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Physicochemical Characterization and Sensory Analysis of Yeast-leavened and Sourdough Soy Breads

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Abstract

Sourdough fermentation has been shown to have numerous beneficial effects on bread quality, and nutritionally enhance soy-supplemented bread by altering isoflavone chemical forms. Given this, the objective of this study was to compare the loaf quality and shelf life of sourdough and yeast-leavened soy breads by various physical, thermal, and sensorial methods, and to assess the effects of fermentation by various microorganisms on isoflavone profile in dough and breads using high-performance liquid chromatography analysis. Sourdough fermentation yielded a less extensible dough compared to yeast-leavened soy dough ($P < 0.001$), and resulted in a harder bread crumb ($P < 0.05$) and lighter crust color ($P < 0.001$), compared to yeast-leavened soy bread (Y-B). Sensory analysis revealed a significantly higher overall liking of Y-B compared to sourdough soy bread (SD-B) ($P < 0.001$). Segmentation analysis of the cohort suggests that overall liking and bread consumption frequency may be determinants of Y-B or SD-B preference. SD-B and Y-B exhibited similar shelf-life properties. Despite significantly different enthalpies associated with the melting of amylose-lipid complexes, thermal analysis of the 2 soy breads stored for 10 d (ambient conditions) demonstrated no significant difference in water distribution and starch retrogradation ($P < 0.05$). Lastly, SD-B was determined to have 32% of total isoflavones occurring in the aglycone form compared to 17% in Y-B. These findings warrant further investigation of sourdough fermentation as a processing technique for quality and nutritional enhancement of soy-based baked goods.

Keywords

bread; fermentation; isoflavone; sourdough; soy

Introduction

Sourdough fermentation is an ancient practice, in which a symbiotic culture of bacteria and yeast (approximate ratio of 100:1) is used to ferment grain (typically wheat or rye), resulting in the production of acids and other flavor molecules, as well as CO₂ for leavening (Corsetti and Settanni 2007). Sourdough fermentation, however, requires more time for CO₂ production compared to baker's yeast (*Saccharomyces cerevisiae*), which led to the industrialization of yeast-leavened bread at the turn of the 20th century and reduced sourdough bread to a niche, artisan market (Carnevali and others 2007). Experts in the field, however, believe that sourdough fermentation can impart enhancing effects on loaf quality, such as the generation of favorable flavor and aroma compounds (Gobbetti and others 2005) and improvement of gas retention and loaf volume (Gänzle and others 2007). Additionally, sourdough fermentation may result in shelf-life extension by the generation of antimicrobial

compounds, retarding of starch retrogradation, and the formation of acids during the fermentation process (Gobbetti and others 2005).

Differences in loaf quality can be quantified by numerous methods. For example, dough extensibility is frequently utilized to quantify large deformations that correlate to baking performance, such as gas retention and loaf volume, as well as gluten composition of doughs and the effect of mixing time on dough extensibility (Anderssen and others 2004; Abang Zaidel and others 2008). Additionally, texture and thermal analyses of bread are frequently utilized to study key events of the bread staling process, such as increasing bread crumb hardness, moisture migration from bread crumb to crust, and amylopectin recrystallization occurring with storage time (Davidou and others 1996; Baik and Chinachoti 2000; Vittadini and Vodovotz 2003; Lodi and Vodovotz 2008).

Sourdough fermentation of alternative flours, such as soy, may offer nutritional benefits in addition to physical benefits on loaf quality. Soy phytochemicals such as isoflavones (genistein, daidzein, and glycitein) are hypothesized to mimic the action of 17- β -estradiol, and their consumption has been correlated with reducing the risk of various diseases such as hormone-related cancers (Messina and others 2006), cardiovascular disease, and osteoporosis (Scheiber and others 2001). Soy-supplemented bread has been shown to be an effective carrier of soy isoflavones compared to other soy-based foods, delivering as much as 31.6 ± 2.1 mg isoflavones/slice (Ahn-Jarvis and others 2013).

