Available online at www.sciencedirect.com

ScienceDirect

Food Chemistry 110 (2008) 554–561

FOOD CHEMISTRY

www.elsevier.com/locate/foodchem

Physical properties and water state changes during storage in soy bread with and without almond

Alessia Lodi [a], Yael Vodovotz [b,*]

[a] *CRUK Institute for Cancer Studies, The University of Birmingham, Edgbaston, Birmingham, B15 2TT, UK*
[b] *Department of Food Science and Technology, The Ohio State University, Parker Food Science and Technology Building, 2015 Fyffe Road, Columbus, OH 43210, United States*

Received 9 October 2007; received in revised form 11 January 2008; accepted 11 February 2008

Abstract

Soy bread was formulated to impart different isoflavone profiles by addition of almond: a natural source of β-glucosidase. In this study, the influence of almond powder on loaf quality and storage stability of soy bread was investigated using thermal analysis, texture, loaf volume and air cell imaging techniques. Lipids introduced with the almond fraction, served as mediators to strengthen the interaction between wheat and soy protein, thereby increasing dough extensibility. Loaf quality of the almond soy bread was therefore improved since the collapse of air cells during bread preparation was prevented. This in turn favored better loaf quality. Additionally, water binding strength and changes thereof during storage, probed by thermal analysis, were found to significantly hinder the staling rate during prolonged storage of soy-containing breads.

Keywords: Soy bread; Almond; Differential scanning calorimetry; Thermogravimetric analysis; Air cell imaging

1. Introduction

The addition of soy to food products has generated growing interest in the last decade due to the health promoting activity of soy protein, and possibly other components, such as isoflavones, found in soybean derived products (Potter, 1998). Soy ingredients are commonly added to white bread in low amounts (1–3%) to improve whitening and lower the staling rate (Bohn & Favor, 1945; Cotton, 1974; Hoseney, Rao, Faubion, & Sidhu, 1980; Maga, 1975). Additionally, others have studied the effects of a greater quantity of soy ingredients (up to about 15%) added to bread, intended at producing food products rich in high quality protein (Chen & Rasper, 1982; Fleming & Sosulski, 1977; Pomeranz, Shogren, & Finney, 1969a, 1969b). Soy has also been considered as a possible substitute for gluten in gluten-free bread products for a select population affected by celiac disease (Sanchez, Osella, & de la Torre, 2004).

The addition of various soy ingredients at levels higher than 6–8% was shown to dramatically affect loaf quality, causing significant decrease in volume and increase in air cell density (Brewer, Potter, Sprouls, & Reinhard, 1992). These deleterious effects were attributed to the lack of interactions between the gluten proteins due to dilution with the soy fraction (Brewer et al., 1992; Knorr & Betschart, 1978), the lack of interactions between wheat and soy proteins (Hyder, Hoseney, Finney, & Shogren, 1974; Ryan, Homco-Ryan, Jenson, Robbins, Prestat, & Brewer, 2002), the dissociation of the high molecular weight protein complexes (Lampart-Szczapa & Jankiewicz, 1982) and the competition for water molecules between the wheat and the non-gluten proteins (Knorr, & Betschart, 1978). Weakened interactions between proteins prevent the formation of an elastic bread matrix, critical in maintaining intact air cells (Brewer et al., 1992; Lampart-Szczapa, & Jankiewicz, 1982) and promoting gas retention (Fleming, & Sosulski, 1977) during fermentation, resulting in inferior crumb

---

* Corresponding author. Tel.: +1 (614) 247 7696; fax +1 (614) 292 0218.
E-mail address: vodovotz.1@osu.edu (Y. Vodovotz).

0308-8146/$ - see front matter © 2008 Elsevier Ltd. All rights reserved.
doi:10.1016/j.foodchem.2008.02.032

structure and loaf volume. However, Fleming and Sosulski (1977) showed that dough containing 15% soy flour has a higher gassing power than traditional wheat bread, which indicates appropriate fermentation. When combined with other minor dough formulation changes (e.g. inclusion of larger amount of water), levels of soy flour use up to about 10% were shown to have minimal impact on loaf quality (Lampart-Szczapa, & Jankiewicz, 1982; Mizrahi, Zimmermann, Berk, & Cogan, 1967).

In early studies, it was also observed that additions of up to 30% of full fat soybean flour did not significantly affect loaf quality nor water absorption and best results were obtained when higher levels of yeast and shortening were used. However, when defatted soybean flour was used, only about 10% soybean flour could be added successfully to the product, when incorporated simultaneously with higher amounts of water (Bailey, Capen, & LeClerc, 1935), indicating the importance of the lipid fraction in the formation of the soy-containing dough.

Improvements in loaf quality were observed in various studies involving addition of lipids in various forms (such as dough conditioners, lecithin, glycolipids, etc. (Mizrahi et al., 1967; Pomeranz et al., 1969a, 1969b; Tsen, Hoover, & Phillips, 1971)) to soy-containing bread formulations. One explanation proposed to explain this behavior relates to lipids disrupting direct interactions between glutenin and gliadin. Therefore, upon addition of polar lipids to the wheat-soy system, lipids and soy protein could interact concurrently with glutenin and gliadin to form a stronger network (Aidoo & Tsen, 1973a, 1973b).

