



Effect of whole amaranth flour on bread properties and nutritive value

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ABSTRACT

This study investigated the effect of replacing wheat flour by whole *Amaranthus cruentus* flour (up to 40 g/100 g) to evaluate its potential utility as a nutritious breadmaking ingredient. The incorporation of amaranth flour significantly increased protein, lipid, ash, dietary fibre and mineral contents. Breads with amaranth have significantly higher amounts of phytates and lower *myo*-inositol phosphates, which could predict low mineral bioavailability at high levels of substitution (30–40 g/100 g). An increase in crumb hardness and elasticity was observed, and tristimulus colour values were significantly affected when the amaranth concentration was raised. Mineral contents, both micro- and macroelements, were increased significantly by the wheat flour substitution. Whole amaranth flour could be used as a partial replacement for wheat flour in bread formulations, increasing the product's nutritional value and raising dietary fibre, mineral and protein levels, with a significant slight depreciation in bread quality when used in proportions between 10 and 20 g/100 g. Thus, the inclusion of amaranth flour could be limited to a maximum proportion of 20 g/100 g, thereby maintaining both product quality as well as the nutritional benefit of this ingredient.

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1. Introduction

Whole grain may increase the nutritional value of bakery products made with refined wheat flour (Marquart, Asp, & Richardson, 2004; Miller Jones, 2009; Sanz-Penella, Collar, & Haros, 2008). One possibility would be to include whole amaranth grain in bread formulations or bakery products. Amaranth is one of the most important pre-Hispanic crops and was part of the diet of the Aztecs, Mayas, Incas and other pre-Colombian civilizations. It belongs to the family of pseudocereals as it has similar properties to those of cereals but botanically does not belong to that family. The genus *Amaranthus* includes more than 60 species that are grown in various parts of the world, such as Central and South America, India, Africa and China (Budín, Breene, & Putnam, 1996). There is increasing interest in the consumption of this genus in Europe, the

USA and Japan, and it is already grown in some parts of these regions. Most species are considered as opportunistic weeds and only three of them, *Amaranthus caudatus*, *Amaranthus cruentus* and *Amaranthus hypochondriacus*, are commonly consumed by humans as a seed or used as a functional ingredient in foods (Gamel, Linssen, Mesallam, Damir, & Shekib, 2006). The amaranth grain can be toasted, popped, extruded or milled into flour and can therefore be consumed as such or included in other cereal products such as bread, cakes, muffins, pancakes, cookies, dumplings, crepes, noodles and crackers. The nutritional quality of amaranth seed is higher than that of most cereal grains, owing to its high protein content and balanced essential amino acid composition (Oszwald et al., 2009). Moreover, amaranth grain protein is rich in lysine, which is usually deficient in cereal grains. The total mineral content has been reported to be generally higher than that observed in cereal grains, especially calcium and magnesium (Alvarez-Jubete, Auty, Arendt, & Gallagher, 2010). On the other hand, it is characterized by higher dietary fibre and lipid content than most cereals and also contains between 50 and 60 g of starch per 100 g of grains (Alvarez-Jubete et al., 2010). Amaranth oil is reported to have high levels of tocotrienols and squalene, which are natural organic compounds that are involved in the metabolism of cholesterol and that could play an important role in lowering LDL-cholesterol in blood (Bodroza-Solarov, Filiocev, Kevresan, Mandic, & Simurina,

Abbreviations: *a** and *b**, colour-opponent dimensions; DRIs, dietary reference intakes; *E*₁, energy during first compression; *E*₂, energy during second compression; *F*₁, force during first compression; *F*₂, force during second compression; *InsP*₆, phytic acid or *myo*-inositol hexakisphosphate; *InsP*₅, *myo*-inositol pentakisphosphate; *InsP*₄, *myo*-inositol tetrakisphosphate; *InsP*₃, *myo*-inositol triphosphate; *InsP*₂, *myo*-inositol diphosphate; *InsP*₁, *myo*-inositol monophosphate; *L**, lightness; VRC, volume recovery coefficient; WAF, whole amaranth flour.