Some research suggests that conjugated isoflavones must be deglycosylated by the action of β -glucosidase to their aglycone forms for absorption via passive diffusion across human enterocytes and, therefore, derivation of their purported health benefits (Xu and others 1995; Scalbert and Williamson 2000). The fermenting culture of sourdough typically contains autochthonous lactic acid bacteria (LAB), and several strains of LAB have been shown to exhibit β -glucosidase activity (Di Cagno and others 2010). Additionally, fermented soy foods have been shown to contain significantly greater aglycone forms compared to nonfermented soy foods, which is generally attributed to the β -glucosidase activity of the fermenting culture (Coward and others 1993, Pyo and others 2005).

The objectives of this study were to compare the bread quality and shelf life of sourdough and yeast-leavened soy breads by various physical, thermal, and sensorial methods, and to assess the isoflavone profile of the fermenting doughs and final baked products by HPLC analysis.

Materials and Methods

Materials

Ingredients (% wet basis) for yeast-leavened soy bread (Y-B) and sourdough soy bread (SD-B) formulation are detailed in Table 1. HPLC-grade acetonitrile (ACN), acetic acid, methanol (MeOH), and water were purchased from Fisher Scientific (Fair Lawn, N.J., U.S.A.). Dimethyl sulfoxide (ACS spectrophotometric grade) and isoflavone standards daidzein, genistein, glycitein, daidzin, and glycitin were purchased from LC laboratories division (PKC Pharmaceuticals, Inc., Woburn, Mass., U.S.A.) and malonyldaidzin, acetyldaidzin, malonylgenistin, acetylgenistin, malonylglycitin, acetylglycitin from Wako Chemicals USA, Inc. (Richmond, Va., U.S.A.), and glycitein from Indofine (Hillsborough, N.J., U.S.A.).

Bread production

Y-B was prepared by a sponge-dough method (Vodovotz and Ballard 2009). SD-B was generated using a modified formulation of Y-B, in which the preferment (dough yield = 200)

inoculated with *Lactobacillus brevis*, *Lactobacillus plantarum*, and *S. cerevisiae* at 0.1% of the preferment flour weight (Lallemand, Montréal, QC, Canada) was incorporated at 21.2% of the total dough weight (wet basis). Final proofing of Y-B was at 40 °C and 95% relative humidity for 1 h (CM2000 combination module, InterMetro Industries Corp., Wilkes-Barre, Pa., U.S.A.), and 30 °C for 9 h for SD-B (Isotemp® Oven 200 Series, Model 215F, Fisher Scientific, Fair Lawn, N.J., U.S.A.), and dough analyses immediately followed. Proofed dough was baked for 50 min at 152 °C (Jet air oven, JA14, Doyon, Linière, QC, Canada), cooled at room temperature for 3 h, sliced (Doyon SM302 bread slicer, Linière, QC, Canada), and sealed in polyethylene bags prior to analyses of fresh bread samples. Y-B and SD-B were stored unsliced in polyethylene bags for 10 d under ambient conditions (approximately 20 °C) prior to slicing and analyses.

Physical properties

Dough extensibility—Dough extensibility methods were based on work conducted by Frazier and others (1985). The Instron Universal Testing Machine 5542 (Instron Corp., Norwood, Mass., U.S.A.) equipped with a 100N load cell was used to test uniaxial dough extensibility. Proofed dough was flattened to 5-mm sheets using a Carver press (Fred S. Carver, Inc., Summit, N.J., U.S.A.), which applied approximately 4000 lbs/in² over a 2-h period. Sample strips (3.0 × 0.5 cm) were placed on the Instron stage, secured by clamps, and pulled at a crosshead speed of 150 mm/min by a dough hook attachment. Bluehill® 2 software version 2.17 (Instron Corp., Norwood, Mass., U.S.A.) recorded maximum force (N) and extension at maximum load (mm), and the test was stopped upon dough rupture.

