Available online at www.sciencedirect.com
ScienceDirect
Food Chemistry 109 (2008) 33–41
Food Chemistry
www.elsevier.com/locate/foodchem

Influence of added bean flour (*Phaseolus vulgaris* L.) on some physical and nutritional properties of wheat flour tortillas

Alex A. Anton [a,*], Kelly A. Ross [a], Odean M. Lukow [b], R. Gary Fulcher [a], Susan D. Arntfield [a]

[a] Department of Food Science, University of Manitoba, Winnipeg, MB, Canada R3T 2N2
[b] Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, MB, Canada R3T 2M9

Received 16 August 2007; received in revised form 28 November 2007; accepted 4 December 2007

Abstract

Composite flours containing 15%, 25%, or 35% of small red, black, pinto, or navy bean flours (BF) and wheat were made into tortillas. Dough rheology, firmness, cohesiveness, rollability, and some physical properties of tortillas were negatively affected as BF concentration increased regardless of bean cultivar. Nutritionally, all bean tortillas had significantly higher levels of crude protein, total phenols, 2,2-diphenyl-1-picrylhydrazyl (DPPH$^{\cdot}$) and 2,2′-azinobis-(3-ethylbenzothiazoline-6-sulphonic acid) (ABTS$^{\cdot+}$) *in vitro* antioxidant activity (AA) and antinutritional compounds such as phytic acid (PA) and trypsin inhibitors (TI) than the wheat control. Tortillas to which 35% of small red, pinto and black BF was added had the highest levels of phenols, which were significantly correlated with both DPPH$^{\cdot}$ ($r = 0.99$) and ABTS$^{\cdot+}$ ($r = 0.99$) AA. Compared to raw flours, PA and TI were reduced from 37.37% to 43.78% and from 50% to 66%, respectively, in the tortillas. Overall analysis indicated that tortillas with acceptable texture and improved nutritional profile were produced at 25% substitution.

Keywords: Beans; Tortillas; Texture; Antioxidant activity; Phytic acid; Trypsin inhibitors

1. Introduction

Flour tortillas are unique baked products that have been produced in Mexico for centuries. The major ingredient for production of flour tortillas is wheat flour and the final product can be defined as a flat, circular, light-coloured bread. Tortillas are generally eaten with beans, meats, cheese, avocados, spreads, and other ingredients (Waniska, 1999).

Tortillas are now more popular in United States than bagels, croissants, English muffin, pitas, or any other type of ethnic bread. In 2005, the baking industry in United States showed a consistent increase in the flour tortilla market. While the consumption of fresh bread was up 0.3%, the increase in tortilla sales was up 3.5% in comparison to the previous year (Kuk, 2006). The reconstruction of the tortilla with new nutritional attributes and new formats has been part of this growth (Berne, 2005).

Nutritionally, wheat based flour tortillas are rich in carbohydrates that generate a high glycemic response after ingestion, similar to white bread (Saldana & Brown, 1984). On the other hand, common bean (*Phaseolus vulgaris* L.) is low in fat and rich in proteins, vitamins, complex carbohydrates and minerals. Besides supplementing basic nutritional requirements, dry bean consumption has been associated to reduced risk of heart disease (Anderson et al., 1984; Winham & Hutchins, 2007), obesity (Geil & Anderson, 1994) and cancer (Azevedo et al., 2003; Garcia-Gasca, Salazar-Olivo, Meniola-Olaya, & Blanco-Labra, 2002).

Economically, beans are an important crop in North America, since their production and export has increased significantly in the last decade (Agriculture & Agri-Food Canada, 2005). However, widespread use of beans as a staple food is negatively affected by the presence of

* Corresponding author. Tel.: +1 204 474 9866; fax: +1 204 474 7630.
E-mail address: umanton@cc.umanitoba.ca (A.A. Anton).

0308-8146/$ - see front matter © 2007 Elsevier Ltd. All rights reserved.
doi:10.1016/j.foodchem.2007.12.005

antinutritional factors that might cause important adverse effects for human and animal nutrition. Some of these compounds include phytic acid and trypsin inhibitors (Martin-Cabrejas et al., 2004). While trypsin inhibitors have been reported to be thermolabile and therefore easily inactivated during thermal processing, phytic acid is known to be quite stable in thermal treatments, undergoing only partial hydrolysis (Adb El-Hady & Habiba, 2003; Estévez, Castillo, Figuerola, & Yánez, 1991; Rehman & Shah, 2005).

Nonetheless, consumption of coloured beans may play an important role against oxidative stress due to the presence of polyphenols that possess *in vitro* antioxidant activity (Anton, Ross, Beta, Fulcher, & Arntfield, 2007; Beninger & Hosfield, 2003; Madhujith & Shahidi, 2005). Antioxidants are accredited to scavenge free radicals and reactive oxygen species, playing an important role in preventing oxidative DNA damage and cellular transformation that lead to degenerative diseases (Madhujith & Shahidi, 2005). These compounds have been promoted by health authorities, thus stimulating the consumption of foods rich in antioxidants (USDA, 2005). Increasing the supply of antioxidants in staple foods such as bread and tortillas may provide a safety net for those who cannot or do not want to consume fruit and vegetables. As a result of this trend, attempts to fortify commonly consumed white bread with antioxidants have been reported (Park, Seib, & Chung, 1997; Park, Seib, Chung, & Seitz, 1997).

Because of their nutritional and health promoting properties, the development of value-added bean-based products for new market opportunities in the functional food and nutraceutical industry is being promoted (Singh, 1999). Attempts aimed to improve the nutritional profile of bread by adding hulls and cotyledons of legumes have been reported (Dalgetty & Baik, 2006; Doxastakis, Zafiriadis, Irakli, Marlani, & Tananaki, 2002), however few publications have focused on improving the nutritional profile of wheat based flour tortillas (Friend, Serna-Saldivar, Waniska, & Rooney, 1992; Gonzales-Agramon & Serna-Saldivar, 1988; Serna-Saldivar, Guajardo-Flores, & Viesca-Rios, 2004).

The purpose of the current study was to examine the effect of adding flours from different bean cultivars at varied levels on some physical and nutritional properties of wheat based flour tortillas. Physically, dough rheology and characteristics of the final product such as firmness, cohesiveness, colour, diameter, thickness, and rollability were investigated. Protein content, total phenolics, antioxidant activity, and levels of phytic acid and trypsin inhibitors were determined in order to verify the nutritional changes.

