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Influence of α-amilase, trehalose, sorbitol, and polysorbate 80 on the quality of gluten-free bread

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Abstract

Obtaining gluten-free bakery products is technologically difficult and the combination of several ingredients and change of traditional processes is often required. The gluten free dough does not have the ability to retain the gas generated during fermentation and baking, yielding bread with low specific volume and firm and rubbery breadcrumb. Hydrocolloids, emulsifiers, milk products, proteins, gelatinized starch and enzymes have been used to improve the rheological quality of the dough, final volume, structural and textural characteristics and the shelf life of gluten-free breads. The objective of this study was to evaluate the effects of α - amylase, trehalose, sorbitol and polysorbate 80 on the technological characteristics and aging of gluten-free breads made from rice flour. Gluten-free breads made with different concentrations of sorbitol, trehalose, α - amylase and polysorbate 80 were evaluated by the following tests: specific volume (SV), score (1 and 48 h), hardness (1, 24 and 48 h), hardness rate (g / day), moisture from the crumb and crust (1 and 48 h) and hydration capacity of the crumb (1 and 48 h). The use of α- amylase, trehalose, sorbitol and polysorbate 80 influenced the technological characteristics of gluten free breads. Improvement was observed in specific volume, score and hardness with the use of sorbitol, trehalose, α - amylase and lower concentrations of polysorbate 80. The bread that got the best characteristics was that added with 0.1% polysorbate 80 and reduction of vegetable oil, presenting a great potential to expand the shelf life of gluten-free breads, due to low hardness and lower hardness rate, in addition to showing little variation in the values of crumb and crust moisture and hydration capacity of the crumb.

Introduction

Celiac disease (CD) is an autoimmune disease, triggered in genetically predisposed individuals, resulting from a permanent intolerance to gluten (Rodrigo, 2006). The reaction to gluten ingestion by patients with celiac disease is the inflammation of the small intestine leading to malabsorption of several important nutrients, including iron, folic acid, calcium and fat-soluble vitamins (Feighery, 1999; Kelly *et al.*, 2004). The toxic fraction of gluten responsible for the clinical manifestations of the disease is gliadin. It is demonstrably present in grains like wheat, rye, barley, triticale and possibly in oats. (Moreira, 2007).

The only effective treatment for celiac disease is strict adherence to a gluten-free diet for life resulting in clinical and intestinal mucus recovery (Kotze, 2006). The only cereals considered safe for celiacs are rice and corn, rice is the most suitable for the production of gluten-free products, due to features such as mild flavor, white color, and hypoallergenicity of proteins (Neumann and Bruemer 1997).

The difficulty of maintaining a gluten-free diet can be attributed mainly to the lack of alternative gluten-free ready foods, in the Brazilian market (Sdepanian *et al.*, 2001). The Brazilian Celiac Association - ACELBRA (2013) reports that the gluten-free product that celiacs wish to find is bread (47%), followed by biscuits and crackers (21%), pasta (21%) and pizza (11%).

Gluten is responsible for the properties of extensibility, elasticity, viscosity and gas retention of the dough and contributes to the appearance and crumb structure of the bread. Therefore, obtaining gluten-free products becomes technically difficult often requiring a combination of various ingredients and the modification of traditional elaboration processes. The gluten-free dough does not have ability to retain the gas generated during fermentation and baking, yielding bread with low specific volume and firm and rubbery crumb (Capriles and Arêas, 2011). According to Ahlborn *et al.* (2005) sensory attributes like crumb structure and flavor, as well as problems in storage have also been verified in gluten free baked products.

According to the legislation, RDC Resolution No.

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263 of 22 September 2005, breads are the products obtained from wheat flour and / or other flours, added with liquid, resulting from fermentation or not and cooking process, and may contain other ingredients, as long as there is no disfiguring of products (Brazil, 2005).

In the dough for the manufacture of bread, basic ingredients like water, flour, salt and yeast are commonly used. However, Brazilian law allows the use of certain auxiliary components, known as additives, which may be incorporated into the dough, to correct certain deficiencies in quality, especially flour. Typically, these additives act in order to balance the enzyme activity of the flour or the dough improving the strength and tolerance to baking process (Aquarone *et al*., 2001).

The gluten-free breads are unable to develop a protein network similar to gluten. Therefore, additives such as hydrocolloids and emulsifiers, as well as dairy products, proteins, gelatinized starch and enzymes have been used to improve the rheological quality of the dough, the final volume, structural and textural characteristics and the life of the rolls (Gallagher *et al.*, 2004;. Nunes *et al*., 2009; Onyango *et al*., 2009; Demirkesen *et al*., 2010; Sciarini *et al.*, 2012).

Enzymes such as transglutaminase (Renzetti *et al*., 2008; Storck *et al.*, 2009; Shin *et al.*, 2010), the α- amylase (Sciarini *et al.*, 2012) and cyclodextringlycosyltransferase (Gujral *et al*., 2003) have been studied and presented positive effect on some properties of gluten-free breads.