Bread crumb hardness—Bread crumb hardness was determined using the Instron Universal Testing Machine 5542, Bluehill 2 software version 2.17, and a modified AACC method 74-09 (AACC 2000), utilizing a 57-mm dia plunger, as conducted by Crockett and others (2011). A uniaxial compression with crosshead speed of 100mm/min was applied to 25 × 25 × 25 mm samples to mimic mastication, with crumb hardness corresponding to the force (N) required for 40% compression.

Specific loaf volume—The specific loaf volume (cm³/g) was determined using the AACC method 10-05 (rapeseed displacement) and the loaf mass (g) (AACC 2000). Measurements of loaf mass were taken immediately following a 3-h cooling period.

Crust color—The crust color of SD-B and Y-B was determined using a Chroma Meter CR-300 (Konica Minolta Sensing Americas, Inc., Ramsey, N.J., U.S.A.), which provided measures of lightness (L), ranging from 0 (black) to 100 (white), and the chromatic components, a* and b*, which range from green to red and blue to yellow, respectively, on a scale of −120 to +120.

Sensory analysis

Ohio State Univ. Institutional Review Board exemption was approved (Protocol Nr: 2012E0320) and consumers who had allergies to wheat and/or soy were excluded from participation. A total of 60 participants were enrolled and 55 adults (> 18 y) completed all aspects of the sensory evaluation. Although untrained, panelists were familiar with product sensory evaluations and most had participated in previous related projects (Wszelaki and others 2005; Kamerund and Delwiche 2007; Ahn-Jarvis and others 2013). Panelist evaluated overall liking and preference of a single pair of Y-B and SD-B using a 9-point hedonic scale and paired difference test, respectively. Y-B and SD-B samples were baked and prepared the day prior to testing. Bread samples were cut into cubes (1.0 × 1.0 × 0.5 in), included bread crumb and crust, stored in tightly covered plastic soufflé cups labeled with 3-digit codes, and counterbalanced and randomized in their presentation. Sensory tests were conducted in a

facility where temperature, airflow, and noise were controlled. Ambient fluorescent light and incandescent light illuminated each sensory booth. Overall liking and demographic information was collected using Compusense® 5.2 software (Compusense, Inc., Guelph, ON, Canada), and preference testing was conducted using paper ballots.

Thermal analyses

Thermogravimetric analysis (TGA)—Thermogravimetric Analyzer Q5000 (TA Instruments, New Castle, Del., U.S.A.) was used to evaluate moisture content of Y-B and SD-B bread crumbs. Samples (15 to 20 mg) were placed on platinum pans (PerkinElmer Life and Analytical Sciences, Inc., Boston, Mass., U.S.A.) and a linear heat ramp of 5 °C/min from approximately 20 °C to 180 °C was applied. Thermograms were analyzed using Universal Analysis™ 2000 software (TA Instruments) and moisture content of samples was determined as the difference between initial and final weights of the sample (Fessas and Schiraldi 2001).

Differential scanning calorimetry (DSC)—Y-B and SD-B bread crumb samples (approximately 10 mg) and references (empty) were hermetically sealed in stainless steel pans (PerkinElmer Life and Analytical Sciences, Inc.). DSC Q100 (TA Instruments) was calibrated with indium and purged with nitrogen prior to loading the cell with food samples. Samples were first cooled to −50 °C and held isothermally for 3 min before being heated to 150 °C at a linear rate of 5 °C/min.