2. Materials and methods

2.1. General

Small red (AE: AC Earlired), black (BV: Black Violet), pinto (AP: AC Pintoba), and navy (GTS: GTS 531) beans were obtained from the Agriculture and Agrifood Canada Research Station in Morden, MB, Canada. The cultivars were grown and harvested in 2005 and exposed to the same environmental conditions in order to avoid external variation. The weight of 100 seeds was determined gravimetrically and expressed as mean ± SD of three determinations. Crude protein content of bean samples were: 22.26% for AE, 25.29% for BV, 25.61% for AP, and 26.42% for GTS.

Whole seeds were ground in a Jacobson pilot scale hammer mill (Model No 120-B, Minneapolis, MN, USA) to pass a 500 μm sieve (35 mesh US Standard Sieve Series). Ground samples were added at different levels (15%, 25%, and 35%) to Canadian hard red spring wheat (Laura variety, crop year 2005; 13.55% crude protein) flour and the composite flours were stored at 5 °C in opaque, closed containers for further use.

2.2. Dough rheology

The effect of adding bean flour on dough-mixing properties was determined using a Brabender Farinograph (Diusburg, Germany) according to the constant flour weight procedure (Method 54-21; AACC, 2000). The parameters determined were % water absorption, dough development time (DDT), and dough stability.

2.3. Tortillas preparation

Flour (100 g) (either wheat or composites) with weight adjusted to 14% of moisture content was added to 9 g of vegetable shortening (Crisco, Markham, ON, Canada), 1.5 g of baking powder (Magic Baking Powder, Kraft Canada Inc., Don Mills, ON, Canada), 1.5 g of sodium chloride (Food Grade, Fisher, Ottawa, ON, Canada) and distilled water. The amount of added water was determined based on the Farinograph Water Absorption (FAB) and previous tests that determined the optimum water addition for making tortillas with the best cohesiveness possible (results not shown). The tests indicated that the final product would achieve its best by adding the FAB value minus 12 g of water. The dry ingredients plus shortening were placed onto a 200 g mixer (National MFG. Co, Lincoln, NB, USA) and mixed for 2 min before water addition. Once water was added, the batter was further mixed for 2 min (35% composites), 3 min (25% composites), or 4 min (15% composites and wheat flour). Mixing time was determined by previous tests that identified the average mixograph peak stability time for each level of substitution (results not shown). After mixing, dough was cut into pieces of 35 g. These pieces were rounded, pinched on the bottom and placed in separate plastic containers, covered with a damp cloth and allowed to rest for 5 min at room temperature. After resting, each 35 g ball was slightly flattened by hand and pressed on a hot press (Doughpro Proprocess Corporation, Paramount, CA, USA) previously heated to 93 °C (top and bottom platens) for 8 s. Thereafter, tortillas were transferred to an electric hot plate preheated at 218 °C and baked for 30 s on the first side, flipped and baked for 40 s on the second side, and flipped and baked for another 10 s on first side. Baked tortillas were cooled on a rack for 1 min and packed in open polyethylene plastic bags. The bags were sealed after 3 h and left at 25 °C overnight. Physical properties of tortillas were evaluated 24 h after production. Each batch yielded four tortillas. Chemical analysis and moisture of tortillas were determined after tortillas were dried at −50 °C, 5 Pa, for 48 h in a freeze dryer (Genesis 25 and 35 Freeze Dryer, SP Industries, Warminster, PA, USA) with samples previously frozen for 16 h at −40 °C.

The raw composite and wheat flours, as well as the freeze-dried tortillas, were analyzed for their moisture content by AOAC method 925.10 (AOAC, 1990). Due to the different moisture content of samples, all calculations were made on a dry matter basis.

2.4. Physical analysis

Tortilla firmness was determined by a puncture test with a TA.XT2 Texture Analyzer (Texture Technologies Corp., Scarsdale, NY/Stable Micro Systems, Godalming, Surrey, UK) equipped with a cylindrical probe (TA 108, 18 mm diameter) with a force of 40 g. Tortillas were placed blistered side down on a tortilla burst rig (in accordance with the manufacturer's instructions) and firmness was measured as the resistance to puncture (peak force). Cohesiveness was calculated as the work during compression (rupture force multiplied by the distance at peak force).

Diameter was measured with a ruler and thickness with a caliper at three different places in each tortilla and the mean was calculated for each (one value was considered for each tortilla). Rollability was evaluated by wrapping a tortilla around a dowel (1.0 cm diameter) and rating the cracking and breakage of the tortilla. Rollability was rated in a scale of 1–6, where 1 = no signs no cracking (best), 2 = edge cracking only, 3 = edge cracking and/or cracking in the center, 4 = cracking and breaking on one side, 5 = cracking and breaking on both sides (clean break) but still rollable, 6 = unrollable.

Colour measurements (CIE L^* a^* b^* colour space) were performed using a Minolta CM-3600d model spectrophotometer. The colour of tortillas was expressed as the average of two L^*, a^*, and b^* readings, where L^* stands for brightness, $+a^*$ redness, $-a^*$ greenness, $+b^*$ yellowness, and $-b^*$ blueness. A white calibration plate was used to standardize the equipment prior to colour measurements.

2.5. Chemical analysis

Nitrogen content was determined by using the Kjeldahl method and was multiplied by a factor of 5.7 to estimate protein content (AOAC, 1990).

For determination of total phenol content and antioxidant activity, 100 mg of finely ground sample was extracted in 2.5 mL of acetone/water (80:20, v/v) (Fisher, Ottawa ON) for 2 h in a rotary shaker. After this period, the samples were centrifuged at 3000*g* in a table centrifuge (GLC-1, Sorval, Newton, CT, USA) for 10 min. Thereafter the supernatant was transferred to a 3 ml syringe (Fisher) and filtered through a 0.45 μm sterile PVDF filter unit (Fisher). The filtrate was collected for further analysis. Preliminary tests on extraction solvents revealed that methanol was unsuitable for extraction of phenolic compounds under such conditions, producing extracts with high degree of turbidity and unacceptable standard deviation in these tests.

The total phenolic content was determined using the Folin–Ciocalteau method (Singleton & Rossi, 1965) as modified by Gao, Wang, Oomah, and Mazza (2002). An aliquot (0.2 mL) of extract was added to 1.5 mL of freshly diluted 10-fold Folin–Ciocalteau reagent (BDH, Toronto, ON, Canada). The mixture was allowed to sit for 5 min and then 1.5 mL of sodium carbonate solution (60 g/L) (Sigma, St. Louis, MO, USA) was added. Afterwards, the mixture was incubated for 90 min and the absorbance read at 725 nm. Acetone/water (80:20, v/v) was used as a blank and ferulic acid (Sigma, St. Louis MO) was used as standard. The results were expressed in mg of ferulic acid equivalents per 100 g of sample. Linearity range of the calibration curve was 20–200 μg ($r = 0.99$).