Aside from fresh loaf quality improvements, lipid addition may also impact storage stability. One of the most apparent changes occurring in bread during staling, which has been traditionally used to characterize staling rate, is firming. Factors such as retrogradation of amylopectin and changes in water states have been linked to the increased firming during prolonged storage. Various methods have been developed to measure these parameters such as the Instron universal testing machine or the texture analyzer (Ponte & Ovadia, 1996) to evaluate firmness and differential scanning calorimetry (DSC), X-ray crystallography, Fourier transform infrared (FTIR) (Osborne, 1998; Schiraldi, Piazza, & Riva, 1996) to quantify extent of crystalline amylopectine. Water binding states and in particular the “freezable” water fraction and its changes during storage (Baik & Chinachoti, 2000; Vodovotz, Hallberg, & Chinachoti, 1996) have also been probed by DSC while other thermal techniques, such as dynamic mechanical analysis (DMA) and thermalgravimetric analysis (TGA) have been used by many authors to characterize water states and populations having different binding strength to the solid bread matrix (Baik & Chinachoti, 2000; Fessas & Schiraldi, 2001; Vodovotz et al., 1996).

Recently patent pending procedure and formulation of soy bread of fully acceptable quality were developed in our laboratory (Vodovotz & Ballard, 2002). A variant of the soy bread was also produced by incorporating 5% (w/w dry ingredients) raw ground almond to the regular soy bread formulation (Vodovotz, Zhang, & Schwartz, 2004). Almond addition both altered the isoflavones profile (almonds are a rich source of β-glucosidase which helps cleave the sugar moiety from glucosides yielding aglycones, (Vodovotz, 2007)) and the lipid content (increased by about 2.5%) of the final product. Therefore a study to characterize the differences between soy breads with and without almond, both fresh and during storage was designed to ascertain whether and eventually how the almond addition affects water states and staling behavior in soy bread.

2. Materials and methods

2.1. Samples preparation

Soy bread samples were prepared according to Patent Pending procedures and formulations (Vodovotz et al., 2002, 2004). Briefly, the ingredients were mixed in a Hobart mixer until a homogeneous dough was obtained, hand-kneaded for about 5 minutes or until ready and formed into ~910 g (2 lb) loaves of proper shape, proofed (CM2000 combination module, InterMetro Industries Corp, Wilkes-Barre, PA) at 48 °C for about 40–45 min, and baked (jet air oven, model: JA14, Doyon, Liniere, Quebec, Canada) at 160 °C for 50 min. Formulation of soy bread and source of the ingredients are included in Table 1. Five percent raw, ground almond (Wild Oats Markets, Inc., Boulder, CO) was added to the regular soy bread (SB) to produce the almond enriched variant (ASB). After baking, the loaves were allowed to cool to room temperature for about 4 h and were then sealed in polyethylene bags to prevent moisture loss. One set of experiments was

Table 1
Formulations of the regular soy bread (SB) (footnotes include the manufacturers of ingredients)

Ingredients	Soy bread formulation (%w)
Water	45.35
Soy milk Powder[a]	6.61
Soy Flour[b]	19.92
Wheat Flour[c]	17.52
Gluten[d]	2.30
Dough Conditioner[e]	0.20
Sugar	4.50
Yeast[f]	1.00
Salt	0.90
Shortening[g]	1.70

[a] Soy Milk Powder, Devansoy Farms, Carrol, IA.

[b] Baker’s Soy Flour, ADM Protein Specialties Division, Decatur, IL.

[c] Bakers High Gluten Enriched Bromated Wheat Flour Bleached, General Mills Operations, Inc Minneapolis, MI.

[d] Vital Wheat Gluten with Vitamin C, Hodgson Mill, Inc., Teutopolis, IL.

[e] Dough Conditioner, Caravan Products Company Totowa, NJ.

[f] Red Star Instant Active Dry Yeast, Universal Foods Corporation, Milwaukee, WI.

[g] Crisco All-Vegetable Shortening, 50% less saturated fat than butter, Procter & Gamble, Cincinnati, OH.

immediately run after cooling and was designated as "day 0" of the storage study. Samples were stored under accelerated staling conditions (4 °C) between analyses and allowed to equilibrate for 3 h at room temperature prior to conducting the experiments on small crumb portions obtained from the loaf center, unless otherwise noted.

2.2. Specific loaf volume measurement

Loaf volume was determined using the standard rapeseed displacement method (method 10-05 (AACC, 2000)) on intact loaves. Each loaf was weighed and the specific loaf volume was obtained from the ratio of volume and weight. The reported data are the result of at least five loaves per type of bread.

2.3. Moisture content

Moisture content in bread crumb samples was measured by weight difference before and after drying of the samples (initial sample weight about 2 g) in a vacuum oven (modified method 44-40 (AACC, 2000)). The results obtained from the vacuum oven procedure were also compared to the thermogravimetric analysis (TGA) results.

Samples from each day of the storage study were analyzed at least in triplicate.

2.4. Crumb air cells evaluation

The C-Cell Bread Imaging System (Calibre Control International LTD, Warrington, UK) was used to acquire, under standardized conditions, high resolution images of bread slices (loaf sliced into ~1 cm thick slices; fourth and fifth slices were considered). Image analysis of the slices was accomplished using the dedicated software (C-Cell, Version 2 Software, Calibre Control International LTD, Warrington, UK), for quality evaluations (e.g., determination of the number and size of the air cells, thickness of the air cells walls).

2.5. Crumb firmness measurement

Firmness of bread crumb was determined using the Instron universal testing machine (Instron Corp., Canton, MA) according to the procedure described in the AACC method 74-09 (AACC, 2000). Samples of 20 × 20 × 25 (height) mm^3 were compressed by 40% of their height and the maximum compressive load was considered as a measure of firmness.

