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# Effect of incorporation of amaranth on the physical properties and nutritional value of cheese bread

*Efeito da incorporação de amaranto nas propriedades físicas e no valor nutritivo do pão de queijo*

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## Abstract

At the present celiac disease has no known cure, and its only treatment is a strict lifelong adherence to a gluten-free diet. Cheese bread is a traditional Brazilian product and a safe option for celiacs. However, like other gluten-free breads, it has inherent low levels of fibers and minerals. The objective of this study was to evaluate the effect of incorporation of whole amaranth flour on the physical properties and nutritional value of cheese bread. Amaranth flour was incorporated at 10, 15, and 20% proportions in different formulations. The increasing amaranth levels darkened the product, reduced specific volume, and increased compression force. Ten percent amaranth-content cheese breads exhibited slight differences in physical properties compared with the controls. These results demonstrated the possibility of incorporating 10% of whole amaranth flour in the formulation of cheese bread resulting in a product with higher dietary fiber and iron contents and the same level of acceptance as that of the conventional formulation. The aim of this approach is to increase the availability of gluten-free bakery products with added nutritional value contributing to increase the variety of the diet of celiac patients.

**Keywords:** gluten-free bread; *Amaranthus cruentus*; celiac disease.

## Resumo

A doença celíaca não tem cura e apresenta como único tratamento a dieta isenta de glúten. O pão de queijo, produto tradicional brasileiro, é uma opção para pessoas com doença celíaca. Entretanto, assim como os demais pães sem glúten, possui baixos teores de fibras e minerais. O objetivo deste estudo foi avaliar o efeito da incorporação de farinha de amaranto integral nas propriedades físicas e no valor nutritivo do pão de queijo. A farinha de amaranto foi incorporada em 10, 15 e 20% das formulações. O aumento dos níveis de amaranto ocasionou: escurecimento do produto, redução do volume específico e aumento da força de compressão. Pães contendo 10% de amaranto apresentaram ligeiras diferenças nas propriedades físicas em relação ao controle. Os resultados mostram a possibilidade de incorporar 10% de farinha de amaranto integral na formulação de pão de queijo, resultando em um produto com maiores níveis de fibra alimentar e de ferro e com o mesmo nível de aceitação quando comparado ao convencional. Este tipo de abordagem visa o aumento da disponibilidade de produtos de panificação sem glúten e com valor nutricional agregado, podendo contribuir para uma maior variação da dieta dos celíacos.

**Palavras-chave:** pão sem glúten; *Amaranthus cruentus*; doença celíaca.

## 1 Introduction

Celiac disease (CD) is a genetically-based autoimmune disease characterized by life-long intolerance to the ingestion of gluten, a term used to encompass specific storage proteins in wheat (gliadin), rye (secalin), and barley (hordein). Occasionally, oats are contaminated by these other grains during processing and consequently contain gluten (KAGNOFF, 2005). Historically, CD has been considered an uncommon condition, but recent screening studies have indicated that the prevalence of CD could be up to 1% in populations of European ancestry. Nonetheless, the majority of cases remain undiagnosed (REWERS, 2005).

Celiac disease as yet has no known cure, and the only scientifically proven treatment is strict lifelong adherence to a gluten-free diet. Early recognition of the disease is important because associated disorders and complications,

such as nutritional loss, impaired growth and psychomotor development, osteoporosis, anemia, and gastrointestinal cancers can be prevented by adherence to dietary therapy (REWERS, 2005). Compliance with dietary treatment is often difficult due to the need for change in eating habits, acquisition and preparation of products that not part of family dietary habits, and lack of gluten-free industrial products (LEE; NEWMAN, 2003).

Amaranth grain is a highly nutritious gluten-free pseudocereal. Its use in food products is desirable to increase provision of nutrients usually lacking in the celiac diet (KUPPER, 2005). *Amaranthus cruentus* L. grain contains 60% starch, 15% protein, 13% fiber, 8% lipids, and 4% ash (CAPRILES et al., 2008a). Amaranth grain also contains minerals in higher amounts than those of most cereal grains

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(GAMEL et al., 2006). Some research has shown the feasibility of amaranth grain usage in gluten-free products such as biscuits (TOSI; CIAPPINI; MASCIARELLI, 1996), cookies (MARCÍLIO; AMAYA-FARFAN; SILVA, 2005), snacks, cereal bars, and breads (CAPRILES, 2009).

