Evaluation of extruded-expelled low-fat soybean flour in flour blends and the effects on bread and dough development

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Evaluation of extruded-expelled low-fat soybean flour in flour blends and the effects on bread and dough development

by

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A dissertation submitted to the graduate faculty in partial fulfillment of the requirements for the degree of

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2006

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ABSTRACT

The research detailed in this document focuses on the performance of a partially defatted soybean flour called low-fat soybean flour (LSF). This soybean flour type has not received widespread application as a food ingredient. This has resulted from a lack of information available on the performance of LSF in a variety of food systems. Based upon its compositional and functional properties, LSF has shown potential for use in a flour blend. Past studies have focused on the usage of flour blends containing wheat flour partially replaced by an oilseed or legume flour. Wheat-soy flour blends have mostly been studied for use in yeast-leavened bread production. These blends have been able to enhance the protein content of yeast-leavened breads; however, favorable and unfavorable alterations to dough and bread development have been observed. In an effort to determine whether LSF could make favorable contributions to bread systems when used in a flour blend with wheat, several studies were performed in comparison to blends of wheat and defatted soy flour (DSF). For all experiments, wheat flour was replaced by soybean flour up to 12% based on weight and/or equivalent protein.

To understand the protein-water interaction character of LSF in a flour blend, water-holding capacity (WHC) was examined. WHC was evaluated for blends at 2, 4, 6, and 12% replacement of wheat flour based on weight and equivalent protein. Findings showed that wheat-LSF blends had very similar WHC character to wheat-DSF blends. Dough development testing using a 10-g mixograph was performed on flour blends at the replacement levels of 2, 6, and 12%. The parameters of mix (peak) time, rate of development, and shear force during development were quantified. Dough development testing was conducted with and without the use of the dough conditioners, sodium stearoyl-lactylate and calcium stearoyl-2-lactylate. Wheat-LSF blend required longer mixing; however, these mix times were decreased after dough conditioner use.
Effects on the physical and sensory properties of yeast-leavened bread were evaluated after making bread using the specified wheat-soybean flour blends at 2, 6, and 12% replacement of wheat flour. The physical properties loaf weight, loaf volume, crumb firmness, and crust/crumb color were evaluated. Sensory evaluation of bread crumb was accomplished using an 11-member sensory panel. Panelists provided sensory perception data for the perceived intensity of the following bread attributes: bread aroma, nutty aroma, crumb firmness, bread flavor and nutty flavor. Nutty flavor was associated with breads that contained soybean flour. Overall wheat-LSF breads had similar physical and sensory properties to wheat bread that did not contain any soybean flour. Most results from this research have been attributed to the presence of approximately 10% oil in the LSF.
CHAPTER 1. GENERAL INTRODUCTION

Introduction

As a result of the Food and Drug Administration’s approval of a health claim related to soy protein and its reduction in coronary heart disease, soy protein has experienced widespread use as a food ingredient in a variety of edible products in the United States. Prior to the health claim era, soy protein could be found in the form of traditional soy foods such as tofu, tempeh, soy milk, and miso (1). Currently in the United States, soy protein products are consumed in conventional food systems including meat, dairy, breakfast cereal, beverages, infant formula, dairy and meat analogs, and bakery (2).

For years, the incorporation of certain types of soybean flour into pan (white) bread has been practiced. In developing countries such as India, soybean and other legume flours have been utilized as protein supplements in products such as bread. For these breads, wheat flour, the staple ingredient of bread is totally or partially replaced by an oilseed, legume, or other cereal flour to form what are known as flour blends or composite flours. The bread products made using flour blends have enhanced nutritional quality and may aid in resolving the problem of protein-calorie malnutrition experienced in some countries (3).

In some instances, the addition of soy flour has affected the physical and sensory properties of pan bread. Some of the sensory property changes in pan bread have included off flavor that has been characterized as beany-like and a darker colored crust and crumb. Speculated influences on these changes include the amount and type of soybean flour used in the formulation, enzyme activity of the soybean flour, and interactions of soybean flour components with other constituents in the bread formula.

Defatted and full fat are two types of soybean flours that have typically been used in the baked goods industry and research studies on bread made from flour blends. Both types have caused shortcomings that pose challenges to the production of baked goods containing soybean flour that meet the sensory properties that consumers will accept. A third type of
flour made from soybeans, low-fat soybean flour, is now being produced and may have some of the functional characteristics that will allow it to also be considered in the formulation of baked goods.

The low-fat soybean flour in this study was processed with an extrusion-expelling (EE) process that utilizes both expelling and extrusion to remove oil from the oilseed. The soy meal that results from this process is then used to produce soy flour with a residual oil content of 4-12% (4). The oil content varies according to differences in processing factors. Information is limited on the use of low-fat soybean flour as an ingredient in food systems in general and baked products in particular. One study has evaluated its effect on fat absorption during the frying of donuts and water-binding when used in a beef patty (5). Research on the functionality of EE low-fat soybean flour in baked goods in a flour blend with wheat has not been extensively explored.

More studies have been performed on the use of full fat and defatted soy flours in baked goods. In a study on biscuits supplemented with soy flour or kimena, a traditional fermented soy food (India), soy flour-containing biscuits were harder and tougher than those containing kimena (6). Kulkarni et al evaluated the use of soy flour (8.52% oil) for bread fortification (7). Twelve percent (by weight) of the wheat flour in the formula was replaced with medium fat soy flour. Properties of bread indicated higher water absorption and lower volume compared to standard white bread.

The research outlined in this dissertation was conducted to determine the effects of flour blends containing low-fat soybean flour and wheat flour on dough development and bread quality. Wheat flour was substituted up to 12% in flour blends by wheat flour. These blends were then evaluated for water-holding capacity and time required to reach optimum dough development with and without the use of dough conditioners. Bread was prepared using these flour blends to assess the effects of low-fat soy flour blends on physical bread quality using objective and subjective testing.
Dissertation organization

This dissertation contains 3 papers. Each paper constitutes a chapter and will comprise the following sections: abstract, introduction, raw materials and experimental procedures, results and discussion, and references. Papers 1-3 found in chapters 3-5 will be formatted for submission to the Journal of American Oil Chemists Society. Chapter 6 will be a comprehensive conclusion that will unify the findings from all 3 papers. References will be located at the end of each chapter and will follow the work-cited format for the Journal of the American Oil Chemists Society.
CHAPTER 2. LITERATURE REVIEW

Soybeans

The first recorded acknowledgement of the shu or soybean plant dates back to 2838 B.C. in a book written about native plant life in China (8). Described among rice, wheat, barley, and millet, as one of the five sacred grains, soybeans have been utilized in China for centuries as the staple raw material of soy-foods such as soymilk, tofu, soy paste, soy sauce and soy sprouts. Cultivation and use of soybeans spread throughout Asia and to North America, where a variety of soy-foods and products have been developed (9). Soybeans are currently most prized for its crude oil that is processed for edible and non-edible uses.

The fruit of the soybean plant, the pod, contains the soybean seeds. The seed has three main parts; the hull (8%), cotyledons (90%), and germ (2%) (9). The seed parts most important to the processing of soybeans are the cotyledons. The majority of the seed’s oil fraction is found in the cotyledons within oil bodies. The germ and hull contain small amounts of protein and oil.

Extraction methods of soybean oil

The two main foodstuffs derived from soybeans are oil and soybean protein products. In order to isolate the oil of soybeans from the solid material a series of steps can be employed. These steps lead to the successful production of good quality oil and soybean meal. Several mechanisms for oil excision are available and choosing which type for use may depend on several factors, including soybean production, yield, desired process efficiency, desired oil quality, and desired meal quality. The oil removal technologies of hexane extraction, screw pressing, and extruding-expelling will be discussed in some detail.

The majority of soybeans grown in the United States are processed by hexane oil extraction to remove approximately 99% of the oil from soybeans (9). Oil extracted soybean flakes are one of the resultant co-products of this process which is also utilized as a base
material for the manufacture of animal feed and protein products used in animal feedstuffs and human foodstuffs.

Since the oil extracted flakes contain residual hexane, desolventizing is employed to remove the hexane prior to making human foodstuffs. For flakes destined for food consumption applications, a flash desolventization process is often employed. Cooking, drying, and sizing processes are often performed after desolventizing (10).

Edible soybean oil and protein products can be produced from oil extraction methods that do not involve the use of organic solvents, such as hexane or petroleum ether. In the early years of soybean oil production, 77% of soybean oil was extracted by mechanical processes. The solvent extraction method is routinely practiced in the United States; however, mechanical-type processing remains a viable industry in other parts of the world (11).

Screw pressing is one type of mechanical oil removal system that was popularly used before the creation of solvent extraction methods. In conventional screw pressing operations, the beans are cracked, and subjected to dry heating until the temperature of the material reaches 116-132°C and moisture reaches 2-5% (11). This process has relatively low oil removal efficiency, leaving 4-8% residual oil. This mechanical method was largely replaced by the solvent extraction method whose efficiency is 97-99% (2).

As the demand for soybean meal as a feed and foodstuff has heightened, diminishing the levels of soybean anti-nutritional factors in the meal has become important. Anti-nutritional factors are components within foods that have an adverse effect on the nutritional and metabolic well-being of a person that consumes that food (12). The most effective means of eliminating the activity of anti-nutritional factors would prove to be by heat treatment of the meal. This notion may be credited to the lessened use of screw pressing as a means of oil removal for soybeans. In order to achieve the decline in anti-nutritional factor presence, soybean meal must be routed through the screw press process several times. During this time,
excessive heating occurs which compromises the meal’s appearance and flavor characteristics (11).

Another mechanical method of oil removal is via extrusion technology. This process is involved in processing many of today’s fabricated and/or convenience foods (13). This technology is applied through an industrial device called an extruder. Early applications of extruders were mainly for the production of pasta products and ready-to-eat (RTE) breakfast cereals. A wide variety of raw materials can be processed in extruders including defatted soy flour or flakes, soy concentrates, and even flakes of whole soybeans (13). New extruder-based processes are now being used for the power-driven outtake of oil from soybeans. One such type is called extruding-expelling.

The extruding-expelling process of coupling dry extrusion and expelling was developed through the collaborative research effort of a group of researchers at the University of Illinois-Urbana Champaign and Triple F, Inc., an extruder equipment manufacturer. The initial goal for the development of this technology was to establish a new concept of coupling dry extrusion with mechanical expelling of oil from soybeans in order to obtain both oil and cake (meal) suitable for human food. In doing so, a semi-fluid extrudate would be produced by appropriate extrusion process parameters and expelled while still in the fluid state as quickly as possible (11).

Extruding-expelling is favored for several reasons. The process is essentially performed in a single step, eliminates certain capital equipment costs including the purchase of steam dryers and conditioners, enhances oil extraction over simple screw pressing, and eliminates the use of organic solvents (4). Another important benefit of extruding-expelling is the incorporation of temperature conditions that effectively reduce anti-nutritional factors. This reduction is effectively done in a high temperature-short time environment that also allows retention of the meal’s nutritional value (11). The meal produced, extruded-expelled (EE) meal, is described as a low-fat, high-protein, high-energy and desirable starter material.
for animal feedstuffs (4,14). Some studies have explored the functionality of meals produced under varied EE process parameters and the utilization of these meals in such food products as meat patties and doughnuts.

**Soy Protein Products**

Soybeans and soybean co-products are widely used for diverse food, feed, and industrial (non-food) applications. The nutritional composition and functional properties of soybeans have allowed this edible seed to have several uses as animal feed and as a food ingredient. Several food ingredients made from soybeans are available and are collectively termed soy protein products. Soy protein products refer to processed, edible dry soybean products other than animal feed meals (15). The four major types of soy protein products in human food formulations are texturized, isolates, concentrates, and flours. One or more of these products can be found in virtually every type of food system (2). The selection of the appropriate protein product for a given application is based on the desired protein content and/or functional attributes such as solubility, emulsification, viscosity, film formation, etc. The functional attributes mentioned and others are derived from soy proteins. The physicochemical properties of the storage proteins, glycinin and β-conglycinin provide soy protein products with functional ability (16).

Soy protein isolates (SPI) and concentrates (SPC) are highly refined soy protein products; SPI being more refined than SPC. SPC has a protein content of 70% (db). Soluble carbohydrates are usually removed by an aqueous alcohol (60-90%) wash method, acid leaching (~ pH 4.5), or a combination of moist heat denaturation and water extraction. SPC can be found as a key ingredient in meatless products and in meat products as a fat and water binder (2). Alkaline extraction and acid precipitation remove soluble and insoluble carbohydrates from defatted soy flakes during processing of SPI production, which results in a 90% (dry basis; db) soy protein product. SPI has found use in a broad range of food systems including processed meat, meat analogs, soups/sauces, nutritional beverages, infant
formulas and dairy replacements. When soy flakes, soy meal or dehulled whole soybeans are ground so that 97% of the solids pass through a No. 100 mesh sieve, flour is the result (15).

Soy flour is the least refined of the soy protein products because it contains both soluble and insoluble carbohydrate components. (2). These flours typically will have a protein content of up to 55% (db). There are 3 main commercially available soy flour varieties based on fat content, full-fat, defatted, and low-fat. Re-fatted and re-lecithinated flours are also available. Though the term flour is used, none of these varieties have dough forming capabilities like that of wheat flour.

**Defatted Soy Flour**

The aforementioned desolventization process performed on oil extracted soybean flakes can give rise to cooked, toasted, or white flakes, which are distinguished by The Soy Protein Council based on nitrogen solubility index (NSI). NSI is a solubility test that provides an estimation of the potential and limitations of applications of proteins. The index of nitrogen solubility over a range of pH values, is used as a guide to protein functionality (17). White flakes serve as the starting meal for making the majority of soy protein products. When milled, white flakes are termed defatted soybean flour (2).

Defatted soy flour contains about 38% total carbohydrates, including 15% soluble mono- and oligosaccharides, and 13% polysaccharides that are later removed if soy protein concentrates or isolates are made (15). The protein content of defatted soy flour ranges from 52-54% (as is) and 56-59% (moisture free basis) (4). Defatted soybean flour is the most commonly used type of soybean flour. It has been valued by the food industry for its functionality in the areas of water and fat absorption capacity and adhesiveness. (2, 9).

A protein dispersibility index (PDI) value is assigned to indicate degree of solubility potential of the protein product. This value may serve as an indirect measurement of the level of heat treatment by which the product has been exposed. Defatted soybean flour is commonly available in 20 (low), 70 (medium), and 90 (high) PDI forms. A PDI of 90
designates that the particular flour has undergone a low degree of heat treatment and thus should demonstrate high solubility and functional characteristics. Low and medium PDI defatted soy flour varieties are mainly used in the baking industry. For bread specifically, these flours have been able to improve moisture retention during processing (2).

**Extruded-expelled Low-fat Soybean Flour**

Partially defatted soy flour produced from the meal of soybeans that have been extruded-expelled for oil removal is also termed low-fat soybean flour (LSF) (2). Extruding-expelling (EE) is a relatively new process to mechanically displace oil from soybeans (4,11). Low-fat or partially defatted soybean flour result once the extruded-expelled meal has been sieved to the particle size of flour.