2.6. Thermogravimetric Analysis (TGA)

About 20 mg of bread crumb, obtained from the center of the loaves, was placed in a previously tared stainless steel pan (PerkinElmer Life And Analytical Sciences, Inc., Boston, MA) inside a thermogravimetric Analyzer, model 2950 (TA Instruments, New Castle, DE). Samples were heated from room temperature (~20 °C) up to 150 °C at the rate of 5 °C/min, and held isothermally for 5 min. Thermograms of the sample weight as a function of temperature and its first derivative were considered for the analysis.

The moisture content of the sample on a wet basis was determined by weight loss from the beginning to the end of the experiment.

The first derivative curve was deconvoluted to a sums of three Gaussian peak functions to determine the presence of one or more water populations (weight loss was presumed to be entirely due to water loss (Fessas et al., 2001)) and the range of temperature of vaporization for each population. Fitting procedures were accomplished using Matlab 6.5 (The MathWorks, Inc., Natick, MA).

2.7. Differential scanning calorimetry (DSC)

Samples were prepared, immediately before analysis, by weighing and sealing about 10 mg of the bread crumb in large volume stainless steel sample pans and lids fitted with O-rings (PerkinElmer Life And Analytical Sciences, Inc., Boston, MA), to prevent moisture loss during analysis. Samples and reference pans (left empty) were placed inside a Differential Scanning Calorimeter, model 2920, equipped with a Refrigerated Cooling System (TA Instruments, New Castle, DE).

The experimental procedure entailed cooling of the sample from room temperature (about 22 °C) to −50 °C at a rate of 5 °C/min. The sample was then held isothermally for 5 min, heated at the rate of 5 °C/min up to 140 °C, held again isothermally for 5 min and cooled down to 25 °C at the rate of 5 °C/min.

The transitions observed in the thermograms were analyzed using the Universal Analysis™ software (TA Instruments, New Castle, DE). Enthalpies of transition were estimated by integration of each peak found in the thermogram (heat flow (W/g of sample) vs. time (s)) and were expressed in J/g of sample.

The percent "freezable" water (FW; per unit weight of water in the sample) was calculated from the peak about 0 °C using the following equation:

$$\%\mathrm{FW} = \frac{A}{\lambda \cdot mc} \tag{4.2}$$

where A is the integrated area under the endothermic peak of water fusion, λ is the specific latent heat of fusion of water (334 J/g) and mc is the moisture content of the sample under analysis.

2.8. Dynamic mechanical analysis (DMA)

Bread samples were prepared for the analysis by cutting a 1 cm thick slice and compressing (Carver press, Carver Inc., Summit, NJ) the crumb to about 3 mm. Rectangular strips (about 25 × 9.5 mm) were cut from the pressed crumb using a custom built cutter and placed inside a dynamic mechanical analyzer, model 2980 (TA Instruments, New

Castle, DE) equipped with a Dual Cantilever clamp used in the Single Cantilever mode.

The experimental procedure consisted of a linear ramp of temperature performed in concurrence with an oscillating, constant frequency (1 Hz) small deformation (10 μm). The sample was initially equilibrated to −80 °C and held isothermally for 2 min. Subsequently, the temperature was ramped up to 50 °C at the rate of 2 °C/min.

The analysis of the results was performed using the Universal Analysis™ software (TA Instruments, New Castle, DE).

To investigate thermomechanical properties changes occurring in soy breads during storage, the position of the flex and the slope in the flex point of the storage modulus transition about 0 °C were determined for each sample at various days of storage. The temperature corresponding to the flex point was determined as the temperature of the first derivative minimum, and the slope of the storage modulus curve in the flex point was the value of the first derivative in its minimum about 0 °C.

3. Results and discussion

3.1. Improved loaf macroscopic quality upon almond addition

Loaf specific volume (Table 2) of ASB was found to be significantly greater than SB (an increase of almost 10%). This outcome is most likely due to the lipid fraction (Mizrahi et al., 1967; Pomeranz, Shogren, & Finney, 1969; Pomeranz et al., 1969) that is included into soy bread upon almond addition. In fact, 50% of almond kernels' weight is comprised of lipids. Additional support for the role of lipids in the increased loaf volume was derived from the comparable loaf volume of the ASB (Table 2) and that of the traditional soy bread with added almond lipid extract (instead of whole ground almond) (2.32 ± 0.03 cm^3/g (Lodi, 2006)).

Image analysis of pictures of bread slices, obtained using the C-Cell Bread Imaging System showed thicker air cell walls for SB compared to ASB. The number of cells was not significantly different between SB and ASB; however, the variability of both the number of cells per slice and the air cell area was much lower for the almond containing breads (Table 2). A larger number of holes were observed within the crumb of the SB images (data not shown), likely leading to the higher variability of the air cell area data. It can be hypothesized that the addition of almond improved dough extensibility, which prevented air cells from collapsing and thus resulted in a more homogenous air cell size distribution (lower variability than SB) and improved loaf quality (e.g. loaf specific volume). Similar results were reported upon addition of dough conditioners to breads containing relevant amounts (up to 15%) of non-wheat proteins (Fleming et al., 1977).

Table 2
Physical properties measured for soy bread (SB) and almond soy bread (ASB) samples

	SB	ASB
Moisture content[a] (%)	44.8 ± 0.3	42.8 ± 0.5
Specific loaf volume (cm^3/g)	2.13 ± 0.06	2.33 ± 0.05
Air cells wall thickness (pixels)	3.60 ± 0.07	3.45 ± 0.05
Number of cells per slice	5442 ± 317	5782 ± 166
Air cell area (pixels)	55.9 ± 3.1	58.1 ± 1.5

[a] Wet basis.