Thermograms were generated by plotting heat flow (J/g) against temperature and were analyzed using Universal Analysis 2000 software (TA Instruments) to determine the enthalpies (J/g) associated with thermal transitions. Endothermic peaks occurring below 0 °C were attributed to ice melting (Reid and others 1993; Vittadini and Vodovotz 2003), allowing for the determination of the percent “freezable” water (%FW) (Eq. 1). Percent “unfreezable” water (%UFW) was defined as the difference between the sample moisture content (obtained from TGA) and %FW. Endothermic peaks occurring between 40 and 60 °C were attributed to amylopectin melting, and endothermic peaks observed between 100 and 130 °C were associated with melting of amylose-lipid complexes (Lodi and Vodovotz 2008).

$$\%FW = \frac{\text{Enthalpy at } 0^{\circ}\text{C (J/g)}}{\text{Latent heat of fusion of water (333.2J/g)}} \times 100 \quad (1)$$

Isoflavone analyses

Dough formulations for isoflavone analysis—To investigate the effects of sourdough compared with straight-dough techniques on isoflavone profiles, 4 dough formulations were examined. SD-B variants were soy sourdough (SD-D), containing *S. cerevisiae*, *Lb. brevis*, and *Lb. plantarum*, and LAB soy sourdough (LAB-D), containing *Lb. brevis* and *Lb. plantarum*. The other 2 straight-dough (Y-B) variants were yeast-leavened soy dough (Y-D), which contained *S. cerevisiae*, and control soy dough (C-D), which contained no fermenting microorganisms.

Isoflavone extraction from bread and dough—Bread crumb (0.5 g) was homogenized in 5 mL of ACN (60% aq, v/v) using a PT 3100 Polytron homogenizer (Kinematica, Inc., Bohemia, N.Y., U.S.A.), whereas in dough, to adjust for the higher content of water, 1.0 g sample was homogenized with 5 mL of ACN (100%). Homogenized samples were vortexed, sonicated for 15 min (Mechanical Ultrasonic Cleaner FS30H, 100

watts at 42 kHz output, Fisher Scientific, Fair Lawn, N.J., U.S.A.), and centrifuged at 3000 rpm for 30 min (IEC HN-SII Centrifuge, Damon/IEC Div., Needhamhts, Mass., U.S.A.). The extraction was repeated twice and supernatant pooled for each sample. Extracts (2.0 mL aliquot) were dried under nitrogen and stored at -25°C until HPLC analysis. Each sample was redissolved in 80% MeOH, sonicated, and filtered with $0.2\ \mu\text{m}$ (13 mm dia) Grace syringe filter (Grace Davison Discovery Sciences, Deerfield, Ill., U.S.A.) prior to HPLC analysis. Triplicate batches of dough and bread were analyzed in triplicate.

Isoflavone separation and quantification—Bread and dough extracts were analyzed utilizing Waters model 2690 HPLC equipped with Waters 2996 photodiode array detector, autosampler (10°C), and column heater at 30°C (Waters Assoc., Milford, Mass., U.S.A.). Reversed phase separation was performed using a $3.0 \times 100\ \text{mm}$, $3.5\ \mu\text{m}$ particle, Symmetry C₁₈ column (Waters Assoc.). The secondary mobile phase (1% aqueous acetic acid: ACN) gradient began at 90:10 progressing linearly to 65:35 in 25 min, 25:75 by 26 min, and 90:10 by 27 min for a total run time of 27 min. Injection volume was $10\ \mu\text{L}$.

Stock solutions of isoflavone standards were prepared as described by Ahn-Jarvis and others (2012). In brief, retention times of standards in HPLC chromatograms and previously published UV-visible spectral signatures were used for identification of unknowns (Murphy and others 2002). A working mixture was used to generate calibration curves with correlation coefficients ($R^2 = 0.998 \pm 0.002$). HPLC peak area of each isoflavone was analyzed and quantified using Empower Pro (version 5.0 Waters Assoc.) software.

Statistical analyses

Statistical analyses were performed using Minitab 15 Statistical Software (Minitab Inc., State College, Pa., U.S.A.) on data collected from sensory evaluation, physicochemical characterization, and isoflavone profiles from dough fermentation with various microorganisms and in finished soy breads. Mean $\pm$ standard deviation was reported and reflect replicate samples (3 replicates/batch, 3 batches). Significant differences (P -value 0.05) in the physical attributes of the Y-B and SD-B were discriminated using an independent t -test, whereas a paired t -test was used to discern significant differences of sensory evaluation tests. A repeated measures analysis of variance was used to model the data for thermal analysis measures with respect to storage duration and bread type as well isoflavone chemical forms in various fermentation strategies in dough and in finished bread. If significant effects were found, then multiple comparisons were performed using Tukey's for pairwise comparisons. Segmentation analysis of soy preference was performed using an Euclidean, single linkage, hierarchical clustering procedure. Analysis of variance (ANOVA) was used to evaluate any significant differences among the various consumer segments.