For measuring the antioxidant activity two methods were employed. Antioxidant activity was initially measured using a modified version of Chen and Ho (1995). For this assay, 200 μL of extract was reacted with 3.8 mL of 2,2-diphenyl-1-picrylhydrazyl ($DPPH^{\cdot}$) solution (6.34×10^{-5} M in methanol). The decreasing absorbance was monitored at 517 nm (Ultraspec 200, Pharmacia Biotech Piscataway, NJ) in the dark at 30 min against methanol blank. The control consisted of 200 μL of acetone/water (80:20, v/v) in 3.8 mL of $DPPH^{\cdot}$ solution. The results were obtained as a percent of discoloration according with the formula:

$$\left[1 - \left(\frac{\text{Absorbance Sample}}{\text{Absorbance Control}}\right)\right] \times 100$$

Simultaneously to the samples, 6-hydroxy-2,5,7,8-tetramethyl chroman-2-carboxylic acid (Trolox) (Sigma) was used as a standard and the results were expressed as μmol of Trolox equivalents per 100 g of sample. Linearity range of the calibration curve was 0.25 to 2.0 μmol ($r = 0.99$).

The second method for determining antioxidant activity was the 2,2′-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid) ($ABTS^{+\cdot}$) radical cation decolorization assay (Re et al., 1999). The ABTS radical cation ($ABTS^{+\cdot}$) was produced by reacting 7 mM ABTS (Sigma) stock solution with 2.45 mM potassium persulphate and allowing the mixture to stand in the dark at room temperature for 16 h before use. The $ABTS^{+\cdot}$ solution (2 days stability) was diluted with methanol to an absorbance of 0.70 ± 0.02 at 658 nm. After addition of 100 μl of extract or Trolox standard to 2.9 mL of diluted $ABTS^{+\cdot}$ solution, tubes were incubated for 15 min at 30 °C and absorbance was measured at 734 nm. Solutions of known Trolox concentrations in acetone/water (80:20, v/v) were used for calibration. Linearity range of the calibration curve was 0.25 to 2.0 μmol ($r = 0.99$).

Phytic acid levels were determined in flours and tortillas by the method of Latta and Eskin (1980). This analysis was done with a chromatographic column (0.7 cm × 15 cm) containing 0.5 g of an anion-exchange resin (100–200 mesh, chloride form; AG1-X8, Bio-Rad Co.). The process was the same as the AOAC method, and only the digestion step was omitted. The Wade reagent (1 mL, 0.03% $FeCl_3 \cdot 6H_2O$ and 0.3% sulfosalicylic acid in distilled water) was added into the extract (3 mL), and the mixture vortexed for 30 s. The absorbance of the supernatant was measured at 500 nm with a UV–vis spectrophotometer.

Trypsin inhibitor activity was measured following the procedure by Kakade, Rackis, McGhee, and Puski (1974), using α-*N*-benzoyl-DL-arginine-*p*-nitroanilidehydrochloride (BAPNA) (Sigma) as the substrate for trypsin. 500 mg of finely ground sample was extracted with 25 mL of 0.01 N sodium hydroxide for 3 h at room temperature in a rotatory shaker. Extracts were centrifuged at 17,500*g* (RC5C, Sorval, Newton, CT, USA) at 4 °C for 20 min, and the supernatants filtered through Nb 1 Whatman filter paper. Thereafter extracts were diluted to 30% in distilled water so that 1.0 mL could inhibit 50% of trypsin activity in the conditions presented herein. Five portions of extracts (0, 0.6, 1.0, 1.4, and 1.8 mL) were pipetted into test tubes and the final volume was adjusted to 2 mL with distilled water. Trypsin solution (2 mL, 20 mg/L in 0.001 M HCl) was added and the tubes were placed in the water bath at 37 °C, followed by addition of 5 mL of BAPNA solution (0.4 mg/mL in Tris-buffer 0.05 M, pH 8.2) previously warmed to 37 °C. After exactly 10 min the reaction was stopped by adding 1 mL of 30% acetic acid to each test tube. The absorbance was read at 410 nm and the reagent blank prepared by adding 1 mL of 30% acetic acid to a test tube containing trypsin and water (2 mL of each) before the BAPNA solution was added. One trypsin unit was arbitrarily defined as an increase of 0.01 absorbance unit at 410 nm per 10 mL of the reaction mixture under the conditions used herein.

2.6. Statistical analysis

All data were recorded as means ± SD and analyzed by GraphPad Instat for Windows (ver. 3). One-way analysis of variance (ANOVA), Tukey tests and two-tail *t*-tests were carried out to test any significant differences between treatments and cultivars. Pearson's correlation coefficient (r) was also applied to establish specific correlations. All tests were performed using $\alpha = 0.05$.

3. Results and discussion

3.1. Effect of added bean flour on dough rheology

The effect of bean flour addition on tortilla dough rheology is summarized in Table 1. Water absorption was increased by addition of bean flour as a function of increased rate of substitution. Substituting wheat flour with 15% bean flour resulted in increases in water absorption of 3–5%. This effect was more pronounced in 35% substitutions, where the increase in water absorption was up to 8%. Since most bean proteins are water soluble (Deshpande, Rangnekar, Sathe, & Salunkhe, 1983; Morales-de-Léon, Vázquez-Mata, Torres, Gil-Zenteno, & Brezan, 2007) and gluten, the most relevant wheat protein, is mostly water insoluble, the higher water absorption of the composites could be related to the high water absorption of the beans (Deshpande et al., 1983). The time required for dough development or time necessary to reach 500 BU of dough consistency (DDT) clearly decreased as bean concentration increased. During this phase of

Table 1
Effect of added bean flours on tortillas dough rheology properties

Flour	Concentration (%)	WA (%)	DDT (min)	Stability (min)
Wheat	100	65 ± 0.1	8.7 ± 0.2	13.7 ± 0.1
AE	15	68.3 ± 0.2a*	8.5 ± 0.5a	6.8 ± 0.25c*
	25	68.85 ± 0.25b*	8.05 ± 0.35a	4.6 ± 0.1b*
	35	69.95 ± 0.25c*	7.85 ± 0.35a	2.7 ± 0.06a*
BV	15	68.3 ± 0.13a*	6.8 ± 0.35a*	5.9 ± 0.1c*
	25	69.2 ± 0.25b*	6.7 ± 0.25a*	3.7 ± 0.1b*
	35	69.8 ± 0.1c*	6.85 ± 0.15a*	2.1 ± 0.1a*
AP	15	67.5 ± 0.58a*	8.40 ± 0.17a	5.93 ± 0.06c*
	25	67.7 ± 0.17a*	8.27 ± 0.40a	4.33 ± 0.29b*
	35	67.76 ± 0.12a*	9.27 ± 0.23b	3.63 ± 0.31a*
GTS	15	68.83 ± 0.46a*	7.1 ± 0.17a*	5.27 ± 0.12c*
	25	69.2 ± 0.52a,b*	6.67 ± 0.29a*	2.73 ± 0.06b*
	35	70.1 ± 0.4b*	7.2 ± 0.25a*	1.8 ± 0.15a*