Depending on the processing parameters used, partially defatted soybean flour can have a broad range of nutritional and functional characteristics and composition. The protein content of low-fat soybean flour may vary according to oil content. LSF typically has a protein content of 48-50% (2). Due to the nature of the EE process, LSF has a much higher residual oil content than defatted soybean flour. Heywood et al (18) quantified residual oil in LSF and placed them in 3 categories based on oil content. High LSF had a residual oil content of around 10.4%, mid-range LSF had a 7.4% residual oil content and low range LSF had a 6.5% oil content. Protein dispersibility index (PDI) for LSF has been reported to range from 12-69 (4).

The functional properties of low-fat soybean flour have been examined (5,18,19,20). Reports show that the solubility of LSF was minimal at pH 4.0. Emulsification, emulsification activity index, emulsion stability index, foaming capacity, foam stability, fat-binding and water-holding capacities of LSF tended to be influenced by PDI and the presence and level of residual oil (20). Based on these properties of LSF, it is possible to include LSF in a variety of food applications for functional purposes.
LSF has been studied as an ingredient in select food systems. Textured LSF has been examined in meat systems. Protein texturization is a process that imparts structure to a proteinaceous material like LSF. (8). Adding 30% EE processed textured soy protein (TSP) to ground beef patties containing 20% fat did not negatively alter specific quality and sensory attributes. In addition, moisture retention, fat retention, and cooking yields were higher in TSP-containing beef patties compared to an all-beef control (5).

In doughnuts, LSF was shown to have an effect on fat absorption during frying. Study results showed that doughnuts containing LSF with low and mid residual oil contents reduced fat absorption as the amount of soy flour increased from 5% to 8% (18). These findings were similar to defatted soybean flour. LSF containing doughnuts also had consistent hardness to that of the control (5). There are few reports in the literature on the use of LSF in bread. In one study, medium fat soy flour (7-9% oil) processed by EE technology was evaluated for use in bread fortification. Baking tests identified breads with added medium fat soy flour to have higher water absorption and lower bread volume compared to standard white bread (7).

**Wheat Flour**

Wheat flour is derived from the cereal grain, wheat. The food staple is best defined by its standard of identity set forth in the Federal Food Drug and Cosmetic Act as “flour, white flour, wheat flour, plain flour, as the food prepared by grinding and bolting cleaned wheat kernels (21,22). Wheat maintains preeminence as a food cereal due to its adaptation to soil and climatic conditions common to large temperate regions of the world, high-yield performance and ease of cultivation (21).

Wheat flour is milled from the wheat grain, also termed the wheat fruit. During milling, the 3 main parts of the wheat grain, germ, endosperm, and bran, are divided. The aim of flour milling is twofold: (a) to separate and remove as completely as possible the bran covering and germ from the endosperm and (b) to obtain the maximum extraction of the
endosperm from the wheat fruit and to achieve this without excessive damage to the starch granules (21).

The milling process involves several steps. During cleaning, a wide range of impurities typically found in wheat are removed according to size, specific gravity, shape, magnetism and air resistance (23). Conditioning is usually the next step and is a critical stage in the milling process. Cleaned wheat grains are allowed to rest in a predetermined level of tempered water for 24 hours. This step hydrates the bran making it easier to remove from the endosperm, thus allowing greater milling efficiency. Wheat grain is then ready for milling into flour. After successive grinding and separation stages, various streams of flour are produced. These streams are blended in various ratios to produce different commercial flour grades (21).

Much of the nutrient composition of flour is dependent on the wheat variety, its prior growing conditions and the streams of flour blended for its production. Protein content typically ranges form 9-13%. Lipids tend to be present at a level of \( \leq 2\% \). Minerals total 1.6-1.8%, while vitamins are mostly of the B-complex and vitamin E (21). Carbohydrates are the constituent of greatest quantity. Flour is mostly composed of the endosperm portion of the wheat grain. Starch containing cells of the endosperm make up 85\% of the whole kernel and starch makes up 65\% of ordinary flour (14\% moisture basis) (23).

Although wheat flour contains a fair amount of nutrients, many commercial flour and bread products are further enhanced via nutrient enrichment. As previously mentioned, the milling process separates the germ and bran portions from the endosperm as efficiently as possible. The germ and bran parts contain much of the wheat kernel’s mineral and protein fractions. The bran fraction contains 60\% of the ash and 90\% of the protein. The germ contains relatively high levels of protein, ash, and lipids (21).

Wheat flour provides functional and nutritive components to bread. Vitamins, proteins, edible fat and carbohydrates are supplied to bread via the endogenous components
of wheat flour and other ingredients. During milling of wheat flour some nutrient components are lessened and/or depleted. This aspect of wheat flour prompted the institution of flour enrichment mandates defined in the Code of Federal Regulations by the Food and Drug Administration. Based on the standard identity of enriched (wheat) flour, folic acid, niacin, riboflavin, iron, and thiamin must be added (21,22,24). From a functional standpoint, wheat flour contains proteins that are important to its performance in food systems. The proteins gliadin and glutenin are responsible for the gluten-forming properties of wheat flour. Formation of gluten gives rise to dough and bread.

**Dough (Gluten) Development**

Dough is viewed as a complex visco-elastic system. At the molecular level, dough is a three-dimensional network made up of protein chains that are linked together by various types of chemical bonds and linkages (28). This three-dimensional network is unique to formation by the storage proteins of wheat and to a lesser degree, rye.

Gliadin and glutenin are the main wheat proteins that together form gluten. Gliadins are storage proteins that give dough its viscous properties. Glutenin together with gliadin are found in discrete protein bodies in grain and flour. These proteins provide dough with its characteristic elastic properties (46). Gluten properties are often influenced by the wheat flour from which it is produced. The strength of the developed dough depends initially on the variety of wheat. Wheat flour can be processed from hard or soft wheat varieties. Hard wheat, having a higher protein content, forms dough that has stronger rheological characteristics than dough made with soft wheat. Gluten quality is also affected by ingredients within the bread formula and soluble and insoluble flour components. Wheat flour components that have a role are starch, storage (gluten) proteins, pentosans (non-starch polysaccharides), lipids, water soluble proteins, and inorganic compounds (ash).

The most accepted explanation of dough development relates to the sulfhydryl-disulfide interchange. The thiol or sulfhydryl group (-SH) of the amino acid, cysteine, is able
to form disulfide bonds (-SS) with other thiol groups along the same or adjacent polypeptide chains when oxidized. These bonds establish disulfide cross-links that produce a rigid network of protein chains. Sulfhydryl groups are allowed to contact when wheat flour is hydrated. The water disperses the proteins (location of –SH groups) and subsequent mixing manipulates these groups into closer proximity. An example of this phenomenon is shown in Figure 2.1. This network forms the gluten (dough) matrix. The rigidity of this matrix reduces the tendency of the dough to flow and increases gas retention capacity (21). As dough is mixed, disulfide cross-links are broken and reformed at other locations on the polypeptide chain. This ebb and flow of cross-linking imparts mobility or viscous flow to the dough (21).

![Figure 2.1. Line drawing of intramolecular (within a polypeptide chain) and intermolecular (between polypeptide chains) disulfide bridges (crosslinks) formed during sulfhydryl-disulfide interchange (47).](image)

**Mixograph Testing**

The mixing of wheat flour, water and other ingredients into a dough can be characterized by 3 stages. The first brings dough ingredients into intimate contact and disperses them into a uniform mass. In the second stage, dough consistency reaches a peak
value. In the third and last stage, dough becomes fully developed. At full development, thin sheets of gluten are present (48). Collectively, all these stages characterize dough rheology.

Dough rheology is a term that describes the responses of dough to shear and extensional stresses. Rheological characteristics of dough are established by molecular and physical activities that occur during mixing and serve to predict how a given dough will behave as it is being processed into a finished product. Rheology of dough also influences the quality of the finished product. Scientists have developed sensitive and precise dough testing equipment that allow for objective measurement of dough rheology. These test apparatuses are either torque or viscosity measuring instruments. Results from these instruments can be correlated with results from baking tests. These correlations can provide useful conclusions for baking behavior of a flour under investigation, such as flour blends (23).

The mixograph (AACC Method 54-40) is a torque-measuring instrument developed by Swanson and Working in 1933 that can relay the mixing requirements of a given flour (28,49). This instrument is described as a recording dough mixer since it essentially is a small-scale mixer with a recorder attachment. Mixographs are easy to use, relatively inexpensive, and require less space than other recorder mixers (49).

Recordings can be performed in a 35 g or 2 g mixing bowl, although a 10 g bowl is used most often. The mixing action occurs in the bowl. Two pairs of thin pins (4 total attached to the mixer head) rotate in a planetary motion to continually mix flour and water samples. Three pins located in the bowl oppose the action of the moving pins (49).

As gluten develops, dough consistency increases. This gradual increase causes a greater amount of force to be required to push the revolving pins through the dough (28). This force is then recorded by the mixograph via computer software or the recorder attachment. The bowl rests on a horizontal plate or lever system that is attached to a pen. As the pins manipulate the developing dough, a twisting motion of the horizontal plate ensues. The plate’s motion causes the attached pen to illustrate a mixograph curve or mixogram on
chart paper moving at constant speed (28). From the mixogram, rate of dough development, optimum development time (point of minimum mobility; curve peak), maximum resistance of the dough to mixing, dough strength and the duration of resistance to mechanical over-mixing can be determined. These and other parameters are calculated when specialized computer software is used to produce and analyze the mixograph curve.

The mixograph can be used to study effects of added ingredients on mixing properties, dough rheology, blends and quality control and evaluation of various types of wheat (50). Some studies have utilized the mixograph to characterize dough development in flour blends (34,35,43,51). For bran addition to wheat flour, dough development time increased from 1.5 to 15 min with addition of 40% bran (51). Fleming and Sosulski found that wheat-soy blends required the longest time (4 min) to reach mixograph peak height while faba-bean and field pea reached maximum peak in the shortest time (2.5 min) (35). The mixing times of different blended flours revealed that a control dough, made of whole wheat flour had the highest mixing time, which decreased upon blending with barley and soy flour (full-fat and defatted) (34).

**Overview of Bread**

Bread is a part of the food category, baked goods. Among the various baked products, bread by far is the most popular and is produced in many shapes, sizes, textures and palates (tastes) (25). Bread can be considered a staple foodstuff that is consumed throughout the world (23). Wheat-based breads provide sources of protein, complex carbohydrates (mainly starch), fiber, vitamins and minerals (23). Bread is a product made with flour in combination with various amounts of liquid (water or milk), sugar, edible fat, and salt. Others ingredients, such as eggs are also included in bread. The mixture of these ingredients can produce several kinds of breads products, depending on the ratio of ingredients and leavening agent employed.
The flour:water ratio in a bread is the most important factor in determining whether gluten or water is the continuous phase (26). When water is the continuous phase, a batter is the result, while the continuous phase of dough is gluten. Batters have varying degrees of fluidity while a dough tends to be stiff. Leavening agents are typically of chemical or biological origin and are used to make dough or batter rise or increase in size (27). This rise or increase in size is the result of gas production catalyzed by a leavening agent and its reactants.

To produce bread, the aforementioned ingredients can be processed by one of two popular bread-making methods. These are the sponge-and-dough and straight dough methods. Although new bread-making technologies have been developed, the sponge-and-dough and the straight-dough methods have remained the most used methods. In the sponge-and-dough method, a sponge (dough) is allowed to develop from 50 to 70% of the total dough flour (28). The sponge is then mixed with the remaining dough flour for further processing.

As the name, straight-dough implies, bread ingredients are combined in a single-step to produce one batch of dough using the straight-dough method. The batch undergoes fermentation for 2 to 4 hr (28). Subsequent processing is similar to the sponge-and-dough method. The advantages of this method over the sponge-and-dough method are lower requirements in processing time, labor, power, and equipment and reduced fermentation losses because shorter fermentation time is employed (28). A proposed advantage of the straight-dough method has been related effects on bread flavor. Although not widely accepted, this method has been thought to enhance bread flavor because all ingredients experience the same fermentation treatment (28). Overall, bread produced by the straight-dough method has good quality and acceptability.

Bread quality is often used to provide reliable information about flour performance in bread production. For consumer acceptability purposes, the determination of optimum quality
bread is highly subjective and influenced by the person eating the bread. To minimize variability in quality interpretation, bread quality observations are made based on three categories: external, internal, and texture/eating (flavor) properties. These properties of finished (test) bread are usually compared to a bread standard for objective quality determination. The factors, volume, appearance, and crust color and formation are external parameters that are observed during quality measurement.

Loaf volume (cubic centimeters; cc) is defined as the amount of space a loaf occupies (28). This parameter is often measured by a seed displacement method. The loaf is placed in the lower compartment of a calibrated rapeseed (or poppy seed) displacement apparatus in which the compartment volume is known. After the seeds are released into the container, the amount of seed displaced equals the loaf volume.

The color of bread crust can be measured by objective means using standard color charts such as the Munsell system or a tristimulus-type instrument (2,13,23) like a HunterLab spectrophotometer. Crust color can range in the intensity of golden brown hue on the top of the bread. The internal bread character can be interpreted by the crumb grain and color. Properties of the bread crumb, such as color can be measured objectively and subjectively using the same analytical tools as with the crust color. Crumb structure is a combination of the sizes, number and distribution of cells within the crumb grain (23). These properties are often assessed by visual aids, such as photographs.

Objective measurements of texture are often carried out using a texture analyzer instrument. The texture of the bread crumb is often characterized by the terms firmness and softness. These terms are also often associated with eating quality. Compression tests are mainly used to quantify firmness and softness. Methods for the tests have been compiled by the American Association of Cereal Chemists (St. Paul, MN) and the American Institute of Baking (Manhattan, KS). For compression testing, the force required to deform the bread
crumb is measured. The applied force is insufficient to rupture the crumb, which usually returns to its uncompressed state once the force is removed (23). Eating quality may be best described by using sensory evaluation.

Sensory evaluation techniques are utilized to measure human responses to foods (29). Texture, eating quality, and flavor are often evaluated by sensory techniques. Sensory panelists are instructed to score these bread attributes according to the descriptive terms provided by the investigator. With regard to the eating quality of the bread crumb, most consumers prefer a soft, resilient, and short (moist) crumb character; these are characteristics that most consumers associate with bread freshness (28). Bread flavor is experienced through taste. Fresh bread typically has a slightly sweet-sour-taste, with a very light salty note, and a barely perceptible bitter element derived from the crust (28). The science of bread flavor is complex. Since crumb and crust both contribute to bread flavor, it is best to assess these parts independently (23).

Aroma cannot be overlooked as a contributor to quality as it affects consumer appeal for a given bread product. Pyler suggests conducting bread aroma testing by holding the loaf close to the nose and squeezing air out of the loaf as smelling takes place. The United States consumer typically prefers a bread aroma that incorporates wheaty, nutty, malty, and sweet diacetyl notes (28).

Dough is used to make a variety of bread products. When dough is processed using yeast, a biological leavening agent, it is termed yeast-leavened bread. There are four essential ingredients required for yeast-leavened breads; wheat flour, water, yeast, and salt. Typical yeast-leavened bread formulas may also contain, sugar, edible fat, milk, dough conditioners, and preservatives. An example of a standard white bread formula is provided in Table 2.1.
### TABLE 2.1

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Weight (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flour</td>
<td>100.0</td>
</tr>
<tr>
<td>(14% moisture basis)</td>
<td>Variable*</td>
</tr>
<tr>
<td>Water</td>
<td>Variable*</td>
</tr>
<tr>
<td>Yeast</td>
<td>8.6</td>
</tr>
<tr>
<td>Sugar</td>
<td>2.0</td>
</tr>
<tr>
<td>Salt</td>
<td>1.5</td>
</tr>
<tr>
<td>Shortening</td>
<td>3.0</td>
</tr>
</tbody>
</table>

*Water amount is variable according to % flour absorption, % protein content, and % moisture content.