Results and Discussion

Physical analysis

Dough extensibility was measured to assess differences between sourdough fermentation (SD-D) and yeast-leavening (Y-D) on the gluten performance of soy-supplemented wheat dough. Y-D displayed a significantly greater maximum load and extension at maximum load compared to SD-D ($P < 0.001$) (Table 2), suggesting a greater polymer network and, therefore, greater dough strength in Y-D compared to SD-D (Abang Zaidel and others 2008). These observations are in accordance with similar studies in which the effects of sourdough fermentation or acidification on wheat dough extensibility was investigated, with these observations generally being attributed to microbial proteolysis and the reduction of intra- and intermolecular disulfide bonds of the gluten macropolymer (Thiele and others 2002; Clarke and others 2004).

The dough pH of SD-D was significantly less than that of Y-D ($P < 0.001$) (Table 2), therefore, the net positive charge of the sourdough system may be partially responsible for the observed decreased dough extensibility. Furthermore, the denser crumb of SD-B, as implied by the decreased dough extensibility of SD-D, may have been responsible for a significantly harder bread crumb of SD-B compared to Y-B ($P < 0.05$) (Anderssen and others 2004). The lower pH of SD-D compared to Y-D may have, additionally, been the causative agent for the lighter crust color (L) observed for SD-B compared to Y-B ($P < 0.001$) (Table 2). Maillard browning, the reaction responsible for the formation of brown pigments of bread crust during baking, is largely influenced by pH, with an optimum Maillard browning rate occurring at pH 10 and decreasing with increasing acidity (Wolf from and others 1974).

Sensory analysis

The overall liking of Y-B and SD-B was determined using a 9-point hedonic scale (9 = like extremely; 5 = neither like nor dislike; 1 = dislike extremely), and a paired difference test was used to determine preference between Y-B and SD-B. Hedonic scores indicated that participants moderately liked Y-B (6.9 ± 1.3) and neither liked nor disliked SD-B (4.9 ± 2.0) ($P < 0.001$). Using a paired preference test, 46 out of 55 subjects with significant probability of less than 0.1% selected preference for Y-B (Roessler and others 1978). Sensory evaluation of Y-B and SD-B was designed to assess the overall liking and preference of the 2 soy breads and not sufficiently powered for segmentation analysis. However, segmentation analysis was performed and discernible differences between those with Y-B preference compared to those with SD-B preference were found in our cohort of 55 adults. Those with SD-B preference consumed bread less than once/wk and their bread variety of preference was sourdough breads. This behavior was significantly distinct ($P < 0.01$) from those who preferred Y-B. Moreover, of those who preferred SD-B, hedonic scores for both SD-B and Y-B were very distinct ($P = 0.005$). Hedonic scores for either SD-B or Y-B was greater than 6 in 89% (8/9) of those who preferred SD-B, whereas of those who preferred Y-B, hedonic scores greater than 6 was observed in 15% (7/46). Although our segmentation analysis may be limited in representing analyzable behavior of the general population, findings warrant the need for a larger sample size of sourdough bread consumers and suggest that a participant base that equally prefers sourdough and yeast-leavened bread varieties may provide a more level comparison of these 2 different bread types.