WA: water absorption; DDT: dough development time.
AE: small read bean variety AC Earlired; BV: black bean variety Black Violet; AP: pinto bean variety AC Pintoba; GTS: navy bean variety GTS 531. All the values are Mean ± SD of three determinations. Data followed by the same character in the same column, within the same bean flour, are not significantly different ($P > 0.05$).
* Significantly different comparing to wheat control using a two-tail *t*-test ($P < 0.05$).

mixing, the water hydrates the flour components and the dough is developed. Dough-mixing studies showed that inclusion of bean flour highly impacted the time the dough maintains its best consistency at the 500 BU line. Dough stability decreased abruptly as bean flour was added. This is logical since beans are absent of proteins that give wheat dough its viscoelastic properties (Deshpande et al., 1983). Therefore, as the concentration of wheat flour, and consequently its gluten content, is decreased by the addition of bean flour the dough rheological properties are negatively affected.

Dough from black (BV) and navy (GTS) bean composite flours demonstrated higher water absorption, and lower DDT and stability. This was probably due to the higher amount of fiber from the seed coats in these flours, which is based on the weight of 100 seeds (AE: 24.46 ± 2.65 g; BV: 17.74 ± 1.65 g; AP: 42.83 ± 1.75 g; GTS: 15.97 ± 0.29 g); when the mass of the seeds is less, the seed coat comprises a larger area relative to one whole seed. Such an effect has been reported by Dalgetty and Baik (2006), who discussed the addition of hulls from legumes on dough rheology.

3.2. Effect of added bean flour on some physical properties of tortillas

The effect of added bean flour on some physical characteristics of flour tortillas is shown on Table 2. Addition of bean flour, regardless of cultivar or concentration, significantly affected firmness and cohesiveness of tortillas. As observed with the dough from composite flours, firmness and cohesiveness decreased as a function of increased substitution of wheat with bean flour. Firmness was reduced from 23.8% to 28.73%, in flours containing 15% bean flour, 37.32–43.66% for 25% substitutions, and 52.02–62.15% for 35% substitutions. Although the effect of level of substitution was significant for all bean cultivars, the effect of bean cultivar on firmness was significant only in tortillas to which 35% bean flour was added (Table 3). Cohesiveness, which accounts for the time and force necessary for rupture

Table 3
Effect of bean cultivar on some physical and nutritional properties of flour tortillas

	Bean flour concentration (%)		
	15	25	35
WA	–*	–**	–***
DDT	–***	–***	–***
Stability	–***	–***	–***
Firmness	NS	NS	–***
Cohesiveness	NS	–*	–***
Diameter	NS	NS	–**
Thickness	–**	–*	NS
Rollability	NS	NS	NS
L^*	–***	–***	–***
a^*	–***	–***	–***
b^*	–***	–***	–***
Protein	–*	–**	–*
TP	–***	–***	–***
AOX[1]	–***	–***	–***
AOX[2]	–***	***	–***
PA Raw Flours	–*	**	–**
PA Tortillas	NS	***	–***
TI Raw Flours	–***	***	–***
TI Tortillas	–***	***	–***

WA: water absorption; DDT: dough development time; L^*: black/white; a^*: green/red; b^*: blue/yellow; TP: total phenol content; AOX[1]: DPPH$^{\cdot}$ antioxidant activity; AOX[2]: ABTS$^{+\cdot}$ antioxidant activity; PA: phytic acid; TI: trypsin inhibitors.
NS: no significant effect ($P > 0.05$);
* $P < 0.05$.
** $P < 0.01$.
*** $P < 0.001$.

Table 2
Effect of added bean flours on some physical properties of wheat tortillas

Flour	%	Firmness (g)	Cohesiveness (g/s)	Diameter (cm)	Thickness	Rollability	L^*	a^*	b^*
Wheat	100	993.44 ± 40.81	10185.57 ± 116.95	16.02 ± 0.62	1.33 ± 0.25	1	81.87 ± 0.43	-0.85 ± 0.2	23.65 ± 3.48
AE	15	757.11 ± 14.93c*	4749.26 ± 82.72c*	16.4 ± 0.32a	1.6 ± 0.11b	1a	74.58 ± 1.22c*	3.8 ± 0.42a*	18.35 ± 0.72b*
	25	622.73 ± 18.53b*	3680.46 ± 126.63b*	16.55 ± 0.48a	1.4 ± 0.15a,b	1a	72.58 ± 0.54b*	4.27 ± 0.33a*	16.85 ± 0.29a*
	35	480.02 ± 5.3a*	2280.86 ± 26.97a*	16.62 ± 0.38a	1.28 ± 0.17a	2b*	68.13 ± 1.13a*	5.46 ± 0.38b*	15.8 ± 0.61a*
BV	15	733.57 ± 6.29c*	4602.63 ± 52.1c*	16.63 ± 0.08a	1.28 ± 0.15a	1a	68.89 ± 1.4b*	-0.26 ± 0.12a	11.16 ± 0.51b*
	25	559.67 ± 7.25b*	2738.55 ± 71.72b*	17.18 ± 0.15b*	1.07 ± 0.1a	1a	59.99 ± 4.61a*	-0.1 ± 0.34a,b*	8.29 ± 0.76a*
	35	413.8 ± 20.41a*	1586.81 ± 134.19a*	17.43 ± 0.15b*	1.07 ± 0.12a	2.5 ± 0.71b*	57.45 ± 1.77a*	0.29 ± 0.26b*	7.43 ± 0.93a*
AP	15	708 ± 38.18c*	4171 ± 222.03c*	16.27 ± 0.15a	1.55 ± 0.08b	1a	76.82 ± 0.93c*	2.36 ± 0.46a*	21.17 ± 0.89a
	25	568.5 ± 9.19b*	2846 ± 61.22b*	16.9 ± 0.29b	1.27 ± 0.15a	1a	73.84 ± 0.75b*	3.04 ± 0.16b*	20.8 ± 0.32a
	35	376 ± 11.31a*	1388.5 ± 2.12a*	17.05 ± 0.23b*	1.13 ± 0.12a	2b*	71 ± 0.42a*	3.76 ± 0.19c*	20.25 ± 0.46a
GTS	15	748.22 ± 21.77c*	4493.48 ± 68.98c*	16.7 ± 0.39a	1.25 ± 0.18a	1a	82.87 ± 0.49a	-0.62 ± 0.16a	22.63 ± 1.21a
	25	588.57 ± 18.36b*	2896.99 ± 97.59b*	16.93 ± 0.34a*	1.13 ± 0.12a	1.5 ± 0.71a,b*	82.69 ± 0.58a	-0.78 ± 0.15a	24.54 ± 1.95a,b
	35	516.8 ± 27.87a*	2337.64 ± 85.75a*	17.2 ± 0.1a*	1.11 ± 0.1a	2.5 ± 0.71b*	82.44 ± 0.36a	-0.8 ± 0.16a	26.13 ± 1.31b