Yeast-leavened bread experiences leavening action via the biological leavening agent, *Saccharomyces cerevisiae*. Cells of *Saccharomyces cerevisiae* are the most prevalently used yeast type for leavening. These microorganisms produce carbon dioxide during the metabolism of sugars, such as glucose. Carbon dioxide gas that is entrapped by the dough is produced according to the following main chemical reaction.

\[
C_6H_{12}O_6 \rightarrow 2C_2H_5OH + 2CO_2
\]

Glucose Ethyl alcohol Carbon Dioxide

Yeast cells are also able to convert disaccharides such as sucrose into its monomers of glucose and fructose for further metabolism into carbon dioxide gas and ethyl alcohol because of the enzyme, invertase (sucrase) (27).

As the dough expands during fermentation and baking due to the trapping of more carbon dioxide gas, a rising effect in the dough can be observed. In addition to the benefits of gas production by yeast, some contributions to the rheological properties of dough and flavor of the finished bread product occur. The intensity of the bread flavor depends on the bread-making process (26). The role of yeast in the rheology of dough is similar to that of added chemicals known to have similar effects (25). The action of yeast is described as having a softening or mellowing of dough (gluten) (28).
The Bread-making Process

Several steps are involved in the production of bread. Mixing and kneading manipulate the bread ingredients until a smooth, homogenous, visco-elastic semi-solid is formed. Kneading is the development of the dough (gluten) structure by “work done” after the initial mixing (23). Prior to the invention of electric powered mixers that mix and knead, these steps were performed by hand. Mixing also causes the incorporation of air bubbles into the developing dough. These air bubbles serve as nuclei for the accumulation of carbon dioxide as the dough ferments and for gasses evolved and expanded as the dough bakes (27).

Dividing is a small step within the bread-making process, yet no less important than mixing. In order for the dough to generate shape and size, it is divided into portions in preparation for fermentation. Fermentation is described as a series of complex, interlinked reactions. It is during this time that yeast acts upon carbohydrates and converts them to carbon dioxide and alcohol (28). Fermentation is best conducted under controlled temperature and humidity to allow for optimum dough fermentation. Temperature during fermentation is generally held at 25°C - 30°C (77°F - 86°F) and the humidity kept at 80-85%. In a commercial-scale facility, dough fermentation is usually performed in a special room or compartment in which these parameters can be maintained under tight control. Duration of fermentation is variable according to how long it takes the divided dough, now termed sponge, to attain the degree of maturity needed for optimum quality characteristics (volume and crumb) of the end product (28). At that point the expanded dough is ready for punching or proofing.

In the straight dough method of bread-making, all ingredients are mixed into a dough ball. The dough ball receives periodic turning or punching that releases the carbon dioxide gas. This action reduces the dough volume (28). Punching keeps the film of gluten around formed gas cells from being overstretched and subdivides the gas cells that have enlarged
during fermentation. The subdividing of gas cells increases the distribution of carbon dioxide in the dough which causes the bread crumb to have a more even grain (27).

This step must be performed with caution to avoid collapse of the sponge. Punching is performed for other reasons as, redistribution of sugars in the vicinity of yeast, dispersion of yeast to other parts of the sponge, and equalization the sponge’s temperature by extraction of heat (27). During fermentation, some dough’s are punched more than once and allowed to ferment thereafter before the sponge is molded into a loaf and proofed. Proving or proofing occurs during the last phase of fermentation. It involves the relaxation of the loaf-shaped dough prior to baking. The shaped loaf is allowed to double in size once again before being placed in the oven for baking.

Baking is the last stage in bread-making. The process is carefully controlled in order to influence the final quality of bread. Baking essentially transforms dough with the aid of heat into a light, porous, readily digestible, and flavorful product. The entire process usually lasts for 22-23 minutes. During this time frame, several changes occur to the external and internal of character of the raw dough.

Entrance of the panned, loaf-shaped dough into the oven (heated environment) initiates a temporary rise in yeast activity and carbon dioxide production. This occurs because of a rise in dough temperature and results in an increase in bread volume by about 33% (28). This action of increased volume is termed oven spring. Externally, a skin is slowly developed on the dough’s surface. This skin will become the bread crust. At about 122°F to 140°F (50°C-60°C), this skin thickens, loses elasticity, and begins to become brown in color. An understanding of crust formation was explored by Jefferson et al., who determined that crust thickness is particularly sensitive to oven temperature and dough vapor pressure, but relatively insensitive to pre-oven dough conditions and dry crust properties (30). It is also at this point in the baking process that most yeast activity has ceased and thermal death begins to occur. This happens as various parts of the dough reaches 140°F (60°C).
The key structural properties and quality of the crumb are dictated by processing conditions prior to baking. Baking establishes the tactile sensory perceptions of this structure and quality. Throughout baking, the dough interior is gradually transformed into a crumb structure. Bread crumb consists of a protein-starch-lipid matrix that encloses in a honeycomb network, minute gas cells that make up most of the loaf volume. Crumb structure development occurs as the interior temperature increases to a maximum temperature of 210°F (99°C) (28). During this time, starch granules swell and gelatinize. Meanwhile, dough proteins undergo heat denaturation that converts the dough into a semi-rigid structure by interaction with the swollen starch granules. At the end of baking, the crumb crust becomes a stabilized arrangement of gelatinized strings of starch granules colloidally dispersed in a denatured protein matrix. At the termination of baking, breads are de-panned and allowed to cool.

**Use of Soy Flour in Bread**

The discovery of soy's health benefits and recent fervor for high-protein diets has driven the application of soy flour in various food systems (2). Soy provides more than just nutritional benefits. In general, economical and functional reasons for soy protein product use in bakery products have been noted.

Defatted soy flour is the most widely used soy ingredient in bread. The flour has aided in the replacement of the high cost ingredient, non-fat dry milk (NFDM). In breads, buns, rolls, cakes, and pancakes, soy flour improves moisture retention (2). At 1-3% usage, defatted soy flour has been observed to improve crumb body and resiliency, enhance crust color, and improve toasting characteristics. Shelf-life is also increased as a result of heightened moisture retention caused by the soy flour (31).

Porter and Skarra (32) reported that soy flours with different protein dispersibility index values (PDI) increased dough absorption when included in a sandwich bread formula. The additional water absorbed and inclusion of defatted soy flour rendered a 1% savings on
bread production which equated to a potential savings greater than $7.5 million (32). Full fat soy flour has been utilized in bread formulations with some success. In comparison, breads formulated with extrusion cooked full fat soy flour had no differences in baking absorption, loaf volume, texture, crust color or odor and flavor to bread containing NFDM.

Research cited in the literature provides insight and information concerning the effect of select soy flours when added or used to partially replace wheat flour in a bread formulation. Soy flour, when used at conservative levels (5%-10% based on weight) has some negative effect on bread loaf volume. Many of the effects of soy flour on bread development when used at higher levels are thought to be overcome by adjustments to formula water and fermentation time (33).

**Use of Flour Blends in Bread Products**

Research efforts on protein enhanced foods, and more specifically bread, has provided important information to assist in finding ways to buffer the protein deficiency problems experienced by populations in developing countries. This human nutrition concern has lead to the inclusion of soybean flour and flours from other oilseed, legume and cereals as a means of increasing nutritive quality of bread. In developing countries, the occurrence of protein deficiencies among human populations for a time had become a common problem. This detrimental problem with regard to human welfare sparked a quest by researchers to find ways to enhance the availability of protein rich foods in these countries. India was one of these countries that did not have a significant availability of animal-based protein sources (high quality protein). Populations within this country utilized proteins of plant origin to meet most nutritional needs.

The goal of researchers was to increase the protein content of commonly consumed foods in countries with such problems (3,6,34,35,36). During this time, one of the key means for protein enhancement of a plant-based food was performed through the use of flour blends. Flour blends or composite flours combine cereal, oilseed, and legume flours as a
mixture in which one flour is totally or partially replaced by the other. This is performed in a ratio that is usually conducive to the target food’s nutritional and functional performance. Many studies have examined the physicochemical properties and nutritional profile of flour blends that contain wheat and other plant-based protein products. Wheat is often the flour in greatest quantity with the non-wheat protein product replacing wheat in some cases up to 40% (37). Food products such as breads, cookies, and texturized products the food types most often used in these studies (6,38,39).

There have been numerous studies investigating a variety of high protein flours to replace wheat. Rao and Rao (40) discovered that wheat-sorghum flour blends had an increase in ash content with an increase in the level of sorghum. This same trend was not observed for protein content. Having high lysine content, made cowpeas a potential enhancer of protein quality when combined with cereal grain products. Much like the previous study, flour blends of wheat and cowpea flour had higher ash and protein contents and crust/crumb color (41). The color increases were attributed to more non-enzymatic browning (Maillard) reactions taking place during baking of the wheat-cowpea loaves.

Other non-wheat protein products that have been evaluated in flour blends with wheat for bread production include, triticale flour (42), faba-bean and field-pea concentrate (35), cottonseed flour (43), barley flour (3), lupin (44), fenugreek legume (36) and pinto and navy bean protein fractions (45).

**Dough Conditioners**

Dough conditioners are used as processing aids in bread production to ensure finished product quality. Dough conditioners are surfactants or emulsifiers whose role is to complex with the protein and starch portions of the dough, to strengthen the extensile gluten-starch film, and to delay the setting of the dough during baking (21). These functions of the dough conditioners increase loaf volume and tolerance to mixing and handling can also be observed (52).
Two such dough conditioners are sodium-stearoyl-lactylate (SSL) and calcium-2-lactylate (CSL). Both belong to the acyl lactylate group and are widely used in the baking industry. As acyl lactylates, SSL and CSL are reaction products of stearic acid and lactic acid neutralized to sodium and calcium salts, respectively (21). These conditioners are usually distributed as free-flowing powders that are insoluble in water and readily soluble in non-polar solvents. SSL and CSL possess high complexing reactivity with gluten proteins and the amylase fraction of starch (21). Maximum usage level based on flour weight is 0.5% according to the code of federal regulations Title 21CFR136.110 (24).

D’Appolonia noted that the production of bread with composite flours or incorporation of protein additives in general requires the use of chemicals such as SSL. In a study on bread production using pinto and navy bean flours, incorporation of SSL increased dough strength and stability which indicated an increase in mixing tolerance (53). Fleming et al added CSL and SSL to wheat-gluten-soy bread doughs to improve the dough and loaf characteristics of the high protein breads. The acyl lactylates were markedly superior to the other dough conditioners as improvers of the soy-supplemented bread. The characteristics of loaves treated with SSL were superior to those with CSL (54).

Water-holding Capacity Testing

Protein-water interactions of soybean proteins relates to many key functional properties demonstrated by soy protein products. Among these is water-holding capacity which is sometimes referred to interchangeably with water retention capacity (55). Water-holding capacity (WHC) is an index of the amount of endogenous and/or added water retained within a protein matrix against external forces (55) (56). The external force often applied is centrifugal force. WHC relates to the protein-water interactions established when wheat flour is manipulated in the presence of water.

Retention of water by protein materials in foods is due to interactions of water with macromolecules and other solutes, and different structures of the material (55). A protein is a
macromolecule that will have an affinity for water according to its amino acid composition. Ionic groups along a polypeptide chain will be more hydrated than non-ionic sites, with an average of 4-8 water molecules bound per ionic group. Amide groups bind one molecule of water and exposed polar atoms (O or N) bind one and six water molecules, respectively. The ability of soy flour and other soy protein products to hold water equates to water related benefits in dough and bread development. Hafner suggested that negative effects to soy-containing, wheat bread quality may be overcome when additional water is incorporated into the bread formulation (33). Research in the literature reports the water holding capacities of 2.6, 2.75, and 6.25 g/g of solids for soy flour, concentrate, and isolate, respectively (8,57). The investigation by Lin et al of WHC revealed an increase in water-holding with increased protein content of sunflower meal products (58). WHC data tends to vary according to the method of testing utilized. An attempt has been made to develop a test method that accurately measures WHC for a variety of protein materials (59). The protein fraction of wheat flour responsible for holding water is glutenin.

REFERENCES


CHAPTER 3. EVALUATION OF WATER- HOLDING CAPACITY FOR WHEAT - SOAP FLOUR BLENDS

A paper accepted for publication in The Journal of the American Oil Chemists Society Toshiba L. Traynham, Deland J. Myers, Alicia L. Carriquiry, and Lawrence A. Johnson

ABSTRACT: The water-holding capacities (WHC) of wheat flour when partially replaced with defatted soybean flour (DSF) or low-fat soybean flour (LSF) were evaluated. Wheat flour was replaced at 2, 4, 6, 8, 10, and 12% levels with DSF or LSF based on sample weight and/or soybean flour protein content. WHC (g water/g flour) was quantified after centrifuging hydrated samples at 1592 × g (3000 rpm) and/or 4424 × g (5000 rpm) for 30 min. Results showed that at both centrifuge speeds, all wheat-soybean flour blends had WHC greater than wheat flour with the exception of 2% blends based on weight. Wheat-soybean flour blends had lower WHC at 5000 rpm than at 3000 rpm. In general, WHC increased as the proportion of soybean flour increased. Differences in WHC were greatest between samples containing 2% and 12% soybean flour. WHC values among 6, 8 and 10% samples were not significantly different for both wheat-DSF and wheat-LSF blends. Blends containing LSF were observed to have comparable WHC to wheat-DSF blends.

KEY WORDS: flour blends, low-fat soybean flour, water-holding capacity, wheat flour

INTRODUCTION

Flour blends are mixtures of cereal, root, or oilseed flours. The most commonly studied flour blends are made by partially replacing wheat flour with non-wheat flour. This practice arose from a need to increase the nutritional quality of wheat products, such as bread, that are consumed in developing countries. Soybean flour has been identified as a suitable

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complement to wheat flour for blends used in cereal based products (1, 2) based on protein quality (3, 4). This type of flour can compensate for the lysine deficiency of wheat flour. Wheat flour contains more sulfur containing amino acids and is able to supplement the low amount present in soybean flour.

Soy protein products, such as soybean flour, are now staple ingredients in almost all food systems (5). The prevalence of these protein products has been seen in bakery, meat, beverage, meat analog, and dairy items. The successful use of soy protein products in such diverse food applications can be attributed to the functional properties displayed by soy proteins. These proteins are known for such functional capabilities as fat binding, emulsification, gelling, foaming, viscosity, and water holding.

Water-holding capacity (WHC) is an important protein-water interaction that occurs in various food systems. WHC is the ability of a protein matrix to absorb and retain bound, hydrodynamic, capillary, and physically entrapped water against gravity (6). Studies have compared the WHC of soy protein products to other protein ingredients such as egg white solids and nonfat dry milk and found that soy protein products are able to hold up to approximately 3 times more water than these ingredients (7).