Cheese bread is a traditional Brazilian product which has high acceptability in the market and presents international market expansion (SILVA; GARCIA; FERREIRA, 2003). Cheese bread has no gluten in its composition (PEREIRA et al., 2005). However, like other gluten-free bakery products, it is made from refined flour and starches, and therefore has low levels of fiber and minerals (GALLAGHER; GORMLEY; ARENDT, 2004). The objective of this study was to evaluate the effect of the incorporation of whole amaranth flour on the physical properties and nutritional value of cheese bread.

## 2 Materials and methods

### 2.1 Formulation and processing of cheese bread

*Amaranthus cruentus* grains were purchased from a local producer in Brasília, Brazil. Amaranth flour was prepared by grinding grains with a hammer mill (MML 100 - Astecma, Grinding Equipment Ltd.) to a particle size of <0.250 mm. The ingredients for preparing the cheese bread were purchased from the local market.

The control formulation was defined after preliminary tests. Subsequently, 10.6, 16.8, and 21.5 g.100 g<sup>-1</sup> of whole amaranth flour were added to the control formulation in order to obtain 10, 15, and 20% amaranth-enriched baked loaves, respectively. These quantities were defined taking into account the total weight of each formulation and 10% bake loss (observed in baking preliminary tests).

The amount of ingredients in the formulations of cheese breads was proportionally reduced to allow the addition of amaranth flour, except for the formulations containing 15 and 20% amaranth, which required an increase in the amount of water resulting in homogeneous dough.

As shown in Table 1, the difference in cheese breads formulations was the amount of all ingredients.

**Table 1.** Cheese bread formulation with different whole amaranth flour content.

Ingredients (g.100 g <sup>-1</sup> )	Whole amaranth flour content (%)			
	0	10	15	20
Cassava starch	28.8	26.0	22.9	21.5
Fermented cassava starch	12.4	11.1	9.8	9.2
Whole amaranth flour	0	10.6	16.8	21.5
Water	5.7	4.5	8.5	8.3
Soy oil	15.1	13.7	11.8	11.2
Salt	1.7	1.5	1.3	1.2
Eggs	15.2	13.6	12.0	11.3
Cheese	21.1	19.0	16.9	15.8

0, 10, 15 and 20% refer to cheese bread prepared with different whole amaranth flour content.

Cassava starch, fermented cassava starch, whole amaranth flour, and salt were weighed, homogenized, and scalded with a mixture of boiling water and oil. The dough was left at room temperature for 25 minutes until the mixture reached a temperature of around 25-30 °C. Subsequently, previously beaten eggs, and cheese were added to the dough. The dough was divided into small cylindrical portions of 25 g, molded by hand, and baked at 180 °C for 15 minutes in a preheated oven. After baking, the cheese breads were removed from the oven and left to cool at room temperature for 1 hour. Next, it was packed in plastic bags and stored at room temperature in a dry place until analysis.

### 2.2 Experimental design

A total of 20 breads were prepared for treatments with 0 and 10% of amaranth and 23 breads for treatments with 15 and 20% of amaranth. Three random units were used for specific volume and color analysis. A further three random units were used for moisture evaluation, and five random units were used in texture evaluations. One unit was used for the photographs.

### 2.3 Physical properties

After cooling, the loaves were weighed, and the loaf volume measured by millet-seed displacement. The loaf specific volume (volume [cm<sup>3</sup>]/weight [g]) was calculated.

Texture was evaluated by compression strength testing, 1.5 hours post baking using a texture analyzer (TA-XT2i, Stable Micro Systems, Surrey, UK). A cylindrical probe (75 mm in diameter) was used at a speed of 2 mm/s and strength of 10 g to compress the cheese bread to 50% its original height.

The color of cheese bread was evaluated using a ColorQuest XE equipment (Hunter Lab, USA) according to the CIE-Lab system. The reading angle was 10°, and the standard illuminant was D65, which corresponds to natural daylight. Measurements were taken for *L* (lightness/darkness), *a* (redness/greenness), and *b* (yellowness/blueness). Total color difference ( $\Delta E^*_{ab}$ ) between the standard and the novel formulations was calculated according to the following Equation 1:

$$\Delta E^*_{ab} = [(L^*_1 - L^*_2)^2 + (a^*_1 - a^*_2)^2 + (b^*_1 - b^*_2)^2]^{1/2} \quad (1)$$

Moisture content was evaluated according to the AACC Approved Method 44-15A (AMERICAN..., 2000).

### 2.4 Sensory evaluation

Ethical procedures were approved by the Ethic Committee of the Faculty of Public Health, University of São Paulo, São Paulo, Brazil (experimental protocol n° 1404).