Recent studies have shown that the addition of emulsifiers, such as polysorbate 80, can help to maintain the crumb softness and thus contribute to the extension of the useful life of gluten-free breads (Nunes *et al*, 2009; Onyango *et al*., 2009; Demirkesen *et al*, 2010; Sciarini *et al*, 2012).

The marketing period for the bread is relatively short, since it is a perishable food. One of the factors limiting the lifetime of the bread is the aging that occurs due to the retrogradation and that adds to the crumb firmness, giving a sensation of a dry product when ingested (Gutkoski *et al.*, 2005). The objective of this study was to evaluate the effects of different additives on technological characteristics and shelf life of gluten-free breads, enabling the development of gluten -free products of good quality.

Material and Methods

Raw material and ingredients

The raw material used in the preparation of gluten-free breads was rice flour provided by Ceralle Indústria e Comércio de Cereais Ltda., located in the city of Pelotas, Brazil. The proximate composition of rice flour provided was as follows: 8.93% moisture, 6.85% protein, 0.37% fat, 0.28% ash and 83.57% carbohydrate.

The formulation of gluten-free bread was 100 g rice flour, 2 g yeast (Fleischmann), 5 g sugar (Caravelas), 2 g salt (Diana), 6 g vegetable oil (Leve), structural agents, 2 g of hydrocolloid methylcellulose (MC) Methocel A4M®, 0,5 g of transglutaminase enzyme (TGase) Activa WM® (81-135 Units/g) that were supplied by Tovani Benzaquen Industries and Ajinomoto Co. respectively, and 0,009 g of P.A. ascorbic acid (L +) used was of Synth brand.

The components tested in the formulations of gluten-free breads were D (+) Trehalose from Sigma-Aldrich Company, α-amylase Termamyl 120L (500-1000 units / mg protein) from Novozymes, sorbitan monooleate (Polysorbate 80, PS 80) from Oxiteno, and sorbitol of the Vetec brand.

Preparation of the breads

The gluten-free breads were prepared as follows: first, the rice flour, salt, sugar, dry yeast and oil were weighed on a precision balance (Marte, model AS200), ascorbic acid, methylcellulose, and transglutaminase were weighed on an analytical balance (Bioprecisa model FA2104N). The dry ingredients were placed in a planetary mixer (“Stand Mixer” 300W) for 1 min at medium speed, then the vegetable oil and the water were added and blended for 9 min and maintained at the same speed. The resulting dough was placed in a container and underwent a first fermentation for 60 min in an stove (Biopar model S150BA) at 30°C. Subsequently, 175 g of dough molds were placed in baking forms with 13.3 × 5.5 cm base (length and bottom width), 15.5 × 7.4 cm of the top (length and top width) and height of 4.5 cm, and fermented for another 55 min at 30°C and then baked at 200°C for 20 minutes in an electric oven (Fischer, Diplomat model).

The breads were removed from the oven as soon as they were baked and cooled to room temperature for an hour after which they were then taken for specific analyses. The bread used in the evaluations during the aging process were stored in polyethylene plastic containers after cooling and kept at room temperature (20ºC).

Evaluation of the breads

To check the influence of α- amylase, trehalose, sorbitol and polysorbate 80 in aging of gluten-free bread elaborated from rice flour, the formulations contained in Table 1 were prepared. They were evaluated through the following analyses: specific

Table 1. Components employees in the formulation of gluten-free breads

Components	Concentration (%) [1]	Code
Sorbitol	0.01	S1
	0.05	S2
Trehalose	1	T1
	2	T2
α-amylase	0.0005	A1
	0.001	A2
Polysorbate 80 [2]	0.1	P1
	0.2	P2

[1]Percentages calculated on the basis of rice flour.
[2]Formulations with 1% vegetable oil.

volume (mL.g^{-1}) score (1 to 48 hours after baking), hardness (g) (1, 24 and 48 h after baking), hardening rate (g/day), humidity (%) of the crumb and crust (1 to 48 h after baking), hydration capacity of the crumb (1 to 48 h after baking).

The specific volume (SV) of the breads was determined by the 10-05 method (AACC, 2000). The breads were weighed on a precision balance and their volume determined by displacement method of millet seeds, where the displaced volume of seeds was measured in a test tube. The specific volume was calculated according to the ratio of the volume and weight of the baked bread.

Internal and external characteristics of the breads were evaluated by 3 trained judges according to El-Dash (1978), which assigns a score to the bread, with a maximum of 100 points distributed in the volume parameters (SV x 3.33), color of the crust, breaking, symmetry, characteristics of the crust, crumb color, crumb cell structure, crumb texture, aroma and taste.