Low-fat soybean flour is partially defatted soy flour that varies in protein dispersibility index (PDI) and residual oil content. The low-fat soybean flour of interest in this study is produced from soybeans that have been processed by an extruded-expelled (EE) method developed in 1987 by Nelson et al (8). During the application of the EE method, soybeans encounter heat and pressure before undergoing the mechanical excision of oil. Partially defatted soybean flour is derived from EE processed soybean meal and reportedly contains 4.5-13% residual oil depending upon processing parameters (9). EE processing is favored for its non-use of hazardous solvents, feasibility for processing small quantities of soybeans, and low capital investment costs (8). The meal produced from the EE method is
reported to have higher digestible energy and amino acid availability in comparison to defatted soybean meal (9).

As a potential new food ingredient, it is customary to identify an ingredient’s functional performance in food systems. Previous studies have examined the utilization of full fat and defatted soybean flours in flour blends and the effect on bread properties (10, 11). Research is sparse on the functionality of EE low-fat soybean flour in flour blends. Noting the importance of protein-water interactions in bread systems, the focus of this research is to understand the water-holding characteristics of EE low-fat soybean flour in a flour blend with wheat flour. These results will provide information for future studies on flour blends containing EE low-fat soybean flour and its use in other food systems.

EXPERIMENTAL PROCEDURES

Materials. Wheat flour (WHT) was obtained from Horizon Milling (Wayzata, MN); defatted soybean flour (DSF) and low-fat soybean flour (LSF) were obtained from Cenex Harvest States Co. (Mankato, MN) and Insta-Pro International (Des Moines, IA) respectively.

Proximate analysis. Crude protein (AOAC 990.03) (12), moisture (AOAC 925.10) (12), fat (AOAC 922.06) (12), and ash (AOAC 925.25) (12) content were performed in triplicate for proximate analysis of the individual flours. The percentage of carbohydrate content was determined by difference. Protein dispersibility index (PDI) was performed using the fast stir method (AOCS Ba10-65) (13) by Woodsen-Tennent, Des Moines, IA.

Solvent retention capacity (SRC). SRC was determined using method AACC 56-11 (14) to quantify potential contributions to WHC by other flour components having water uptake capabilities. The water-based solvents used were a sucrose solution (50%; w/w), sodium bicarbonate solution (5%; w/w), and a lactic acid solution (5% w/w). Twenty-five milliliters of prepared solvent were added to 5 g of flour (14% mb) in 30 mL centrifuge tubes. Centrifugation at 1239 × g (3000 rpm) was performed for 15 minutes. After
decanting, the remaining gels were weighed and SRC value (%) calculated as 
\[ \%SRC = \left[ \frac{(gel \text{ wt}/flour \text{ wt}) \times \left(\frac{86}{100 - \% \text{ flour moisture}}\right) - 1}{100} \right] \times 100 \]
for each water-based solvent.

**Water-holding capacity (WHC).** WHC was determined using methods modified from Heywood *et al.* (15) and Lin and Zayas (16). Wheat-soybean flour blends were formulated for evaluation by replacing wheat flour with LSF or DSF at 2, 4, 6, 8, 10, or 12%. Replacement of wheat flour was performed in two ways: 1) wheat flour was replaced based on the sample weight, and 2) because LSF and DSF had differing protein contents, replacements were made based on protein content to equalize the amount of protein contributing to WHC. Fifteen grams of total flour was dispersed in 285 mL of distilled water in a 500 mL centrifuge bottle. Bottles were agitated for 10 minutes then centrifuged at either 1592 \( \times \) g or 4424 \( \times \) g (3000 rpm and 5000 rpm, respectively) for 30 min. After decanting the supernatant, each bottle was weighed and WHC \((\text{g of water/g flour})\) was calculated as:

\[ \text{WHC} = \frac{[\text{wt of bottle after decanting} - \text{wt of dry bottle}] - \text{total flour wt (g)}}{\text{total flour wt (g)}} \]

**Statistical Analysis.** Water-holding capacity testing followed a complete randomized design. Data from 4 replicates were subjected to ANOVA using the general linear model (GLM) procedure in the Statistical Analysis Software Program version 9.1 (SAS Institute, Inc., Cary, NC). Tukey’s test was used for multiple comparisons. Differences for the interaction of flour and percentage of soy flour were deemed significant at the \( p < 0.05 \) level.

**RESULTS AND DISCUSSION**

**Proximate analysis.** Table 3.1 shows the proximate composition for the flours tested. DSF and LSF contained substantially higher protein contents than did wheat flour. Wheat flour production involves milling, which frees the endosperm from the bran and germ portions of the wheat kernel. Wheat typically contains about 9-13% protein and most wheat flours have a
protein content of approximately 10-13%. These values tend to fluctuate based on variety, environmental conditions during growth and flour type (17).

Variability in the composition of soy flour has been attributed to the variety and composition of the original soybean, geographic environmental growing conditions, and processing methods applied (17). Soy flours contain protein in the range of 40 to 54% (10). Comparing LSF and DSF, the protein content differences were most likely influenced by the processing method used to remove the oil from soybeans prior to flour production and/or soybean growing conditions and variety. Oil content is directly influenced by the oil removal regime employed.

Almost all defatted soybean flour is produced from soybeans that have oil removed via solvent extraction. The oil fraction of LSF was mechanically removed by the EE method in which residual oil content can vary (4.5-13%) depending on processing parameters (10). LSF contained 9.23% more fat than DSF. Based on the study conducted by Heywood et al. (9), EE low-fat soy flour containing residual oil at this level was labeled high LSF.

Mineral losses that occur during milling account for the low ash content of wheat flour compared to the soy flours (17). Carbohydrate data can be attributed to the natural differences among the carbohydrate compositions of cereals (70%) and legumes (26-38%) (5, 17). In general, wheat flour would be expected to have a dissimilar proximate analysis profile than soybeans because it can be characterized as high in carbohydrate content, relatively low in protein content, and minute in lipids, fiber, minerals and vitamins (17).

**Solvent Retention Capacity.** The normal contribution of wheat flour components that have water-holding potential to WHC cannot be overlooked. Solvent retention capacity was evaluated to determine if the wheat flour used for WHC testing had a normal composition of components that could contribute to the uptake of water. Solvent retention capacity (SRC) is typically used to establish a practical quality/functionality profile of flour (15). Several flour constituents are noted as having influence on water retention potential and include pentosans,
damaged starch, glutenin. In SRC testing, the amounts of glutenin (gluten protein), damaged starched, pentosans (water extractible arabinoxylans) based on flour percentage are estimated using lactic acid, sodium carbonate, and sucrose respectively. For flour typically used to produce bread by the sponge-dough method, optimal SRC profile values would be ≥ 100% glutenin, ≤ 96% pentosans, ≤ 72% damaged starch (15). All mean values for water retention components of WHT were within the typical range for a sponge-dough bread system. This SRC profile obtained for WHT (Table 3.2) indicated that the wheat flour evaluated would be suitable for bread production and has a normal composition of water uptake constituents.

**Water-holding Capacity.** Past studies have utilized various centrifuge speeds for WHC determination (9,16,19). Since speed could potentially influence WHC, performing WHC testing at two centrifuge speeds allowed its effect on WHC of flour blends to be assessed at a numerically similar and dissimilar speed to other studies. Mean values of WHC for wheat flour and wheat-soy blends at 1592 × g (3000 rpm) and 4424 × g (5000 rpm) are shown in Figures 3.1 and 3.2. On average, WHC values for all wheat-soy flour blends were lower at 5000 rpm than 3000 rpm, as would be expected, because of the greater centrifugal force being applied to samples. Speed was deemed a significant (p<0.05) variable by statistical analysis; however, no significant interaction of speed with flour or replacement level was noted. This finding suggests that speed does not impact WHC character of the flour types and levels of replacement used in this study. When replacement was performed based on weight (Fig 3.1), WHC values increased with the level of soy flour, except for the 6% blends; however, this exception was not statistically significant. These results were similar to those observed for water uptake capacity of wheat-legume blends. D’Appolonia found that the water uptake capacity for blends containing wheat flour and pinto, navy, or mung bean flour increased as bean flour level increased (19).

Protein-water interactions are related to WHC, or water-binding capacity; therefore, the amount of protein within blends may have influence on the WHC values observed
The amount of protein in DSF and LSF was not the same. Thus, for flour blends based on weight, the contribution of protein-water interactions may have been innately disproportional. This is why the amount of LSF used in blends based on protein was increased to reflect a protein content equivalent to DSF. Mean WHC for WHT-LSF blends (based on protein) ranged from 0.63 to 0.87 g water/g flour. Those for WHT-DSF blends were 0.62 to 0.81 g water/g flour. These WHC values are higher than those observed for blends based on weight. This may indicate that increasing the amount of total soy protein in the flour blends allowed more interactions with water to occur.

In general, results showed that at both centrifuge speeds, all wheat-soybean flour blends had WHC greater than wheat flour with the exception of 2% blends based on protein content (Figure 3.2). For almost all flour blends, WHC values based on protein content were slightly higher than those based on weight. This may be attributed to the presence of more protein available for protein-water interactions.

WHC values for WHT-LSF blends were comparable to those of WHT-DSF blends. This finding was initially not expected due to differences in processing methods. Oil content of LSF is about 10% higher than that of DSF. Oil exhibits hydrophobic characteristics in solutions containing water; therefore, it was suspected that the residual oil of WHT-LSF blends would inhibit water-binding and cause lower WHC values.

During the EE process, soybeans are exposed to high temperature and pressure, both of which will cause some level of protein denaturation. Processing of this sort would render the protein less soluble in an aqueous solution. PDI is a measure of protein-water interaction and an indirect measure of the degree of heat treatment imposed to a protein material (20). LSF had a PDI of approximately 27 indicating a lower solubility potential in comparison to DSF. With a lower PDI and higher residual oil/hydrophobic constituent content, WHT-LSF blends were expected to have low WHC values compared to WHT-DSF blends. Despite this, some WHT-LSF blends achieved higher WHC than blends containing DSF (PDI=67) at the
same level (2, 6, 8% based on sample weight; 2, 4, 10, 12% based on protein content). This
difference may indicate that the physicochemical changes imposed by the EE process on the
proteins of this LSF may not have had caused deleterious effects on WHC. These changes
possibly may include conformational alterations that allow these proteins to form favorable
interactions for water holding.

The results indicated that when the LSF investigated in this study is used to replace
up to 12% wheat flour by weight or protein content, mean WHC in the range 0.60 to 0.87 g
water/g flour can be expected under the utilized testing parameters. Among the replacement
levels and centrifuge speeds tested, the greatest capacity of water held by these blends was
achieved at 12% replacement. For the flour blends tested, decreased WHC was observed at
higher centrifugal force.

For comparisons among replacement levels, significant differences in WHC were
consistently observed between the 2 and 12% replacement level. Figures 3.1 and 3.2 show at
which replacement levels WHC of WHT-LSF began to exceed that of WHT-DSF based on
sample weight and protein content. The figures also indicate that for every 2% increase in the
amount of soy flour used to replace wheat flour, an approximate 1% increase in WHC was
demonstrated on average for the wheat-soybean flour blends used in this study. This may
serve as a gauge for the amount of LSF required to alter water-holding in foods formulated
with a WHT-LSF flour blend. Overall, WHT-LSF blends were comparable to blends
containing DSF in water-holding capabilities. These results may imply that WHT-LSF blends
could be used interchangeably in bread systems and other select food applications in which
water-holding or retention are of importance.

ACKNOWLEDGMENTS

This work was partially supported by the Center for Crops Utilization Research
Center at Iowa State University (Ames, IA) and Alliance for Graduate Education and the
Professoriate-Iowa (AGEP). Technical assistance with raw materials was provided by Regina Butler (presently) and Elaine Wolf (formerly) of Soy Innovations (Des Moines, IA).

REFERENCES


### TABLE 3.1
Proximate Analysis Data for Wheat Flour (WHT), Defatted Soy Flour (DSF), and Low-fat Soy Flour (LSF)

<table>
<thead>
<tr>
<th>Constituent (%)</th>
<th>WHT&lt;sup&gt;b&lt;/sup&gt;</th>
<th>DSF&lt;sup&gt;b&lt;/sup&gt;</th>
<th>LSF&lt;sup&gt;b&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein</td>
<td>13.31</td>
<td>51.71</td>
<td>45.34</td>
</tr>
<tr>
<td>Ash</td>
<td>0.50</td>
<td>6.14</td>
<td>5.79</td>
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<tr>
<td>Residual oil</td>
<td>2.23</td>
<td>0.99</td>
<td>10.22</td>
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<tr>
<td>Moisture</td>
<td>12.36</td>
<td>5.93</td>
<td>5.73</td>
</tr>
<tr>
<td>Carbohydrates&lt;sup&gt;c&lt;/sup&gt;</td>
<td>71.60</td>
<td>35.23</td>
<td>34.94</td>
</tr>
<tr>
<td>PDI</td>
<td>n.d.</td>
<td>67</td>
<td>27</td>
</tr>
</tbody>
</table>

<sup>a</sup>Results are expressed on an as-is basis.<br>
<sup>b</sup>Mean data based on 3 replicates.<br>
<sup>c</sup>Values determined by difference.<br>Abbreviation n.d. denotes “not determined”.
**TABLE 3.2**  
**Solvent Retention Capacity Profile of Wheat Flour**

<table>
<thead>
<tr>
<th></th>
<th>WHT(^a)</th>
<th>Flour for sponge and dough system(^b)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lactic Acid (%)</td>
<td>115.50</td>
<td>≥ 100</td>
</tr>
<tr>
<td>Sodium carbonate (%)</td>
<td>68.47</td>
<td>≤ 72</td>
</tr>
<tr>
<td>Sucrose (%)</td>
<td>73.45</td>
<td>≤ 96</td>
</tr>
</tbody>
</table>

\(^a\)Mean data based on 3 replicates.  
\(^b\)AACC (15)
FIG 3.1. WHC based on sample weight of wheat-soy blends at different centrifuge speeds. × WHT-DSF 3000 rpm; ■ WHT-LSF 3000 rpm; + WHT-DSF 5000 rpm; □ WHT-LSF-5000 rpm
FIG 3.2. WHC based on equivalent protein of wheat-soy blends at different centrifuge speeds. × WHT-DSF 3000 rpm; ■ WHT-LSF 3000 rpm; + WHT-DSF 5000 rpm; □ WHT-LSF-5000 rpm
CHAPTER 4. EVALUATION OF DOUGH DEVELOPMENT PROPERTIES FOR WHEAT-SOYBEAN FLOUR BLENDS

A paper to be submitted to The Journal of the American Oil Chemists Society

Toshiba. L. Traynham\textsuperscript{5,6} And Deland Myers\textsuperscript{5}

**ABSTRACT:** The effects of low-fat soybean flour (LF) on dough development when used in a flour blend were investigated. Flour blends contained wheat flour (W) that was partially replaced by LF or defatted soybean flour (DF) based on sample weight and equivalent protein content at 2, 6, and 12\%. Dough development was recorded by a 10–g mixograph. Mix (peak) time, rate of development and work input were determined. Mix (peak) time ranged from 5.40–6.56 min. W-LF blends required the most time to achieve optimum dough development. This effect may result from the oil present in LF that may have caused reductions in shear force during the mixing of the dough in the mixograph. Dough development time increased as the amount of soybean flour increased for W-LDF blends. The relationship of shear force during dough development was independent of the level of wheat flour replacement for W-LF and W-LP blends. In general, the use of dough conditioners caused strengthening of the gluten network as shown by shortened mix times and steady changes in shear force during dough development. The rate of development was quickened after dough conditioner use for all blends.