Crumb firmness changes during 10 days of storage, measured using Instron Universal Testing Machine, are reported in Fig. 1. Maximum compression load (N) was lower (~2.7 N) for fresh ASB samples as compared to fresh SB (~6.5 N). These results are, at least in part, due to the lower specific loaf volume (~2.1 cm^3/g for SB vs ~2.3 cm^3/g for ASB, Table 2), as well as to the larger cells wall thickness of SB (Table 2): a larger load is required to compress the sample by 40% of its height because of the denser crumb structure of the soy bread sample. During storage the maximum compression load value increased for all the considered samples, as expected. The maximum compression load of ASB remained lower than that of SB during the entire duration of the storage study. After 10 days of storage the absolute compression load value increased by ~5 N for SB, and ~4 N for ASB.

3.2. The addition of almond to soy bread does not affect the amylopectin recrystalization rate, nor the formation of amylose – lipid complexes

The staling process associated with baked goods has been shown to involve amylopectin recrystallization: during storage amylopectin molecules, which in part gelatinized during the baking process, tend to recrystallize with the incorporation of water (Gray & Bemiller, 2003). The amount of retrograded amylopectin in fresh and stored soy breads with (SB) and without almond (ASB) was

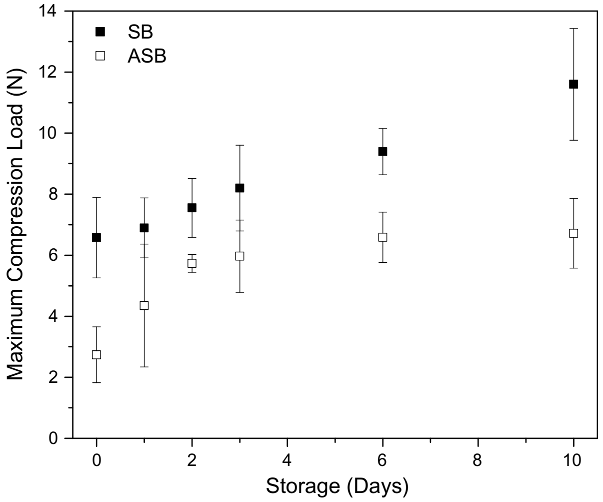

Fig. 1. Firmness changes in soy bread (SB) and almond soy bread (ASB) during 10 days of storage, obtained using the Instron.

evaluated based on the integrated area of the broad DSC endothermic peak, between 40 °C and 60 °C, associated with melting of amylopectin crystals. The amount of retrograded amylopectin was found to increase in both soy breads (Fig. 2). The amount of retrograded amylopectin in soy bread with and without almond was not found to be significantly different (other than on day 3 of storage). It is worth noting that, in traditional wheat bread, amylopectin recrystallization processes occur to a much greater extent and higher rate than in the samples containing soy (Vodovotz, 2007), although both types of bread have moisture content in the range 35–0% which has previously been shown to be the optimal moisture range for the amylopectin retrogradation process.

Another transition observed in DSC thermograms of carbohydrate rich system is a peak found between 100–130 °C, attributed to the dissociation of the amylose–lipid complex (Davidou, LeMeste, Debever, & Bekaert, 1996). The specific melting temperature of these complexes has been shown to depend on the ligands forming the complex, on the conditions of crystallization (Liu, Arntfield, Holley, & Aime, 1997), and on the moisture content of the samples under analysis (Davidou et al., 1996). The area of the peak (normalized per unit weight of wheat flour used in the formulation) observed between 100–110 °C in DSC thermograms of soy breads (both SB and ASB) was measured to be about 0.3 J/mg (of wheat flour in the soy breads) and did not significantly changed during storage for any of the samples. These findings suggest that the additional lipid fraction, introduced in the ASB formulation through almonds, is not involved in the complex formation or at least does not promote the formation of supplemental complexes. Moreover these results are in accordance with previous findings of Czuchajowska and Pomeranz (1989), who hypothesized that these complexes are formed during or immediately after baking.

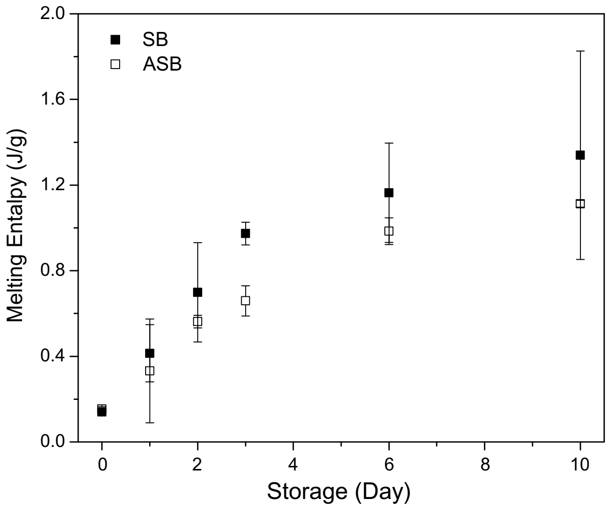

Fig. 2. Heat of melting of amylopectin crystals (normalized per unit weight of wheat flour) in soy bread (SB) and almond soy bread (ASB) obtained from the DSC endothermic peak at ~50 °C during 10 days of storage.