L^*: black/white; a^*: green/red; b^* blue/yellow.
AE: small read bean variety AC Earlired; BV: black bean variety Black Violet; AP: pinto bean variety AC Pintoba; GTS: navy bean variety GTS 531. All the values are Mean ± SD of four determinations. Data followed by the same character in the same column, within the same bean flour, are not significantly different ($P > 0.05$).
* Significantly different comparing to wheat control using a two-tail t-test ($P < 0.05$).

of tortilla, showed more visible differences amongst all the levels of substitutions and cultivars. This parameter was decreased by 53.37–59.05% in 15% bean tortillas, 63.81–73.11% in 25% substitutions, and 77–86.37% in 35% substitutions. The effect of cultivar was significant when tortillas contained 25% and 35% bean flour. In general, small red and navy beans (AE and GTS) composite flours were shown to produce tortillas with the best firmness and cohesiveness, however these parameters were not correlated with dough rheological properties. This negative impact on textural parameters due to addition of increasing levels of alternative ingredients on wheat tortilla formulations have been reported for oat bran and rice bran (Friend et al., 1992) and triticale (Serna-Saldivar et al., 2004). Physical properties such as diameter showed an increasing trend as the level of substitution increased, whereas thickness showed the opposite, indicating thinner tortillas at higher bean flour concentrations. Dalgetty and Baik (2006) showed a reduction in volume of bread resulted from increased levels of substitution of wheat with legume fibers. In the case of tortillas, as puffiness is decreased, demonstrated by decreased thickness, diameter is increased. Factors such as those described for the negative dough properties of bean composite flours may justify the changes in diameter and thickness. Furthermore, it is hypothesized that bean flour addition is greatly affecting the gluten network of flour tortillas. Tortillas containing 35% of bean flour were less rollable than those added of 0%, 15%, or 25% bean flour regardless of cultivar, i.e., tortillas broke more easily during rollability test.

Except for tortillas to which navy bean flour was added, colour was highly affected by incorporating small red, black, or pinto beans in flour tortilla formulations. Black bean flour addition caused the highest change in colour in comparison to the wheat control. Although different than the control, small red and pinto bean flour tortillas did not differ significantly between each other in regards to lightness ($P > 0.05$).

3.3. Effect of added bean flour on some nutritional properties of tortillas

Changes in selected nutritional properties of tortillas are summarized in Table 4. The addition of bean flour to wheat flour was expected to increase the protein content of the final product, since legumes generally contain more proteins than cereals (Tharanathan & Mahadevamma, 2003). In fact, even at the lowest concentration bean tortillas were 13.6% on average richer in protein than the control. Significant increases were observed at all levels of substitution; however, 25% and 35% substitutions resulted in more evident changes. More relevant than that is the consequent enhancement on the amino acid profile of wheat tortillas. Although amino acids were not evaluated in this study, the literature shows that addition of legume flour on wheat flour baked products improves the essential amino acid balance of such foods (Koehler, Chang, Scheier, & Burke, 1987; Shehata, Darwish, El-Nahry, & Razek, 1988; Tharanathan & Mahadevamma, 2003).

Levels of total phenols and antioxidant activities demonstrated significant variations with respect to bean flour concentration and bean cultivar. Tortillas made out of composite flours containing coloured dry beans (AE, BV, and AP) had significantly higher ($P < 0.001$) levels of total phenols and antioxidant activity than those formulated with navy bean flour (GTS) or wheat flour alone; yet navy bean flour tortillas were significantly higher in these parameters than the control, the difference was remarkably higher when comparing navy bean and wheat flour tortillas to those added of coloured beans. The colour of dry beans

Table 4
Effect of added bean flours on some nutritional properties of wheat tortillas

Flour	%	Protein (g/100 g)	TP (mg FAE/100 g)	AOX[1] (μmol TE/100 g)	AOX[2] (μmol TE/100 g)
Wheat	100	10.98 ± 0.5*	16.88 ± 0.3*	37.27 ± 0.27*	297.02 ± 6.69*
AE	15	12.31 ± 0.02a	96.08 ± 3.55a	281.62 ± 11.32a	673.3 ± 8.89a
	25	12.48 ± 0.81a	136.76 ± 3.35b	391.93 ± 6.71b	925.87 ± 24.13b
	35	13.04 ± 0.34a	158.62 ± 1.14c	483.48 ± 2.51c	1128.01 ± 23.89c
BV	15	12.83 ± 0.02a	55.12 ± 6.88a	171.31 ± 7.79a	418.74 ± 9.81a
	25	13.83 ± 0.01b	78.25 ± 3.57b	317.2 ± 9.41b	766.86 ± 10.49b
	35	13.93 ± 0.18b	103.2 ± 5.72c	408.53 ± 6.72c	965.52 ± 11.89c
AP	15	12.45 ± 0.2a	73.77 ± 5.07a	216.38 ± 1.32a	538.42 ± 17.40a
	25	14 ± 0.34b	114.62 ± 5.25b	364.65 ± 12.18b	829.63 ± 26.7b
	35	14.38 ± 0.94b	142.01 ± 5.66c	463.09 ± 1.35c	1082.99 ± 21.9c
GTS	15	12.29 ± 0.45a	23.21 ± 0.41a	44.39 ± 1.06a	407.84 ± 7.54a
	25	13.63 ± 0.57b	24.72 ± 0.78a	55.06 ± 2.05b	412.46 ± 7.84a
	35	14.24 ± 0.04b	27.48 ± 1.29b	79.76 ± 1.99c	417.08 ± 13.06a

TP: total phenol content; FAE: ferrulic acid equivalents; AOX[1]: DPPH$^{\cdot}$ antioxidant activity; TE: trolox equivalent; AOX[2]: ABTS$^{+\cdot}$ antioxidant activity. AE: small read bean variety AC Earlired; BV: black bean variety Black Violet; AP: pinto bean variety AC Pintoba; GTS: navy bean variety GTS 531. All the values are Mean ± SD of four determinations adjusted to dry matter. Data followed by the same character in the same column, within the same bean flour, are not significantly different ($P > 0.05$).