**KEY WORDS:** low-fat soybean flour, flour blends, dough development, mix (peak) time

**INTRODUCTION**

The low-fat soybean flour observed in this study is derived from soybeans that have undergone oil removal by a technology called extruding-expelling. This unique process of oil

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\textsuperscript{6} Principal investigator and author.
removal is performed without the popularly used organic solvent, hexane. Extruding-expelling involves the extrusion cooking of dehulled soybeans resulting in an extrudate that is exposed to a screw press for the releases of oil. This process excises up to 12% residual oil from the extrudate leaving behind a high quality meal (1). After sieving, the remaining extrudate becomes what is termed extruded-expelled (EE) low-fat soybean flour. This method of oil removal is favored by some for its environmental friendliness and low capital investment for (1) small-scale soybean processors (2).

EE low-fat soybean flour is viewed as a viable food ingredient due to its nutritional and functional properties. Reports have indicated EE soybean flour has a protein content ranging from 42.5% to 50.2% (3,4). The protein content will vary according to processing parameters and residual oil content. The EE process reportedly produces meal that has higher amino acid availability and digestible energy compared to solvent-extracted meal and these nutritional benefits are transferred to the flour (1,5). Research by Heywood et al (4) that examined the functionally of EE soybean flours showed that the flours had minimal solubility and demonstrated increased emulsification capacity with increased pH. Water holding capacity, fat binding capacity and foaming capacity were influenced by residual oil content and protein dispersibility index (PDI) (4,5,6). Based on EE low-fat soybean flour’s nutritional composition and functional capacity, it has the potential to be used in a variety of food products; however, widespread use has not occurred. Use of this flour may be heightened after studies are able to provide more technical information on its performance in select food systems. Food categories in which this flour type has been successfully utilized include meat (beef patties) and bakery products (doughnuts) (3). It can be used as the starting material for textured soy protein as well as an ingredient for co-processing with cereal grains into snacks.

The primary application that soybean flour has been cited for use is in bakery products. Most bakery applications of soybean flour in the past have been as a bleaching
agent and a milk protein or non-fat dry milk ingredient replacer (1). Products where the protein quality of the baked good is important have sparked the use of flour blends containing wheat and soybean flour. Combining the two flours serves to remedy the lysine deficiency of wheat flour with a higher lysine containing protein source (6).

Previous studies have produced discordant findings on bread made from flour blends of wheat and soybean flours with various residual oil contents. Kulkarni et al reported that breads prepared with flour blends containing EE processed soybean flour (6-9% residual oil) had similar quality characteristics to flour blends prepared with soybean flour produced from pre-conditioned extruded soybeans (7). In contrast, Dhingra and Jood found that volume and overall acceptability were lowest for breads made from blends of wheat and full-fat soybean flour (8). Although researchers have reported on the effects of soy flour on final bread quality (volume, appearance, and taste perceptions), the literature is devoid of research that focuses on an understanding of its performance and functionality during bread dough development. Bread dough properties usually have a direct impact on finished bread quality (9).

Physical dough testing provides information about the rheological properties of wheat flour doughs. These rheological properties, as recorded by a mixograph give information related to bread dough formation such as mixing time (mix (peak) time), rate of dough development (left of peak slope), and shear force during dough development (peak integral). These values and others can be collected during dough development testing using a mixograph. The mixograph is a high speed recorder-mixer that is equipped with four vertical pins attached to a rotating head (6). As the pins rotate within a flour-water mixture, gluten develops. The pins record the force required to manipulate the dough; the force will vary according to the developing dough’s rheological character and changes in consistency. Mixograph testing can be used to predict effects of additives, such as non-gluten components on dough rheology and final product quality (10).
Past studies have evaluated the rheological properties of wheat when combined with a variety of non-gluten flours and additives. In an evaluation by Fleming and Sosulski, mix (peak) times for flour blends of field pea, faba bean and sunflower concentrate were lower than the control. Wheat-defatted soy flour blends in the same study had the longest mix (peak) time (11). For wheat flour blends containing up to 10% defatted soy flour and various wheat flour varieties, slight differences in mix peak times were attributed to the gluten content of the wheat flours. The mixograph data showed that the addition of soy flour up to 6% did not significantly affect mixing time of either of the wheat flours studied (12).

There is limited information available on the effect of EE low-fat soybean flour on the development of wheat flour in dough systems. In addition, the rheological properties of wheat-EE low-fat soybean flour have yet to be evaluated. It is the focus of this study to investigate alterations (if any) to the mixing time, rate of development, and shear force during dough development for bread dough made from wheat-EE low-fat flour blends. Effects on dough development time will be evaluated with and without the use of dough conditioners.

Since the soybean flour being investigated contains residual oil, it is proposed that the oil has a lubricating effect on flour components. The lubricating effect will cause less shear force to be required during mixing and extend mix (peak) time. The surfactant properties of the selected conditioners will aid in dough development by strengthening the gluten matrix. By defining the rheological character of dough developed from wheat-EE lowfat soybean flour blends, this flour’s use as a workable bread ingredient can be evaluated. Information should also result from this study that help define optimum dough development for these blends and provide a better understanding of what reactions may be occurring differently in this dough system as compared to one of a wheat-defatted soybean flour blend.
EXPERIMENTAL PROCEDURES

Raw materials. Wheat flour (WHT) and defatted soybean flour (DF) were obtained from the commercial sources, Horizon Milling (Wayzata, MN) and Cenex Harvest States Co. (Mankato, MN) respectively. Low-fat soybean flour (LF) was obtained from Insta-Pro International (Des Moines, IA). Flour blends were formulated by mixing wheat flour with low-fat or defatted soybean flour in the following ratios (wheat:soybean flour): 98:2, 94:6, 88:12 (wt basis). For LF, samples were also formulated based on equal protein content (LP) with DF according to the ratios specified above. Since LF (~45%) contained less protein than DF (~51%), samples based on weight may not have been indicative of an equal potential based on the protein of these soybean flours to effect dough development. The dough conditioners, sodium stearoyl lactylate and (SSL) calcium stearoly-2-lactylate (CSL) were used individually and in combination (DSL) (1:1) at 0.5% (wt basis). The conditioners were added to the flour in dry form prior to water addition.

Proximate composition analyses. Wheat-soybean flour blends (1:1) were analyzed for the following: protein (AOAC 990.03) (13), moisture (AACC 44-40) (10), fat (AOAC 922.06) (13), and ash (AACC 08-03) (10). Proximate composition of wheat flour was also quantified by the same methods. The percentage of carbohydrate content was calculated by difference. Flour blends consisted of a 1:1 ratio of wheat flour:soy flour.

Mixograph analysis. A 10-g mixograph was used to record dough mixing characteristics by the AACC method 54-40A. Flour (14% mb) and water amounts were acquired from the Absorption, Flour, and Water Amount Table in the Mixograph Handbook based on the protein and moisture content of wheat flour. Mid-line analysis values for left of peak (% torque), peak (min), and integral (% torque-min) were derived from computer software analysis (Mixsmart, v. 4.1, National Mfg. Lincoln, NE) of mixograms. The values of left of peak, peak, and integral were recorded for each sample after a 10 min mix period as rate of dough development, mix time to optimum development, and work input, respectively.
Data analysis. Data was collected following a 2x3 (without dough conditioners) and a 2x3x3 (with dough conditioners) factorial design and subjected to Analysis of Variance (ANOVA) using the GLM procedure by the Statistical Analysis Software Program (version 9, SAS Institute, Inc., Cary, NC). Tukey’s Honestly Significant Difference (HSD) Test was used to compare the main effect of independent variables deemed significant at p<0.05. For rate of development, differences among means for the main effect of flour were deemed significant at p <0.05.

RESULTS AND DISCUSSION

Proximate composition analyses. The composition of flour blends can be found in Table 4.1. Blends containing soybean flour had higher protein and fat content than the wheat flour. Differences in carbohydrate content can be attributed to the replacement of wheat flour with soybean flour.

Mixograph analysis. Mean mix (peak) times for flour-blends tested are listed in Table 4.2 Based on the results, time (min) required for optimum dough development was affected by the level of soybean flour replacement (p=0.016) and dough conditioner use (p<0.001). The addition of high levels of soybean flour and various types of dough conditioners have been shown to cause alterations to dough development, water absorption and bread quality. Past studies have demonstrated that these two components have a bearing on the rheological properties of wheat-soy dough (10,13,14). The incremental addition of soybean flour in this study is viewed as increasing the availability of soy proteins for interaction with gluten-forming proteins. It has been suggested that the presence of non-gluten forming proteins dilute gluten structure (15). Dilution of gluten structure could equate to a weakened dough matrix.

Without the use of dough conditioners (Table 4.2), W-DF blends results showed that as the level of soybean flour inclusion increased, mix peak time decreased. It is probable that the interactions of DF with gluten-forming proteins caused the exertion of stronger shear
forces during mixing as compared to the other blends. However, this may not signify the formation of good quality dough. Stronger binding of water by DF proteins may have also had a role, making water less accessible to wheat flour for hydration and gluten formation.

The majority of W-LF and W-LP blends demonstrated higher mean mix (peak) times in comparison to WHT and W-DF when dough conditioners were not employed. Slight increases in these delays were observed as the level of LF or LP increased. Statistical comparison of these means revealed that the majority of these mix times were not significantly different. This result showed that the time needed for optimum dough development of blends consisting of WHT and LF based on equivalent protein and weight would typically be the same. It may also be possible to eliminate the need to adjust mix time for blends in which 2, 6, or 12% WHT is replaced with this LF based on equivalent protein or weight. The exception, 98:6 W-LF blend may be attributed to natural variation.

The presence of residual oil in LF (~10%) may have contributed to lower dough strength and lengthened mix time. The residual oil may have had a lubricating effect that caused dough character to be less elastic and more viscous. This would impose less shear force on the mixograph pins. This action may potentially result in decreased shear force during dough development, hastened rates of dough development and longer time requirements to reach mix (peak) time as measured by the mixograph, all of which were observed for W-LF blends. W-DF blends had steeper slopes for rate of development. Shear force decreases with an increase in percentage of soy flour were more defined for W-DF than W-LP.

When used in yeast-leavened products, dough conditioners serve as surface active agents to flour components. Dough conditioners interact with gluten proteins to form a reinforced dough matrix. In general, dough conditioners aid in increasing dough mixing tolerance to ingredient variations (6). Noticeably, the mean mix time for W increased after
dough conditioner use. This signifies that a strengthening effect had occurred due to dough conditioner presence.

Dough conditioners were able to minimize differences in mix time for W-LF and W-LP blends (Table 4.2). Mix (peak) times for W-LF and W-LP blends were not significantly different from the control. There were also no significant differences in mix time for W-LP and W-LF. This may mean that the employment of dough conditioners may have effectively strengthened the dough matrix by emulsifying water, protein, and oil for W-LF and W-LP blends. SSL and CSL are widely used dough conditioners in the baking industry. As reaction products of stearic acid and lactic acid neutralized to sodium and calcium salts, these conditioners have different emulsifying capabilities. CSL is reported to demonstrate limited emulsifying properties in a bread dough as compared to SSL (6).

Use of dough conditioner was statistically significant for all observed measures except rate of dough development. Based on the overall means for the main effect of flour, samples containing added CSL required the least shear force during dough development (155.37 % tq*min). SSL required the most shear force for both measures. This may demonstrate its strengthening and emulsification power. When a 1:1 ratio of dough conditioners was employed, necessary work input was significantly different from the individual conditioners and had values in between that of SSL and CSL as SSL ≥ DSL ≤ CSL.

By observing Fig 4.1 and Fig. 4.2, it is apparent that the slopes were different for the trend lines when dough conditioners were and were not used. It is possible that the conditioners had some effect on dough strength that equated to increased tolerance to mixing. Steeper slopes observed for the relationship between shear force during development and percentage of soy flour without dough conditioners may mean that dough demonstrated less tolerance to mixing. Increased tolerance as demonstrated by slope values closer to zero may
be an indication of more steady changes in dough consistency being displayed by the developing dough as a result of conditioner use.

Overall the wheat-LF blends observed required more time for dough development than wheat flour alone as measured by the mixograph. W-LF and W-LP blends potentially required the most time due to lubrication of developing dough proteins and starch and lessened dough strength by residual oil present. Rate of dough development was more gradual for these blends as compared to W-DF blends (Table 4.1). Dough conditioners lowered mix (peak) time and in general, decreased in shear force during dough development.

For W-DF blends there were stronger correlations of level of soy flour replacement with the observed measures. Overall, the dough development properties of W-LF and W-LP blends were not comparable to W-DF blends. Dough produced from these blends displayed greater mixing tolerance and shear force requirements during dough development. In general, increasing the level of soy flour used in W-LF and W-LP blends did not have deleterious effects on the measures of mix time, rate of development, and shear force during dough development. It is important to note that these slight effects on dough character may not disqualify W-LF blends from producing good quality bread and thus warrants investigation.

ACKNOWLEDGMENTS

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REFERENCES


<table>
<thead>
<tr>
<th>Constituent (%)</th>
<th>W</th>
<th>W-DF</th>
<th>W-LF</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein</td>
<td>13.01</td>
<td>29.88</td>
<td>26.68</td>
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<tr>
<td>Moisture$^a$</td>
<td>12.36</td>
<td>9.13</td>
<td>8.75</td>
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<tr>
<td>Fat</td>
<td>1.68</td>
<td>2.24</td>
<td>5.27</td>
</tr>
<tr>
<td>Ash</td>
<td>0.5</td>
<td>1.83</td>
<td>2.83</td>
</tr>
<tr>
<td>Carbohydrate$^a$</td>
<td>72.45</td>
<td>56.92</td>
<td>56.35</td>
</tr>
</tbody>
</table>

W, W-DF, W-LF denote wheat flour, wheat-defatted soy blends, wheat-low-fat soy blends respectively.

$^a$Mean values based on 3 replicates.

$^b$Ratio of wheat flour:soybean flour = 1:1 on an as-is basis.

$^c$Based on 4 replicates; all other constituents based on 3 replicates.