Thermal analysis

TGA and DSC were utilized to determine the moisture content (%) and water distribution of fresh and stored Y-B and SD-B. Additionally, DSC thermograms of fresh and stored bread samples were analyzed for transitions associated with bread crumb staling, including the melting of amylopectin crystals and amylose-lipid complexes. The moisture properties and thermal transitions of interest were fairly similar between fresh Y-B and SD-B, and exhibited similar trends upon storage (Table 3). Exceptions to this generalization were the lower peak temperature associated with ice melting in fresh SD-B compared to fresh Y-B ($P < 0.05$), and lower enthalpy associated with melting of amylose-lipid complexes in fresh and stored SD-B compared to fresh and stored Y-B ($P < 0.05$).

Amylose-lipid complex formation is hypothesized to occur during and/or immediately after baking (Czuchajowska and Pomeranz 1989), and remains stable with storage time (Davidou and others 1996; Lodi and Vodovotz 2008). Given that the initial ingredient composition of SD-D was minimally modified from that of Y-D, the discrepancy in enthalpies associated with the melting of amylose-lipid complexes of SD-B and Y-B is likely attributable to the sourdough fermentation process. The decreased concentration of amylose-lipid complexes in SD-B may have occurred as a result of amylolytic activity of the LAB present in the

sourdough starter culture. While amylolytic activity of LAB is strain specific, several strains of *Lb. plantarum* have been shown to exhibit this activity (Giraud and others 1991; Corsetti and others 1998); therefore, *Lb. plantarum* present in the sourdough starter culture may have resulted in a smaller amylose pool available for complexation with lipids.

The complexation of lipid with amylose is believed to result in a decrease in starch retrogradation; however, this has only been shown to be effective upon extended storage (Davidou and others 1996). This theory is supported by the similar trends observed for amylopectin recrystallization occurring over short-term (10 d) storage in 2 breads (Y-B and SD-B) with significantly different concentrations of amylose-lipid complexes (Figure 1). Furthermore, amylopectin recrystallization observed for Y-B and SD-B was much less than that observed for wheat breads not containing soy following storage (Vittadini and Vodovotz 2003). This observation was previously noted by Vittadini and Vodovotz (2003), who reported an increase in the enthalpy associated with the melting of amylopectin crystals from 0.6 to 3.8 W/g after only 7 d of ambient temperature storage in a control wheat bread.

Isoflavone analysis in soy bread dough

β -glucosidase is the primary enzyme responsible for the chemical conversion of isoflavone glycosides to their respective aglycones, which are hypothesized to be more bioavailable by the body for derivation of their purported health benefits. Given this, changes in the isoflavone profile of soy bread doughs were examined using sourdough fermentation and straight dough techniques (Figure 2). β -glucosidase activity has been observed in previous studies in wheat flour, soy ingredients (soy flour and soy milk powder), yeast (*S. cerevisiae*), and LAB (Zhang and others 2004; Riedl and others 2005). Of the dough types analyzed, the major effects on isoflavone profile were from fermentation type and duration of fermentation. Among the chemical forms examined, time and fermentation interaction was observed only in malonylglucoside forms. LAB-D resulted in the greatest proportional increase in isoflavone aglycones and correspondingly, the greatest proportional decrease in isoflavone simple β -glucosides during proofing (Figure 3A). This change in isoflavone profile may be attributed to the β -glucosidase activity of the particular *Lb. brevis* and *Lb. plantarum* strains present in the preferment. Several strains within each of these species have been identified as possessing β -glucosidase activity (Pyo and others 2005; Di Cagno and others 2010), with *Lb. plantarum* strains frequently displaying the highest activity of LAB species analyzed (Di Cagno and others 2010).

The conversion of isoflavone conjugates to aglycones was less pronounced in the remaining dough types (C-D, Y-D, and SD-D) over the 9 h fermentation period (Figure 3). These findings suggest that the fermentative microorganisms are not the primary catalysts of the observed conversion, since the dough containing no fermenting culture (C-D) also displayed substantial increases in isoflavone aglycones during proofing. Furthermore, the addition of *S. cerevisiae* to the preferment containing LAB appears to hinder the conversion of conjugated isoflavones to aglycones, evidenced by a significantly lower increase in isoflavone aglycones observed in SD-D compared to LAB-D.