* Significantly different comparing to added bean flours in the same column using a two-tail t-test ($P < 0.05$).

is determined by the concentration of phenolic compounds, such as flavonol glycosides, anthocyanins, and condensed tannins (proanthocyanidins) in the seed coat (Feenstra, 1960). This may explain the differences observed in this study as the colour of navy beans seed coats is cream white, whereas the seed coats of the other cultivars are dark red (AE), black (BV) or predominantly brown (AP). In all tortillas levels of total phenols were significantly ($P < 0.01$) correlated with both DPPH$^{\cdot}$ ($r = 0.975$) and ABTS$^{+\cdot}$ ($r = 0.956$) antioxidant activities. Antioxidant activity determined by the ABTS$^{+\cdot}$ radical (Re et al., 1999) was to a great extent higher than the results from the DPPH$^{\cdot}$ radical assay. Similar observations have however been described by Sánchez et al. (2007) and Wang and Ballington (2007). Also, our findings are in accordance with Madhujith and Shahidi (2005) and Espinosa-Alonso, Lygin, Widholm, Valverde, and Paredes-Lopez (2006), who suggested that while coloured dry beans may be an important source of dietary antioxidants, the method of determination of antioxidant activity plays an important role in the quantification of antioxidant capacity of these foods. As expected, tortillas containing the highest concentration of small red (AE), black (BV), or pinto (AP) bean flours showed the highest antioxidant activities regardless of antioxidant assay. The effect of bean cultivar was clearly visible and significant at all levels of substitution (Table 3). Analysis of variance showed that among coloured bean tortillas, small red had significant ($P < 0.05$) higher levels of total phenols and antioxidant activities (both DPPH$^{\cdot}$ and ABTS$^{+\cdot}$) in all levels of substitution, followed by pinto and black bean tortillas.

The presence of antinutritional compounds has limited the use of bean for human and animal consumption. The phytic acid contents of the bean tortillas varied from 18.15 to 43.97 mg/10 g (Table 5). Tortillas containing higher levels of bean flour, and consequently higher protein content, had higher phytic acid levels. This compound has been reported to be quite stable in thermal treatments, undergoing only partial hydrolysis (Adb El-Hady & Habiba, 2003; Estévez et al., 1991; Rehman & Shah, 2005). Nonetheless, our data shows that a consistent reduction in phytic acid levels occurred in composite flours processed into tortillas. Phytic acid levels were reduced to levels ranging from 37.37% to 43.78% in flours subjected to the type of heat treatment usually employed in hot-press flour tortilla production (considering only the composite flour portion of tortilla formulation, equal to 90%; i.e., excluding shortening, salt and baking powder). Such reductions may be explained on the basis that during cooking inositol hexaphosphate could have been hydrolyzed to lower molecular weight forms, which is in agreement with the work of Alonso, Aguirre, and Marzo (2000), who reported a significant reduction in phytic acid content in beans submitted to extrusion cooking. Similar reductions in phytic acid levels in whole beans processed under various conditions, including soaking, roasting, autoclaving, and pressure-cooking have been reported (Alonso et al., 2000; ElMaki et al., 2007; Shimelis & Rakshit, 2007), however a direct comparison with these studies is difficult since the biochemical reactions involved in the processing of whole seeds and flour of legumes occur in a different manner (Alonso et al., 2000).

Of the protease inhibitors present in legumes, the most important are the trypsin inhibitors, whose action has been thoroughly studied (Adb El-Hady & Habiba, 2003; Deshpande et al., 1983; Estévez et al., 1991). Navy bean

Table 5
Effect of added bean flours on some antinutritional factors of wheat flour and tortillas

Flour	(%)	Phytic acid (mg/10 g)		Trypsin inhibitors (TIU/100 mg)	
		Raw flour	Tortilla	Raw flour	Tortilla
Wheat	100	22.06 ± 0.66*	4.95 ± 0.31*	ND	ND
AE	15	34.68 ± 1.57a	18.76 ± 0.83a	222.73 ± 2.75a	83.76 ± 3.43a
	25	51.12 ± 2.57b	28.07 ± 0.82b	298.93 ± 3.8b	128.86 ± 3.8b
	35	69.5 ± 2.83c	39.07 ± 0.62c	387.35 ± 8.31c	151.08 ± 2.59c
BV	15	36.12 ± 1.17a	18.15 ± 0.34a	263.89 ± 3.4a	77.74 ± 1.59a
	25	55.58 ± 2.13b	29.39 ± 0.57b	423.24 ± 1.95b	119.43 ± 2.4b
	35	73.23 ± 2.25c	41.18 ± 1.53c	588.62 ± 6.02c	146.64 ± 6.29c
AP	15	37.02 ± 1.29a	19.44 ± 0.75a	259.59 ± 2.6a	83.65 ± 1.5a
	25	57.89 ± 2.83b	31.03 ± 1.05b	344.43 ± 5.04b	132.51 ± 4.1b
	35	75.89 ± 1.49c	42 ± 1.4c	518.7 ± 6.66c	183.46 ± 3.94c
GTS	15	38.71 ± 1.84a	19.79 ± 1.36a	364.14 ± 5.05a	149.51 ± 2.21a
	25	58.72 ± 2.01b	33.78 ± 1.87b	588.62 ± 13.52b	207.04 ± 3.15b
	35	77.04 ± 2.83c	43.97 ± 1.06c	828.9 ± 21.97c	301.17 ± 8.13c

TIU: trypsin inhibitory units; ND: non detectable; AE: small read bean variety AC Earlired; BV: black bean variety Black Violet; AP: pinto bean variety AC Pintoba; GTS: navy bean variety GTS 531.

All the values are Mean ± SD of four determinations adjusted to dry matter. Data followed by a different character in the same column, within the same bean flour, are significantly different ($P < 0.05$).