$^d$Calculated by difference.
<table>
<thead>
<tr>
<th>Flour Blend</th>
<th>Replacement Level</th>
<th>Mix (peak) time (min) Without dough conditioner</th>
<th>Mix (peak) time (min) With Dough conditioner</th>
</tr>
</thead>
<tbody>
<tr>
<td>W</td>
<td>0</td>
<td>5.39&lt;sup&gt;c&lt;/sup&gt;</td>
<td>5.67&lt;sup&gt;ac&lt;/sup&gt;</td>
</tr>
<tr>
<td>W-DF</td>
<td>2</td>
<td>5.95&lt;sup&gt;ac&lt;/sup&gt;</td>
<td>5.30±&lt;sup&gt;ac&lt;/sup&gt;</td>
</tr>
<tr>
<td>W-DF</td>
<td>6</td>
<td>4.57&lt;sup&gt;b&lt;/sup&gt;</td>
<td>4.95&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>W-LF</td>
<td>2</td>
<td>4.02&lt;sup&gt;b&lt;/sup&gt;</td>
<td>4.01&lt;sup&gt;b&lt;/sup&gt;</td>
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<td>W-LF</td>
<td>6</td>
<td>5.89&lt;sup&gt;cd&lt;/sup&gt;</td>
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<tr>
<td>W-LF</td>
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<td>5.49&lt;sup&gt;c&lt;/sup&gt;</td>
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</tr>
<tr>
<td>W-LP</td>
<td>2</td>
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<tr>
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<td>6</td>
<td>6.21&lt;sup&gt;ad&lt;/sup&gt;</td>
<td>5.74&lt;sup&gt;ac&lt;/sup&gt;</td>
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<td>5.96&lt;sup&gt;ac&lt;/sup&gt;</td>
<td>5.37&lt;sup&gt;ac&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

*Mean values for the main effect of flour are based on 5 replicates; mean ± standard error.
*Means within a column with the same letter are not significantly different (p>0.05).
*Standard errors for W, W-DF and W-LF are ±0.17 and ±0.22, respectively.
*Standard error is ±0.14 for all means within the column.
Table 4.3
Mean Rate of Dough Development for Wheat-soy Flour Blends With and Without Use of Dough Conditioners

<table>
<thead>
<tr>
<th>Flour Blend</th>
<th>Rate of dough development(^#) (%/min)</th>
<th>Rate of dough development(^#) (%/min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>W</td>
<td>2.67±0.20(^a)</td>
<td>2.69±0.49(^a)</td>
</tr>
<tr>
<td>W-DF</td>
<td>2.00±0.25(^b)</td>
<td>2.95±0.23(^a)</td>
</tr>
<tr>
<td>W-LF</td>
<td>1.39±0.18(^b)</td>
<td>2.95±0.23(^a)</td>
</tr>
<tr>
<td>W-LP</td>
<td>0.60±0.26(^c)</td>
<td>3.00±0.24(^a)</td>
</tr>
</tbody>
</table>

\(^a\) Mean values for the main effect of flour are based on 5 replicates.
\(^b\) Means with the same letter within a column are not significantly different (p>0.05).
Figure 4.1. Relationship of shear force during dough development and percentage of soy flour without use of dough conditioners.

Equations:

- $y = -8.1335x + 197.08$, $R^2 = 0.8207$
- $y = 0.306x + 195.93$, $R^2 = 0.0062$
- $y = -2.5106x + 212.68$, $R^2 = 0.1272$
Figure 4.2. Relationship of shear force during dough development and percentage of soy flour after dough conditioner addition.

The graph shows the relationship between shear force (% kg/min) and percentage of soy flour with the following equations and R² values:

- For W-DF: 
  \[ y = -5.0831x + 192.69 \]
  \[ R^2 = 0.2984 \]

- For W-LF: 
  \[ y = 1.8025x + 189.13 \]
  \[ R^2 = 0.0287 \]

- For W-LP: 
  \[ y = -0.7077x + 196.74 \]
  \[ R^2 = 0.0056 \]
CHAPTER 5. BREAD-MAKING PERFORMANCE OF FLOUR BLENDS CONTAINING EXTRUDED-EXPELLED LOW-FAT SOYBEAN FLOUR

A paper to be submitted to The Journal of the American Oil Chemists Society
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ABSTRACT: The performance of low-fat soybean flour in a flour blend with wheat (W-LF) and its effects on bread-making were investigated. Low-fat and defatted (W-DF) soybean flour was used to partially replace 2%, 6%, and 12% of wheat flour in flour blends. The physical properties of bread as affected by W-LF were determined by measuring loaf weight, volume, crumb/crust color, and crumb texture. The flavor and aroma characteristics were assessed by members of a trained sensory panel. These properties were also evaluated for wheat bread and flour blends with defatted soybean for comparison. Loaf weight and volume ranged from 139.03 g to 142.58 g and 683 cc to 740 cc respectively. Weight and volume were not significantly different among breads with and without wheat flour replacement with soy flour. W-LF and W-DF produced breads with a darker crust and crumb than the control. W-LF bread crumb had less greenness and than the control and more perceived yellowness than W-DF. In general, the firmness of bread crumb for flour blend breads increased with storage and the amount of soy flour used. Sensory evaluation data showed that panelists were able to recognize differences in bread aroma, flavor, and firmness among wheat bread and flour blend breads. Panelists perceived W-LF breads to be firmer and have more intense bread aroma and flavor than W-DF breads. Overall, W-LF produced bread with up to 12% replacement of wheat flour with indistinguishable crumb firmness and bread flavor to a wheat bread not containing soy flour. These results will provide direction to assist bakers in formulating and processing breads with increased use of low-fat soybean flour produced using the extruding-expelling method.

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8 Principal investigator and author
KEYWORDS: low-fat soybean flour, flour blends, loaf volume, bread crumb, bread crust

INTRODUCTION

The use of soybean flour is not new to the baking industry and bread formulation. Soybean protein products such as flours are versatile and can be incorporated into almost all food systems (1). In bread systems, there are several proposed benefits from the use of soy flour. Soy flour has been shown to prolong shelf life, improve crumb structure, aid in the increase of crumb consistency, and upgrade nutritional quality (2).

The exploration of protein-enhanced bread has stemmed from the need to curtail nutritional deficiencies in developing countries. It is for this reason among others, that soybean flour is often combined with wheat flour in a flour blend or composite flour for bread-making purposes. Wheat flour is innately low in the essential amino acid, lysine. Coupling wheat flour with a protein material that is high in lysine allows a bread product to be viewed as having an improved amino acid profile (3). One such protein material that has a higher lysine content than wheat flour, is soybean flour. Soybean flour is considered an excellent complement to wheat flour for protein enhancement.

The interest in protein-enhanced bread has lead to many studies involving the use of flour blends. Past research has mainly focused on partially replacing wheat flour with an oilseed, legume, or another cereal protein product. When soy flour or the plant protein concentrates of sunflower, faba bean, and field-pea were investigated in flour blends up to a 25% replacement of wheat flour, loaf volume, specific volume, crumb grain, crumb compressibility, and loaf shape and loaf shape were compromised (4). Barley, full-fat and defatted soybean flours used in blends caused breads to have varied loaf weight, volume and sensory characteristics (5).

Low-fat soybean flour (LF) having a residual oil content of 4-12% can be prepared by an extruding-expelling (EE) process (1,6). For this high temperature, short time process,
soybeans are sheared and heated to release oil from cells. The product is then passed through an expeller or screw press to mechanically press out the oil (7,8). The EE method is a successful oil excision technology that does not utilize organic solvents. Soybean meal produced by EE processing is cited as having unique characteristics compared to that of solvent-extracted meal, such as lower protein dispersibility index. (7). However, these unique characteristics have not been evaluated in a variety of food systems. With little information (other than product specifications provided by commercial suppliers) concerning food uses of EE prepared low-fat soybean flour available, it is less likely that this flour will be positioned for widespread use.

The residual oil present in EE-LF may provide benefits to bread-making in addition to those previously stated for soybean flour in general. Previous studies have shown that residual oil present in EE-LF had a lubricating effect on dough components (9). Dough generated from flour blends of wheat flour and EE-LF (up to 12%) had longer mixing requirements, maintained slower rates of development and exhibited less shear force during dough development. This implied that the dough’s consistency changed more gradually as compared to flour blends containing defatted soy flour. These observed characteristics could have positive implications for bread volume and crumb texture. Edible fats are known to impart tenderness, darker crust color, and enlarged loaf volume to bread (9). It is the objective of this study to explore the performance of EE-LF in a flour blend system used to produce bread. Evaluation of bread weight, volume, crumb/crust color, and crumb texture will provide information on the contribution of EE-LF to the physical characteristics of bread. Sensory evaluation of breads will determine if EE-LF provides any distinctive characteristics to bread aroma and flavor. The results of this study will provide a baseline understanding of the functionality of EE-LF in this food system. This information could be used to make bread formula recommendations when added EE-LF is desired and further the widespread use of EE-LF.
EXPERIMENTAL PROCEDURES

*Bread composition.* Six to ten slices were randomly picked from 2 loaves per sample. Slices were laid on racks and allowed to air dry for 24 hrs. Breads were prepared for proximate analyses by grinding using AACC method 62-05. Moisture, ash and protein content were quantified by AACC 44-15A (10), AACC 08-01 (10), and AOAC 990.03 (11), respectively. Method AOAC 954.02 (11) performed by Woodsen-Tennent, (Des Moines, IA) was used to measure edible fat (residual oil) content. Carbohydrate content was determined by difference. All proximate analyses were performed in triplicate.

*Bread preparation.* Bread was prepared according to the straight dough method outlined by AACC method 10-10B (10) with some modifications. Bread ingredients were combined based on the formula percentages of 53.98% wheat flour (14% mb), 35.44% water, 4.64% yeast (cake), 3.24% sugar, 0.81% salt, 1.62% shortening, and 0.27% sodium stearoyl lactylate (SSL; dough conditioner). Flours used were wheat flour (W; Horizon Milling, Mankato, MN), defatted soy flour (DF; Cenex Harvest States, Makato, MN), and low-fat soy flour (LF; Soy Innovations, Des Moines, IA). Protein contents for flour were 13%, 51% and 45% respectively (9). Oil contents were 2%, 1%, and 10%, respectively (9). Flour blends of wheat-low-fat soy flour (W-LF) or wheat-defatted soy flour (W-DF) in the following ratios of wheat flour:soybean flour, 98:2, 94:6, and 88:12 were prepared and employed in the bread formulation. The formula produced 2 mini loaves (5 x 4 x 5 in.) per sample.

Dough was developed in a Hobart mixer for 5 min 30 sec. Preliminary mixograph testing results were used to determine an average mix time for all samples. Dough was divided and placed in a fermentation cabinet at 30°C and 85% relative humidity (RH) for a total fermentation period of 60 min. Dough was passed through a sheeter to release developed gas (punch). A molder was applied to roll the dough into a loaf. The loaves were transferred to a pan (5 x 6 in.) and allowed to proof (30°C and 85% RH) for 24 min at.
Loaves were baked in a rotary oven for 24 min at 415°C. Finished breads were cooled on racks for 10 min prior to volume and weight analyses and allowed to rest for an additional 35 min before storage. Bread loaves were double bagged for storage at 25°C ± 1°C for 24 hrs.

**Weight and volume measurements.** Bread loaves were weighed after 10 min of cooling. The seed displacement method using a rapeseed displacement apparatus was performed to determine bread volume in cubic centimeters (cc).

**Color measurements.** The color of bread crust and crumb were recorded by a HunterLabscan XE Spectrophotometer (Hunter Associates, Reston, VA) as $L$ (lightness), $a$ (redness to greenness), $b$ (blueness to yellowness) values. The $L$ scale ranges from 0 to 100 with 100 denoting perfect white and 0 denoting black. Positive values for $a$ and $b$ indicate redness and yellowness respectively. Negative $a$ and $b$ values specify greenness and blueeness, respectively. Configurations were light source, D65; instrument geometry, 45°/0°; view area, 0.5 cm; port size, 0.70 cm. Crust color was determined for 3 lateral locations along the top-side of bread loaves. After slicing bread (6-7 mm thickness), 3 non-overlapping spots of the crumb were evaluated: top left, top right, and bottom middle. Two loaves were evaluated for crust color measurements and 2 slices per sample for crumb color analysis.

**Textural measurement.** Bread firmness on days 1, 3, and 7 was determined according to the American Institute of Baking (AIB) White Pan Bread Firmness Measurement using a TA-XT2i texture analyzer (Texture Technologies Corp, Scarsdale, NY). Two slices were stacked and impacted by a 25 mm plastic cylinder probe to record maximum peak force (g) of compression. Configurations included 25 kg load cell, 1.7 mm/s test speed, 6.2 mm distance, and 10 g trigger force.

**Sensory evaluation.** A panel of 11 members (7=female, 4=male; ages 21-50) was compiled for sensory evaluation of bread samples among constituents of Iowa State University. Panelists participated in 4, 1-hr training sessions. Descriptors for the attributes of aroma and flavor were developed by consensus. Sample presentation and ballot revisions
were also finalized during this time. Panelists were trained to identify samples that were considered to have extreme intensities of the given attributes. For example, training samples without soy flour were used to train for intense bread flavor and aroma.

Samples were given 3-digit codes and presented to panelists for evaluation as 2 1/8 in. diameter discs of only the crumb. Since the evaluation of crust and crumb together could not be effectively standardized, crusts were not included in test samples. All sensory assessments were made under red light to mask any perceived color differences. Five bread samples were evaluated by panelist at each test session. These were W-LF 2%, W-LF 12%, W-DF 2%, W-DF 12% and a control, wheat (W) bread, without soy flour replacement. For crumb firmness evaluation, panelists were instructed to break a dime size piece of the bread disc, place the sample on middle of the tongue, and compress against the palate. Three testing sessions were held every other day for 1 week.

Data analysis. Physical and sensory property data followed a 2 x 3 factorial design. Data from 3 replicates were subjected to analysis of variance (ANOVA) using the general linear model (GLM) procedure by the Statistical Analysis Software Program (version 9, SAS Institute, Inc., Cary, NC). Tukey’s Honestly Significant Difference (HSD) Test were used to compare the main effect of independent variables that were deemed significant at p<0.05. Pair-wise comparisons (Tukey’s) were performed for means based on interactions between independent variables; significance was established at p<0.05.

RESULTS AND DISCUSSION
Bread composition. Breads demonstrated varied compositions as shown in Table 5.1. Breads made of wheat flour (W) alone had the lowest protein content. Protein content of breads made from wheat-low-fat soy flour blends (W-LF) and wheat-defatted soy flour blends (W-DF) ranged from 12.85% to 16.03%, while wheat only bread had a protein content of 11.49%. This verifies that the soybean flour added to blends was able to aid in increasing the
protein content of these breads. This has been cited as one of the main nutritional benefits of processing wheat-soybean flour blends into bread.

Moisture content and retention is an important factor in bread production and provides bakers with a prospective of soy flour’s potential contribution to bread yield and shelf-life. Breads containing soy had higher moisture content than wheat-only bread. This may have resulted from the water-holding capabilities of soybean flour. Soybean flour is said to hold 2.6 times its weight in water (12). Usually, the more moisture a bread loaf is able to retain, the greater the potential yield and shelf-life duration (13). Overall, moisture retention may be influenced by the amount of water flour blends are able to entrap and retain during the bread-making process. Moisture losses can occur from the loaf in the oven and during cooling. Crumb moisture losses can occur during slicing (14). In addition, variation in moisture contents may be attributed to non-uniformity in the randomly selected dried slices used for compositional testing, as well as moisture retention by fiber present in the soy flours.

Edible fat contents ranged from 5.25% to 5.92%. The LF used had a residual oil content of ~ 10%. Thus breads containing LF had higher edible fat content compared to those containing DF. Wheat flour typically contains about 1.5% edible fat, therefore; shortening used in the bread formula would explain the increased fat content of the wheat bread.