3.3. Moisture retention and distribution during storage contributes to mitigate the staling processes in soy breads

Minimal moisture loss and moisture migration occurred during 10 days of storage under the conditions studied. Moisture content obtained by total weight loss during TGA experiments were compared to vacuum oven values (Table 2) and found not to be statistically different. We have shown a similar trend in these breads using magnetic resonance imaging (MRI) (Lodi, Abduljalil, & Vodovotz, 2007) where a homogeneous water distribution was attributed to the lack of moisture migration, since no driving force was present even immediately after baking. Furthermore it can be inferred that, although the moisture content values included in Table 2 were measured on the central part of the crumb, these can be regarded as being representative of the overall moisture content in the crumb. Compared to standard wheat bread formulas, the homogeneous water distribution in the crumb and the lack of moisture migration suggest that the inclusion of high amounts of soy in bread formula may hinder staling rate of bread (Baik et al., 2000).

3.4. Water binding

Significantly larger amounts of water need to be added to the formulation of soy-containing breads, compared to traditional wheat bread formulations, to improve dough handling and loaf homogeneity. The binding state of water molecules in the bread matrix is important as it affects storage stability of the product. Thermogravimetry, differential scanning calorimetry and dynamic mechanic analysis were used to investigate changes of binding strength and state of the different water populations to bread macromolecules during storage.

3.4.1. Thermogravimetric analysis

The analysis of weight loss first derivative curves, acquired using a thermogravimetric analyzer helped to discriminate and quantify different water populations. These populations were characterized by varying binding strength to the macromolecular bread matrix resulting in water evaporating at different temperature ranges (Fessas & Schiraldi, 2005). Deconvolution with 3 Gaussian peaks (which were found to provide the best fitting; an example is included in Fig. 3a) of the TGA weight loss first derivative data of SB and ASB samples was performed and the temperature of peak maximum and the peak area are reported in Fig. 3b and c, respectively. The area under each peak obtained from the deconvolution procedure is proportional to the amount of water in each population, evaporating in the correspondent temperature range (Schiraldi et al., 1996). Peak temperatures obtained for the different water populations of SB were found to be about 48, 68 and 81 °C, while for ASB these were about 52, 69 and 81 °C. Although the values of temperature and area at each day of storage (Fig. 3b and c, respectively) were different

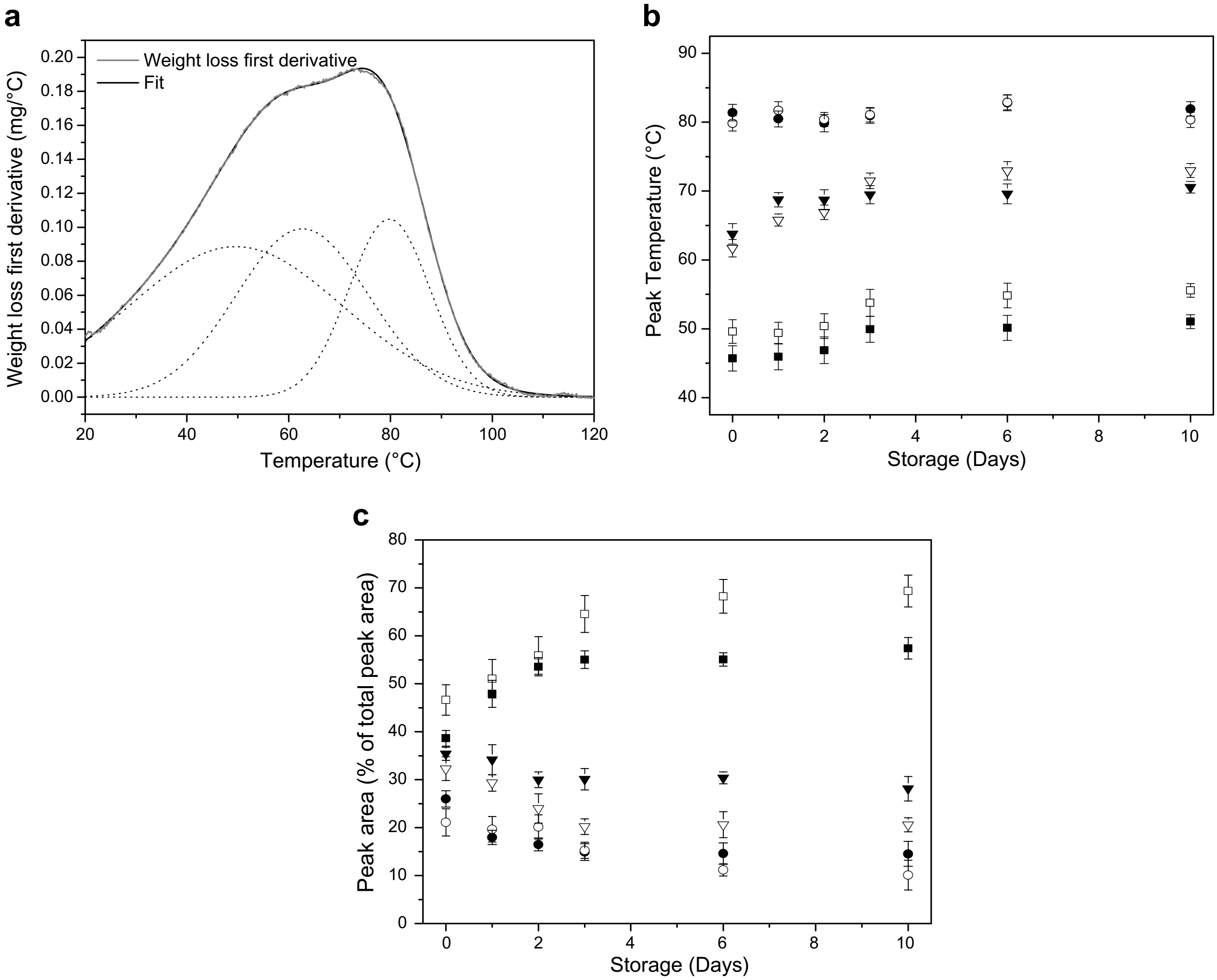

Fig. 3. (a) Deconvolution (solid black line) of the TGA weight loss first derivative curve (grey solid line) for soy bread at day 1 of storage) with 3 Gaussian peaks (black dotted lines), (b) Temperature of maximum and (c) percent of the total area under the curve of peaks obtained from deconvolution of weight loss first derivative (from TGA) curves with 3 Gaussian peak functions for soy bread (SB; full symbols) and almond soy bread (ASB; empty symbols).