Sensory acceptability of the cheese bread was determined 20 minutes after baking, at the temperature of 40 °C. Fifty-four untrained judges recruited from staff and students of the Faculty of Public Health - USP evaluated the color, texture, flavor, and overall acceptability of the control and the 10% amaranth formulation, which obtained the best results in the physical analysis, on a 9-point hedonic scale (STONE;

SIDEL, 1985) - (1 = extreme dislike, 5 = neither like nor dislike, 9 = extreme like). To be considered accepted, the cheese bread had to reach the established cut off point (values equal or greater than 6) (MUÑOZ; CIVILLE; CARR, 1992).

The samples were presented as “cheese bread” for the control and “wholemeal cheese bread” for the product containing amaranth. The samples were offered in a monadic and sequential way according to a balanced block design. The samples were placed on white plates and identified with three random numbers. The judges evaluated the samples in a testing area and were instructed to rinse their mouths with water between samples to minimize any residual effect.

### 2.5 Estimation of nutritional value

The nutritional value of the products was calculated based on the composition of the whole amaranth flour and the other ingredients, for which data were obtained from the Brazilian Table of Food Composition – TACO (UNIVERSIDADE..., 2006).

### 2.6 Statistical analysis

The results were expressed as mean values and standard deviation. Student t test or analysis of variance (ANOVA) followed by the Tukey test was performed to determine statistical differences among samples. Pearson's correlation coefficients were calculated between amaranth levels and physical properties, and the correlation significance was assessed using the two-tailed Student t test. Statistical analyses were performed using the SPSS software (SPSS Institute Inc., Cary, NC, USA) adopting a significance level of  $p < 0.05$ .

## 3 Results and discussion

### 3.1 Physical properties

The physical properties of cheese breads are shown in Table 2.

Increasing amaranth levels were shown to decrease the specific volume of breads ( $r = -0.99$ ,  $p < 0.05$ ). The effect

of formulations on cheese bread volume is also shown in Figure 1. The addition of amaranth resulted in increased compressive strength of the product (from 34 N in the control up to 240 to 265 N in formulations with amaranth). There was no significant difference in texture between the amaranth formulations.

The addition of amaranth flour and consequently the reduction of the remaining ingredients reduced specific volume and increased compression strength of cheese bread. This is due to the fact that cassava starch, eggs, and cheese are important ingredients for the texture and expansion cheese bread. Cassava starches favor cheese bread expansion during baking (PEREIRA et al., 1998, 2004). Fats from eggs and cheese provide greater softness and uniformity of cells and structure of the crumb, which helps in obtaining softer texture (PEREIRA et al., 1998; MACHADO; PEREIRA, 2010).

The greater the amount of amaranth flour in the formulations, the lower the L values ( $r = -0.97$ ,  $p < 0.05$ ) showing that the breads prepared with higher amount of amaranth were darker than the control. Adding amaranth flour also reduced the b values of bread resulting in a significant difference in total color ( $\Delta E_{ab}$ ) compared with the control. These effects are shown in Figure 1.

The intrinsic color of whole amaranth flour may have caused the darkening of cheese breads, as observed in cakes (CAPRILES et al., 2008b) and gluten-free sandwich breads (CAPRILES, 2009; ALVAREZ-JUBETE et al., 2010).

### 3.2 Sensory acceptability

Although texture was not significantly different between the amaranth-enriched samples (Table 2), the 10% amaranth-enriched bread was selected for sensory acceptability because, in addition to an attractive appearance, it presents specific volume and color values closer to those of the control cheese bread and (Figure 1).

Regarding the acceptability test, the control and the 10% amaranth formulations were offered in a monadic way to panelists. Figure 2 shows the mean values of acceptability for the evaluated attributes.

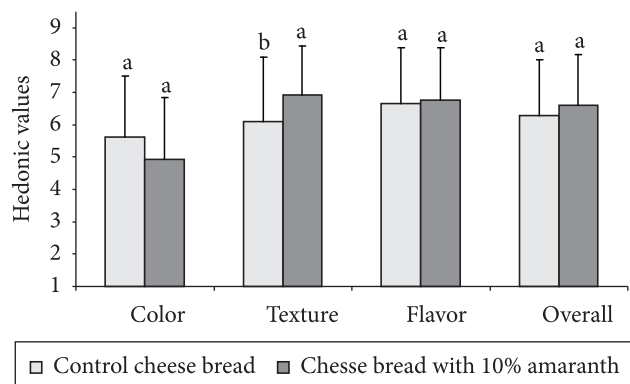
**Table 2.** Physical properties of cheese breads with different whole amaranth flour content.