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2008; Budin et al., 1996). The optimal nutritive composition of this seed has made its use attractive as a blending food source to improve the nutritional value of some cereal by-products. Protein content was significantly increased by up to 4.4 g/100 g by using popped amaranth grain or amaranth flour in bread, with maximum levels of substitution of 20 g/100 g (Bodroza-Solarov et al., 2008; Tosi et al., 2002). Mineral and dietary fibre contents in bread and pasta were also significantly increased by flour substitution at levels up to 20 g/100 g (Dyner et al., 2007). With regard to sensory appreciation, bakery products incorporating amaranth have been accepted at levels up to 15–25 g/100 g (Bodroza-Solarov et al., 2008; Sindhuja, Sudha, & Rahim, 2005). Despite all the virtues attributed to amaranth grain, there have been reports of the presence of some anti-nutritional factors, such as phenolic compounds, trypsin inhibitors and phytic acid (*myo*-inositol hexakisphosphate, InsP_6) or its salts, the phytates (Gamel et al., 2006). Phenols and trypsin inhibitors are at such low levels that they do not present a risk to the nutritional status (Bodroza-Solarov et al., 2008). Phytate content in various whole grains of the *Amaranthus* genus has been published, ranging from 4.8 to 9.4 $\mu\text{mol/g}$ (Colmenares de Ruiz & Bressani, 1990; Lorenz & Wright, 1984; Teutonico & Knorr, 1985). Phytic acid intake has been reported to have favourable effects, such as antioxidant function, prevention of heart diseases and anticarcinogen effect, which it performs through its hydrolysis products (Haros et al., 2009; Kumar, Sinha, Makkar, & Becker, 2010). Phytic acid is strongly negatively charged and thus has a great potential for complexing positively charged multivalent cations such as calcium, magnesium, zinc, copper and iron. This has adverse effects on mineral bioavailability, owing to the formation at physiological pH values of insoluble complexes which are non-absorbable in the human gastrointestinal tract (Lopez et al., 2001; Sandberg, Hulthen, & Türk, 1996). The negative health effects of phytates are more significant in developing countries and in risk populations owing to their higher incidence of undergoing mineral deficiencies (Hurrell, Reddy, Juillerat, & Cook, 2003). During the breadmaking process phytate is sequentially hydrolysed by the action of the cereal's own phytate-degrading enzymes. However, whole grain breads still contain high phytate levels owing to a slow and inefficient enzymatic dephosphorylation (Haros, Rosell, & Benedito, 2001; Türk & Sandberg, 1992). Some strategies to reduce or eliminate phytate in breadmaking processes include increasing fermentation time, lowering process pH by the inclusion of sourdough, or adding exogenous phytase (Lopez et al., 2001; Sanz-Penella et al., 2008; Sanz-Penella, Tamayo-Ramos, Sanz, & Haros, 2009; Türk & Sandberg, 1992).

Much of the published research on phytate content in baking products has focused on whole grain breads made from wheat, rye, rice or mixtures of them, but no data are available for the amount of phytate in bread made with amaranth flour and there is a lack of scientific reports regarding this field. Therefore the purpose of the present work was to provide further information on how replacing wheat flour by whole amaranth flour from *A. cruentus* (up to 40 g/100 g) affects the phytate content of bread and its performance, and to evaluate its potential utility as a nutritious breadmaking ingredient.

2. Materials and methods

2.1. Materials

The commercial grain amaranth and flour were purchased from the local Spanish market. *A. cruentus* was used in this research, whose colour was yellow-gold. The characteristics of the commercial wheat and amaranth flours used were (g/100 g): moisture 15.28 ± 0.01 and 11.04 ± 0.01 ; protein ($N \times 5.70$)

11.70 ± 0.06 and ($N \times 5.85$) 14.04 ± 0.01 dry matter (d.m.); fat content 1.11 ± 0.01 and 6.04 ± 0.01 d.m.; and ash 0.53 ± 0.01 and 2.44 ± 0.08 d.m., respectively. Mineral content and the amount of *myo*-inositol phosphates are summarized in Table 1. Compressed yeast (*Saccharomyces cerevisiae*, Lesaffre, Wołczyn, Poland) was used as starter.