for SB and ASB, trends were similar between these samples. Moreover, for both sets of data, the peaks resulting from the deconvolution were found to be broad for the lowest temperature peak (peak width at half height between 40 and 45 °C) and narrower for the higher temperature peaks (24–28 °C for the mid peak and 12–17 °C for the highest temperature peak), as shown in Fig. 3a for soy bread at day 1 of storage. The two peaks at lower temperature (<69 °C) exhibited a shift towards significantly higher temperatures (Fig. 3b, square and triangle symbols) during storage, while the evaporation temperature of the most strongly bound population (circles in Fig. 3b) did not change significantly. The amount of water contributing to each of these populations, shown as percentage in Fig. 3c, depicts a significant decrease in the amount of water evaporating at the higher temperatures (triangles and circles in Fig. 3c), and an increase for the least strongly bound population (squares in Fig. 3c). The curve areas plateau by day 3 of storage. Moreover, in ASB water molecules that were easily removed (peak at lowest temperature, square symbol in Fig. 3b) evaporated later (at higher temperature) than in SB indicating stronger binding of the most easily removed water population to the bread matrix in ASB than in SB.

3.5. Differential scanning calorimetry

Percent “freezable” water (FW) levels in fresh SB and ASB (Fig. 4) were not significantly different ($p < 0.05$). During the first 10 days of storage, the ASB “freezable” water content increased from ~78% to ~83% (expressed as percent weight of total moisture content in the sample), while %FW levels in fresh SB did not significantly change ($p < 0.05$). These results suggest that, during storage, bulk water progressively increases in ASB and more water becomes available for freezing. This is unlike wheat based breads, where “freezable” water levels are usually observed to decrease during storage (Rasmussen & Hansen, 2001). However, Baik and Chinachoti (2000) observed that upon storage of wheat bread without the crust (to minimize moisture content gradients within the loaf section and thus prevent moisture migration from the crumb to the crust)

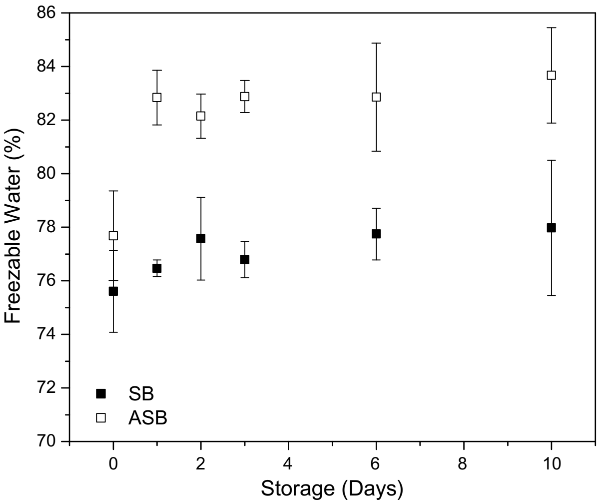

Fig. 4. "Freezable" water in soy bread (SB) and almond soy bread (ASB) obtained from the DSC endothermic peak at ~0 °C during 10 days of storage. "Freezable" water is expressed as percent weight of total water content in bread samples.

"freezable" water levels remained constant during storage. As previously remarked, the addition of soy to bread formulations was found to promote a very homogeneous distribution of water molecules throughout the loaf (Lodi, Abduljalil, et al., 2007; Lodi, Tiziani, & Vodovotz, 2007). Therefore, lack of change in "freezable" water for soy bread can be attributed to the minimal moisture migration from the localized area tested. Furthermore, the addition of soy, which has high affinity for water molecules, may have an effect on "freezable" water, as well (Zhang, 2004). In ASB, "freezable" water increased significantly during the first day of storage and then remained fairly constant. This increase in "freezable" water may be due to the displacement of the water associated with the matrix by the lipid fraction due to the increased lipid content of ASB. We have previously noted (Lodi, Tiziani et al., 2007) that the amount of lipid bound in the matrix of ASB is different and more mobile than that of SB affecting the quality of the product.

3.6. Dynamic mechanical analysis

During the DMA experiments the temperature was not increased beyond 50 °C since significant water loss could change the surface properties of the sample. Moreover, by 50 °C the moduli have reached a low value compared to the initial values and the tan(δ) is noisy. Any appreciable transition above 50 °C would therefore more likely be due to case-hardening artifacts than to real thermomechanical transitions in the sample.

A major transition was observed from the tan(δ) curve about 0 °C as was seen previously in wheat bread (Vodovotz et al., 1996). Changes in slope of E′ and E″ also suggested the existence of another transition at lower temperature (about −50 °C).