Physical properties	Whole amaranth flour content (%)			
	0	10	15	20
Specific volume ( $\text{cm}^3 \cdot \text{g}^{-1}$ )	1.48 <sup>a</sup> $\pm$ 0.06	1.30 <sup>b</sup> $\pm$ 0.04	1.09 <sup>c</sup> $\pm$ 0.08	0.96 <sup>c</sup> $\pm$ 0.08
Compressive strength (N)	34.51 <sup>b</sup> $\pm$ 9.91	240.49 <sup>a</sup> $\pm$ 14.73	264.96 <sup>a</sup> $\pm$ 10.18	263.83 <sup>a</sup> $\pm$ 20.01
Bread color				
L*	79.99 <sup>a</sup> $\pm$ 0.38	71.88 <sup>b</sup> $\pm$ 0.39	69.68 <sup>c</sup> $\pm$ 0.24	68.75 <sup>d</sup> $\pm$ 0.23
a*	1.03 <sup>c</sup> $\pm$ 0.08	5.56 <sup>a</sup> $\pm$ 0.97	3.59 <sup>b</sup> $\pm$ 0.10	3.60 <sup>b</sup> $\pm$ 0.12
b*	25.95 <sup>a</sup> $\pm$ 0.20	25.75 <sup>a</sup> $\pm$ 0.80	20.54 <sup>b</sup> $\pm$ 0.08	20.11 <sup>b</sup> $\pm$ 0.60
$\Delta L^*$	-	8.11	10.31	11.24
$\Delta a^*$	-	-4.53	-2.55	-2.57
$\Delta b^*$	-	0.20	5.41	5.84
$\Delta E_{ab}$	-	9.29	11.92	12.93

0, 10, 15 and 20% refer to cheese bread prepared with different whole amaranth flour content. Same letters in row indicate no significant difference between samples ( $p < 0.05$ ).



**Figure 1.** Cheese breads with different whole amaranth flour content.



**Figure 2.** Mean hedonic values assigned for color, texture, flavor, and overall acceptability of 0 and 10% amaranth-content cheese breads. Different letters indicate significant differences between samples ( $p < 0.05$ ).

The results demonstrated that it is possible to incorporate 10% whole amaranth flour in cheese bread while retaining product color, flavor, and overall acceptability. The sample containing 10% amaranth showed higher acceptability in terms of texture. According to the panelists, the formulation was less oily than the standard, probably due to the lower amount of oil, cheese, and egg used; ingredients which contribute to the oiliness of cheese bread.

Muñoz, Civile and Carr (1992) consider a score of six on the nine point hedonic scale as a quality threshold for food products. Although the product containing amaranth did not attain this score for color, there was no statistically significant difference in color acceptability between the formulation with 10% amaranth and the control. Based on these criteria, 10% amaranth-content cheese bread has market potential, as shown in Figure 2.

### 3.3 Nutritional value

Table 3 presents the estimation of the nutritional value of the conventional product and 10% amaranth-content bread.

The incorporation of 10% amaranth resulted in an increase in some nutrients: 17% of protein, 18 times the amount of dietary fiber, and triple the amount of iron compared to the control.

According to the Brazilian technical regulation regarding additional nutritional information (BRASIL, 1998), this new product can be considered a source of protein for all population

**Table 3.** Nutritional value of cheese bread with different whole amaranth flour content.

	Whole amaranth flour content (%)	
	0	10
Calories (Kcal)	356	356
Moisture (g.100 g <sup>-1</sup> )	29.6	28.7
Protein (g.100 g <sup>-1</sup> )	5.82	6.80
Lipids (g.100 g <sup>-1</sup> )	20.7	19.5
Cholesterol (mg.100 g <sup>-1</sup> )	36.8	33.0
Carbohydrates (g.100 g <sup>-1</sup> )	36.6	38.5
Dietary Fiber (g.100 g <sup>-1</sup> )	0.08	1.51
Calcium (mg.100 g <sup>-1</sup> )	140	145
Iron (mg.100 g <sup>-1</sup> )	0.64	2.42

0 and 10% refer to cheese bread prepared with different whole amaranth flour content.

groups since it provides at least 10% of the recommended daily intake/100 g of solid food. Similarly, it is a recognized source of iron since it contains 2.42 mg of iron/100 g (higher than that established by legislation, 2.25 mg.100 g<sup>-1</sup>).

These results show that is possible to develop a tasty and nutritious gluten-free product enriched with amaranth.

## 4 Conclusions

Based on the aforementioned results, the introduction of 10% amaranth in cheese breads proved successful, which is evidenced by slight changes in physical properties and absence of negative effects in the sensory quality. The product presented higher amounts of protein, fiber, and iron than those of the conventional formula.

The aim of this approach is to increase the availability of gluten-free bakery products with added nutritional value contributing to increase the variety of the diet of celiac patients.

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