* Significantly different comparing to added bean flours in the same column using a two-tail t-test ($P < 0.05$). All tortillas were significantly different than their respective raw flours for both phytic acid and trypsin inhibitors ($P < 0.05$).

composite flours and tortillas showed the highest levels of trypsin inhibitors, while small red had the lowest (Table 5). These inhibitors are thermolabile and their inhibitory activity can be reduced considerably by an appropriate thermal treatment (Alonso et al., 2000; Shimelis & Rakshit, 2007). In fact, trypsin inhibitors were reduced ranging from 50% to 66% in composite flours made into tortillas (considering only the composite flour portion of tortilla formulation, equal to 90%; i.e., excluding shortening, salt and baking powder) (Table 5). These reductions are in agreement with the findings of Estévez et al. (1991), who found that levels of trypsin inhibitors were reduced from 57% to 62% in beans soaked for 16 h at room temperature and cooked for 60 min. This study reported that the cooked bean samples showed *in vitro* protein digestibility of nearly 90%, as well as good net protein ratios (3.2 in comparison to 4.2 of casein). It has been suggested that inactivation of trypsin inhibitors depends on the physical state of the material. Carvalho and Scarbieri (1997) reported that bean flours submitted to soaking and autoclaving had significantly lower levels of inactivation than whole beans. Our results are comparable with bean flours soaked for 12 h and autoclaved at 121 °C for 5 and 20 min resulting in 55% and 65% reductions, respectively. Since inactivation of trypsin inhibitors in bean tortillas has not been complete, it appears that a thermal treatment longer than that usually applied in flour tortillas manufacturing may be necessary. Nevertheless, based on the levels of substitution studied and on the degree of inactivation reached, it seems that the present bean tortillas would be safe for human nutrition.

4. Conclusions

The effect of bean cultivar on some physical and nutritional properties of flour tortillas was more significant for 25% and 35% substitutions. Composite flours containing 25% bean flour demonstrated acceptable textural parameters and improved nutritional profile in comparison to the wheat control. Tortillas containing navy bean flour (GTS) had no impact on colour, however the levels of phenolics and antioxidant activity were not changed to a great extent in such formulations. Levels of antinutritional compounds were significantly reduced in all levels of substitution, indicating that although these compounds have not been entirely inactivated, bean tortillas could still be safely used for human nutrition.

Further sensory studies and biological trials must be carried out in order to evaluate the acceptability of bean tortillas by a consumer panel and to verify the impact of such foods on animal and human nutrition. Additionally, a modified manufacturing procedure and the application of texture enhancers, such as hydrocolloids, may help improve the nutritional profile and the texture of tortillas added at higher levels of bean flour. Nonetheless, our results appear to indicate that production of bean tortillas is feasible and that their consumption can play an important role in the maintenance of a healthy life style.

Acknowledgements

The authors acknowledge the Agri-Food Research and Development Initiative in care of Manitoba Association of Agricultural Societies and the Saskatchewan Pulse Growers Association for their project sponsorship.

References

AACC (2000). *Approved methods of the American association of cereal chemists* (10th ed.). St. Paul, MN: The Association, Method 54-21.

Adb El-Hady, E. A., & Habiba, R. A. (2003). Effect of soaking and extrusion conditions on antinutrients and protein digestibility of legume seeds. *Lebensmittel Wissenchaft und Technology, 36*, 285–293.

Agriculture and Agri-Food Canada. Canadian pulses and special crops industry: situation and outlook. Retrieved August 20, 2006, from http://www.agr.gc.ca/mad-dam/index_e.php?s1=pubs&s2=bi&s3=php&page=bulletin_18_02_2005-01-28 .

Alonso, R., Aguirre, A., & Marzo, F. (2000). Effect of extrusion and traditional processing methods on antinutrients and in vitro digestibility of protein and starch in faba and kidney beans. *Food Chemistry, 68*, 159–165.

Anderson, J. W., Story, L., Sieling, B., Chen, W. J. L., Petro, M. S., & Story, J. (1984). Hypocholestrolemic effects of oat-bran or bean intake for hypercholestrolemic men. *American Journal of Clinical Nutrition, 48*, 749–753.

Anton, A. A., Ross, K. A., Beta, T., Fulcher, R. G., & Arntfield, S. D. (2007). Effect of pre-dehulling treatments on some physical and nutritional properties of navy and pinto beans (*Phaseolus vulgaris* L.). *LWT – Food Science and Technology*, in press, doi: 10.1016/j.lwt.2007.05.014.

AOAC (1990). *Official methods of analysis*. Washington, DC: Association of Official Analytical Chemists.

Azevedo, A., Gomes, J. C., Stringheta, P. C., Gontijo, A. M. C., Padovani, C. R., Riberio, L. R. Z., et al. (2003). Black bean (*Phaseolus vulgaris* L.) as a protective agent against DNA damage in mice. *Food and Chemical Toxicology, 41*, 1671–1676.

Beninger, C. W., & Hosfield, G. L. (2003). Antioxidant activity of extracts, condensed tannin fractions and pure flavonoids from *Phaseolus vulgaris* L. seed coat colour genotypes. *Journal of Agricultural and Food Chemistry, 51*, 7879–7883.

Berne, S. (2005). Activity building. In Baking and snack, pp. 28–40.

Carvalho, M. R., & Scarbieri, V. C. (1997). Heat treatment and inactivation of trypsin–chymotrypsin inhibitors and lectins from beans (*Phaseolus vulgaris* L.). *Journal of Food Biochemistry, 21*, 219–233.

Chen, C. W., & Ho, C. T. (1995). Antioxidant properties of polyphenols extracted from green and black teas. *Journal of Food Lipids, 2*, 35–46.

Dalgetty, D. D., & Baik, B. K. (2006). Fortification of bread with hulls and cotyledon fibers isolated from peas, lentils, and chickpeas. *Cereal Chemistry, 83*, 269–274.

Deshpande, S. S., Rangnekar, P. D., Sathe, S. K., & Salunkhe, D. K. (1983). Functional properties of wheat–bean composite flours. *Journal of Food Science, 48*, 1659–1662.

Doxastakis, G., Zafiriadis, I., Irakli, M., Marlani, H., & Tananaki, C. (2002). Lupin, soya and triticale addition to wheat flour doughs and their effect on rheological properties. *Food Chemistry, 77*, 219–227.

ElMaki, H. B., AbdelRahaman, S. M., Idris, W. H., Hassan, A. B., Babiker, E. E., & El Tinay, A. H. (2007). Content of antinutritional factors and HCl-extractability of minerals from white bean (*Phaseolus vulgaris*) cultivars: Influence of soaking and/or cooking. *Food Chemistry, 100*, 363–368.