*Weight and volume measurements.* These two measurements are important to predicting bread quality. The most desirable texture and grain for a given bread variety can often be achieved with an ideal relationship between dough weight and loaf volume (13). Bread weight ranged from 139.03 g to 142.58 g (Figure 5.1). Dissimilarities in weight between W and flour blend breads may have been caused by differences in water retention and water loss during various bread-making steps. Based on previous water-holding capacity observations by Traynham *et al* (9), W-LF and W-DF blends demonstrated a 1% increase in
water holding capacity for each 2% replacement of wheat flour with LF or DF (9). For this reason, it was projected that breads containing soy flour would have noticeably higher weight due to increased moisture retention than bread containing no soy flour. Similar results were not found under the current bread-making conditions possibly because of the affinity for water by other ingredients that occurred and moisture losses throughout the bread-making process.

Bread volume is the space occupied by the loaf (13). Several studies have viewed a volume diminishing effect when non-wheat flours were used in combination with wheat to generate bread (4,15,16). Hallen et al (3) observed negative effects to volume for breads containing a combination of fermented/germinated cowpea flour and wheat flour. When wheat-soy-barley flour blends were evaluated for bread baking performance, volume decreased with increasing amount of non-wheat flour substitution (5). Wheat-defatted soy flour breads caused a 44 cc drop in bread volume on average. Similar results were observed in this study.

W-LF and W-DF breads had lower volumes than the control at all replacement levels (Figure 5.2). Loaf volume decreased as much as 57 cc for flour blend breads compared to wheat-only breads. W-LF breads were projected to have comparable loaf volume to control bread. If the residual oil present was able to lubricate dough components and aid in dough expansion, it was proposed that compromises to volume due to soy protein addition would be counteracted. This may have occurred for W-LF 2% bread but not W-LF 6% and 12%. These breads may have had more soy protein in the dough matrix than the residual oil lubrication could remedy. Soy flour type was not a statistically significant variable for volume measurements, meaning either LF or DF had little effect on bread volume. Visual appearance of bread loaves (Figures 5.4 and 5.5) did not subjectively give an indication of drastic volume decreases between W and W-LF breads. Greatest differences in bread volume
could be observed visually between W-DF and W. Volume was also influenced by loaf symmetry.

**Color measurements.** Overall, crust and crumb $L a b$ color values (Table 5.2) varied according to samples with and without wheat flour replacement. W-LF (43.83) and W-DF (44.23) breads had darker crusts than the control (48.89). Darker crust for breads containing soy may be explained by a higher degree of Maillard (non enzymatic) browning. The activity of non-enzymatic browning may have been enhanced by the increased protein and presence of sugars contributed by the soy flours in the bread formula. When reducing sugars chemically react with the amino group of an amino acid on a protein chain or a free amino acid, Maillard browning occurs (17). With regard to crust redness/greenness, W-DF breads (+14.31) had the highest mean for redness. W-LF bread crusts were no different from the control in redness. Mean $b$ values (+yellowness/-blueness) of crust color ranged from +16.54 to +19.88. W-LF 12% had significantly less yellowness than all other bread samples.

The bread crumb is utilized as a major quality determinant. Flour is the ingredient that has the most impact on crumb color; more importantly the grade of flour will have a directive effect. Higher grade flour will have less wheat bran particulates and thus will be lighter in color. This lightness will effect crumb lightness. Lightness for bread crumb followed the same outcomes for bread crust. Crust $a$ values were -1.12, -0.31 and -0.39 for W, W-DF, and W-LF, respectively. W-LF bread crumb had less perceived greenness than the control and W-DF and more perceived yellowness for the control. LF flour was considered more yellow then DF by subjective appearance which would explain this result. Both soy flours experienced some degree of heat treatment during processing. The color produced by heat treatment of soy flour can vary from pale cream to pale yellow (2).

**Textural measurement.** Evaluation of bread crumb firmness was projected to show differences in crumb texture among the control and breads made with flour blends. Soy flour, soy protein concentrate and soy protein isolates derived from it have a long history as quality
improving additives that also retard the staling process (13). It was probable that wheat-soy blend breads would have less firm crumbs throughout the test period for this reason. W-LF blends were proposed to display a less firm crumb than W-DF after 1, 3, and 7 days of storage due the potential tenderizing effect of its residual oil.

The interaction of flour and level of wheat flour replacement had a statistically significant effect on bread firmness for days 1, 3, and 7 of textural analysis (p=0.003, 0.03, and 0.01, respectively). On day 1, firmness for W-LF breads at all replacement levels were not significantly different from the control (Figure 5.3). This finding was the same on days 3 and 7. This result indicates that these breads would potentially have similar textural properties to typical white bread for up to 7 days after processing. Overall, W-DF blend breads were more firm than the other breads up to 7 days after processing. DF had a higher protein content than LF and may therefore contribute more interaction in the dough matrix. A greater availability for interaction would equate a firmer crumb texture, as soy flour is credited for imparting “body” or firmness to the crumb while reducing tendency toward dough-like consistency (2). This may also explain the occurrence of increased firmness with increased percentage of defatted soy flour used.

Sensory evaluation. After 4 training sessions, panelists were able to define descriptors for aroma and flavor for wheat only and flour blend breads. Nutty flavor and aroma were determined to be attributes that may be ascribed to soybean flour addition. For testing, aroma and flavor attributes were assigned an intensity level on a 15 cm line scale by each panelist (15 = intense); bread firmness was assessed as firm at 15 cm.

Sensory evaluation test results are shown in Table 5.3. Bread aroma was perceived to be the most intense for the control (W) bread, which was expected. W-LF 12% (8.44) was determined to have more intense bread aroma than W-DF 12% (6.92); however, this greater degree of bread aroma was not statistically significant. Differences in nutty aroma and flavor could be recognized between flour blend breads and the control by panelists. W-LF breads
had a higher mean bread flavor score over W-DF breads. This may indicate that the residual oil in these breads has some effect on bread flavor and may be enhancing bread flavor slightly.

Breads containing soy flour were perceived to have more intense nutty aroma and flavor than the control. Previous studies on the sensory properties of wheat-soy bread have quantified grain, beany, or grassy flavors (1,2,18). Shrogen *et al.* found that composite flours of whole wheat flour and defatted soy flour (up to 30%) had higher perceived beany flavor than other breads (16). Grain flavor was significantly lower for the same composite flour breads. Ryan *et al.* found no significant differences in grassy or grain-like flavor for breads made with non-solvent and solvent-extracted texturized soy flours (19). The LF used underwent a non-solvent oil extraction process. Grassy flavor was not identified by panelists during training. Grain-like aroma and flavor were perceived by some panelist during training but were deemed attributes characteristic to that of fresh bread.

Crumb firmness results varied among samples and ranged from 1.61 to 9.52 (Fig. 5.3). The greater the value the more firm the sample was rated. Soy flour type and level of replacement had the most effect on this attribute. Breads at 2% replacement were not significantly different from the control. W-LF 12% was given a lower crumb firmness score than W-DF 12%. It is likely that the residual oil may have been able to have a tenderizing effect on crumb texture.

In general, level of replacement had minimal effect on the physical and sensory properties studied. The greatest differences in properties were observed at the 12% level of replacement. As with findings from other researchers, most characteristics of bread may demonstrate a direct or inverse relationship as the level of soy flour present is adjusted. The physical properties of weight and volume were shown by others to be highly altered due to wheat flour replacement with soy flour. The anticipated drastic effects on bread volume were not observed for W-LF breads which may relate to the method of oil extraction for LF. The
residual oil and potential denaturation of protein that results from this process may have caused LF to have favorable bread-baking potential.

Crust color was most affected by LF use while crumb color remained very similar to the control. Aside from volume differences, this indicates that when comparing the appearance of W-LF and W breads it would potentially be difficult to tell the breads apart. The same would be true for W-LF bread crumb. Based on sensory evaluation data, W-DF and W-LF had similar nutty flavor and aroma. This refutes differences that were expected in nutty flavor due to differences in the manufacture of the respective soy flours. Both may be contributing only mild characteristic soy aroma and flavor. Textural properties determined by objective measurement found that W-LF breads remained softer than breads containing DF for up to seven days and were not different from the control. Based on the bread formulas used, similar firmness to a standard white bread can be maintained by W-LF at 2%, 6%, and 12% replacement of wheat flour. Sensory observations revealed the same result. In general, W-L breads were rated approximately 65% less firm than W-DF breads by subjective measurement.

Based on the results of this study, LF is a suitable non-gluten replacement for use in a flour blend with wheat flour for the production of bread. Incorporation of LF in a blend up to 12% was able to produce bread of comparable weight, volume, crumb color, and firmness to a typical wheat bread. This indicates that the methods employed for oil removal during the processing of LF has a positive effect on its bread baking performance and sensorial contributions in bread.

ACKNOWLEDGMENTS

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REFERENCES


### Table 5.1
Proximate Composition of Wheat and Wheat-soy Flour Blend Breads

<table>
<thead>
<tr>
<th>Flour blend</th>
<th>% soy flour</th>
<th>Moisture&lt;sup&gt;b&lt;/sup&gt; (%)</th>
<th>Protein&lt;sup&gt;b&lt;/sup&gt; (%)</th>
<th>Fat&lt;sup&gt;b&lt;/sup&gt; (%)</th>
<th>Ash&lt;sup&gt;c&lt;/sup&gt; (%)</th>
<th>Carbohydrates&lt;sup&gt;d&lt;/sup&gt; (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>W</td>
<td>0</td>
<td>11.69</td>
<td>11.29</td>
<td>5.42</td>
<td>2.16</td>
<td>69.44</td>
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<tr>
<td>W-DF</td>
<td>2</td>
<td>12.77</td>
<td>13.24</td>
<td>5.29</td>
<td>1.99</td>
<td>66.71</td>
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<td>W-DF</td>
<td>6</td>
<td>11.47</td>
<td>13.45</td>
<td>5.30</td>
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<td>67.46</td>
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<tr>
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<td>11.95</td>
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<td>66.48</td>
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<td>2</td>
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<td>5.43</td>
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<td>10.75</td>
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<td>11.89</td>
<td>16.03</td>
<td>5.77</td>
<td>2.45</td>
<td>63.86</td>
</tr>
</tbody>
</table>

<sup>a</sup>W, W-DF, W-LF denote wheat flour, wheat-defatted soy flour blend, wheat-low-fat soy flour blends.

<sup>b</sup>Results are expressed on an as-is basis.

<sup>c</sup>Mean of 3 replicates.

<sup>d</sup>Mean of 2 replicates.

<sup>e</sup>Calculated by difference.
Table 5.2
Hunter L a b Color Values for Crust and Crumb of Wheat and Wheat-soy Flour Blend Breads

<table>
<thead>
<tr>
<th>Flour*</th>
<th>Bread crust#</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>L</td>
<td>a</td>
<td>b</td>
<td></td>
</tr>
<tr>
<td>W</td>
<td>48.89±0.28 x</td>
<td>+13.41±0.15 y</td>
<td>+19.89±0.20 x</td>
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<tr>
<td>W-DF</td>
<td>44.23±0.45 y</td>
<td>+14.31±0.40 x</td>
<td>+18.48±0.54 y</td>
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</tr>
<tr>
<td>W-LF</td>
<td>43.83±0.57 y</td>
<td>+13.84±0.11 y</td>
<td>+18.18±0.29 y</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Bread crumb##</th>
<th></th>
<th></th>
<th></th>
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</thead>
<tbody>
<tr>
<td>L</td>
<td>a</td>
<td>b</td>
<td></td>
</tr>
<tr>
<td>W</td>
<td>73.91±0.28 x</td>
<td>-1.12±0.04 x</td>
<td>+12.36±0.19 x</td>
</tr>
<tr>
<td>W-DF</td>
<td>72.05±0.28 y</td>
<td>-0.31±0.10 y</td>
<td>+13.76±0.26 x</td>
</tr>
<tr>
<td>W-LF</td>
<td>72.12±0.22 y</td>
<td>-0.39±0.08 y</td>
<td>+15.14±1.91 x</td>
</tr>
</tbody>
</table>

#Mean ± standard error.
*Means for the main effect of flour within a column with the same letter are not significantly different (p>0.05); mean values based on 3 replicates.
Table 5.3
Sensory Evaluation Data for Wheat and Wheat-soy Flour Blend Breads

<table>
<thead>
<tr>
<th>Flour</th>
<th>Bread Aroma</th>
<th>Nutty Aroma</th>
<th>Bread Flavor</th>
<th>Nutty Flavor</th>
<th>Firmness</th>
</tr>
</thead>
<tbody>
<tr>
<td>W</td>
<td>11.07±0.44&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.97±0.40&lt;sup&gt;b&lt;/sup&gt;</td>
<td>10.86±0.57&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.15±0.26&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.61±0.24&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>W-DF</td>
<td>8.99±0.41&lt;sup&gt;b&lt;/sup&gt;</td>
<td>4.76±0.48&lt;sup&gt;a&lt;/sup&gt;</td>
<td>9.05±0.37&lt;sup&gt;b&lt;/sup&gt;</td>
<td>3.62±0.46&lt;sup&gt;a&lt;/sup&gt;</td>
<td>6.04±0.56&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>W-LF</td>
<td>9.60±0.32&lt;sup&gt;b&lt;/sup&gt;</td>
<td>4.73±0.55&lt;sup&gt;a&lt;/sup&gt;</td>
<td>9.62±0.33&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>3.31±0.42&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.97±0.44&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Mean scores for attributes based on responses from 11 panelists per test session; 3 test sessions were conducted.

Mean values for the main effect of flour within a column with the same letter are not significantly different (p>0.05).
Figure 5.1. Bread loaf weight data for wheat (W) and wheat-soy flour blend breads (W-DF and W-LF). Bars represent means for the main effect of flour. Bars with the same letter are not significantly different at p>0.05.
Figure 5.2. Bread loaf volume data for wheat (W) and wheat-soy flour blend breads (W-DF and W-LF). Bars represent means for the main effect of flour. Bars with the same letter are not significantly different at $p>0.05$. 
Figure 5.3. Firmness measurement of wheat and wheat-soy flour blend breads on days 1, 3, and 7. Bars with the same letter within a day are not significantly different (p > 0.05). Means are based on 2 firmness measurements per bread for 3 replicates.
Figure 5.4. Front view of wheat, wheat-low-fat soy flour blend (12% soy flour), and wheat-defatted soy flour blend (12% soy flour) breads.
Figure 5.5. Side view of wheat, wheat-low-fat soy flour blend (12% soy flour), and wheat-defatted soy flour blend (12% soy flour) breads.
CHAPTER 6. GENERAL CONCLUSIONS

The objective of this research was to evaluate the performance of low-fat soybean flour (LSF) in a flour blend. Blends were studied at up to 12% replacement of wheat flour. The effects of wheat-LSF blends on dough and bread development were observed in comparison to a wheat flour individually (control) and wheat-defatted flour blends.