Temperature of the flex point of the storage modulus curve (the flex point of the storage modulus curve corresponding to the minimum in the first derivative curve, Fig. 5, inset) was found not to be significantly different between soy breads with and without almond (about −10.0 ± 0.4 °C for fresh SB and −10.5 ± 0.3 °C for ASB) and did not significantly change during storage (data not shown). The change in the slope (absolute value) of the storage modulus curve in its flex point for SB and ASB during 10 days of storage is shown in Fig. 5. The decrease in slope during storage, observed in both SB and ASB corresponds to a slower decrease of storage modulus with temperature in stored bread and may indicate a less homogeneous distribution of frozen water in the bread matrix (Vittadini & Vodovotz, 2003). The melting process occurring over a wider range of temperature may in fact cause a decrease of the rate of change of the thermomechanical properties. Changes in slope during storage were found to be slightly larger in ASB than in SB.

In combination, the results obtained from thermogravimetric, calorimetric and thermomechanical experiments indicate that the amount of bulk/"freezable" water, loosely bound to the bread matrix, increases more appreciably during storage of ASB than SB, as indicated by the increase of "freezable" water (DSC), the amount of the easily removed water molecules (TGA), and the slower thermomechanical changes occurring during ice crystal melting (DMA). The water binding strength of the easily removed molecules progressively strengthens while those at higher temperatures tended to converge to an intermediate condition. Compared to a traditional wheat bread product, water molecules in both soy bread varieties appear to be much more easily removed. In fact, previous reports (Zhang, 2004) identified two water populations (by deconvolution with two Gaussian peaks) at about 80 °C (~30% of the total area) and 100 °C in the fresh sample, which were observed to increase up to about 90 °C and 107 °C after 6 days of storage (Zhang, 2004).

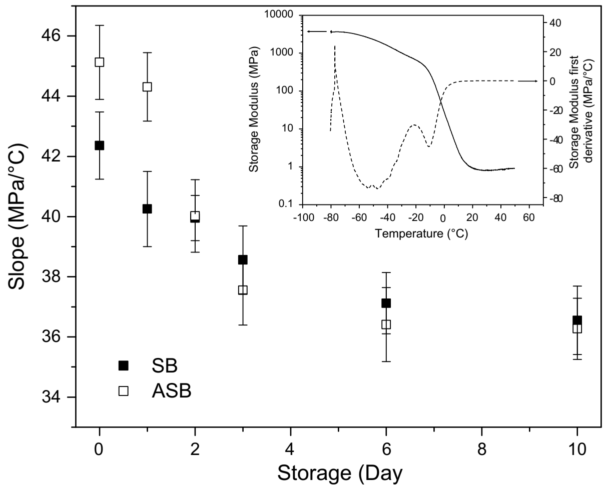

Fig. 5. Slope (absolute value) of the storage modulus (DMA) curve in its flex point in soy bread (SB) and almond soy bread (ASB) during storage. Inset: DMA storage modulus (solid line) and its first derivative curve (dashed line) as function of temperature.

In summary the homogeneous distribution of water contributes to hinder the staling rate in both soy bread varieties, as demonstrated by the various thermal techniques discussed above. Moreover, the addition of almond to soy bread improves loaf quality, probably due to a stronger interaction between proteins of wheat and soy origin favored by the higher lipid content, as indicated by the loaf specific volume and the air cells characteristics (C-cell).

Acknowledgements

The authors wish to thank Margo Caley, Grain Quality and Structure Research, USDA, Manhattan, Kansas for helping with the C-cell experiments.

References

AACC (2000). *Approved methods of the American Association of Cereal Chemists* (10th ed.). MN: St. Paul.

Aidoo, E. S., & Tsen, C. C. (1973a). Influence of surfactants or soy proteins on the extractability, gel filtration, and disc electrophoretic patterns of wheat proteins. *Cereal Science Today, 18*(9), 302.

Aidoo, E. S., & Tsen, C. C. (1973b). Surfactant-protein interactions in model systems. *Cereal Science Today, 18*(9), 301.

Baik, M. Y., & Chinachoti, P. (2000). Moisture redistribution and phase transitions during bread staling. *Cereal Chemistry, 77*(4), 484–488.

Bailey, I. H., Capen, R. G., & LeClerc, J. A. (1935). The composition and characteristics of soybeans, soybean flour and soybean bread. *Cereal Chemistry, 12*, 441–472.

Bohn, R. T., & Favor, H. H. (1945). Functional properties of soybean flour as a bread ingredient. *Cereal Chemistry, 22*, 296–311.

Brewer, M. S., Potter, S. M., Sprouls, G., & Reinhard, M. (1992). Effect of soy protein isolate and soy fiber on color, physical and sensory characteristics of baked products. *Journal of Food Quality, 15*(4), 245–262.

Chen, S. S., & Rasper, V. F. (1982). Functionality of soy proteins in wheat flour/soy isolate doughs. II. Rheological properties and bread making potential. *Canadian Institute of Food Science and Technology Journal, 15*(3), 211–220.

Cotton, R. H. (1974). Soy products in bakery goods. *Journal of the American Oil Chemists' Society, 51*(1), 116A–119A.

Czuchajowska, Z., & Pomeranz, Y. (1989). Differential scanning calorimetry, water activity, and moisture contents in crumb center and near-crust zones of bread during storage. *Cereal Chemistry, 66*, 305–309.

Davidou, S., LeMeste, M., Debever, E., & Bekaert, D. (1996). A contribution to the study of staling of white bread: Effect of water and hydrocolloid. *Food Hydrocolloids, 10*(4), 375–383.

Fessas, D., & Schiraldi, A. (2001). Water properties in wheat flour dough. I: Classical thermogravimetry approach. *Food Chemistry, 72*(2), 237–244.

Fessas, D., & Schiraldi, A. (2005). Water properties in wheat flour dough. II: Classical and Knudsen thermogravimetry approach. *Food Chemistry, 90*, 61–68.