Espinosa-Alonso, L. G., Lygin, A., Widholm, G. M., Valverde, M. E., & Paredes-Lopez, O. (2006). Polyphenols in wild and weedy Mexican common beans (*Phaseolus vulgaris* L.). *Journal of Agricultural and Food Chemistry, 54*, 4436–4444.

Estévez, A. M., Castillo, E., Figuerola, F., & Yánez, E. (1991). Effect of processing on some chemical and nutritional characteristics of pre-cooked and dehydrated legumes. *Plant Foods for Human Nutrition, 41*, 193–201.

Feenstra, W. J. (1960). Biochemical aspects of seed coat colour inheritance in *Phaseolus vulgaris* L. *Meded Landbouwhogeschool Wangeningen, 60*, 1–53.

Friend, C. P., Serna-Saldivar, S. O., Waniska, R. D., & Rooney, L. W. (1992). Increasing the fiber content of wheat flour tortillas. *Cereal Foods World, 3*, 325–328.

Gao, L., Wang, S., Oomah, B. D., & Mazza, G. (2002). Wheat quality: Antioxidant activity of wheat millstreams. In P. Ng & C. W. Wrigley (Eds.), *Wheat Quality Elucidation* (pp. 1–24). St. Paul. MN: AACC International.

Garcia-Gasca, T., Salazar-Olivo, L. A., Meniola-Olaya, E., & Blanco-Labra, A. (2002). The effects of a protease inhibitor fraction from tepary bean *Phaseolus acutifolius* on in vitro cell proliferation and cell adhesion of transformed cells. *Toxicology in Vitro, 16*, 229–233.

Geil, P. B., & Anderson, J. W. (1994). Nutrition and health implications of dry beans: A review. *Journal of the American College of Nutrition, 13*, 549–558.

Gonzales-Agramon, M., & Serna-Saldivar, S. O. (1988). Effect of defatted soybean and soybean isolate fortification on the nutritional physical chemical and sensory properties of wheat flour tortillas. *Journal of Food Science, 53*, 793–797.

Kakade, M. L., Rackis, J. L., McGhee, J. E., & Puski, G. (1974). Determination of trypsin inhibitor activity of soy bean products: A collaborative analysis of an improved procedure. *Cereal Chemistry, 51*, 376–382.

Koehler, H. H., Chang, C., Scheier, G., & Burke, D. W. (1987). Nutrient composition, protein quality, and sensory properties of thirty-six cultivars of dry beans (*Phaseolus vulgaris* L.). *Journal of Food Science, 52*, 1335–1340.

Kuk, T. (2006). A look into the future: The US baking industry in the 21st century. *Cereal Foods World, 5*, 306–310.

Latta, M., & Eskin, M. (1980). A simple method for phytate determination. *Journal of Agricultural and Food Chemistry, 28*, 1313–1315.

Madhujith, T., & Shahidi, F. (2005). Antioxidant potential of pea beans (*Phaseolus vulgaris* L.). *Journal of Food Science, 70*, S85–S89.

Martin-Cabrejas, M. A., Sanfiz, B., Vidal, A., Mollá, E., Esteban, R., & López-Andreu, F. J. (2004). Effect of fermentation and autoclaving on dietary fiber fractions and antinutritional factors of beans (*Phaseolus vulgaris* L.). *Journal of Agriculture and Food Chemistry, 52*, 261–266.

Morales-de-Léon, J. C., Vázquez-Mata, N., Torres, N., Gil-Zenteno, L., & Brezan, R. (2007). Preparation and characterization of protein isolate from fresh and hardened beans (*Phaseolus vulgaris* L.). *Journal of Food Science, 72*, C96–C102.

Park, H., Seib, P. A., & Chung, O. K. (1997). Fortifying bread with a mixture of wheat and psyllium husk fiber plus three antioxidants. *Cereal Chemistry, 74*, 207–211.

Park, H., Seib, P. A., Chung, O. K., & Seitz, L. M. (1997). Fortifying bread with each of three antioxidants. *Cereal Chemistry, 74*, 202–206.

Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M., & Rice-Evans, C. (1999). Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radical Biology and Medicine, 26*, 1231–1237.

Rehman, Z., & Shah, W. H. (2005). Thermal heat processing effects on antinutrients, protein and starch digestibility of food legumes. *Food Chemistry, 91*, 327–331.

Saldana, G., & Brown, H. E. (1984). Nutritional composition of corn and flour tortillas. *Journal of Food Science, 49*, 1202–1209.

Sánchez, C. S., González, A. M. T., García-Parrilla, M. C., Granados, J. J. Q., Serrana, H. L. G., & Martínez, M. C. L. (2007). Different radical scavenging tests in virgin olive oil and their relation to the total phenol content. *Analytica Chimica Acta, 593*, 103–107.

Serna-Saldivar, S. O., Guajardo-Flores, S., & Viesca-Rios, R. (2004). Potential of triticale as a substitute for wheat in flour tortilla production. *Cereal Chemistry, 81*, 220–225.

Shehata, N. A., Darwish, N., El-Nahry, F., & Razek, F. A. A. (1988). Supplementation of wheat flour with some local legumes. *Food/Nahrung, 32*, 1–8.

Shimelis, E. A., & Rakshit, S. K. (2007). Effect of processing on antinutrients and in vitro digestibility of kidney bean (*Phaseolus vulgaris* L.) varieties grown in East Africa. *Food Chemistry, 103*, 161–172.

Singh, S. P. (1999). Production and utilization. In S. P. Singh (Ed.), *Common bean improvement in the twenty-first century* (pp. 1–24). Dordrecht, Netherlands: Kluwer.

Singleton, V. L., & Rossi, J. A. (1965). Colorimetry of total phenolics with phosphomolybdic–phosphotungistic acid reagents. *American Journal of Enology and Viticulture, 16*, 144–158.

Tharanathan, R. N., & Mahadevamma, S. (2003). A Review: Grain legumes a boon to human nutrition. *Trends in Food Science and Technology, 14*, 507–518.

. *United States Department Health and Human Services*. *Dietary guidelines for Americans*. Washington, D.C.: U.S. Government Printing Office.

Wang, S. Y., & Ballington, J. R. (2007). Free radical scavenging capacity and antioxidant enzyme activity in deerberry *Vaccinium stamineus* L. *LWT – Food Science and Technology, 40*, 1352–1361.

Waniska, R. D. (1999). Perspectives on flour tortillas. *Cereal Foods World, 44*, 471–473.

Winham, D. M., & Hutchins, A. M. (2007). Baked bean consumption reduces serum cholesterol in hypercholesterolemic adults. *Nutrition Research, 27*, 380–386.