Chapter 3 discussed the water-holding capacity (WHC) of wheat-soy flour blends which indicated that for each 2% increase in soybean flour used in blends, an approximate 1% increase in WHC could be expected for the blends. WHC was greater for flour blends than the wheat only sample. In some instances, WHC for wheat-LSF blends exceeded that of wheat-DSF blends. This result was counter to those expected. LSF had a residual oil content of ~10%. Protein dispersibility index (PDI) of this flour was 27. The presence of a component with hydrophobic properties and a low PDI would signify that this soy flour would have low solubility in solution and thus have limited WHC. Overall, wheat-LSF blends had comparable WHC to wheat-DSF blends. It was proposed from this finding that these blends would display similar character in dough and bread. It is speculated the processing conditions of

Dough development testing results were outlined in chapter 4. It was proposed that the residual oil present in LSF would have a lubricating effect on flour components during dough development. This effect would cause wheat-LSF blend to have longer mixing time and impose less shear forces during dough development. Findings from mixograph observations showed that wheat-LSF blends required more time to reach optimum dough development, rate of development was more gradual and shear force during dough development was independent of percentage of soy flour used. Shear force for all blends decreased as the percentage of soy flour increased. This occurrence was more defined for wheat-DSF blends than wheat-LSF blends. Use of dough conditioners lowered mixing time requirements and caused shear force decreases during dough development to be more
constant. Decreases in mix (peak) time signified that dough conditioners were able to form a strengthened gluten network.

In general, the occurrence of longer mixing and constant shear force during dough development for wheat-LSF blends was a good indicator that a lubricating effect did occur. It is apparent that the residual oil, acting as a lubricator caused dough consistency to be steady throughout mixing. Since the mixograph records changes in dough consistency as it increases to a peak, it would take longer for a peak value to be detected for wheat-LSF blends.

The effects of DSF and LSF presence in flour blends on bread development were discussed in chapter 5. The physical and sensory properties of wheat-soybean flour blend bread are important to the acceptability of these breads as viable food products. For physical property evaluation, wheat-soy breads were not statistically different in loaf weight or volume to the control. Numerically, weight values were higher for wheat-soy flour blend breads than for wheat-only bread. This added weight displays the ability of soy flour to hold more water and aid in increased water absorption in dough. Volume may have been influenced by the symmetry of the bread produce. Wheat-DSF breads have more flat crust tops than wheat-only and wheat-LSF breads.

The results for crumb firmness measurements can be attributed to the level of residual oil present in the soybean flours. Oil present in LSF could have formed a hydrophobic barrier to moisture release, allowing the crumb of wheat-LSF bread to stay softer than wheat-DSF bread for up to 7 days of storage. Findings from the measurement of crust and crumb color were similar to those found in previous studies. Sensory evaluation of wheat-soy flour blend breads revealed that a nutty flavor and aroma could be perceived by panelists. These descriptors were attributed to soy presence but were considered undesirable aroma and flavor properties. Since bread flavor was not statistically different among wheat-only and wheat-LSF breads, it was concluded that the residual oil may have caused flavor enhancement of bread flavor.
Based on the performance data for LSF, flour blends containing this partially defatted soybean flour did not pose objectionable effects on water-holding capacity, dough development or bread development. The dough development properties observed were not diminishing to dough quality as determined by bread quality. Breads produced with wheat-LSF blends had marginal compromise to bread volume, increased bread weight, and indistinguishable crumb firmness and bread flavor to wheat-only bread. Overall, results for these studies may provide a baseline understanding of wheat-LSF functionality that may be used by bakers for bread production purposes.

**RECOMMENDATIONS FOR FUTURE STUDIES**

While the performance of LSF in a flour blend was favorable to bread and dough development, it may still be possible to improve the quality of wheat-LSF dough and bread. As with most food ingredients, processing parameters and conditions can affect finished product quality. Future studies could be performed in which adjustments to processing conditions are made that could have a bearing on improved dough and bread quality. The dough and bread processing of wheat-soy flour blends was done using the same amount of water for all samples. Since it has already been shown that LSF and DSF increase the water-holding capacity of flour blends, dough mixing and bread-making could be conducted with added water using the established premise that for each 2% increase in the level of soy flour used, a 1% increase in water-holding capacity can be observed. Bread volume improvements may be made by evaluating longer fermentation of dough and/or the addition of vital wheat gluten.

An investigation on the WHC results of wheat-LSF blends is warranted. Differences in the soybean flours used for this study include residual oil content and protein dispersibility index (PDI). The PDI of LSF was low indicating that the proteins in this flour had undergone some degree of denaturation and a high level of heat treatment. By performing WHC testing on blends with varying oil contents and high, mid, and low PDI values, it may be possible to
elucidate why LSF was able to have comparable WHC to a soybean flour of less oil content and higher PDI. By comparing the WHC of a defatted soy flour with a low PDI that has and has not been re-fatted to the same level as LSF, two determinations can be made. It is the belief of the investigator that processing conditions used for extruding-expelling of soybeans in the production of LSF caused denaturation of soy proteins that allowed for favorable conformations of proteins for protein-water interactions, such as water-holding. If LSF still has comparable water-holding capacity, this may mean that the proteins have a unique molecular configuration that is responsive to water-holding. If the re-fatted, defatted soy flour with low PDI has dissimilar WHC to LSF, this will imply that the lipids present in the residual oil of LSF have formed protein-lipid interactions that do not interfere with water-holding.
ACKNOWLEDGMENTS

I would like to thank my Lord and Savior, who has provided me with the physical and financial means, brain-power, will-power, determination, health and strength, necessary to attain a Doctorate degree in Food Science and Technology. I have to acknowledge my family and friends for their encouragement and belief in me. Believing that all things can be done once one puts their mind to it truly helped me along the way. I would also like to acknowledge the mentors and colleagues that helped me grow academically and professionally at Iowa State University. I am sure that our relationships will continue. My time in Ames, IA has been a memorable learning experience because of these individuals.

I would especially like to acknowledge Virginia State University (VSU), my alma mater. Some members of the Iowa State University faculty and staff have various opinions about students that receive degrees at historically black colleges and universities (HBCU’s). I can truly attest to their ignorance on the notion that these schools do not equip its students with skills necessary to matriculate in a graduate program at a research 1 institution. I can verify that their ideas have no merit. Without the academic and professional foundation that I received at this HBCU, I do not believe that I could have attained this degree. Thanks to everyone that has had an influence on my accomplishments from my birth to now.
## APPENDIX

**Water Holding Capacity of Wheat-Soy Flour Blends at Different Centrifuge Speeds**

<table>
<thead>
<tr>
<th>Flour Blends</th>
<th>3000 rpm</th>
<th>5000 rpm</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>By sample weight (g water/g flour)</td>
<td>By protein content (g water/g flour)</td>
</tr>
<tr>
<td>WHT</td>
<td>0.65 ± 0.02</td>
<td>-</td>
</tr>
<tr>
<td>WHT + DSF</td>
<td></td>
<td></td>
</tr>
<tr>
<td>98:2</td>
<td>0.60 ± 0.08</td>
<td>0.72 ± 0.01</td>
</tr>
<tr>
<td>96:4&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.71 ± 0.01</td>
<td>0.71 ± 0.02</td>
</tr>
<tr>
<td>94:6</td>
<td>0.66 ± 0.03</td>
<td>0.75 ± 0.01</td>
</tr>
<tr>
<td>92:8</td>
<td>0.71 ± 0.05</td>
<td>0.79 ± 0.03</td>
</tr>
<tr>
<td>90:10</td>
<td>0.77 ± 0.003</td>
<td>0.76 ± 0.03</td>
</tr>
<tr>
<td>88:12</td>
<td>0.79 ± 0.01</td>
<td>0.81 ± 0.01</td>
</tr>
<tr>
<td>WHT + LSF</td>
<td></td>
<td></td>
</tr>
<tr>
<td>98:2</td>
<td>0.61 ± 0.02</td>
<td>0.75 ± 0.01</td>
</tr>
<tr>
<td>96:4&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.67 ± 0.02</td>
<td>0.74 ± 0.01</td>
</tr>
<tr>
<td>94:6</td>
<td>0.68 ± 0.03</td>
<td>0.74 ± 0.02</td>
</tr>
<tr>
<td>92:8</td>
<td>0.72 ± 0.01</td>
<td>0.76 ± 0.03</td>
</tr>
<tr>
<td>90:10</td>
<td>0.75 ± 0.03</td>
<td>0.79 ± 0.03</td>
</tr>
<tr>
<td>88:12</td>
<td>0.74 ± 0.03</td>
<td>0.87 ± 0.02</td>
</tr>
</tbody>
</table>

<sup>a</sup>Values are mean ± standard error of 3 replicates.

<sup>b</sup>Values are mean ± standard error of 4 replicates.
Electron micrographs of wheat-soybean flour blends. (Lipids are encircled)
<table>
<thead>
<tr>
<th>Blend</th>
<th>Loaf weight* (g)</th>
<th>Loaf volume* (cc)</th>
</tr>
</thead>
<tbody>
<tr>
<td>W</td>
<td>140.00±0.24</td>
<td>760±7</td>
</tr>
<tr>
<td>W-DF 2%</td>
<td>142.58±1.86</td>
<td>717±15</td>
</tr>
<tr>
<td>W-DF 6%</td>
<td>141.78±1.22</td>
<td>660±39</td>
</tr>
<tr>
<td>W-DF 12%</td>
<td>140.72±1.94</td>
<td>686±74</td>
</tr>
<tr>
<td>W-LF 2%</td>
<td>139.87±0.45</td>
<td>729±27</td>
</tr>
<tr>
<td>W-LF 6%</td>
<td>142.29±0.24</td>
<td>694±18</td>
</tr>
<tr>
<td>W-LF 12%</td>
<td>141.45±0.51</td>
<td>687±44</td>
</tr>
</tbody>
</table>

*Mean ± standard error based 6 bread loaves.
*No mean values are significantly different within a column.
Sensory Evaluation Results for Wheat and Wheat-soybean Flour Blend Breads Based on Flour Type

<table>
<thead>
<tr>
<th></th>
<th>Nutty aroma*</th>
<th>Bread flavor*</th>
<th>Nutty flavor*</th>
</tr>
</thead>
<tbody>
<tr>
<td>W</td>
<td>1.97±0.40 b</td>
<td>10.86±0.57 a</td>
<td>1.15±0.26 b</td>
</tr>
<tr>
<td>W-DF</td>
<td>4.76±0.48 a</td>
<td>9.05±0.37 b</td>
<td>3.61±0.46 a</td>
</tr>
<tr>
<td>W-LF</td>
<td>4.73±0.55 a</td>
<td>9.62±0.33 ab</td>
<td>3.31±0.42 a</td>
</tr>
</tbody>
</table>

*Means ± standard error based 3 replicates. Mean values with the same letter within a column are not significantly different (p>0.05); significant differences based on means for the main effect of flour.
**Sensory Evaluation Results for Wheat and Wheat-soybean Flour Blend Breads Based on Flour Type and Replacement Level**

<table>
<thead>
<tr>
<th></th>
<th>Crumb firmness*</th>
<th>Bread aroma*</th>
</tr>
</thead>
<tbody>
<tr>
<td>W</td>
<td>1.61±0.24 b</td>
<td>11.07±0.44 a</td>
</tr>
<tr>
<td>W-DF 2%</td>
<td>2.56±0.49 b</td>
<td>11.06±0.42 a</td>
</tr>
<tr>
<td>W-LF 2%</td>
<td>2.77±0.55 b</td>
<td>10.74±0.40 a</td>
</tr>
<tr>
<td>W-DF 12%</td>
<td>9.52±0.54 a</td>
<td>6.92±0.48 b</td>
</tr>
<tr>
<td>W-LF 12%</td>
<td>5.18±0.59 c</td>
<td>8.49±0.43 b</td>
</tr>
</tbody>
</table>

*Significant differences based on mean of the interaction of the independent variables flour and percentage of soy flour; means with the same letter within a column are not significantly different (p>0.05).
**E-mail Script for Recruitment of Sensory Panalists**

Title: Sensory Evaluation of Bread Made From Wheat-Soybean Flour Blends

Greetings,

Here is an opportunity that may interest you!

Dr. Deland Myers, and I (Toshiba Traynham), in the Food Science and Human Nutrition Department, are recruiting panelists to participate in a research study on the sensory characteristics of bread made with wheat flour and soybean flour. All of the products have been prepared in a food grade facility. If you agree to participate, you will be asked to attend up to six, approximately one-hour training sessions and up to ten approximately, 30-minute test sessions. Testing will be conducted over a three-month period with one to two test sessions per week. There will be up to seven samples per session and you will be evaluating aroma and flavor. There will be no names on the ballot. The study will be conducted in Room 2951, Food Sciences Building.

The ingredients are water, wheat flour, compressed yeast, sugar, soybean flour (defatted or low-fat), salt, vegetable shortening, and dough conditioners (sodium stearoyl-lactylate or calcium stearoyl-2-lactylate). You should not participate if you have an allergy or sensitivity to any of the aforementioned ingredients. You will be assigned an identifier code to be used on the ballots. No references will be made to individuals in any presentations or publications resulting from this study.

The general benefit derived from this study for society is that information will be gained regarding the difference (if any) of the sensory properties of bread made with soybean flour compared to each other and bread that does not contain soybean flour. You will receive a food reward after each session (e.g. cheese, fruit, ice cream) and a gift certificate from a store such as Hy-Vee, Target, Wal-Mart or Borders.

Your participation in this study is completely voluntary and you may withdraw from the study at any time without penalty by notifying me, the Principal Investigator or other key personnel listed below.

**Principal Investigator**
Toshiba Traynham, 1251 Food Sciences Bldg., 294-4890, toshibat@iastate.edu

**Key Personnel**
Deland Myers, 1139 Food Sciences Bldg., 294-5216, dmyers@iastate.edu
Ken Prusa, 222C Mac Kay Hall, 294-4323, kprus@iastate.edu

Are you interested in participating? If yes, please email me the weekdays and times you are NOT available from 7/1/06 to 1/31/06. I will let you know the final schedule by phone, email or in person. If you have further questions at any time regarding the study, please contact the principal investigator. If you have any questions about the rights of research subjects or research-related injury, please contact Ginny Austin-Eason, Institutional Review Board Administrator, Office of Research Assurances, 1138 Pearson Hall, (515) 294-4566; austinger@iastate.edu or Diane Ament, Director, Office of Research Assurances, 1138 Pearson Hall, (515) 294-3115; dament@iastate.edu

Thank you!
Toshiba Traynham
Sensory Evaluation Training Consensus Ballot
Aroma Assessment of wheat-soy breads of varying replacement levels

Panelist ______

Instructions: Take 2-3 deep, quick sniffs of a sample. After sniffing, write down the terms that you think describe the samples aroma.

<table>
<thead>
<tr>
<th>Sample Code Color</th>
<th>Aroma Description</th>
<th>Consensus Description</th>
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</thead>
<tbody>
<tr>
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### Sensory Evaluation Training Consensus Ballot

**Flavor assessment of wheat-soy breads of varying replacement levels**

Panelist ______

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<tr>
<th>Sample Code Color</th>
<th>Flavor Description</th>
<th>Consensus Description</th>
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**Sensory Evaluation Training Consensus Ballot**  
Texture assessment of wheat-soy breads of varying replacement levels

Panelist ______

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<tr>
<th>Sample Code Color</th>
<th>Texture Description</th>
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<table>
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<tr>
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<th>Consensus Description</th>
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<tbody>
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</tbody>
</table>
Sensory Evaluation Ballot

Panelist # __________      Date:       /        /

Bread aroma

None                                Intense

Nutty aroma

None                                Intense

Crumb firmness
(Pull a ¼ piece from the bread disc (nickel-sized), place on the center of tongue and press upward against the roof of mouth with the tongue)

Soft                                firm

Bread flavor

None                                Intense

Nutty flavor

None                                Intense

Comments: