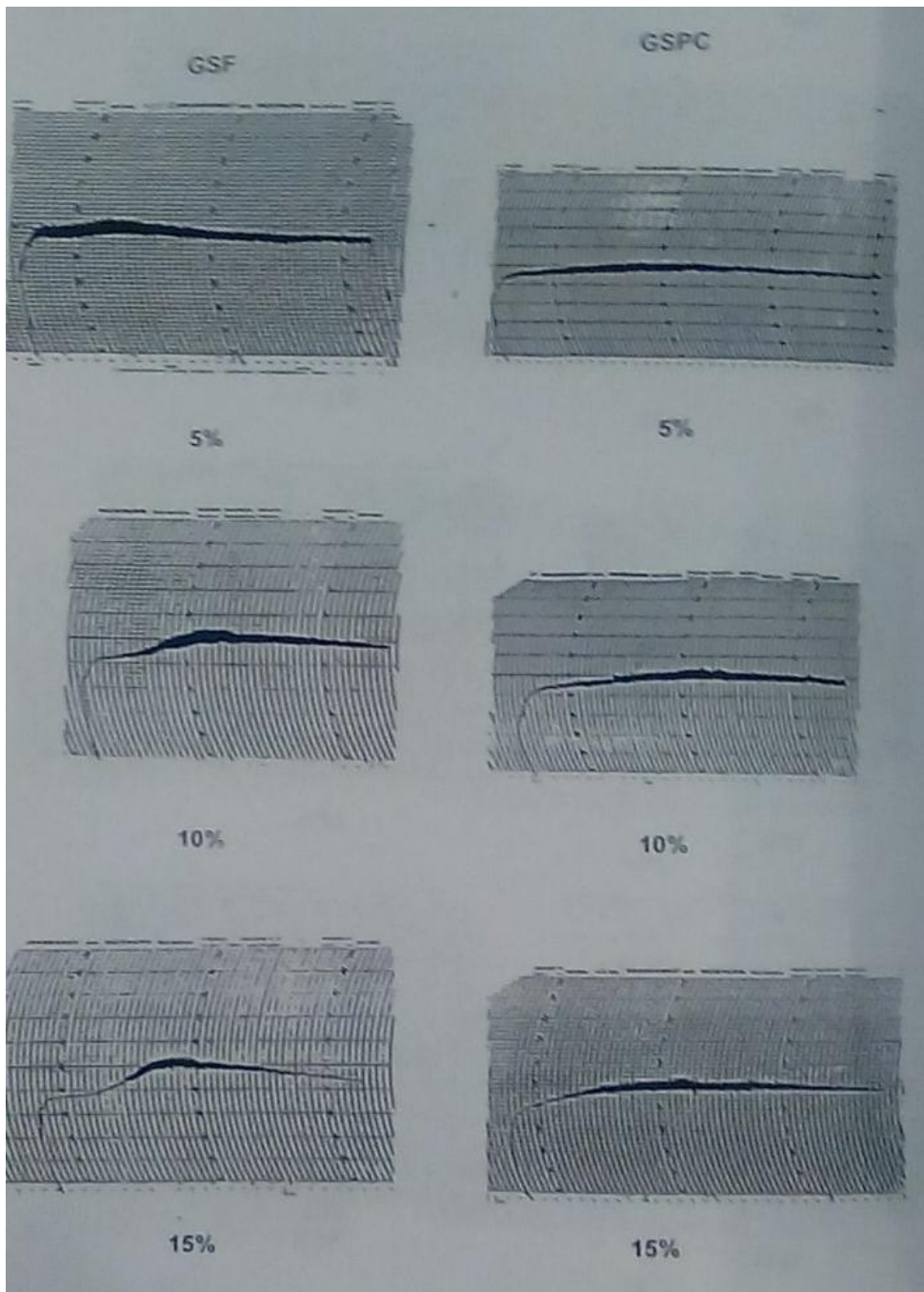


Fig. (9): Farinogram of blends containing wheat flour and different levels of germinated soybean flours (GSF) and protein concentrate (GSPC).

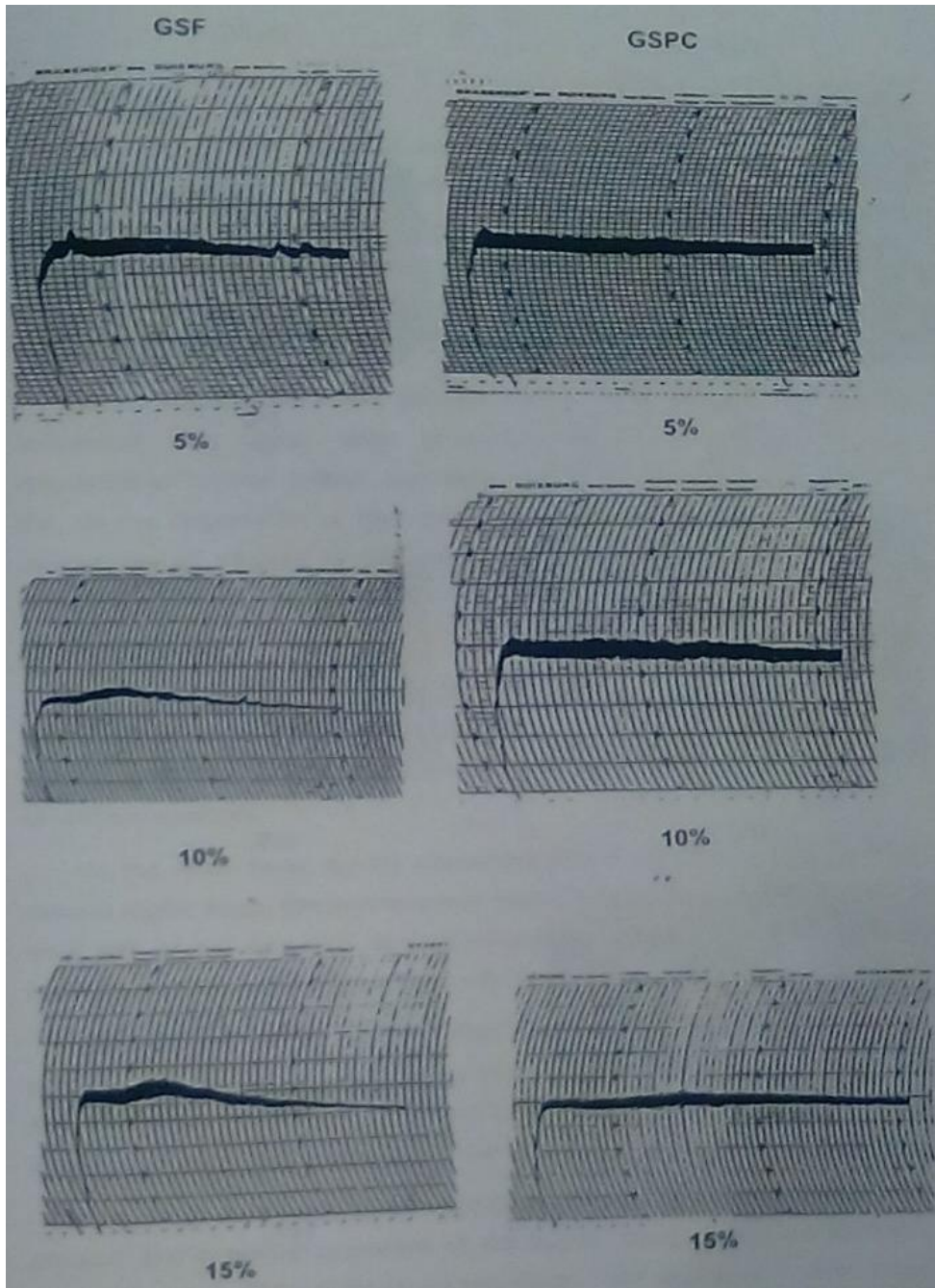


Fig. (10): Farinogram of blends containing wheat flour and different levels of germinated field pea flours (GPF) and protein concentrate (GPPC).

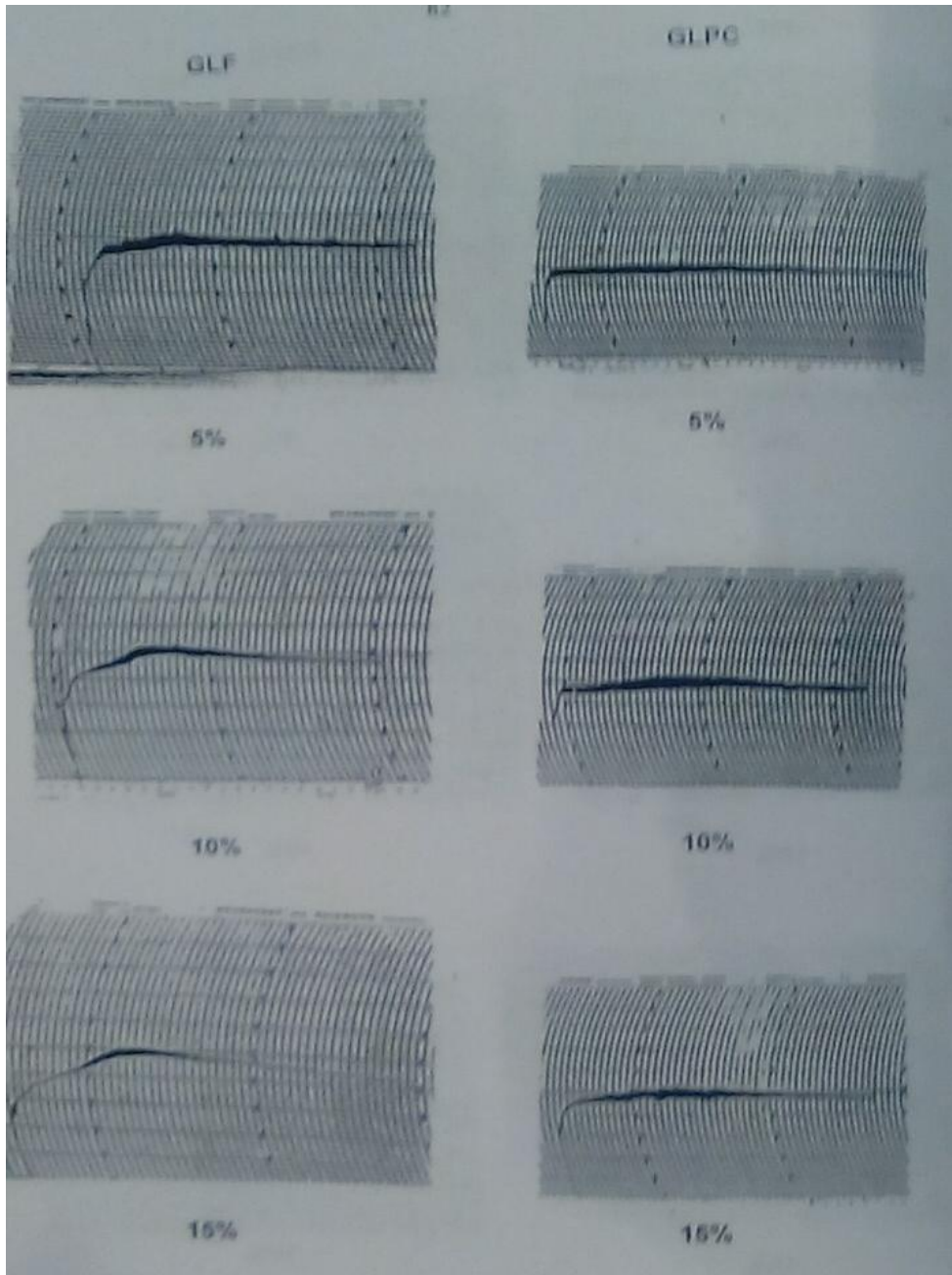


Fig. (11): Farinogram of blends containing wheat flour and different levels of germinated lupine flours (GLF) and protein concentrate (GLPC).

These results are similar to those obtained by **Campos and El-Dash (1978)**. They showed that lupine protein products had higher water holding capacity than different tested legume flours. The increase in water absorption was probably a result of the higher protein content of the blends causing greater hydration capacity. Same findings were obtained by **Soliman *et al.* (1987)**, **Ereifej and Shibil (1993)** and **Wikstrom and Eliassan (1998)**.

Blends supplemented with different levels of germinated legume protein concentrate had higher water absorption values than those contained ungerminated legume protein concentrate with some exception. This might be due to the degradation of high protein molecular weight to low molecular components as affected by proteolytic enzymes, which were activated during germination. These results are in accordance with **Hassan (1980)**, **Sathe *et al.* (1981)**, **Bahnassey and Khan (1986)** and **Mohsen *et al.* (1989)**.

With respect to the arrival time blends containing different levels of legume products showed similar or higher arrival time than control (100% wheat flour). The highest arrival time was obtained for blends supplemented with high levels of soybean products.

On the other hand, blends containing different levels of legume products showed higher dough development time (mixing time) than control (100% wheat flour) with some exception. The highest increase in mixing time was observed when soy products were incorporated with wheat flour.

Khairy *et al.* (1986) reported that dough mixing time increased as the percent of lentil and broad bean protein concentrates incorporated in wheat flour dough progressed. The increase in mixing-time may be due to differences in particle size of protein concentrate and wheat flour.

The increase in mixing time reflected the expected differences in the physical and chemical properties of the legume protein products (**Hassan, 1980; Hsu *et al.*, 1982; Bahnassey and Khan, 1986** and **Hegazy and Fahied, 1991**).

Dough stability is the most important index for dough strength. Dough stability had been attributed to protein poor in sulfhydryl groups which normally caused a softening or degradation action of the dough (**El-Farra et al., 1981**). Consequently the replacement ratio of wheat flour with defatted of ungerminated and germinated legume flours may decrease the dough stability, (Table 13). Also, raising defatted ungerminated soy flour more than 10% depressed dough stability. In general, it could be concluded that the low dough stability of the blends might be due to the higher fiber content which destroyed the gluten matrix (referred to Table 7). These results are in agreement with those obtained by **Foda et al. (1987)**. They found that increasing the replacement level of wheat flour with sorghum or millet flour may decrease the dough stability. Same results were obtained by **D'Appolonia, 1977; Morad et al., 1980; Abd El-Lateef, 1995** and **El-Shatanovi and El-Kalyoubi, 1995**.

On contrary, most of dough mixture containing various amount of germinated legume protein concentrates and ungerminated soy protein concentrate had a higher stability time upon mixing than control. The increase in dough stability could be attributed to the increase in protein level, which could render the dough more stable (**El-Farra et al., 1981**).

Same results are observed by **Khairy et al. (1986), Hegazy et al. (1991)** and **Hafez (1996)**. They showed that dough mixtures containing various amounts of protein concentrate had a higher stability upon mixing than the control.

Supplementation of wheat flour with different levels of legume products led to considerable increase in mixing tolerance index for the most fortified blends. The mixing tolerance index of dough made of wheat flours was 20 B.U. and it was generally raised to 60-65 B.U as a result of adding 15% of GPF and ULPC, respectively.

Matsuo et al. (1972) reported that farinograph characteristics were markedly affected by the increase of protein content, since this increase led to elevating the mixing tolerance index.

Addition of defatted legume flour and their protein concentrates generally increased the softening degree of wheat flour blend with some exception as could be seen in Table (13). The degree of softening of control dough was 20 B.U.

raised to 95 B.U. for 15% GPE. These results are in accordance to those obtained by **Morad *et al.* (1980)** and **Makhlouf (1984)**.

4.3.2. Wet and dry gluten

The technological properties of wheat flour are depended on wheat genotype and growing conditions and are mainly determined by data structure and quantity of gluten (**Wieser *et al.*, 1998**). The data in Table (14) explained the effect of fortification of wheat flour with different levels of legumes products (flour and protein concentrates) on wet and dry gluten contents of the blends.

Wheat flour is generally characterized by a high protein and gluten content. Data in Table (14) showed that wheat flour had a relatively higher wet and dry gluten being (27.58 and 10.41%, respectively). These results are in agreement with those reported by **Boyacioglu and D'Appolonia (1994)** and **Hassan (1998)**. Results also showed that the addition of different legume products to wheat flour significantly reduced gluten values of the blends.

