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Evaluation of Water-Holding Capacity for Wheat–Soy Flour Blends

T. L. Traynham \cdot Deland J. Myers \cdot A. L. Carriquiry \cdot L. A. Johnson

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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 $1592 \times g$ (3,000 rpm) and/or $4,424 \times g$ (5,000 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 5,000 rpm than at 3,000 rpm. In general, the WHC increased as the proportion of soybean flour increased. Differences in WHC were greatest between the samples containing 2 and 12% soybean flour. WHC values among the 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.

T. L. Traynham - D. J. Myers - L. A. Johnson Department of Food Science and Human Nutrition and Center for Crops Utilization Research, Iowa State University, Ames, IA 50011, USA

A. L. Carriquiry Department of Statistics, Iowa State University, Ames, IA 50011, USA

D. J. Myers (\boxtimes) 2312 Food Sciences Bldg., Ames, IA 50011, USA e-mail: dmyers@iastate.edu

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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 complement to wheat flour for blends used in cereal-based products $[1, 1]$ $[1, 1]$ $[1, 1]$ [2](#page-4-0)] based on protein quality [\[3](#page-4-0), [4](#page-4-0)]. 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](#page-4-0)]. 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](#page-4-0)]. 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 three times more water than these ingredients [\[7](#page-4-0)].

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](#page-4-0)]. 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](#page-4-0)]. EE processing is favored for its lack of use of hazardous solvents, feasibility for processing small quantities of soybeans, and low capital investment costs $[8]$ $[8]$. The meal produced from the EE method is reported to have higher digestible energy and amino acid availability compared to defatted soybean meal [\[9](#page-4-0)].

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 their effect on bread properties [\[10](#page-4-0), [11](#page-4-0)]. 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, USA); defatted soybean flour (DSF) and low-fat soybean flour (LSF) were obtained from Cenex Harvest States Co. (Mankato, MN, USA)

and Insta-Pro International (Des Moines, IA, USA), respectively.

Proximate Analysis

Crude protein (AOAC 990.03) [[12\]](#page-4-0), moisture (AOAC 925.10) [[12\]](#page-4-0), fat (AOAC 922.06) [\[12](#page-4-0)], and ash (AOAC 925.25) [\[12](#page-4-0)] 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](#page-4-0)] by Woodsen–Tennent, Des Moines, IA,USA.

Solvent-Retention Capacity (SRC)

The SRC was determined using method AACC 56-11 [\[14](#page-4-0)] to quantify potential contributions to WHC by other flour components having water-uptake capabilities. The solvents used were sucrose (50% v/v), sodium bicarbonate (5% v/v), and lactic acid (5% v/v). Twenty-five milliliters of prepared solvent were added to 5 g of flour in 30 mL centrifuge bottles. Centrifugation at $1,239\times g$ (3,000 rpm) was performed for 15 min. After decanting, a gel remained. Gels were weighed and the SRC value (%) calculated as % $SRC = [[gel wt/flow wt] \times (86/(100 - % flow time)$ ture)) – 1)] \times 100] for each solvent.

Water-Holding Capacity (WHC)

The WHC was determined using methods modified from Heywood et al. [\[15](#page-4-0)] and Lin and Zayas [\[16](#page-4-0)]. 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 min, then centrifuged at either $1,592\times g$ or $4,424\times g$ (3,000 and 5,000 rpm, respectively) for 30 min. After decanting the supernatant, each bottle was weighed and WHC (g of water/g flour) was calculated as:

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

Statistical Analysis

Water-holding capacity testing followed a complete randomized design. Data from four replicates were subjected to analysis of variation (ANOVA) using the general linear model (GLM) procedure in the Statistical Analysis Software program version 9.1 (SAS Institute, Inc., Cary, NC, USA). Tukey's test was used for multiple comparisons. Differences were deemed significant at the $P < 0.05$ level.

Results and Discussion

Proximate Analysis

Table 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](#page-4-0)].

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 [\[18](#page-4-0)]. Soy flours contain protein in the range of 40–54% [[10\]](#page-4-0). 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 oilremoval regime employed.

Table 1 Proximate analysis data for wheat flour (WHT), defatted soy flour (DSF), and low-fat soy flour (LSF)

WHT ^b	DSF ^b	LSF ^b
13.31	51.71	45.34
0.50	6.14	5.79
2.23	0.99	10.22
12.36	5.93	5.73
71.60	35.23	34.94
ND	67	27

ND not determined

Results are expressed on an as-is basis

b Mean data based on three replicates

^c Values determined by difference

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 the residual oil content can vary (4.5–13%) depending on the processing parameters [[10\]](#page-4-0). The LSF contained 9.23% more fat than the DSF. Based on the study conducted by Heywood et al. [\[9](#page-4-0)], 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\]](#page-4-0). Carbohydrate data can be attributed to the natural differences among the carbohydrate compositions of cereals (70%) and legumes $(26-38\%)$ [\[5](#page-4-0), [17](#page-4-0)]. In general, wheat flour would be expected to have a dissimilar proximate analysis profile than soybeans because it can be characterized as high in carbohydrate content, relatively low in protein content, and minute in lipids, fiber, minerals and vitamins [[17\]](#page-4-0).

Solvent-Retention Capacity

The contributions of other flour components to WHC cannot be overlooked. Solvent-retention capacity (SRC) testing is used to establish a practical quality/ functionality profile of flour [[15\]](#page-4-0). Several flour constituents are noted as influencing water-retention potential, including 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 $\geq 100\%$ glutenin, $\leq 96\%$ pentosans, \leq 72% damaged starch [[15\]](#page-4-0). All mean values for waterretention components of WHT were within the typical range for a sponge–dough bread system. This SRC profile obtained for WHT (Table 2) indicated that the wheat flour evaluated would be suitable for bread production and has a normal composition of wateruptake constituents.

^a Mean data based on three replicates

 b AACC(15)

Water-Holding Capacity

Past studies have utilized various centrifuge speeds for WHC determination [[9,](#page-4-0) [16,](#page-4-0) [19](#page-4-0)]. Since speed could potentially influence the 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 $1,592 \times g$ (3,000 rpm) and $4,424 \times g$ (5,000 rpm) are shown in Figs. 1 and 2. On average, WHC values for all the wheat–soy flour blends were lower at 5,000 rpm than 3,000 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 the WHC character of the flour types and levels of replacement used in this study.

When replacement was performed based on weight (Fig. 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 [\[19](#page-4-0)] found that the water uptake capacity for blends containing wheat flour and pinto, navy, or mung bean flour increased as the bean flour level increased.

Protein–water interactions are related to WHC [\[20](#page-4-0)], or water-binding capacity [[15\]](#page-4-0); therefore, the amount

Fig. 1 WHC based on sample weight of wheat–soy blends at different centrifuge speeds. cross WHT–DSF 3,000 rpm; filled square WHT-LSF 3,000 rpm; plus WHT–DSF 5,000 rpm; open square WHT–LSF 5,000 rpm

Fig. 2 WHC based on the protein content of wheat–soy blends at different centrifuge speeds. cross WHT–DSF 3,000 rpm; filled square WHT–LSF 3,000 rpm; plus WHT–DSF 5,000 rpm; open square WHT–LSF 500 pm

of protein within blends may influencethe 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 0.62–0.81 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. 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 applied 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 lower WHC values than the 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 caused deleterious effects on WHC. These changes may possibly 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, a mean WHC in the range 0.60–0.87 g water/g flour can be expected under the testing parameters used. 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 [1](#page-3-0) and [2](#page-3-0) show at which replacement levels the 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 water-retention are of importance.

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