Fleming, S. E., & Sosulski, F. W. (1977). Breadmaking properties of four concentrated plant proteins. *Cereal Chemistry, 54*(5), 1124–1140.

Gray, J. A., & Bemiller, J. N. (2003). Bread staling: molecular basis and control. *Comprehensive Reviews in Food Science and Food Safety, 2*, 1–21.

Hoseney, R. C., Rao, H., Faubion, J., & Sidhu, J. S. (1980). Mixograph studies. IV: The mechanism by which lipoxygenase increases mixing tolerance. *Cereal Chemistry, 57*(3), 163–166.

Hyder, M. A., Hoseney, R. C., Finney, K. F., & Shogren, M. D. (1974). Interactions of soy flour fractions with wheat components in breadmaking. *Cereal Chemistry, 51*, 666–675.

Knorr, D., & Betschart, A. A. (1978). The relative effect of an inert substance and protein concentrates upon loaf volume of breads. *Lebensmittel-Wissenschaft Technologie, 11*, 198–201.

Lampart-Szczapa, E., & Jankiewicz, M. (1982). Changes in the protein complex of wheat dough affected by soybean 11 S globulin: Part 1. The effects of soybean 11 S globulin addition on the technological properties of wheat dough. *Food Chemistry, 9*(4), 307–314.

Liu, H., Arntfield, S. D., Holley, R. A., & Aime, D. B. (1997). Amylose-lipid complex formation in acetylated pea starch-lipid systems. *Cereal Chemistry, 74*(2), 159–162.

Lodi, A. (2006). Physico-chemical and molecular characterization of soy bread containing almond. Ph.D. thesis, The Ohio State University, Columbus, OH.

Lodi, A., Abduljalil, A.M., & Vodovotz, Y. (2007). Characterization of water distribution in bread during storage using magnetic resonance imaging. *Magnetic resonance imaging*.

Lodi, A., Tiziani, S., & Vodovotz, Y. (2007). Molecular changes in soy and wheat breads during storage as probed by nuclear magnetic resonance (NMR). *Journal of Agricultural and Food Chemistry, 55*(14), 5850–5857.

Maga, J. A. (1975). Bread staling. *CRC Critical Reviews in Food Technology, 5*, 443–492.

Mizrahi, S., Zimmermann, G., Berk, Z., & Cogan, U. (1967). The use of isolated soybean proteins in bread. *Cereal Chemistry, 44*(2), 193–203.

Osborne, B. G. (1998). NIR measurements of the development of crystallinity in stored bread crumb. *Analusis, 26*, M55–M57.

Pomeranz, Y., Shogren, M. D., & Finney, K. F. (1969). Improving breadmaking properties with glycolipids. I. Improving soy products with sucroesters. *Cereal Chemistry, 46*(5), 503–511.

Pomeranz, Y., Shogren, M. D., & Finney, K. F. (1969b). Improving breadmaking properties with glycolipids. II. Improving various protein-enriched products. *Cereal Chemistry, 46*(5), 512–518.

Ponte, J. G., & Ovadia, D. Z. (1996). Instrumental methods. In *Baked goods freshness: Technology, evaluation, and inhibition of staling*. New York: Marcel Dekker.

Potter, S. M. (1998). Soy protein and cardiovascular disease: The impact of bioactive components in soy. *Nutritional Reviews, 56*(8), 231–235.

Rasmussen, P. T., & Hansen, A. (2001). Staling of wheat bread stored in modified atmosphere. *Lebensmittel-Wissenschaft und Technologie, 34*(7), 487–491.

Ryan, K. J., Homco-Ryan, C. L., Jenson, J., Robbins, K. L., Prestat, C., & Brewer, M. S. (2002). Lipid extraction process on texturized soy flour and wheat gluten protein-protein interactions in a dough matrix. *Cereal Chemistry, 79*(3), 434–438.

Sanchez, H. D., Osella, C. A., & de la Torre, M. A. (2004). Use of response surface methodology to optimize gluten-free bread fortified with soy flour and dry milk. *Food Science and Technology International, 10*(1), 5–9.

Schiraldi, A., Piazza, L., & Riva, M. (1996). Bread staling: A calorimetric approach. *Cereal Chemistry, 73*(1), 32–39.

Tsen, C. C., Hoover, W. J., & Phillips, D. (1971). High-protein breads. Use of sodium 2-stearoyl lactylate and calcium 2-stearoyl lactylate in their production. *Baker's Digest, 45*(2), 20–23, 26, 74.

Vittadini, E., & Vodovotz, Y. (2003). Changes in the physico-chemical properties of wheat and soy containing breads during storage as studied by thermal analyses. *Journal of Food Science, 68*(6), 2022–2027.

Vodovotz, Y. (2007). Soy enriched bread. In *Technology of functional cereal products cambridge*. UK: Woodhead Publishing, Ltd.

Vodovotz, Y., & Ballard, C. (2002). Formula and Process for Making Soy-Based Bakery Products. A patent application, Docket No. 10-267845.

Vodovotz, Y., Hallberg, L., & Chinachoti, P. (1996). Effect of aging and drying on thermomechanical properties of white bread as characterized by dynamic mechanical analysis (DMA) and differential scanning calorimetry (DSC). *Cereal Chemistry, 73*(2), 264–270.

Vodovotz, Y., Zhang, Y.C. & Schwartz, S.J. (2004). Methods for Enhancing Soy-Containing Foods and Foods Made Thereby. A patent application, Docket No. 22727.

Zhang, Y.C. (2004). Physicochemical properties and isoflavone content of bread made with soy. Ph.D. thesis, The Ohio State University, Columbus, OH.