The reduction in wet and dry gluten content was increased by increasing the replacement levels of legume products. Wet and dry gluten values were reduced from 25.87, and 9.52% for blends contained 5% USF to 18.96, and 7.21% when substitution level increased to 15%. Fortification of wheat flour with different levels of lupine flours significantly reduced wet and dry gluten values of the blends than those of soy and field pea flours.

Blends containing different levels of germinated legume products had relatively higher wet and dry gluten values than those containing the corresponding levels of ungerminated legume products. Also, most of legume flours-blends had lower values of wet and dry gluten than legume protein concentrates blends.

The supplementation of wheat flour with different amount of legume products influenced significantly the hydration ratio of the dough. However, hydration ratio was decreased by increasing the replacement levels of legume products with some exception.

Same findings were obtained by **Patel and Venkateswara Raot (1995)**, they found that gluten contents were reduced substantially on substitution of more than 5% of the wheat flour with either untreated or germinated black gram flour, which may be attributable to increased proteolytic activity.

Table (14): Wet and dry gluten* of wheat flour-legume products blends as affected by germination process.

Sample	Replacement level	Gluten %		Hydration ratio	Gluten %		Hydration ratio
		Wet	Dry		Wet	Dry	
Control	0	27.58 ^A	10.41 ^A	165.17 ^{PQ}			
Flour		Ungerminated			Germinated		
Soy bean	5	25.87 ^{DE}	9.52 ^C	171.74 ^N	26.11 ^{CD}	8.77 ^{KLM}	197.55 ^A
	10	24.64 ^{H-L}	9.19 ^{FGH}	168.12 ^O	25.36 ^{EFG}	8.83 ^{J-M}	187.20 ^E
	15	18.96 ^S	7.21 ^S	162.89 ^R	20.68 ^R	7.67 ^R	169.79 ^O
Field pea	5	25.36 ^{EFG}	8.92 ^{J-M}	184.30 ^F	26.00 ^D	9.96 ^B	161.04 ^S
	10	24.96 ^{G-J}	8.98 ^{H-K}	177.95 ^{HJ}	24.99 ^{GHI}	9.17 ^{F-I}	172.52 ^N
	15	21.22 ^Q	7.79 ^R	172.13 ^N	22.99 ^{NO}	8.36 ^{op}	175.00 ^{KLM}
Sweet lupine	5	24.29 ^{KL}	8.70 ^{MN}	179.19 ^H	24.35 ^{KL}	8.95 ^{I-L}	172.07 ^N
	10	22.41 ^P	8.19 ^{PQ}	173.63 ^{MN}	22.64 ^{OP}	8.52 ^{NO}	165.73 ^P
	15	15.28 ^U	5.75 ^T	165.90 ^P	17.88 ^T	7.03 ^S	154.08 ^T
Protein concentrates							
Soy bean	5	26.53 ^{BC}	8.95 ^{I-L}	196.29 ^{AB}	26.64 ^B	9.94 ^B	168.04 ^O
	10	25.55 ^{DEF}	8.73 ^{LMN}	192.67 ^C	25.72 ^{DE}	9.43 ^{CDE}	172.64 ^N
	15	24.20 ^L	8.37 ^{OP}	189.29 ^D	24.83 ^{G-K}	9.04 ^{G-J}	174.60 ^{LM}
Field pea	5	25.32 ^{EFG}	9.31 ^{C-F}	171.93 ^N	26.69 ^B	10.22 ^A	161.12 ^S
	10	24.51 ^{I-L}	8.46 ^O	189.72 ^D	24.95 ^{G-J}	9.48 ^{CD}	163.14 ^{QR}
	15	23.63 ^M	8.01 ^Q	195.01 ^B	24.41 ^{JKL}	9.28 ^{DEF}	163.04 ^{QR}
Sweet lupine	5	25.98 ^D	9.24 ^{EFG}	181.09 ^G	26.95 ^B	9.34 ^{C-F}	188.54 ^{DE}
	10	25.16 ^{FGH}	9.03 ^{G-J}	178.58 ^{HI}	25.95 ^D	9.16 ^{F-I}	183.29 ^F
	15	23.19 ^{MN}	8.38 ^{OP}	176.80 ^{IJK}	23.58 ^M	8.54 ^{NO}	176.11 ^{JKL}

* Means in columns followed by the same letter(s) are not significantly different at $P \leq 0.05$.

4.4. Characteristics of some bakery products (pan bread-biscuit) fortified with some legume protein concentrates:

4.4.1. Chemical composition:

The chemical composition of pan bread fortified with different levels (5, 10 and 15%) of defatted flours and protein concentrates of ungerminated and germinated legume seeds were compared statistically and the results are given in Table (15). Unfortified pan bread (wheat bread, control) contained 28.99% moisture, 12.44% protein, 2.78 fat and 1.76% ash. These results are in agreement with those obtained by **Lucisano and Pompei (1981)** and **Foda *et al.* (1987)**.

Moisture content of legume fortified bread were ranged between 31.44 to 38.79%. It can be observed that high protein bread characterized by high water absorption (Table 13) and consequently high moisture content. Similar results have been previously reported by **Lucisano and Pompei (1981)**, who found that presence of lupine flour increased the water required for the optimum bread making absorption. The protein content of the bread continues to increase as the proportion of legume flours and concentrate are increased in the blends. As expected, bread fortified with legume protein concentrates exhibited higher values of protein content than those fortified with legume flours. For example, replacement of wheat flour with 5% USF caused an increase in bread protein content by 6.35% while fortification with 10 and 15% caused 24.04 and 37.70% increases respectively (Fig. 12). On the other hand, fortification of wheat flour with 5, 10 and 15% of USPC improved protein contents by 21.78, 32.39 and 51.53% respectively. Also, soybean and lupine products significantly improved protein content of the fortified bread than field pea products.

Generally, bread fortified with germinated legume products showed higher protein content than those fortified with ungerminated legume products.

Results also showed that bread fortified with 15% of GSPC exhibited the maximal improve in protein content (58.52%).

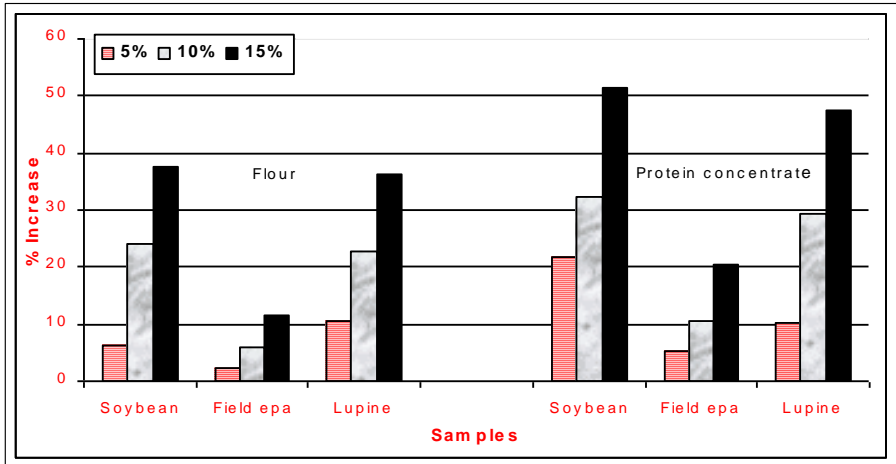
Lucisano and Pompei (1981) found that the protein content of the 20% substituted bread with defatted lupine flour increased by more than 80% compared to the unsupplemented bread. Similar results were obtained by **Foda *et al.* (1987)** and **Hafez (1996)**.

Table (15): Chemical composition of pan bread made from legume products-wheat flour blends.

Products	Replace- ment level %	Moisture %	Chemical composition calculated on dry weight basis			NFE*
			Protein %	Fat %	Ash %	
Control (100% wheat flour)		28.99	12.44	2.78	1.76	83.03
Ungerminated (UG)						
Flour						
Soy bean (USF)	5	32.62	13.23l	2.80	2.04	81.93
	10	32.77	15.43	3.76	2.20	78.61
	15	35.14	17.13	3.96	2.47	76.45
Field pea (UPF)	5	31.44	12.74	3.25	1.85	82.25
	10	32.01	13.19	3.95	2.03	80.83
	15	32.71	13.89	4.26	2.23	79.62
Sweet lupine (ULF)	5	31.56	13.75	2.95	1.78	81.52
	10	31.83	15.26l	2.96	2.02	79.76
	15	32.86l	16.97	3.34	2.15	77.54
Protein concentrate						
Soy bean (USPC)	5	33.60	15.15	2.83	2.07	79.94
	10	33.94	16.47	3.16	2.25	78.12
	15	35.69	18.85	3.49	2.56	75.10
Field pea (UPPC)	5	33.88	13.09	3.57	1.80	81.54
	10	34.01	13.76l	3.87	1.84	80.52
	15	34.25	15.00	4.09	1.97	79.04
Sweet lupine (ULPC)	5	33.82	13.73l	2.23	1.97	82.07
	10	34.23	16.10	2.61l	2.09	79.19
	15	35.49	18.24	4.04	2.18	75.54
Germinated (G)						
Flour						
Soy bean (GSF)	5	32.28	14.63	2.02	2.13	81.22
	10	32.97	17.68	2.09	2.60	77.63
	15	34.99	18.10	2.40	2.87	76.62
Field pea (GPF)	5	31.59	13.37	2.97	2.01	81.31
	10	32.01	13.91	3.95	2.03	80.83
	15	33.69	14.33	4.27	2.22	79.18
Sweet lupine (GLF)	5	31.95	14.72	2.07	1.93	80.95
	10	31.85	15.63	3.52	1.98	78.87
	15	33.98	17.21	3.69	2.11	76.99
Protein concentrate						
Soy bean (GSPC)	5	34.89	15.72	2.19	2.05	80.04
	10	35.64	17.32	2.26	2.19	78.23
	15	38.79	19.72	4.17	2.50	73.60
Field pea (GPCC)	5	33.96	13.77	2.69	1.74	81.80
	10	34.16	14.31	3.23	1.98	80.49
	15	35.57	15.39	3.97	2.08	78.56
Sweet lupine (GLPC)	5	31.74	14.12	1.38	2.03	82.47
	10	32.72	16.94	1.78	2.16	79.13
	15	34.09	18.93	2.33	2.29	76.45
LSD at 0.05		0.914	2.495	0.668	0.027	0.478

* Nitrogen free extract.

Ungerminated



Germinated

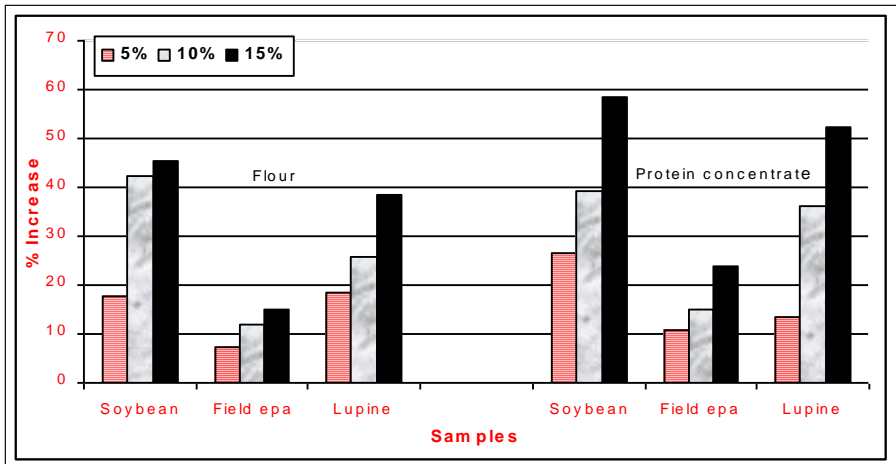


Fig. (12): Percentage increase in protein content of pan bread containing different levels of flours and protein concentrates of legumes (compared to 100% wheat flour bread).

Legume fortified bread contained high levels of fat and ash than the wheat bread (control). Same results were obtained by **Foda et al. (1984c and 1987)**.

From the above mentioned results it can be concluded that fortification of wheat flour with high levels of legume protein concentrates significantly improved the nutritional value of the fortified bread, (**Foda et al., 1987** and **Hafez, 1996**).

The chemical composition of biscuits fortified with different levels of legume products are presented in Table (16).

Moisture content of biscuits supplemented with legume products increased relatively by increasing the supplementation level. This might be due to the water retention capacity of legume products as reported by **McWatters (1978)**.

The protein content of the fortified biscuit increased by increasing the level of replacement of wheat flour with legume products. This is mainly due to the higher protein content of the legume products than of wheat flour. Addition of 15% soy products raised significantly protein content of the biscuit from 8.03% for control to 12.56, 12.85, 13.65 and 14.47% for those fortified with USF, USPC, GSF and GSPC respectively. The increase percentages for protein content reached 56.41, 60.02, 69.99 and 80.19% for the corresponding samples (Fig. 13). Biscuit fortified with germinated legume products showed higher protein content than those fortified with ungerminated products. Field pea-biscuit had lower protein contents than soy and legume-biscuits.

Fat and ash contents of the legume-biscuit exceeded significantly the levels shown for the control. As the percent of replacement increased, the moisture, protein, ash and fat content of legume containing biscuit samples also increased.

The obtained results showed that legume-fortified biscuit contained higher levels of nutrients than the control biscuit (100% wheat flour).

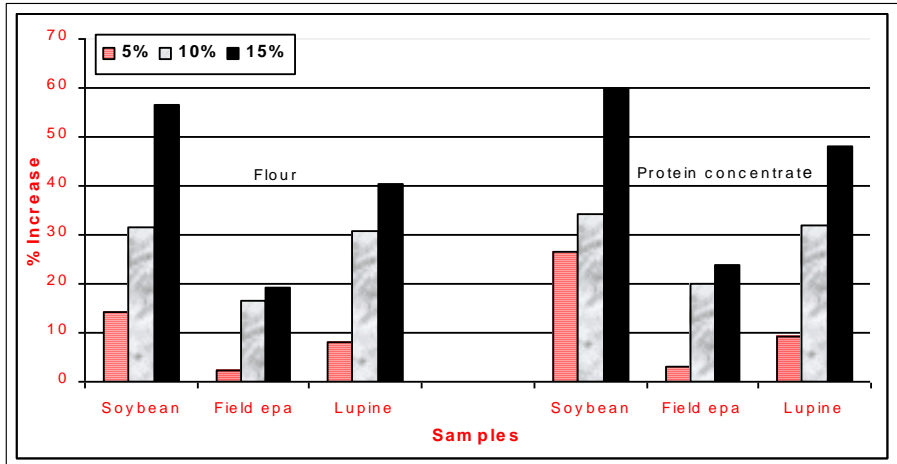
These results are in agreement with those reported by **Mcwatters (1978)**, **Kerolles and Rasmy (1990)**, **Faheid and Hegazy (1991)** and **El-Bahey and El-Sanafiry (1994)**.

Table (16): Chemical composition of biscuit made from legume products-wheat flour blends.

Products	Replace- ment level %	Moisture %	Chemical composition calculated on dry weight basis			NFE*
			Protein %	Fat %	Ash %	
Control (100% wheat flour)	0	3.89	8.03	9.45	0.56	82.05
Ungerminated (UG)						
Flour						
Soy bean (USF)	5	4.80	9.17	9.81	0.83	80.19
	10	4.90	10.55	10.06	1.06	78.34
	15	5.28	12.56	10.73	1.29	75.13
Field pea (UPF)	5	4.45	8.05	10.58	0.64	80.89
	10	4.59	9.36	11.60	0.84	78.19
	15	4.76	9.57	12.24	1.00	77.19
Sweet lupine (ULF)	5	4.34	8.68	10.99	0.81	79.51
	10	4.78	10.51	11.00	0.96	77.53
	15	5.13	11.28	12.12	1.13	75.47
Protein concentrate						
Soy bean (USPC)	5	4.65	10.16	9.98	0.94	78.91
	10	5.05	10.77	10.38	1.11	77.74
	15	5.73	12.85	11.08	1.44	74.93
Field pea (UPPC)	5	4.71	8.28	13.46	0.79	77.47
	10	5.41	9.63	13.79	0.91	75.66
	15	5.72	9.93	14.21	1.09	74.76
Sweet lupine (ULPC)	5	4.42	8.78	11.18	1.01	79.04
	10	5.17	10.59	12.02	1.36	76.03
	15	5.31	11.88	13.26	1.41	73.46
Germinated (G)						
Flour						
Soy bean (GSF)	5	5.33	10.680	11.16	0.87	77.29
	10	5.41	11.08	11.64	1.36	75.92
	15	6.37	13.65	12.58	1.74	72.03
Field pea (GPF)	5	4.62	8.66	11.72	0.82	78.80
	10	5.12	9.77	11.41	0.86	77.96
	15	5.41	10.42	12.52	0.92	76.14
Sweet lupine (GLF)	5	4.77	9.28	11.08	0.79	78.85
	10	5.23	10.61	11.12	0.84	77.42
	15	5.48	12.78	11.64	0.91	74.67
Protein concentrate						
Soy bean (GSPC)	5	5.54	10.86	11.23	1.27	76.64
	10	5.74	12.43	12.42	1.41	73.74
	15	5.79	14.47	13.71	1.46	70.36
Field pea (GPPC)	5	6.08	8.96	12.05	0.84	77.97
	10	6.36	10.17	12.82	1.36	75.65
	15	6.54	10.74	13.34	1.83	74.09
Sweet lupine (GLPC)	5	5.80	9.65	11.26	0.82	78.26
	10	6.51	11.93	11.77	1.13	75.17
	15	7.23	13.93	12.70	1.02	72.33
LSD at 0.05		0.387	2.298	0.602	0.229	0.913

* Nitrogen free extract.

Ungerminated



Germinated

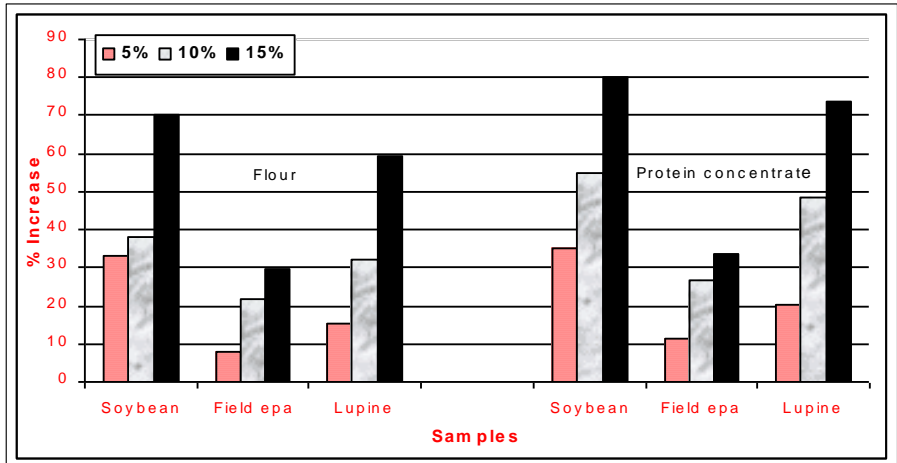


Fig. (13): Percentage increase in protein content of biscuits containing different levels of flours and protein concentrates of legumes (compared to 100% wheat flour bread).

4.4.2. Baking quality

The effect of supplementation of wheat flour with different levels of legume products on loaf weight, loaf volume, and specific volume of pan breads are given in Table (17). The enrichment of wheat flour with different levels of legume products increased significantly loaf weight than control. This is mainly due to the higher water absorption of legume products as mentioned by **Hafez (1996)**. Generally, legume flours-bread showed higher loaf weight than legume- protein concentrates especially at high levels. On the other hand, most of legume-fortified-bread showed higher loaf volume than control except for those contained high levels of protein concentrates. The highest loaf volume was noticed for bread fortified with 5% of either ULF (305 cm³), GLF (290 cm³). The lowest loaf volume was recorded at 15% of GPPC (180.5 cm³) and GLPC (189.3 cm³). Specific loaf volume (cm³/g) showed the same trend of either loaf weight or loaf volume. The best specific volume was noticed at 5% of either ULF or GLF. The specific volumes of the breads decreased as the level of legume product increased, nevertheless, also 15% supplemented bread had a specific volume close to that of control pan bread.

Same findings were reported by **Hafez (1996)** who found that adding 15% or 10% defatted soy flour to wheat flour improved loaves quality. On contrary **Lucisano and Pompei (1981)** showed that the specific volumes of the breads decreased as the level of lupine flour increased; nevertheless, also the 20% supplemented bread had a volume close to that of Italian commercial breads.

Also, **McWatters (1978)** used field pea flour to replace 10, 20 and 30% of the wheat flour in sugar cookies. They found that sensory quality attributes were not affected adversely by use of this flour except at the 30% replacement level.

4.5. Organoleptic properties

Sensory evaluation data of the pan bread fortified with different levels of legume products were statistically analyzed and the means are given in Tables 18 to 21

Results in Table (18) showed that most organoleptic attributes of pan bread were not affected adversely by addition of different levels of defatted soy flour or protein concentrate.

Table (17): Baking quality* of pan bread made from wheat flour and legume products blends.

Product	Replacement level	Loaf weight (g)	Loaf volume (cm ³)	Specific volume (cm ³ /g)	Loaf weight g	Loaf volume (cm ³)	Specific volume (cm ³ /g)
Control	0	90.07 ^M	222.5 ^L	2.473 ^P			
		Ungerminated (UG)			Germinated (G)		
Flour							
Soy bean (SF)	5	97.43 ^{A-G}	245.0 ^I	2.52 ^O	96.47 ^{B-I}	281.7 ^{DE}	2.92 ^E
	10	97.07 ^{A-H}	240.0 ^{IJ}	2.47 ^P	91.06 ^{KLM}	285.3 ^{CD}	3.13 ^B
	15	100.00 ^A	245.7 ^I	2.46 ^P	93.29 ^{JK}	246.2 ^I	2.64 ^M
Field pea (PF)	5	97.08 ^{A-H}	270.0 ^G	2.78 ^J	99.78 ^A	280.0 ^{DEF}	2.80 ^I
	10	97.75 ^{A-D}	275.7 ^{EFG}	2.82 ^{HI}	96.51 ^{B-I}	274.3 ^{FG}	2.85 ^G
	15	95.49 ^{B-J}	240.2 ^J	2.51 ^O	99.83 ^A	255.0 ^H	2.56 ^N
Sweet lupine (LF)	5	95.23 ^{B-J}	305.0 ^A	3.20 ^A	95.00 ^{C-J}	290.0 ^{BC}	3.05 ^D
	10	95.60 ^{B-J}	295.0 ^B	3.08 ^C	98.19 ^{AB}	284.3 ^{CD}	2.89 ^F
	15	95.85 ^{B-J}	280.0 ^{DEF}	2.92 ^E	94.65 ^{E-J}	276.0 ^{EFG}	2.92 ^E
Protein concentrates							
Soy bean (SPC)	5	97.87 ^{ABC}	280.0 ^{DEF}	2.86 ^G	95.69 ^{B-J}	260.0 ^H	2.72 ^K
	10	94.38 ^{HIJ}	220.7 ^L	2.34 ^R	94.86 ^{C-J}	255.0 ^H	2.69 ^L
	15	93.56 ^{IJK}	238.8 ^J	2.56 ^N	94.52 ^{G-J}	211.7 ^M	2.24 ^T
Field pea (PPC)	5	97.66 ^{A-E}	261.0 ^H	2.67 ^L	97.63 ^{A-F}	225.7 ^{KL}	2.31 ^S
	10	94.72 ^{D-J}	205.0 ^N	2.17 ^U	92.93 ^{JKL}	201.0 ^N	2.16 ^U
	15	94.61 ^{F-J}	191.0 ^O	2.01 ^W	90.50 ^{LM}	180.5 ^P	1.99 ^W
Sweet lupine (LPC)	5	97.33 ^{A-H}	275.3 ^{EFG}	2.83 ^H	93.39 ^{JK}	257.5 ^H	2.76 ^J
	10	94.47 ^{G-J}	270.0 ^G	2.86 ^G	89.34 ^M	226.3 ^{KL}	2.53 ^O
	15	94.83 ^{D-J}	230.0 ^K	2.42 ^Q	90.53 ^{LM}	189.3 ^O	2.09 ^V

* Means in columns followed by the same letter(s) are not significantly different at $P \leq 0.05$.

Table (18): Organoleptic properties of pan bread made from soybean products - wheat flour blends.

Product	Suppl. level (100)	Appearance %	Crumb texture (20)	Crumb grain (20)	Crust color (20)	Taste (10)	Odor (20)	Overall acceptability (10)
Control (100% wheat flour)	0	16.18	16.18	16.09	7.27	15.09	6.82	77.64
Ungerminated (UG)								
Soybean flour (USF)	5	17.00	16.27	15.73	7.18	15.82	6.27	78.27
	10	17.55	16.55	16.45	7.46	16.18	6.46	80.64
	15	16.55	15.73	15.64	7.09	16.27	6.27	77.55
Protein concentrate (USPC)	5	15.36	15.82	16.27	6.64	14.82	6.55	75.45
	10	15.55	16.09	15.91	7.00	14.91	6.64	76.09
	15	15.09	16.27	15.55	6.64	15.18	6.46	75.18
Germinated								
Soybean flour (GSF)	5	16.91	18.18	17.09	8.00	16.82	7.64	84.45
	10	17.82	17.45	16.91	8.18	16.64	7.27	84.27
	15	16.91	17.27	17.18	8.09	15.82	7.00	82.27
Protein concentrate (GSPC)	5	15.91	16.73	16.64	7.27	16.27	7.00	79.82
	10	15.91	17.00	16.45	7.36	15.27	6.91	78.91
	15	15.55	16.36	16.45	7.09	15.27	6.91	77.64
L.S.D. at 0.05		1.130	1.164	1.066	0.791	1.449	0.874	6.066

No significant differences were obtained between the sensory attributes of pan bread fortified with either USF or USPC, except that USF bread had significantly higher appearance score than USPC-bread at different substitution levels. On the other hand, bread fortified with germinated soy flour has superior appearance, crumb texture, crumb grain and crust color than those contained germinated soy protein concentrate. The overall acceptability of bread fortified with different levels of soy flour was generally higher than that fortified with soy protein concentrate. At the same time, bread enriched with germinated soy products showed superior organoleptic properties from those contained ungerminated soy products.

Foda et al. (1987) found that the crust and crumb of the bread fortified with defatted soy flour up to 10% were golden in colour. At high levels of supplementation, i.e., more than 10% defatted soy flour, the manufacture of bread samples were found to score lower grades. So the level of the added protein is often limited by residual flavour characteristic of raw soybeans.

Hafez (1996) mentioned that supplementation with defatted soy flour at 5, 10 and 15% caused an excellent color of the bread. This may be due to the protein compounds and free amino acids, which combine the free sugars to produce the bread color.

No significant differences were obtained between organoleptic properties of control bread (100% wheat flour) and those fortified with different levels of field pea products, Table (19). Generally, panel members gave unfortified bread higher organoleptic scores than pea-fortified bread except of crumb grain and odor. However, the overall acceptability of bread contained different levels of field pea products was less than that of control. Also, most of the organoleptic properties were decreased with increasing the supplementation level.

Table (20) presented the results of the sensory evaluation of the pan bread containing different amount of lupine products. Lupine-bread was ranked similarly to whole wheat bread in appearance, crumb texture, crumb grain, taste and overall acceptability attributes. In crust color 5% ULF, 10% ULPC and 10% GLF were ranked as significantly better than the wheat bread. Generally, bread

Table (19): Organoleptic properties of pan bread made from field pea products - wheat flour blends.

Product	Suppl. level (100)	Appearance %	Crumb texture (20)	Crumb grain (20)	Crust color (20)	Taste (10)	Odor (20)	Overall acceptability (10)
Control (100% wheat flour)	0	16.18	16.18	16.09	7.27	15.09	6.82	77.64
Ungerminated (UG)								
Field pea flour (UPF)	5	15.91	15.27	16.64	7.27	14.36	7.82	76.36
	10	15.09	15.73	16.64	7.18	14.82	7.46	76.73
	15	14.55	15.27	16.00	7.00	15.00	7.46	74.45
Protein concentrate (UPPC)	5	15.36	15.27	16.45	7.00	14.36	7.36	74.27
	10	15.00	14.82	16.45	6.64	14.45	6.55	72.00
	15	15.09	14.55	15.64	6.18	14.18	6.36	69.91
Germinated (G)								
Field pea flour (GPF)	5	15.36	15.91	16.45	7.27	15.09	7.09	77.09
	10	15.55	15.73	16.45	7.09	15.09	7.09	76.45
	15	15.09	15.27	15.82	6.73	15.64	6.55	74.36
Protein concentrate (GPPC)	5	16.36	15.91	16.73	7.09	14.82	7.09	77.09
	10	15.45	14.73	16.18	6.36	14.91	7.00	73.27
	15	14.55	14.27	15.64	6.36	14.82	6.73	70.91
L.S.D. at 0.05		1.755	1.590	1.692	0.988	1.992	0.866	8.360

Table (20): Organoleptic properties of pan bread made from lupine products - wheat flour blends.

Product	Suppl. level (100)	Appearance %	Crumb texture (20)	Crumb grain (20)	Crust color (20)	Taste (10)	Odor (20)	Overall acceptability (10)
Control (100% wheat flour)	0	16.18	16.18	16.09	7.27	15.09	6.82	77.64
Ungerminated (UG)								
Lupine flour (ULF)	5	16.36	17.09	16.64	8.27	16.55	8.18	83.09
	10	16.73	16.27	16.64	7.73	16.55	7.91	81.82
	15	15.91	14.91	16.00	6.36	16.27	7.27	76.73
Protein concentrate (ULPC)	5	16.73	16.82	16.45	7.91	16.45	7.55	81.91
	10	17.18	17.27	16.45	8.18	16.55	7.73	83.36
	15	16.45	16.18	15.64	7.36	15.36	7.36	78.36
Germinated (G)								
Lupine flour (GLF)	5	17.09	17.09	16.46	8.00	16.46	7.82	82.91
	10	17.00	17.36	16.46	8.18	16.18	7.73	82.91
	15	15.18	16.00	15.82	7.18	15.91	7.27	77.36
Protein concentrate (GLPC)	5	16.91	17.00	16.73	7.82	16.64	7.73	82.82
	10	16.27	16.91	16.18	7.55	16.27	7.55	80.73
	15	15.55	15.36	15.64	7.36	15.55	7.09	76.55
L.S.D. at 0.05		1.211	1.218	1.425	0.756	1.465	0.919	6.574

containing 5 and 10% of lupine products had higher overall acceptability than control bread.

Lucisano and Pompi (1980) evaluated the colour of the bread crusts containing different amount of lupine flour. Lupine supplementation did not introduce colours extraneous to baking products moreover the new colour resembled the colour of a product containing egg yolk.

Duncan's multiple range test was conducted to compare the sensory attributes of pan bread fortified with different levels of ungerminated and germinated legume products (Table 21).

Sweet lupine bread received significantly higher crust color and odor rating than soybean bread. Lupine bread was ranked similarly to soy bread in appearance, crumb texture, crumb grain, taste and overall acceptability. In all organoleptic properties soy and lupine breads had superior scores than field pea-bread. Our results are similar to those obtained by **Sosulski and Fleming (1978)**.

Ungerminated legume-bread was ranked similarly to germinated legume-bread in sensory attributes except in crumb texture which germinated recorded superior values than ungerminated.

Legume flour-breads were ranked as significantly better than the legume protein concentrate-breads in appearance, crumb grain, crust color, taste and overall acceptability, while legume flour breads was ranked similarly to legume protein concentrate breads in both crumb texture and odor.

On the other hand, bread containing 10% of different legume products showed generally higher organoleptic attributes than control (100% wheat flour). However, increasing substitution level to 15%, overall acceptability was decreased from 78.21 to 77.56.

From the above mentioned results it can be concluded that pan bread fortified with 10% of lupine and soybean products were preferred over field pea. However, soy and lupine-bread were ranked similarly to whole wheat bread in most sensory attributes studied.

Breads containing low (6 to 8%) and high (12 to 15%) levels of concentrated plant proteins (soy, field pea and faba bean) were sensory evaluated using triangle tests by **Sosulski and Fleming (1978)**. They found that

Table (21): Duncan's multiple range test* for organoleptic properties of pan bread made from legume products - wheat flour blends.

Variable	Appearance (100)	Crumb texture (20)	Crumb grain (20)	Crust color (20)	Taste (10)	Odor (20)	Overall acceptability (10)	
Legume effect								
Soybean	16.30 ^a	16.53 ^a	16.29 ^a	7.32 ^b	15.60 ^a	6.79 ^b	78.83 ^a	
Field pea	15.51 ^b	15.47 ^b	15.43 ^b	6.96 ^c	14.87 ^b	6.99 ^b	75.22 ^b	
Sweet lupine	16.38 ^a	16.44 ^a	16.22 ^a	7.56 ^a	15.94 ^a	7.40 ^a	79.94 ^a	
Treatment effect								
Ungerminated	16.02 ^A	15.97 ^B	15.84 ^A	7.19 ^A	15.36 ^A	7.02 ^A	77.42 ^A	
Germinated	16.10 ^A	16.32 ^A	16.11 ^A	7.36 ^A	15.58 ^A	7.09 ^A	78.58 ^A	
Extract effect								
Legume flour	16.23 ^a	16.27 ^a	16.18 ^a	7.41 ^a	15.67 ^a	7.14 ^a	78.91 ^a	
Protein concentrate	15.89 ^b	16.02 ^a	15.78 ^b	7.14 ^b	15.28 ^b	6.98 ^a	77.09 ^b	
Replacement effect								
Control	0%	16.18 ^A	16.19 ^A	16.09 ^A	7.27 ^A	15.09 ^B	6.82 ^B	78.21 ^{AB}
	5%	16.27 ^A	16.45 ^A	16.23 ^A	7.48 ^A	15.70 ^A	7.34 ^A	76.62 ^B
	10%	16.26 ^A	16.33 ^A	16.10 ^A	7.41 ^A	15.65 ^A	7.19 ^A	79.60 ^A
	15%	15.54 ^B	15.62 ^B	15.49 ^B	6.96 ^B	15.44 ^{AB}	6.89 ^B	77.56 ^{AB}

* Means in columns followed by the same letter(s) are not significantly different at $P \leq 0.05$.

some panelists stated that all the breads were acceptable and tasty but were different than the white bread usually consumed and were therefore rated lower for preference. Nutritional value seemed to be of some concern, and many panelists indicated that they would alter buying habits to purchase a bread with higher nutritional value. Some felt that a good marketing campaign could help to sell these breads since people would of course, notice differences from the traditional white bread. Other stated that the differences they noted would be masked by eating bread with other foods, as is generally done.

Means of sensory properties of biscuits fortified with different levels of legume products are presented in Tables (22 to 25).

Results in Table (22) showed that use of different levels of ungerminated and germinated soybean flours or their protein concentrates significantly improved most organoleptic attributes of the biscuits. No significant differences were obtained between the organoleptic properties of biscuits fortified with either ungerminated or germinated soy products. Biscuits contained different levels of soybean flours had similar appearance, color, odor, taste and mouth-feel with those fortified with the same levels of soy protein concentrate. However, taste panelists gave USPC-biscuit superior texture and crispiness than USF-biscuit. Generally, biscuit fortified with soy protein concentrate showed higher overall acceptability than those contained soy flours. Also, overall acceptability of soy-biscuit was decreased slightly with increasing the substitution level.

Hegazy and Fahied (1991) showed that most organoleptic attributes of cookies containing soybean flour were not adversely affected by the addition of 10 or 15% soybean flour.

EI-Bahay et al. (1994) reported that sensory properties of soy-biscuit with 80% wheat flour and 20% soybean flour were rated as good as the control sample (100% wheat flour).

Results in Table (23) revealed that organoleptic properties of wheat-biscuit (control) were not affected significantly by fortification of biscuit with different levels of field pea products. However, ungerminated field pea, containing biscuits received less overall acceptability than control biscuit. On the other hand

Table (22): Organoleptic properties of biscuits made from soybean products - wheat flour blends.

Product	Suppl. level (100)	Appearance %	Color (10)	Odor (20)	Taste (10)	Mouth feel (20)	Texture (10)	Crispiness (15)	Overall accept. (15)
Control (100% wheat flour)	0	7.00	16.82	6.36	12.73	6.73	9.18	10.64	69.45
Ungerminated (UG)									
Soybean flour (USF)	5	7.64	17.73	6.55	16.18	7.91	9.46	12.45	77.91
	10	8.00	17.18	6.82	15.55	7.18	9.82	11.91	76.45
	15	8.18	17.91	8.00	15.73	7.55	9.55	11.45	78.36
Protein concentrate (USPC)	5	8.09	16.55	7.64	17.00	7.91	13.18	13.00	83.36
	10	8.00	16.91	7.18	16.82	7.82	12.82	13.00	82.55
	15	7.91	17.27	7.18	16.27	7.36	12.82	12.82	81.64
Germinated (G)									
Soybean flour (GSF)	5	7.82	17.45	7.73	16.73	7.64	10.18	11.82	79.36
	10	7.91	17.91	7.46	13.73	6.64	9.09	11.09	73.82
	15	7.64	17.27	8.00	14.09	6.73	10.27	10.91	74.91
Protein concentrate (GSPC)	5	8.00	17.27	7.00	17.55	8.00	13.27	13.36	84.45
	10	8.36	17.09	7.73	16.91	7.82	13.36	13.36	84.64
	15	8.18	17.09	7.36	15.91	7.55	12.36	12.45	80.91
L.S.D. at 0.05		0.824	1.033	1.073	2.154	0.985	1.656	1.280	8.353

Table (23): Organoleptic properties of biscuits made from field pea products
- wheat flour blends.

Product	Suppl. level (100)	Appearance %	Color (10)	Odor (20)	Taste (10)	Mouth feel (20)	Texture (10)	Crispi-ness (15)	Overall accept. (15)
Control (100% wheat flour)	0	7.00	16.82	6.46	12.73	6.73	9.18	10.64	69.55
Ungerminated (UG)									
Field pea flour (UPF)	5	6.73	16.18	7.18	12.64	6.46	8.27	10.36	67.82
	10	7.18	15.91	6.91	12.55	6.82	8.91	10.27	68.55
	15	7.46	15.55	6.82	11.09	6.55	8.18	9.91	65.55
Protein concentrate (UPPC)	5	6.64	15.73	6.91	12.09	6.55	9.18	9.64	66.73
	10	6.27	15.82	6.73	12.00	6.09	9.91	9.27	65.09
	15	6.55	15.09	6.55	12.18	6.18	9.18	9.82	65.55
Germinated (G)									
Field pea flour (GPF)	5	7.00	15.73	7.27	13.36	6.64	9.64	10.64	70.27
	10	7.18	15.64	6.73	12.73	6.73	10.09	10.73	69.82
	15	7.00	15.73	6.73	12.00	6.55	9.55	10.91	68.45
Protein concentrate (GPPC)	5	6.82	15.82	7.09	13.91	7.00	9.55	11.55	71.73
	10	6.91	16.00	6.82	12.91	6.82	10.09	10.64	70.18
	15	6.91	16.27	6.64	11.82	6.55	10.36	10.36	68.91
L.S.D. at 0.05		0.807	1.189	0.943	2.428	0.999	2.304	2.054	10.090

biscuit fortified with 5% GPF and 5, 10% GPPC showed superior overall acceptability scores than the control.

McWatters (1978) used field pea flours to replace 10, 20 and 30% of the wheat flour in sugar cookies. They found that sensory quality attributes were not affected adversely by use of this flour except at the 30% replacement level.

Results in Table (24) showed that lupine products used to replace 5, 10 and 15% of the wheat flour in biscuit generally improved all the sensory parameters. No significant differences were obtained between samples fortified with different levels of ungerminated and germinated legume products. At the same time, panel members gave biscuits fortified with 5% ULPC higher acceptability followed with 15% GLF.

Duncan's multiple range test was used to evaluate the organoleptic attributes of biscuits made from legume-wheat flour blends (Table 25).

Taste panelists gave the biscuits fortified with either soybean and lupine products superior total score than field pea. The total score of biscuits contained soy products was 77.26% and those contained lupine and field pea products were 75.79 and 68.55, respectively. The total score of 74.36 for biscuits fortified with germinated products diminished slightly by 0.98 for ungerminated legume products. No significant differences were obtained between biscuits enriched with either legume flour or their protein concentrate.

Results also showed that unfortified biscuits (control) recorded significantly less total score than legume fortified samples. However, the total score of legume fortified biscuits was decreased gradually with increasing the level of substitution with legume product.

From the overall biscuit sensory properties, substitution of soybean and lupine products of wheat flour at different levels were considered optimal for the preparation of biscuits (**Wittig De Penna *et al.*, 1987; Hegazy and Faheid, 1991** and **El-Bahay *et al.*, 1994**).

Table (24): Organoleptic properties of biscuits made from lupine products - wheat flour blends.

Product	Suppl. level (100)	Appearance %	Color (10)	Odor (20)	Taste (10)	Mouth feel (20)	Texture (10)	Crispiness (15)	Overall accept. (15)
Control (100% wheat flour)	0	7.0	16.82	6.46	12.73	6.73	9.18	10.64	69.55
Ungerminated (UG)									
Lupine flour (ULF)	5	7.45	16.45	7.27	14.64	7.27	9.91	11.64	74.64
	10	7.91	17.18	7.09	14.64	6.91	10.64	11.73	76.09
	15	7.91	16.36	7.27	14.18	7.00	10.64	12.45	75.82
Protein concentrate (ULPC)	5	8.64	17.27	7.18	15.09	8.18	12.45	12.55	81.36
	10	7.91	17.00	7.09	15.82	8.09	12.18	12.27	80.36
	15	8.00	16.45	6.91	14.27	7.27	11.27	11.91	76.09
Germinated (G)									
Lupine flour (GLF)	5	7.64	16.55	7.55	15.36	7.55	10.91	11.91	77.45
	10	8.27	17.55	7.09	15.64	7.55	11.55	12.18	79.82
	15	8.27	17.55	7.46	15.73	8.00	11.73	12.18	80.91
Protein concentrate (GLPC)	5	8.00	16.18	7.36	14.09	7.27	12.82	12.00	77.73
	10	8.00	16.00	7.36	15.27	7.91	11.82	12.00	78.36
	15	7.73	15.18	7.18	14.55	7.55	11.64	12.00	75.82
L.S.D. at 0.05		1.100	1.406	1.087	2.193	1.000	1.898	1.331	9.347

Table (25): Duncan's multiple range test* for organoleptic properties of biscuits made from legume products - wheat flour blends.

Variable	Appearance (100)	Color (10)	Odor (20)	Taste (10)	Mouth feel (20)	Texture (10)	Crispi- ness (15)	Overall accept. (15)	
Legume effect									
Soybean	7.73 ^a	17.18 ^a	7.13 ^a	15.21 ^a	7.31 ^a	10.81 ^a	11.89 ^a	77.26 ^a	
Field pea	6.92 ^b	16.05 ^c	6.76 ^b	12.51 ^c	6.61 ^b	9.29 ^b	10.42 ^b	68.55 ^b	
Sweet lupine	7.73 ^a	16.69 ^b	7.04 ^a	14.39 ^b	7.34 ^a	10.89 ^a	11.71 ^a	75.69 ^a	
Treatment effect									
Ungerminated	7.44 ^A	16.64 ^A	6.91 ^A	13.96 ^A	7.06 ^A	10.10 ^B	11.26 ^A	73.38 ^A	
Germinated	7.49 ^A	16.63 ^A	7.05 ^A	14.11 ^A	7.12 ^A	10.56 ^A	11.41 ^A	74.36 ^A	
Extract effect									
Legume flour	7.47 ^a	16.78 ^a	7.02 ^a	13.87 ^a	7.00 ^a	9.73 ^b	11.18 ^a	73.05 ^a	
Protein concentrate	7.46 ^a	16.50 ^b	6.94 ^a	14.20 ^a	7.18 ^a	10.93 ^a	11.49 ^a	74.69 ^a	
Replacement effect									
Control	0%	7.00 ^B	16.82 ^A	6.42 ^B	12.73 ^C	6.73 ^B	9.18 ^B	10.64 ^B	69.52 ^B
	5%	7.54 ^A	16.58 ^A	7.23 ^A	14.89 ^A	7.36 ^A	10.73 ^A	11.74 ^A	76.07 ^A
	10%	7.66 ^A	16.68 ^A	7.08 ^A	14.55 ^{AB}	7.19 ^A	10.77 ^A	11.54 ^A	75.48 ^A
	15%	7.64 ^A	16.48 ^A	7.17 ^A	13.98 ^B	7.07 ^A	10.63 ^A	11.43 ^A	74.41 ^A

* Means in columns followed by the same letter(s) are not significantly different at $P \leq 0.05$.

Part II. Whey products as a source of animal protein

4.6. Chemical composition of whey products

Different levels of sweet whey powder (SW), ultrafiltrated-whey protein concentrate (WPC) and an equal mixture of them were used in this investigation for fortification of some bakery products.

Composition of SW and WPC are presented in Table (26) SW contained 3% moisture, 12% protein, 70% lactose, 1.5% fat and 8% ash, WPC had the following analysis, 4.5% moisture, 78% protein, 4% lactose, 7.5% fat and 2.5% ash. SW characteristics by high lactose content than WPC. The WPC used in this study was high protein because it contained protein between 60% and 80% as mentioned by **Glover (1985)** and **Ottosen (1991)**.

Table (26): Composition of sweet whey powder (SW)* and ultrafiltrated-whey protein concentrate (WPC)**.

Sample	Moisture %	Chemical composition on dry weight basis			
		Protein %	Fat %	Ash %	Lactose %
SW	3.00	12.00	1.50	8.00	70.0
WPC	4.50	78.00	7.50	2.50	4.0

* Spray dried sweet whey powder, ADPI Extra grade, Dutch origin, valid for Human consumption. Taly Establishment (Holland).

** Standards of Espriion 580 (ultrafiltrated-whey protein concentrate) DMV interaccional - veghel- The Netherlands.

4.7. Functional properties

Functional properties of whey proteins encompass those physico-chemical attributes of a protein that make it useful in food products.

4.7.1. Water and oil absorption capacities

Water and oil absorption capacities (WAC, OAC) and water-oil absorption index values (WOAI) of sweet whey powder (SW), ultrafiltrated-whey protein concentrate (WPC) and an equal mixture of them are presented in Table (27). WPC had significantly higher WAC (414.53 g water/100 g sample) than those of SW + WPC (345.81 g water/100 g) and SW (247.19 g water/100 g). The highest

WAC value of WPC may contributed to their high protein content (78.0%). **Fleming et al. (1974)** reported that water absorption was increased by soy products with increased protein content. Interaction of water with proteins are important both to the structure of the proteins and to their behaviour in food systems. Hydrogen bonding between amino acid residues and water, ion dipole and dipole-dipole interactions are all important in protein-water interactions. A part from these molecular interactions between protein and water physicochemical forces (such as adsorption) may also cause water-protein interactions. Water can be contained in capillaries or physically entrapped in particles of proteins (**Kilara, 1994**).

Concerning oil absorption, data in Table (27) clearly showed that WPC absorbed significantly more oil (248.3 g oil/100 g sample) than a mixture of SW + WPC (200.85) and WS (163.16).

Table (27): Water and oil absorption capacities* (WAC and AOC) and water-oil absorption index (WOAI) of sweet whey powder (SW) and ultrafiltrated-whey protein concentrate (WPC).

Products	WAC (g water/100 g sample)	OAC (g oil/100 g sample)	WOAI (g water/g oil)
SW	247.19 ^b	163.16 ^c	1.52 ^a
50% SW + 50% WP	345.81 ^{ab}	200.85 ^b	1.72 ^a
WPC	414.53 ^a	248.30 ^a	1.67 ^a

* Means in the same column followed by the same letter(s) are not significantly different at $P \leq 0.05$.

Oil absorption was mainly attributed to the physical entrapment of oil and was related to the number of non-polar side chains on proteins that bind hydrocarbon chains of fats (**Lin et al., 1974** and **Kinsella, 1979**). Different tested whey products had lower oil absorption compared to their water absorption values suggested that the major protein in these products were predominantly hydrophilic (**Deshpande et al., 1982**).

Water-oil absorption index (WOAI) is a measure of relative simultaneous attraction of a protein to water and oil. However, a suitable balance between hydrophilic and lipophilic (WOAI nearly two) was required for maximal emulsifying capacity (**De Kanterewicz et al., 1987**).

All whey products showed high and similar WOAI, whereas the suitable balance were noted in all tested samples. This indicates that the protein molecule acted as a mediator in the formation of stable emulsion by binding both water and oil molecules to form thick barriers which prevented the oil particles from coalescing (**Okezia and Bells, 1988**).

4.7.2. Emulsion capacity (EC) and stability (ES)

Behaviour of proteins at the oil/water interface are of interest in foods. If the dispersed phase is oil and the continuous phase is water, then an oil-in-water emulsion results. On the other hand, if the continuous phase is oil and the dispersed phase is aqueous, a water-in-oil emulsion is obtained. If the density of the two phases are different, separation of the phases occurs sooner (**Kilara, 1994**).

Emulsion capacity (EC) and emulsion stability (ES) of sweet whey powder (SW), ultrafiltrated whey protein concentrate and an equal mixture of them are given in Fig. (14) and Table (28).

The minimal emulsifying capacity of different samples were measured at pH 4.5, near their isoelectric point with the lower protein solubility (Fig. 15) and it increased below and above this region, reaching its maximum at pH 9.0. Moreover, WPC has a higher emulsification capacity (610 ml oil/g sample at pH 9.0) followed by SW + WPC (450 ml oil/g sample) and (350 ml oil/g sample). Similar observation have been made by **Melachouris (1984)**. He found that WPC has a higher emulsification capacity than nonfat dry milk but not as high as sodium caseinate.

Ottosen (1991) mentioned that whey protein is a good emulsifier. It contains both hydrophilic and lipophilic groups and therefore has the ability to produce the surface tension between oil and water or, in other words, to form oil-in-water and water-in-oil emulsions. The emulsifying properties of WPC are highly dependent on the solubility of the proteins and will diminish with decreasing solubility.

Emulsion stability (ES) of 1% dispersion of SW and WPC were followed during 48 hr and the results are given in Table (28). WPC had considerably higher ES (the lower percentage of aqueous phase separated after 48 hr)

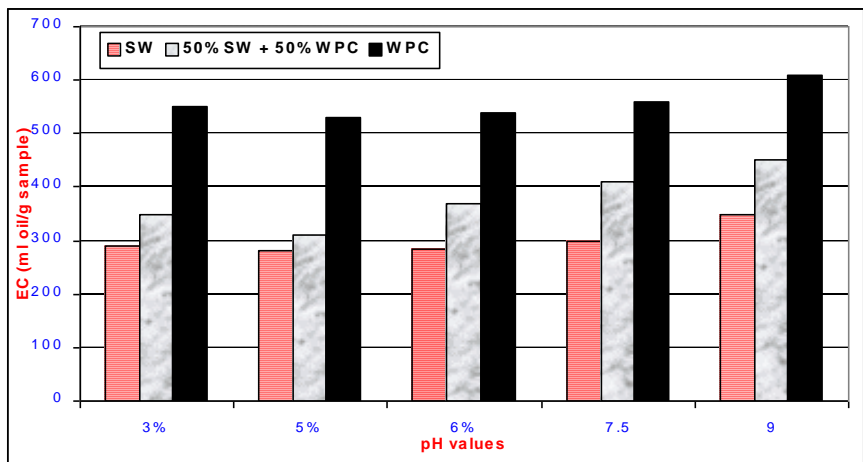


Fig. (14): Emulsion capacity (EC) of 1% dispersion of sweet whey powder (SW) and ultrafiltrated whey protein concentrate (WPC).

Table (28): Emulsion stability (ES) of 1% dispersion of sweet whey powder (SW), and ultrafiltrated-whey protein concentrate (WPC).

Sample	pH value	ES% Water separated after time, hr.					
		0.25	0.50	2.00	3.00	24.00	48.00
SW	3.0	17.95	19.23	20.51	20.51	20.51	23.31
	4.5	29.82	31.58	31.58	32.46	33.33	35.09
	6.0	27.27	27.59	29.31	29.31	29.31	31.03
	7.5	17.14	20.00	20.00	22.86	22.86	24.29
	9.0	6.67	6.67	10.00	13.33	16.67	16.67
50% SW + 50% WPC	3.0	0.00	3.33	4.44	6.67	14.44	15.56
	4.5	10.00	14.29	33.33	33.33	37.04	37.04
	6.0	0.00	26.00	28.00	30.00	30.00	34.00
	7.5	0.00	0.00	15.71	15.71	17.14	20.00
	9.0	0.00	0.00	1.79	10.71	12.50	13.39
WPC	3.0	0.00	2.94	7.06	7.93	13.33	14.44
	4.5	11.11	17.78	20.00	20.00	22.22	22.22
	6.0	0.00	3.77	11.32	15.09	16.98	18.87
	7.5	0.00	1.85	3.70	5.56	12.96	12.96
	9.0	0.00	0.00	0.00	3.70	5.56	7.41

followed by a mixture of SW + WPC and SW. Emulsion stability showed wide variations for the different samples over the pH ranges of 3 to 9. However, the minimum ES was found around pH 4.5. **De Wit (1988)** mentioned that factors affecting whey protein emulsion include pH and ionic strength. Around their isoelectric point whey proteins form poor, unstable emulsion.

4.7.3. Nitrogen solubility index (NSI)

The functional applications of milk proteins are primarily dependent upon their solubility in water and their hydrophilic characteristics (**Smith, 1976**).

The nitrogen solubility index (NSI) of sweet whey powder and ultrafiltered-whey protein concentrate was determined as a function of pH, and the results are given in Fig. (15).

Results showed that different tested samples (SW, SW + WPC and WPC) are highly soluble in water at different pH values. The protein solubility for SW and WPC was shown to be independent of pH. SW had higher nitrogen solubility index (reached 96.93% at pH 9) than WPC (88-62%).

Ottosen (1991) stated that the whey protein in the native state exhibit rapid water solubility, even at low pH. Also, the protein solubility for iron complex, CMC complex and metaphosphate complex WPC was shown to be highly dependent on pH, whereas protein solubility for the other WPCs were essentially independent of pH.

Kilara (1994) reported that proteins are least soluble in the pH range close to their isoelectric point, but whey protein are soluble at these pH values. The wide range of pH values over which whey proteins are soluble make them ideal for use in a variety of products.

4.8. Rheological and physical properties of wheat flour-whey products blends

4.8.1. Farinograph properties

Effect of whey products added at 5, 10 and 15% of wheat flour on farinograph properties of the dough are given in Figures (16 To 19) and Table (29).

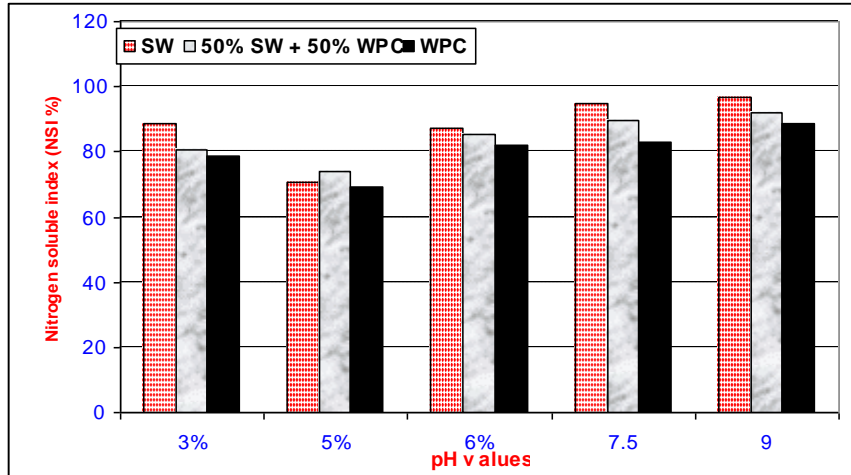


Fig. (15): Nitrogen solubility index (NSI) of sweet whey powder (SW) and ultrafiltrated whey protein concentrate (WPC).

Table (29): Farinogram parameters of whey products-wheat flour blends.

Sample	Replacement level	Farinogram parameters					
		Water absorption (%)	Arrival time (min.)	Dough development (min.)	Dough stability time (min.)	Mixing tolerance index (B.U.)	Degree of softening (B.U.)
Control	0	61.5	1.5	3.0	13.5	20	20
SW	5	56.0	1.0	1.5	24.0	25	10
	10	50.0	1.0	8.5	30.5	25	10
	15	46.0	0.5	11.0	41.5	25	5
50% WP + 50% WPC	5	55.0	3.0	10.0	15.5	30	50
	10	50.0	6.5	11.5	15.5	25	45
	15	48.2	13.0	15.5	13.0	30	40
WPC	5	56.0	6.5	11.0	11.5	40	40
	10	56.8	12.5	14.5	4.5	60	70
	15	55.0	18.0	21.0	4.5	70	80

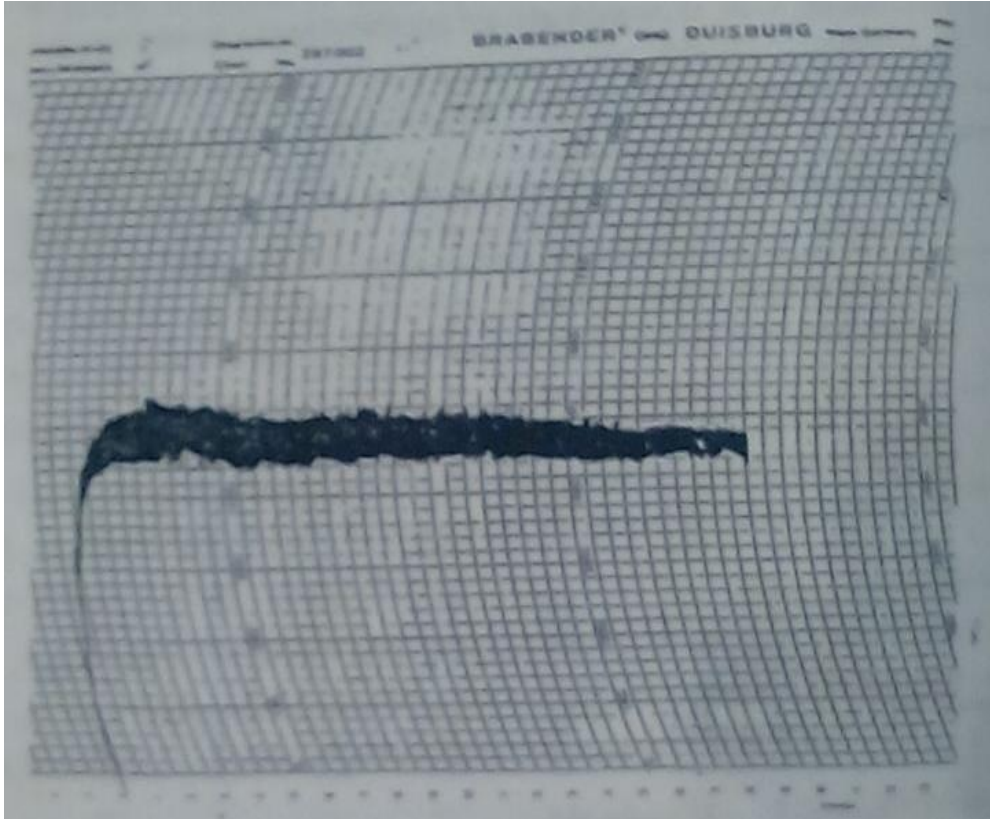


Fig. (16): Farinogram of 100% wheat flour (Control).

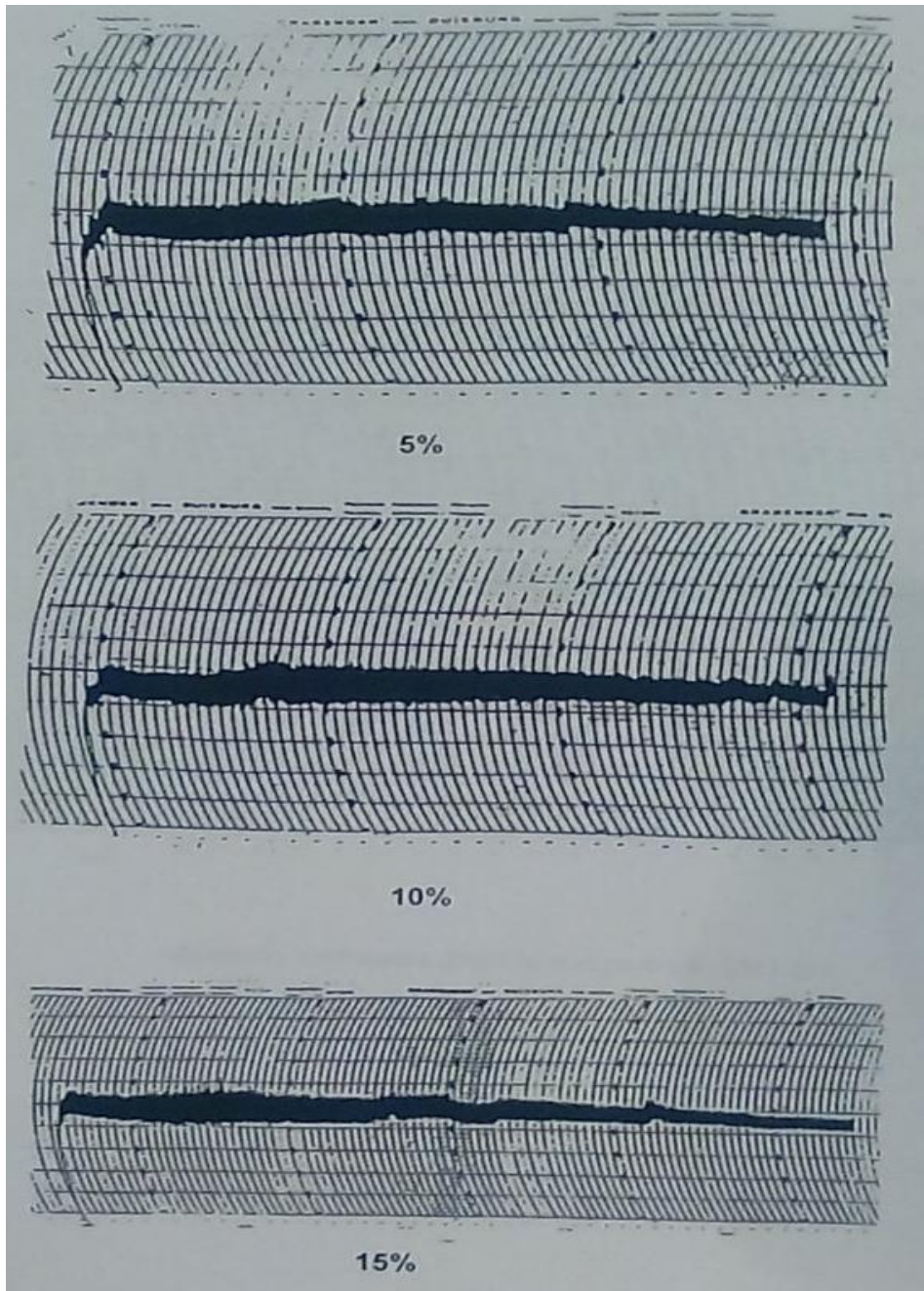


Fig. (17): Farinograms of blends containing wheat flour and different levels of sweet whey powder (SW).

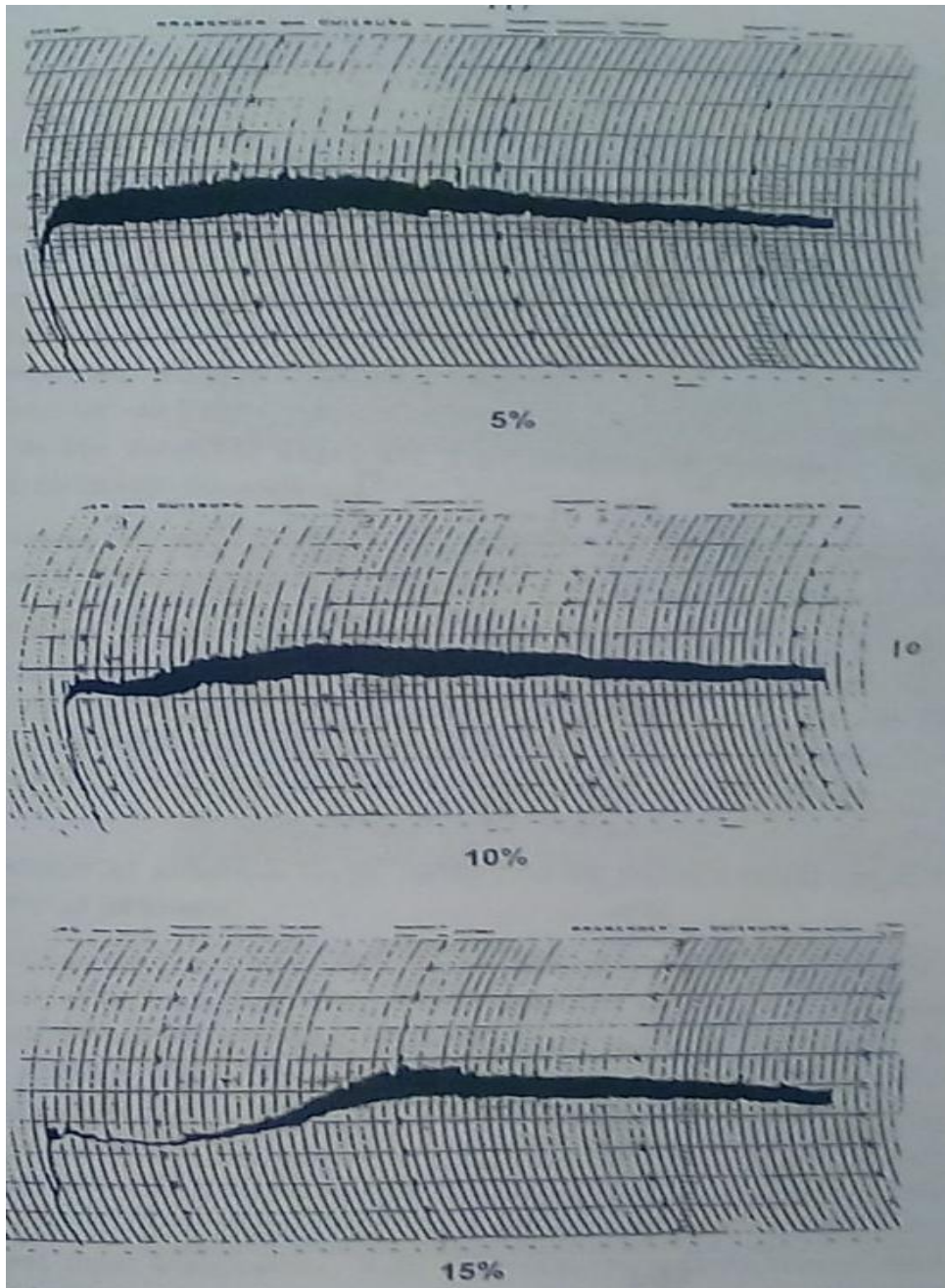


Fig. (18): Farinograms of blends containing wheat flour and an equal mixture of sweet whey powder (SW) and whey protein concentrate (WPC).

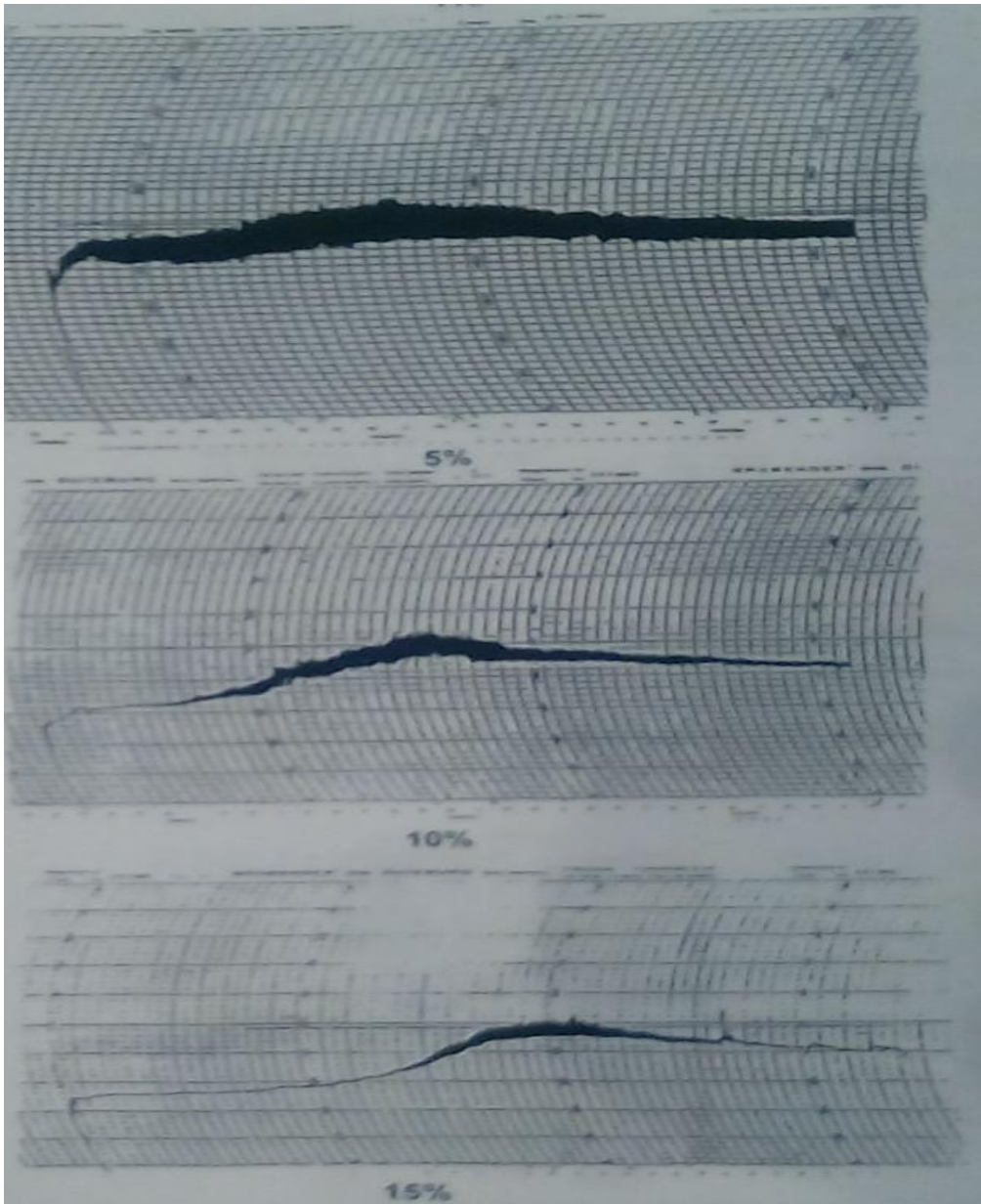


Fig. (19): Farinograms of blends containing wheat flour and different levels of whey protein concentrate (WPC).

It could be noticed that dough water absorption was decreased with increasing the level of whey products. The highest decrease was observed for the dough containing sweet whey powder (SW) and the lower decrease was obtained for whey protein concentrate. **El-Farra et al. (1981)** mentioned that addition of whey caused a decrease in the water absorption which seems to be a function of the whey solids (lactose and lacto-albumins) which are not classified as a high moisture absorbent during dough processing stage. **Sanchez et al. (1984)** found that water absorption is decreased as the level of WPC in the blend is increased, thus demonstrating the importance of wheat dilution on dough consistency.

Barnes et al. (1973) found that lactose and acid whey decrease the water absorption of dough. Same findings were obtained by **Korshid et al. (1994)**.

Addition of different levels of sweet whey (SW) generally reduced the arrival time of the dough. However, dough containing different levels of WPC and a mixture of SW+WPC had a higher arrival time (ranged from 3.0 to 18.0 min) than control (1.5 min) and the arrival time was increased as the amount of the former products increased. Similar results are obtained by **Holsinger (1983)**.

Addition of different levels of WPC to the flour increased arrival time than SW+WPC at all levels.

Results also showed that dough development time (mixing time) was increased with increasing the amount of whey products in the blends. The dough development time was increased from 3.0 min for control to 21.0 min for dough fortified with 15% WPC. The increase in mixing time reflected the expected differences in the physical and chemical properties of the whey protein products. **El-Farra et al. (1981)** found that addition of liquid whey had a significant effect on the mixing time of the dough. The increase in dough mixing time may be due to the differences in molecular weights of between whey solids and wheat flour compounds. With respect to the stability, results showed that blends containing different levels of sweet whey had higher dough stability than control. On contrary, WPC reduced markedly dough stability as can be seen in Table (29).

The combination of SW with WPC had an intermediate stability time between SW and WPC. These results are in accordance with those obtained by **Sanchez et al. (1984)**.

Supplementation of wheat flour with different levels of whey products led to increase mixing tolerance index of the fortified dough and it was more pronounced for those containing high levels of WPC. The mixing tolerance index of wheat flour-dough was 20 B.U., raised to 70 B.U. as a result of adding 15% of WPC. The tolerance index is also influenced in a linear manner by the WPC content at the blend (**Sanchez et al., 1984**). Same findings were obtained by **Matsuo et al. (1972)**, who reported that farinograph characteristics markedly affected by the increase of protein content, since this increase led to evaluating the mixing tolerance index.

The incorporation of WPC and a mixture of SW+WPC in wheat flour dough increased the dough softening. However, dough became softer as the level of WPC was increased in the dough.

Sanchez et al. (1984) found that incorporation of low denaturated whey protein concentrates to wheat flour increased markedly the degree of softening. This may be due to the presence of sulfhydryl groups in whey lacto-albumin, which would cause the dough softening (**El-Farra et al., 1981**).

On the contrary, fortification of wheat flour with different levels of SW markedly reduced degree of softening than the control.

4.8.2. Wet and dry gluten

Gluten is a protein complex which forms during the mixing of flour and water. This formation takes place thoroughly in the dough prepared for bread-making.

Therefore, the effect of replacement of wheat flour with different levels of SW, SW+WPC and WPC on wet and dry gluten values of the blends are represented in Table (30).

Wheat flour-blend contained significantly higher wet and dry gluten (27.58 and 10.40%) and lower hydration ratio (165.17) than SW and WPC-blends.

Table (30): Wet and dry gluten values* of whey products-wheat flour blends.

Sample	Replacement level %	Gluten %		Hydration ratio
		Wet %	Dry %	
Control (100% wheat flour)	0	27.58 ^A	10.40 ^A	165.17 ^F
SW	5	26.12 ^B	9.52 ^B	174.72 ^E
	10	22.25 ^E	7.91 ^E	181.40 ^{BC}
	15	17.75 ^H	6.45 ^G	175.00 ^E
50% SW + 50% WPC	5	27.25 ^A	9.69 ^B	180.99 ^{BC}
	10	25.17 ^C	9.03 ^C	178.84 ^{CD}
	15	19.88 ^G	7.18 ^F	176.95 ^{DE}
WPC	5	25.98 ^B	8.80 ^{CD}	195.26 ^A
	10	24.02 ^D	8.49 ^D	182.99 ^B
	15	21.13 ^F	7.81 ^E	183.24 ^B

* Means in columns followed by the same letter(s) are not significantly different ($P \leq 0.05$).

Consequently increasing SW and WPC levels in the blends was accompanied by a significant decrease in wet and dry gluten values. On the other hand, whey-wheat blends showed significantly higher hydration ratio than wheat flour blend. However, the gluten content of the blends containing up to 15% whey protein concentrate remained acceptable.

Addition of liquid whey into wheat flour doughs especially at high levels decreased both the wet and dry gluten yield. This could be attributed to the effect of lacto-albumin on the gluten network (**El-Farra *et al.*, 1981**).

4.9. Characteristics of pan bread and biscuits fortified with SW and WPC

4.9.1. Chemical composition

The chemical composition of pan bread fortified with different levels of sweet whey (SW) and ultrafortified whey protein concentrate (WPC) and a mixture of them were compared with wheat flour-bread and the results are presented in Table (31).

The protein content of the bread was improved significantly from 12.44% up to 20.50% depending on the amount and the protein content of the added whey products. As expected bread enriched with WPC contained higher values of protein followed by SW + WPC and SW. Protein content was raised significantly from 12.44% for wheat flour-bread (control) to 13.00, 13.51 and 14.08 for bread fortified with 5, 10 and 15% SW respectively. The corresponding increase percentages of protein reached 4.50, 8.60, and 13.18% (Fig. 20). On the other hand, supplementation of wheat flour with 5, 10 and 15% of WPC increased significantly the protein content of the bread to 15.29, 17.87 and 20.50%, respectively (22.91, 43.65 and 64.79% increase).

Renz-Schauen and Renner (1987) found that by adding the WPCs to wheat flour, the protein content of the bread was increased from 12.8% up to 15.9% when 6% of high whey protein concentrate was added.

Fat and ash contents are also higher for whey-fortified bread than for control bread specially at 15% SW. Chemical composition of bread fortified with different levels of SW + WPC recorded intermediate results between those

Table (31): Chemical composition** of pan bread fortified with different levels of sweet whey powder (SW) and ultrafiltrated whey protein concentrate (WPC).

Sample	Replacement level %	Chemical composition calculated on dry weight basis				NFE**
		Moisture %	Protein %	Fat %	Ash %	
Control (100% wheat flour)	0	28.99 ^{BC}	12.44 ^I	2.78 ^C	1.76 ^G	83.03 ^A
SW	5	28.10 ^C	13.00 ^H	2.89 ^C	2.17 ^{DE}	81.94 ^B
	10	28.42 ^C	13.51 ^G	3.33 ^{ABC}	2.45 ^B	80.71 ^C
	15	28.71 ^{BC}	14.08 ^F	3.94 ^A	2.83 ^A	79.16 ^E
50% SW + 50% WPC	5	26.14 ^D	14.11 ^F	2.90 ^C	1.90 ^F	81.08 ^C
	10	29.06 ^{BC}	14.73 ^E	2.82 ^C	2.21 ^D	80.24 ^D
	15	31.62 ^A	15.84 ^C	3.19 ^{BC}	2.37 ^C	78.60 ^F
WPC	5	28.56 ^{BC}	15.29 ^D	2.74 ^C	1.76 ^G	80.21 ^D
	10	28.88 ^{BC}	17.87 ^B	3.64 ^{AB}	1.91 ^F	76.58 ^G
	15	29.73 ^B	20.50 ^A	3.73 ^{AB}	2.13 ^E	73.64 ^H

* NFE = Nitrogen free extract.

** Means in columns followed by the same letter(s) are not significantly different ($P \leq 0.05$).

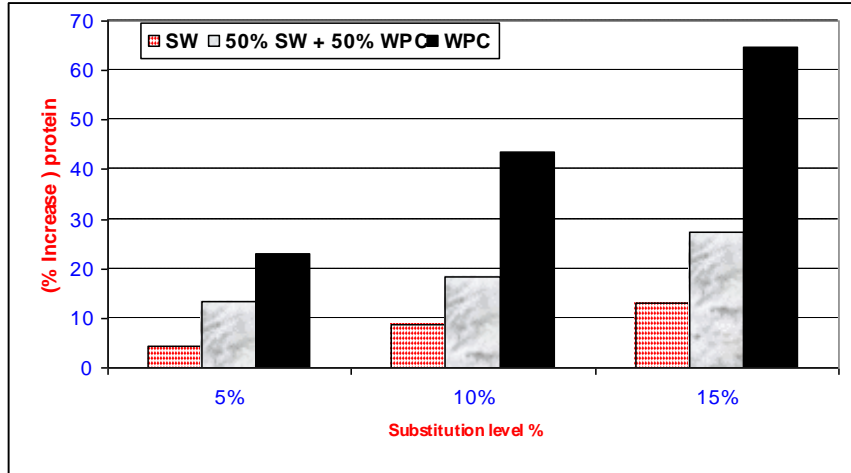


Fig. (20): Percentage of protein increase of pan bread fortified with sweet whey powder (SW) and ultrafiltrated whey protein concentrate (WPC).

contained WPC and SW. Same results were obtained by **Renner (1983)** and **Sanchez et al. (1989)**.

Data presented in Table (32) revealed the effect of fortification of wheat flour with different levels of whey products on the chemical composition of the resultant biscuits. From these data, it becomes evident that particularly the amount of whey products added to wheat flour at manufacturing biscuits has a pronounced effect on the chemical analysis of the resulting product. Wheat flour biscuit contained 3.89% moisture, 8.06% protein, 9.33 fat and 0.56% ash. As whey products have a high protein content, the protein content of the biscuit can be significantly increased when wheat flour is partly substituted by WPC. By adding 5, 10 and 15% of SW the protein content of the control biscuit was increased by 12.16, 31.89 and 50.49%, respectively, (Fig. 21). Addition of 5, 10 and 15% of WPC markedly improved protein content of the biscuit to 11.89, 13.80 and 15.40% corresponding to 47.52, 71.22 and 91.07% increase. However, fortification of the wheat flour with 50:50 of SW+WPC recorded intermediate results between SW and WPC. Biscuits fortified with different whey products showed significantly higher fat and ash content than the control biscuit.

4.9.2. Baking quality

The influence of the fortification of wheat flour with different levels of whey products on loaf weight, loaf volume and specific volume of pan bread are shown in Table (33). By adding different levels of whey product the loaf weight of the bread was improved significantly compared with the control samples except for those contained 5% of either SW or SW+WPC. The loaf weight for 15% WPC bread was 104.30 g. However, fortification of wheat flour with different levels of SW produced bread with low volume compared with control.

Harper et al. (1983) mentioned that lactose is more concentrated in sweet whey (73.5%) than in non fat dry milk (NFDM) 35.9%, and therefore might be a key to the low volume of sweet whey breads. Control bread (no milk) and those contained 4% of either sweet whey or NFDM recorded the following loaf volume 836, 813 and 868 cm³, respectively.

Table (32): Chemical composition** of biscuits fortified with different levels of sweet whey powder (SW) and ultrafiltrated whey protein concentrate (WPC).

Product	Replacement level %	Moisture %	Chemical composition calculated on dry weight basis			NFE*
			Protein %	Fat %	Ash %	
Control (100% wheat flour)	0	3.89 ^F	8.06 ^I	9.33 ^E	0.56 ^D	82.05 ^A
SW	5	5.16 ^E	9.04 ^H	9.67 ^{DE}	1.03 ^C	80.26 ^B
	10	6.40 ^{BC}	10.63 ^F	9.87 ^{CD}	1.14 ^C	78.36 ^D
	15	6.56 ^B	12.13 ^{CD}	10.38 ^{BC}	1.16 ^{BC}	76.33 ^F
50% SW + 50% WPC	5	5.96 ^D	9.75 ^G	9.86 ^{CD}	1.08 ^C	79.31 ^C
	10	6.49 ^B	11.16 ^E	10.27 ^{BC}	1.18 ^{BC}	77.40 ^E
	15	6.65 ^B	12.54 ^C	10.61 ^B	1.40 ^{AB}	75.45 ^G
WPC	5	6.13 ^{CD}	11.89 ^D	9.89 ^{CD}	1.17 ^{BC}	77.06 ^{EF}
	10	6.71 ^B	13.80 ^B	10.47 ^B	1.25 ^{ABC}	74.48 ^H
	15	7.33 ^A	15.40 ^A	11.41 ^A	1.45 ^A	71.74 ^I

* NFE = Nitrogen free extract.

** Means in columns followed by the same letter(s) are not significantly different ($P \leq 0.05$).

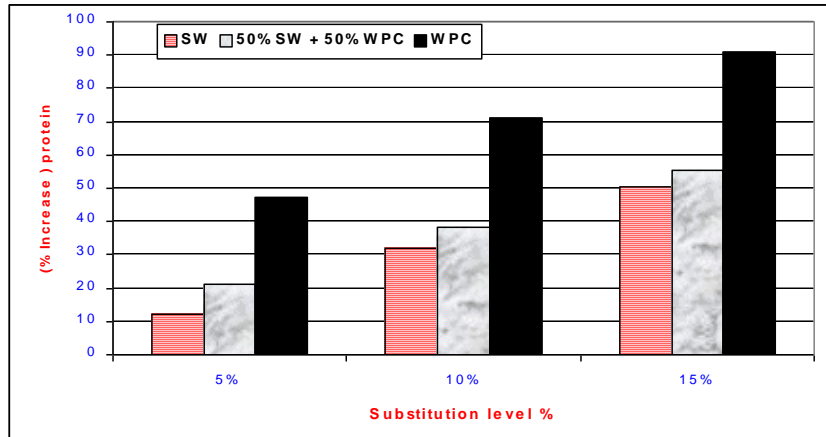


Fig. (21): Percentage of protein increased of biscuits fortified with sweet whey (SW) and ultrafiltrated whey protein concentrate (WPC).

Table (33): Baking quality* of pan bread fortified with different levels of sweet whey powder (SW) and ultrafortified whey protein concentrate (WPC).

Product	Replacement level %	Loaf weight (g)	Loaf volume (cm ³)	Specific volume (cm ³ /g)
Control (100% wheat flour)	0	90.70 ^E	222.5 ^C	2.47 ^D
SW	5	82.00 ^F	131.3 ^F	1.59 ^H
	10	95.56 ^{CD}	120.7 ^G	1.26 ^I
	15	98.34 ^{BC}	101.8 ^H	1.03 ^J
50% SW + 50% WPC	5	79.13 ^G	166.8 ^E	2.11 ^E
	10	94.24 ^D	177.8 ^D	1.88 ^F
	15	95.52 ^{CD}	169.2 ^E	1.77 ^G
WPC	5	94.96 ^D	299.8 ^A	3.16 ^A
	10	100.20 ^B	280.0 ^B	2.79 ^C
	15	104.30 ^A	295.2 ^A	2.83 ^B

* Means in columns followed by the same letter(s) are not significantly different ($P \leq 0.05$).

As mentioned before, (Table 26) sweet whey powder used in this investigation contained high amount of lactose content (70%) and, therefore it explained that it produced bread with low volume.

However, fortification of bread with different levels of WPC gave loaf volume higher than control. Same findings was obtained by **Harper et al. (1983)**. They found that whey protein concentrate tested (34.5% and 39.1% protein) giving loaf volumes, similar to NFDM control loaves and higher than control bread (no milk).

This can be explained that WPC characteristics by high protein content (78%) and low lactose content (4%) than in SW and, therefore might be a key to the high volume of WPC breads (**Harper et al., 1983**).

Specific loaf volume (cm³/g) of WPC breads showed significantly higher values than that of control bread. However, SW and SW + WPC-bread recorded lower specific volume than control and WPC. The best loaf volume and specific volume was observed for bread fortified with 5% of WPC.

4.10. Sensory evaluation

The organoleptic properties of pan bread enriched with different levels of whey products are given in Table (34).

Panel members gave the bread fortified with 5 or 10% of whey products superior scores than that of wheat flour bread (control). At the same time, the organoleptic properties of whey products-bread were not differed significantly with those of bread manufactured from 100% flour except for crust color. Pan bread fortified with either 5% SW or 5 and 10% of WPC showed significantly higher crust colour than control. However, overall acceptability was reduced gradually with increasing the level of fortification with whey products.

From the above mentioned results, it can be concluded that replacement of wheat flour with 5 and 10% whey protein concentrates generally improved organoleptic properties of the resultant breads.

These results were in agreement to **Harber (1973)**, **Huffman and Hewitt (1990)** and **Korshid et al. (1994)**. They found that the addition of concentrated whey to the wheat flour improved the organoleptic properties of bakery products.

Table (34): Organoleptic properties* of pan bread made from whey - wheat flour blends.

Product	Suppl. level (100)	Appearance %	Crumb texture (20)	Crumb grain (20)	Crust color (20)	Taste (10)	Odour (20)	Overall accept. (10)
Control	0	14.56 ^{AB}	16.67 ^A	16.67 ^A	6.22 ^C	15.56 ^A	6.78 ^A	76.44 ^A
SW	5	16.22 ^{AB}	17.11 ^A	16.33 ^A	7.89 ^{AB}	16.22 ^A	6.89 ^A	80.67 ^A
	10	15.67 ^{AB}	16.67 ^A	17.00 ^A	7.44 ^{ABC}	16.89 ^A	7.56 ^A	81.22 ^A
	15	13.78 ^B	15.56 ^A	15.44 ^A	6.56 ^{ABC}	15.56 ^A	6.89 ^A	73.78 ^A
50% SW+50% WPC	5	16.22 ^{AB}	17.00 ^A	15.56 ^A	7.44 ^{ABC}	15.67 ^A	7.33 ^A	79.22 ^A
	10	15.44 ^{AB}	15.89 ^A	15.56 ^A	7.11 ^{ABC}	16.00 ^A	7.00 ^A	77.00 ^A
	15	15.44 ^{AB}	16.33 ^A	16.00 ^A	6.67 ^{ABC}	16.11 ^A	6.78 ^A	77.33 ^A
WPC	5	17.22 ^A	16.56 ^A	17.00 ^A	8.11 ^A	16.89 ^A	8.00 ^A	83.78 ^A
	10	16.78 ^{AB}	16.22 ^A	16.67 ^A	8.11 ^A	16.78 ^A	8.00 ^A	82.56 ^A
	15	14.67 ^{AB}	15.56 ^A	15.56 ^A	6.44 ^{BC}	15.78 ^A	7.56 ^A	75.56 ^A

* Means in columns followed by the same letter(s) are not significantly different ($P \leq 0.05$).

Schaap (1992) found that there are significant taste differences between the bread samples fortified by WPC and the control samples. Such significant differences only could be observed at an amount of 6% added whey protein concentrate but it has to be emphasized that the panelists certified a better taste quality to the protein fortified bread.

Means of sensory properties of biscuit samples fortified with different levels of whey products are presented in Table (35). Results showed that biscuits contained different levels of SW had similar appearance, color, mouth-feel crispiness and overall acceptability with that of control, except for those contained 10% SW which recorded less color score than control. However, SW-biscuits recorded significantly superior odor, taste and texture scores than wheat flour biscuit (control).

With respect to the effect of supplementation of wheat flour with different levels of WPC on sensory properties, results in Table (35) showed that WPC-biscuits had significantly higher appearance, odor, taste, texture and overall acceptability over the control. At the same time, no significant differences were obtained for color, mouth-fell and crispiness between WPC-biscuit and control.

Sensory properties of biscuit fortified with different levels of equal mixture of SW + WPC recorded moderate scores between those contained SW or WPC.

In overall acceptability WPC-biscuit received the highest ranking, however, wheat flour biscuit (control) received the lowest ranking, and SW+WPC and SW-biscuit were taken intermediate scores. The total scores for biscuit supplemented with 15% of SW, SW+WPC and WPC were 75.00, 82.09 and 82.36, respectively and that for control was 65.27.

Generally, biscuit fortified with WPC was preferred by the panelists over SW and SW+WPC.

At the same time, panel members gave the best scores for biscuit contained 15% of either WPC or SW+WPC for all sensory properties followed by those contained 10 to 5% WPC (without significant differences between them).

Table (35): Organoleptic properties* of biscuits made from whey - wheat flour blends.

Variable	Appearance (100)	Color (10)	Odor (20)	Taste (10)	Mouth feel (20)	Texture (10)	Crispi- ness (15)	Overall accept. (15)
Control	0 6.36 ^C	16.46 ^{AB}	5.82 ^B	12.00 ^C	6.27 ^B	8.09 ^B	10.27 ^B	65.27 ^B
SW	5 6.27 ^C	14.91 ^{BC}	7.55 ^A	14.00 ^{BC}	6.73 ^B	11.73 ^A	11.36 ^{AB}	72.55 ^{AB}
	10 6.36 ^C	14.27 ^C	7.18 ^A	14.46 ^B	6.82 ^{AB}	11.73 ^A	11.09 ^{AB}	71.91 ^{AB}
	15 6.82 ^{BC}	15.64 ^{ABC}	7.27 ^A	14.82 ^{AB}	7.00 ^{AB}	12.55 ^A	10.91 ^{AB}	75.00 ^{AB}
50%SW+50% WPC	5 6.36 ^C	14.91 ^{BC}	7.18 ^A	15.27 ^{AB}	6.73 ^B	12.09 ^A	11.91 ^{AB}	74.46 ^{AB}
	10 7.18 ^{ABC}	15.82 ^{ABC}	7.55 ^A	14.91 ^{AB}	6.36 ^B	11.55 ^A	11.00 ^{AB}	74.36 ^{AB}
	15 7.36 ^{ABC}	16.64 ^{AB}	7.73 ^A	16.27 ^{AB}	8.09 ^A	13.27 ^A	12.73 ^A	82.09 ^A
WPC	5 7.18 ^{ABC}	16.18 ^{ABC}	7.73 ^A	16.18 ^{AB}	7.55 ^{AB}	12.46 ^A	12.36 ^{AB}	79.64 ^A
	10 7.82 ^{AB}	17.09 ^A	7.82 ^A	16.09 ^{AB}	7.46 ^{AB}	12.64 ^A	12.55 ^A	81.46 ^A
	15 8.18 ^A	17.27 ^A	7.73 ^A	17.00 ^A	7.55 ^{AB}	12.73 ^A	11.91 ^{AB}	82.36 ^A

* Means in columns followed by the same letter(s) are not significantly different at $P \leq 0.05$.

Sanchez et al. (1989) added nine types of whey protein concentrates (WPC) to wheat flour at 3 substitution levels (5, 10 and 15%), the mixture were manufactured to crackers. They found that WPC with a high denaturation degree and a medium protein content produce the best technological results and the best acceptance of the resulting products.

From the previous results it can be concluded that WPC biscuit was preferred over other whey products fortified biscuits.

5. SUMMARY AND CONCLUSION

The total yield of bread grains in Egypt is not satisfying the needs of the country. The total production of wheat grains cover only about 25% of the total needs. The way to overcome this problem is to find other cereal sources which could be added to wheat flour for bread making.

In Egypt, cost plays a large part in the kind of food consumed and animal protein is beyond the economic means of many people. Hence, it is important to develop protein mixtures that use total local inexpensive sources, such as cereals and legumes. Legumes are considered an important source of different nutrients specially protein and minerals. Baked products (i.e. bread, biscuit, cake, mulleins, cookie, etc.) are consumed on a large scale all over the world. Therefore, fortification of baked products with high protein legume products could provide a good opportunity to improve the nutritional quality of protein consumed by many people.

The present investigation was carried out to produce high-protein bakery products (i.e. pan bread and biscuit). Therefore, the effect of supplementation of wheat flour with different levels of legume flours or their protein concentrates and whey powder or whey protein concentrates on the chemical composition, physical characteristics and sensory properties of the products were studied.

To achieve this purpose, the investigation was aimed to:

1. Prepare legume flours or their protein concentrates (as a source of plant protein) from ungerminated or germinated soybean, field pea and sweet lupine seeds.
2. Determine the chemical composition, functional properties and trypsin inhibitor activities of the legume flours or their protein concentrates as affected by germination process.
3. Determine the chemical composition and functional properties of whey powder and whey protein concentrates as a source of animal protein.
4. Study the effect of fortification of wheat flour with three levels of legume flours or their protein concentrates and whey protein concentrates on rheological properties and wet and dry gluten of the resultant blends.

5. Prepare high protein-pan bread and biscuit by fortification of wheat flour with legume protein concentrates and whey protein concentrates.
6. Evaluate the resultant fortified bakery products from chemical composition and sensory properties points of view.

The obtained results can be summarized as follows:

Part I. Fortification of bakery products with legume protein concentrates

1. Chemical analysis of whole legume seeds showed that sweet lupine seeds contained the highest protein content (43.05) followed by soybean (39.39) and field pea (33.50%). However, soybean seeds had the maximum value of fat and the minimum values of fiber and total carbohydrates. Germination process led to improve the protein content by 19.78, 27.55 and 22.16% for soybean, field pea and lupine flour, respectively. Defatted legume flour had 59.77, 37.64 and 56.18% protein content for soybean, field pea and lupine flours, respectively. However, legume protein concentrates prepared from germinated seeds had higher protein content than those obtained from ungerminated samples. Whole soybean seeds contained high level of trypsin inhibitor activity followed by field pea and lupine. Preparation of legume protein concentrates reduced significantly the level of trypsin inhibitor activity especially for previously germinated samples.
2. The functional properties of the legume products showed that legume protein concentrates absorbed significantly more water than the corresponding legume flours. At the same time, the highest water absorption was obtained for germinated soy protein concentrate. The highest oil absorption was observed for field pea products. Germination process reduced significantly the oil absorption of legume flours and their protein concentrates except for field pea product. The less nitrogen solubility was obtained at pH 4.5 and all legume flours recorded higher nitrogen solubility than protein concentrates at different pH. Germination process increased generally nitrogen solubility of legume flours and their protein concentrates. Legume protein concentrates had significantly higher emulsion capacity than the corresponding samples of legume flours. Emulsion stability of soybean flour and their protein

concentrate was generally higher than those of field pea and lupine products particularly at higher pH value.

3. Rheological characteristics of dough were clearly influenced by the type of legume products and the substitution levels. Water absorption, arrival time, dough development time, mixing tolerance index and degree of softening increased in wheat-legume flour or concentrates blends with some exception. Substitution of wheat flour with legume flour or their protein concentrates led to a decrease in the wet and dry gluten values in the blends.
4. Supplementation of wheat flour with different levels of legume products increased significantly the protein content and ash of the resultant pan bread compared with control. Generally, bread fortified with 15% of germinated soy protein concentrate exhibited the maximal improve in protein content (58.52%). On most cases, legume flours-bread showed higher loaf weight than legume protein concentrates especially at high levels and most of legume fortified-bread showed higher loaf volume than control except for those contained high levels of protein concentrates. The organoleptic properties showed that pan bread containing 10% of different legume products recorded generally higher organoleptic attributes than control (100% wheat flour). However, increasing substitution level to 15% overall acceptability was decreased from 78.21 to 77.56.
5. Supplementation of wheat flour with 15% of germinated soybean, field pea and sweet lupine protein concentrates improved significantly the protein content of the biscuits from 8.03% for control to 14.47, 10.74 and 13.93%, respectively. Test panelists gave the biscuits fortified with either soybean and lupine products superior total score than field pea. No significant difference were obtained between biscuits enriched with either legume flour or their protein concentrate.

Part II. Fortification of bakery products with whey protein concentrates

1. The chemical analysis data of whey products used in this study showed that protein concentrate (WPC) characteristics by high protein content (78%) than sweet whey powder (SW) (12%). On the contrary SW had markedly higher lactose content (70%) than that of WPC (4%).

2. Functional properties of whey products showed that WPC had significantly higher WAC (414.53 g water/100 g sample), OAC (248.3 g oil/100 g sample), emulsification capacity and stability and nitrogen solubility index than SW.
3. Rheological characteristics of dough were clearly influenced by the type of whey products and substitution levels. Addition of different levels of SW decreased water absorption, arrival time and degree of softening and increased dough development time, dough stability and mixing tolerance index of the dough. On the other hand, water absorption, dough stability time were decreased and arrival time, dough development time, mixing tolerance index and degree of softening were increased with increasing the WPC levels in the blends. Wheat flour-blend contained significantly higher wet and dry gluten (27.58 and 10.40%) and lower hydration ratio (165.17) than SW and WPC blends.
4. The protein content of the bread was improved significantly from 12.44% up to 20.50% depending on the amount and the protein content of the added whey products. Fortification of wheat flour with different levels of SW produced bread with low volume compared with control. However, incorporated of WPC gave loaf volume higher than control. The replacement of wheat flour with 5% or 10% WPC generally improved organoleptic attributes of the resultant breads.
5. Addition of 5, 10 and 15% of WPC markedly improved protein contents of the biscuits to 11.89, 13.80 and 15.40% (corresponding to 47.52, 71.22 and 91.07 increase) WPC-biscuits had higher appearance, odor, taste, texture and overall acceptability over the control. At the same time, no significant differences were obtained for color, mouth-feel and crispiness between WPC-biscuits and control.

From the above mentioned results, it can be concluded that fortification of wheat flour with either 10% of soy and lupine protein concentrate or whey protein concentrate produced high protein bakery products (pan bread and biscuit) without reversible effects on their rheological and organoleptic properties.

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الملخص العربي

تدعيم بعض منتجات المخابز بمركزات البروتين

لا تغطي الكمية المنتجة من الحبوب المختلفة كافة الاحتياجات الكلية اللازمة لصناعة الخبز في مصر فالقمح يغطي فقط حوالي 40% من تلك الاحتياجات الكلية وللتغلب على تلك المشكلة توجهت الانظار إلى استخدام بدائل أخرى لتعويض هذا النقص ومن هذه البدائل استخدام البقوليات كمصدر نباتي للبروتين وبروتينات الشرش كمصدر حيواني للبروتين وهذه المصادر منخفضة الثمن ولذا تعد من الطرق الاقتصادية المستخدمة في رفع القيمة الغذائية للمخبوزات وتحسين جودتها.

والهدف الأساسي من هذه الدراسة هو إنتاج منتجات مخابز (خبز، بسكويت) ذات محتوى عالي من البروتين ولذلك تم دراسة تأثير إضافة تركيزات مختلفة من دقيق البقوليات أو بر وتيناتا المركزة وكذلك بروتينات الشرش المركزة على التركيب الكيماوى والخصائص الطبيعية والصفات الحسية للخبز والبسكويت الناتج ولتحقيق ذلك شملت خطة البحث ما يلي:

1. إنتاج الدقيق والبروتين المركز من بذور البقوليات الخام والمنبتة (فول الصويا - بسلة - ترمس حلو).
2. تقدير التركيب الكيماوى والخواص الوظيفية ونشاط مثبط التربسين لمنتجات البقوليات المستخدمة وتأثير عملية الإنبات عليها.
3. تقدير التركيب الكيماوى والخواص الوظيفية للشرش الحلو الجاف ومركزات بروتين الشرش.
4. دراسة تأثير تدعيم دقيق القمح بثلاث تركيزات من دقيق البقوليات أو بروتيناتا المركزة وبروتينات الشرش المركزة على الخواص الريولوجية والمحتوى الرطب والجاف من الجلوتين للخلطات الناتجة.
5. إعداد خبز وبسكويت عالي في البروتين وذلك بتدعيم دقيق القمح بتركيزات مختلفة من بروتينات البقوليات المركزة وبروتينات الشرش المركزة.
6. تقييم الخبز والبسكويت المدعم من ناحية التركيب الكيماوى والخواص الحسية.

ويمكن إيجاز النتائج المتحصل عليها فيما يلي:

الجزء الأول: تدعيم بعض منتجات المخابز ببروتينات البقوليات المركزة (كمصدر نباتي للبروتين)

1. اتضح من التحليل الكيماوى أن البقوليات تحتوى على نسبة عالية من البروتين كانت أعلى نسبة لبذور الترمس 43.05% يليه بذور فول الصويا 39.39% ثم البسلة 33.50%. أدت عملية الإنبات إلى زيادة المحتوى من البروتين وذلك بنسبة 19.78% ، 27.55% ، 22.16%، لدقيق فول الصويا والبسلة والترمس على التوالى واتضح من التحليل الكيماوى لدقيق المنزوع الدهن إحتواءه على نسبة عالية من البروتين 59.77%، 37.64%، 56.18% لدقيق بذور فول الصويا والبسلة والترمس على التوالى. كما أدت عملية الإنبات إلى زيادة نسبة البروتين لمركزات البروتين فكانت أعلى نسبة لبذور فول الصويا 78.66% يليها الترمس 73.94 ثم البسلة 51.20%. بالنسبة لمثبط التربسين كانت أعلى نسبة فى بذور فول الصويا 46.35 وحدة/ملجم يليها البسلة 7.65 وحدة/ملجم ثم الترمس 7.47 وحدة/ملجم وقد أدت عملية الإنبات إلى انخفاض المحتوى من التربسين.

2. بدراسة الخصائص الوظيفية لمنتجات البقوليات وجد أن بروتينات البقوليات المركزة لها قابلية اعلى لإمتصاص الماء عن دقيق البقوليات المقابل وسجلت مركزات الصويا المنبتة أعلى القيم. وبالنسبة لامتصاص الزيت سجلت منتجات البسلة أعلى القيم. وأدت عملية الانبات إلى إنخفاض معنوى فى كمية الزيت الممتص بواسطة كلا من منتجات الصويا والترمس. وسجلت بروتينات البقوليات المركزة سعة إستحلابية معنوية أعلى عن العينات المقابلة

من الدقيق. وكان المستحلب المحضر من منتجات الصويا أكثر ثباتاً مقارنة بالبسلة والتمرسم وخاصة عند القيم المرتفعة لـ pH.

كما أوضحت النتائج لمعامل ذوبان النيتروجين أن أقل ذوبان كان عند pH (4.5) وكانت قيم عينات الدقيق أعلى من عينات البروتين وأدت عملية الإنبات إلى إرتفاع معامل ذوبان النيتروجين لكل من الدقيق والبروتين المركز مقارنة بالعينات الخام.

3. بدراسة منحنيات الفارينو جراف اتضح تأثير الخواص الريولوجية بكل من نوع الإضافة (دقيق، بروتين مركز) ونسبة الإضافة (5، 10، 15%) فقد أدت الإضافة إلى زيادة نسبة امتصاص الماء وزمن الوصول وزمن تكون العجينة ومدى تحمل عملية الخلط ودرجة ضعف العجينة مع وجود بعض الاستثناءات. واتضح انخفاض المحتوى من الجلوتين الرطب والجاف لجميع الخلطات مقارنة بالكنترول (72% دقيق قمح).

4. حدثت زيادة معنوية في كل من البروتين والرماد للخبز المدعم بنسب مختلفة من منتجات البقوليات عن الكنترول (100% دقيق القمح) وسجل الخبز المدعم بـ 15% بروتين الصويا المركزة المنبئة أعلى تحسن وبلغ 58.52% وبصفة عامة زاد وزن الرغيف المدعم بالبروتينات المركزة للبقوليات عن المدعم بالدقيق وأدى التدعيم بنسب مرتفعة من مركبات البروتين إلى حدوث انخفاض في حجم الرغيف الناتج. وأوضحت نتائج التحليل الحسى حصول الخبز المحتوى على 10% من منتجات البقوليات على قيم مرتفعة مقارنة بالكنترول وأدت زيادة نسبة الاستبدال إلى 15% إلى حدوث انخفاض في القابلية العامة للاستهلاك من 78.21% إلى 77.56%.

5. على الجانب الآخر حدث تحسن معنوى فى المحتوى البروتينى للبسكويات المدعم بنسب مختلفة من منتجات البقوليات حيث ارتفعت نسبة البروتين من 8.03 (100% دقيق قمح) إلى 14.47 و 10.74 و 13.93% للبسكويات المدعم بـ 15% من البروتينات المركزة المنبئة لكل من فول الصويا والبسلة والتمرسم على التوالى. وأعطى المحكمين البسكويات المدعم بمنتجات الصويا والتمرسم نتائج أفضل من البسلة ولم يلاحظ وجود اختلاف معنوى فى الخواص الحسية بين البسكويات المدعم بالدقيق والمدعم بالبروتينات المركزة للبقوليات.

الجزء الثانى: تدعيم بعض منتجات المخايز ببروتينات الشرش المركزة (كمصدر حيوانى للبروتين)

1. أظهر التركيب الكيماوى لمنتجات الشرش المستخدمة فى هذه الدراسة ارتفاع نسبة البروتين لبروتينات الشرش المركزة (78%) عن مسحوق الشرش الحلو (12%) بينما انخفض محتواه من اللاكتوز (4%) عن الشرش الحلو (70%).

2. بدراسة الخواص الوظيفية لبروتينات الشرش المركزة اتضح ارتفاع نسبة امتصاص الماء والزيت (14.53 جم ماء/100 جم عينة) و (248.3 جم زيت / 100 جم عينة) على التوالى عن الشرش الحلو ما تميز بارتفاع سعته الاستحلابية 610 جم زيت / جم عينة عند درجة pH 9 عن مسحوق الشرش الحلو (350 جم زيت/جم عينة) وكذلك كانت درجة ثبات المستحلب أفضل مقارنة بالعينات الأخرى. كما رتفعت نسبة البروتينات الذائبة فى الماء عند درجات الحموضة المختلفة مقارنة بالشرش الحلو.

3. تتوقف الخواص الريولوجية للعجائن المدعمة بمنتجات الشرش على نوع المنتج المستخدم ونسبة الاستبدال. اتضح من الدراسة أن إضافة مسحوق الشرش الحلو بنسب مختلفة أدى إلى انخفاض نسبة امتصاص الماء، زمن الامتصاص، درجة الضعف وأدى إلى زيادة مدة تكوين العجينة ومدة الثبات ومعدل الصمود للعجائن بينما عند إضافة مركبات بروتينات الشرش ارتفع كل من زمن الامتصاص، مدة تكوين العجينة، معدل الصمود ودرجة

الضعف وكان لنسبة الإضافة تأثير على ارتفاع هذه الخصائص (كلما زاد التركيز زادت). أدت عملية الاستبدال إلى انخفاض كل من الجلوتين الرطب والجاف بصفة عامة عن الكنترول.

4. أظهرت الدراسة حدوث تحسن معنوي لنسبة البروتين للخبز المدعم حيث ارتفعت نسبة البروتين من 12.44% (للكنترول) إلى 20.5% في الخبز المدعم بـ 15% مركزات بروتينات الشرش. كما تحسن حجم الرغيف عند إضافة مركزات بروتينات الشرش مقارنة بالكنترول. وبصفة عامة فإن العينات المدعمة بـ 5، 10% مركزات بروتينات الشرش حسنت الخواص الحسية للخبز الناتج.

5. أتضح من الدراسة ان تدعيم دقيق القمح بـ 5، 10، 15% من مركزات بروتينات الشرش أدت إلى ارتفاع نسبة البروتين في البسكويت المدعم بصورة ملحوظة إلى 11.89، 13.80، 15.40% على التوالي (وبلغت نسبة الزيادة 47.52، 71.22، 91.07% على التوالي). حصل البسكويت المدعم بـ 15% من بروتينات الشرش المركز على قيم حسية معنوية أعلى من الكنترول من حيث المظهر والرائحة والطعم والقوام ومدى القابلية للاستهلاك.

ومن ذلك يتضح أن تدعيم منتجات المخايز (الخبز، البسكويت) بتركيز 10% سواء من البروتينات المركزة لفول الصويا أو الترمس الحلو أو بروتينات الشرش المركزة أدى إلى تحسن القيمة الغذائية لتلك المنتجات بدون التأثير السلبي على الخواص الريولوجية وتحسين المنتج النهائي.