2.2. Breadmaking procedure

The bread dough formula consisted of commercial wheat flour (500 g) with replacement by different concentrations of amaranth flour, 0, 10, 20, 30 and 40 g/100 g (Control, 10WAF, 20WAF, 30WAF and 40WAF, respectively), compressed yeast (15 g), sodium salt (5 g) and tap water up to optimum absorption (500 Brabender Units), between 51.0 and 58.4 g of water/100 g of flour, conditioned by the formula. The ingredients were mixed (Kitchen Aid, Long Beach, USA) for 4.5–5.5 min, depending on the formulation, and the doughs were fermented (ZBPP, Bydgoszcz, Poland) for 60 min at 30 °C and 65% relative humidity. The doughs were then kneaded, divided into three pieces of 250 g, put into pans and proofed under the above-mentioned conditions for 60 min. After the fermentation step, the doughs were baked in an electric oven with an incorporated proofing chamber (ZBPP, Bydgoszcz, Poland) at 225 °C for 20 min. Finally, the bread loaves were cooled at room temperature for 60 min for their subsequent analysis. The experiments were done in triplicate.

2.3. Bread composition

Starch content was measured by the total starch assay procedure (AOAC, 1996). The resistant starch, considered as the starch fraction not hydrolysed *in vitro* by pancreatic α -amylase, EC 3.2.1.1, from porcine pancreas (Sigma, A-3176, St. Louis, USA), was determined in dried bread crumb according to the Champ, Martin, Noah, and Gratas (1999) method. The products of hydrolysis were extracted with 80 g/100 g (v/v) ethanol and the non-digested material was solubilised in 2 mol/l KOH, and then hydrolysed with amyloglucosidase EC 3.2.1.3 (Novozymes, AMG 300L, Bagsvaerd, Denmark) into glucose. The free glucose was finally quantified with a glucose oxidase/peroxidase analysis kit (Liquick Cor-

Table 1
Mineral and *myo*-inositol phosphates content of flours.

Sample ^a	Units ^b	Wheat flour	Whole amaranth flour
Ash	g/100 g	0.53 ± 0.01	2.44 ± 0.08
<i>Microelements</i>			
Cu	$\mu\text{g/g}$	1.83 ± 0.03	6.94 ± 0.01
Mn	$\mu\text{g/g}$	5.82 ± 0.01	36.55 ± 0.12
Zn	$\mu\text{g/g}$	7.35 ± 0.10	42.08 ± 0.32
Fe	$\mu\text{g/g}$	12.66 ± 0.04	82.13 ± 0.17
<i>Macroelements</i>			
Ca	mg/g	0.22 ± 0.01	2.04 ± 0.01
Mg	mg/g	0.25 ± 0.01	2.69 ± 0.01
P	mg/g	1.11 ± 0.02	5.30 ± 0.02
Na	$\mu\text{g/g}$	112.4 ± 1.4	8.21 ± 0.27
K	mg/g	1.56 ± 0.01	4.70 ± 0.03
<i>Myo</i> -inositol phosphates			
InsP_6	$\mu\text{mol/g}$	n.d.	21.1 ± 2.1
InsP_5	$\mu\text{mol/g}$	n.d.	2.3 ± 0.5
InsP_4	$\mu\text{mol/g}$	n.d.	0.86 ± 0.08
InsP_3	$\mu\text{mol/g}$	n.d.	n.d.

^a Mean \pm SD, $n = 3$; InsP_3 to InsP_6 : *myo*-inositol containing 3–6 phosphates per inositol residue; not detected (n.d.).

^b Units expressed in dry matter.

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