Isoflavone profiles in soy bread

The proportion of aglycones in SD-B was almost double of that in Y-B (Figure 4). Correspondingly, the simple β -glucoside pool of SD-B was significantly smaller than that of Y-B ($P < 0.001$). A similar increase in the aglycone pool of soy bread was observed upon the addition of almond powder (5% w/w), which is a naturally rich source of β -glucosidase (Vodovotz 2007). The deglycosylation of isoflavones by sourdough fermentation, however, is advantageous to the baking industry, as this technique does not require the addition of an allergen.

The proportion of isoflavones reported as aglycones in SD-B was significantly lower than that of SD-D at T9 of fermentation, which is likely attributable to ongoing conversion of isoflavones by β -glucosidase during the lengthy (approximately 4 h) extraction process within the dough matrix. Moreover, the proportions of acetyl- and malonyl-glucosides varied between bread types ($P < 0.001$), with the malonyl-glucosides representing one of the largest pools of isoflavones in both Y-B and SD-B. Notably, the isoflavone profiles of Y-B and SD-B did not change after 10 d of room temperature storage (data not shown). This finding suggests that the isoflavone profile within a dough system is stabilized once baked and sustained over short-term storage.

Conclusion

The physical, thermal, and chemical properties of sourdough and yeast-leavened soy breads, as well as overall liking and preference between these products, were evaluated to determine the effect of sourdough fermentation on the overall loaf quality of a soy-supplemented wheat bread. Physical analyses displayed several significant differences between bread types, including a less extensible dough, harder bread crumb, and lighter crust color of SD-B compared to Y-B, all of which may be attributable to a significantly lower pH of SD-D compared to Y-D. Sensory analysis revealed a higher overall liking and preference for Y-B compared to SD-B, however, this may have been a reflection of overall participant preference for yeast-leavened bread varieties. Thermal analyses of Y-B and SD-B displayed similar moisture and starch retrogradation patterns after 10 d of room temperature storage, despite significantly different enthalpies associated with the melting of amylose-lipid complexes. HPLC analysis of SD-B, however, displayed almost double the proportion of isoflavones occurring in the aglycone form compared to Y-B (32% and 17%, respectively). These findings warrant further investigation of sourdough fermentation as a processing technique for enhancement of isoflavone aglycones in soy-based baked goods.

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Figure 1.

Typical DSC thermograms for fresh and stored SD-B and Y-B displaying the enthalpy associated with the melting of amylopectin, and the melting of amylose-lipid complexes in fresh bread (inset).

Figure 2.

Overlay of chromatograms displaying a representative isoflavone profile of soy sourdough (SD-D) at the beginning (0 h) and end (9 h) of fermentation at 30 °C. The identities of the labeled peaks are as follows: (1) daidzin, (2) glycitin, (3) genistin, (4) malonyldaidzin, (5) malonylglycitin, (6) acetyldaidzin, (7) acetylglycitin, (8) malonylgenistin, (9) daidzein, (10) glycitein, (11) acetylgenistin, (12) genistein.

Figure 3.

Isoflavone profile of 4 chemical forms at start (0 h) and end (9 h) of fermentation. ANOVA and Tukey's *post hoc* test was used to determine significant differences. Letters represent where differences were observed.

Figure 4.

Isoflavone profiles (aglycones and glucosides expressed as percent of total isoflavone concentration) and total isoflavone concentration for Y-B and SD-B. Mean $\pm$ SD of triplicate analyses of 3 batches of soy breads. ANOVA and Tukey's *post hoc* test was used to discern significantly different ($P < 0.05$) values and are annotated by differing letters.

Table 1

Ingredients and formulations (% wet basis) for Y-B and SD-B.

Ingredient	Manufacturer, location	Formulation (% wet basis)	
		Y-B	SD-B
Water	–	36.6	26.2
Bread flour (13% protein)	Bay State Milling Co., Quincy, Mass., U.S.A.	40.7	30.2
Soy flour (53% protein)	ADM, Decatur, Ill., U.S.A.	12.2	12.3
Soy milk powder (49% protein)	Devansoy, Carroll, Iowa, U.S.A.	4.1	4.1
Sugar	Gordon Food Service, Grand Rapids, Mich., U.S.A.	2.0	2.0
Shortening	The J.M. Smucker Co., Orrville, Ohio, U.S.A.	2.0	2.0
Wheat gluten	Bob's Red Mill, Milwaukie, Oreg., U.S.A.	1.0	1.0
Salt	U.S. Foodservice, Columbia, Md., U.S.A.	0.8	0.8
Dough conditioner	Caravan Ingredients, Lenexa, Kans., U.S.A.	0.1	0.1
Yeast	Lallemand, Montréal, QC, Canada	0.4	0.0
Preferment	–	0.0	21.2

Y-B, yeast-leavened soy bread; SD-B, sourdough soy bread.

Table 2

Physical properties for Y-D/Y-B and SD-D/SD-B.

	Y-D/Y-B	SD-D/SD-B
Dough maximum load (N)	0.61 ± 0.11 [§]	0.41 ± 0.13 [§]
Dough extension at maximum load (mm)	29.71 ± 4.76 [§]	17.13 ± 2.45 [§]
Dough pH	6.07 ± 0.05 [§]	4.22 ± 0.14 [§]
Specific loaf volume (cm ³ /g)	2.3 ± 0.1	2.2 ± 0.1
Bread crumb hardness (N)	13.54 ± 1.60 [§]	17.04 ± 1.81 [§]
Crust lightness (L)	33.46 ± 1.03 [§]	36.69 ± 1.65 [§]
Crust chromatic component a*	+14.95 ± 0.95	+15.37 ± 1.18
Crust chromatic component b*	+16.71 ± 1.79 [§]	+21.88 ± 1.05 [§]

Y-D/Y-B, yeast-leavened soy dough and bread; SD-D/SD-B, sourdough soy dough and bread.

Mean ± SD reported of triplicate analyses from 3 batches of dough or bread.

[§]Statistical significance ($P < 0.05$) determined by independent *t*-test.

Table 3

Moisture and thermal properties for fresh and stored Y-B and SD-B.

	Y-B		SD-B	
	Fresh	Stored	Fresh	Stored
Moisture content (%)	42.33 ± 0.55 ^a	41.66 ± 0.59 ^a	41.99 ± 0.98 ^a	41.95 ± 0.52 ^a
Ice melt enthalpy (J/g)	82.45 ± 3.82 ^a	70.61 ± 4.53 ^b	83.62 ± 2.12 ^a	75.28 ± 4.67 ^b
Ice melt peak temperature (°C)	-2.57 ± 0.37 ^a	-3.62 ± 0.53 ^{bc}	-3.45 ± 0.41 ^b	-4.14 ± 0.32 ^c
FW (%)	24.74 ± 1.15 ^a	21.19 ± 1.36 ^b	25.10 ± 0.64 ^a	22.59 ± 1.40 ^b
UFW (%)	17.58 ± 0.99 ^a	20.47 ± 1.88 ^b	16.89 ± 1.11 ^a	19.36 ± 1.71 ^b
Amylopectin melt enthalpy (J/g)	0.38 ± 0.13 ^a	1.51 ± 0.23 ^b	0.40 ± 0.14 ^a	1.54 ± 0.16 ^b
Amylose-lipid melt enthalpy (J/g)	1.39 ± 0.15 ^a	1.09 ± 0.12 ^b	0.68 ± 0.20 ^c	0.71 ± 0.07 ^c

Y-B, yeast-leavened soy bread; fresh (day 0) and stored (day 10).

SD-B, sourdough soy bread; fresh (day 0) and stored (day 10).

Mean ± SD reported of triplicate analyses from 3 batches of bread.

Significant difference is indicated by different superscripts, ANOVA and Tukey's HSD *post hoc* test ($P < 0.05$).