To evaluate the behavior of the water content of bread during aging moisture analysis of the crust and the crumb was performed separately. The humidity was determined according to AACC (2000) method No. 44 - 15A. To evaluate the hardness of the crumb of the bread crumb hardness analyses were performed in a TAXT2 texture analyzer (Stable Micro System, Surrey, UK). The test was performed according to method No. 74-09.01 of the AACC (2000), which consists of placing a slice of 25 mm thick in the Texture Analyzer TAXT2 platform center and compressing it with a cylindrical probe of 36 mm diameter under the following work conditions: pre-test speed: 1.0 mm/s, test speed: 1.7 mm/s, post-test speed: 10.0 mm/s; compression: 40% ; trigger type: 5 g.

The hardness rate (g/day) was determined as the difference between the final (48 h) and initial (1 h) hardness of the crumb divided by storage time (Onyango *et al*., 2009). According to Martin *et al.* (1991) the hydration capacity of the crumb is a method that consists in determining the ability to maintain the crumb of bread moist during the storage period. The analysis was performed as follows: 12.5 g of ground bread was suspended in 75 mL of water for 30 minutes with gentle agitation. The mixture was centrifuged at 2683 G for 12 minutes. The hydration capacity was determined as the weight (g) of wet sediment per gram (dry basis) of bread crumbs. All analyses were performed in triplicate.

Statistical analysis

The results were statistically analyzed using analysis of variance (ANOVA), and compared by Tukey test at 5% significance.

Results and Discussion

According to Table 2, which presents the measures of specific volume (SV) and score (1 to 48 hours after baking) of the breads, it can be seen that there is significant difference in the values of specific volume and score between the control bread and breads with the addition of sorbitol, polysorbate 80, trehalose and α-amylase. The greater specific volume found was 3.5 mL/g of the bread with 0.1% polysorbate 80, followed by 3.38 mL/g of the bread with 0.001% α- amylase and 3.23 mL/g of the bread with 2% trehalose.

It was found that the bread with 0.001% α-amylase had an increase of 36% in the specific volume compared to the control bread. In the study by Sciarini *et al* (2012) an increase of 8.5% in the specific volume of gluten-free breads prepared with 45% rice flour, 45% cassava starch, 10% soybean meal and addition of 0.0006% of α- amylase were reported, where the control bread (no α- amylase) had an SV of 1.98 mL/g.

As reported by Zhou *et al.* (2007), the addition of trehalose in wheat bread can decrease the hardness of the bread crumb and increase water retention and the specific volume of the bread, besides improving the sensory quality. As can be seen in Table 2, the breads with trehalose showed a specific volume 30% greater than the control bread. Therefore, trehalose has a similar effect on gluten-free breads compared to wheat bread.

The addition of 0.1% polysorbate 80 caused a 41.7% increase in specific volume, causing a positive effect on SV of the gluten-free bread prepared from rice flour, but adding 0.2% of polysorbate 80 occasioned a reduction of 15% in the specific volume, acting contrarily, with a negative effect, which may be due to the excess polysorbate 80 added.

Table 2. Measure de specific volume, pontuation, parameters of hardness and hardness rate of gluten-free breads

Bread	Specific volume (mL/g)	Pontuation		Hardness (g)			Hardness rate (g/day)
		1h	48h	1h	24h	48h	
C	2.47 ± 0.07^{e}	79.60 ± 0.89^{Ae}	74.60 ± 0.89^{Be}	371.71 ± 31.37^{Cb}	484.03 ± 51.85^{Bbc}	810.03 ± 46.88^{Ab}	219.16 ± 37.71^{b}
S1	3.11 ± 0.14^{cd}	80.66 ± 1.13^{Ade}	77.66 ± 1.14^{Bd}	177.16 ± 32.64^{Cde}	423.46 ± 42.47^{Bbcd}	519.65 ± 44.41^{Acd}	171.33 ± 33.09^{bc}
S2	3.03 ± 0.09^{d}	81.32 ± 0.50^{Ad}	77.99 ± 0.76^{Bd}	194.93 ± 13.04^{Cde}	421.96 ± 18.81^{Bbcd}	517.21 ± 42.76^{Acd}	161.14 ± 27.75^{cd}
T1	3.22 ± 0.06^{c}	83.37 ± 0.40^{Ac}	81.37 ± 0.40^{Bbc}	222.08 ± 0.42^{Bcde}	396.76 ± 36.74^{Acde}	444.37 ± 31.42^{Ad}	111.14 ± 14.72^{de}
T2	3.23 ± 0.01^{bc}	83.59 ± 1.01^{Abc}	79.42 ± 0.55^{Bc}	261.38 ± 66.04^{Ccd}	494.19 ± 11.63^{Bb}	605.35 ± 33.29^{Ac}	171.98 ± 48.87^{bc}
A1	3.15 ± 0.01^{cd}	84.15 ± 0.60^{Abc}	80.93 ± 0.48^{Bb}	201.37 ± 20.15^{Bde}	447.87 ± 48.89^{Abcd}	507.94 ± 93.18^{Acd}	153.28 ± 36.65^{cd}
A2	3.38 ± 0.04^{ab}	84.93 ± 0.71^{Ab}	80.93 ± 0.48^{Bb}	276.87 ± 27.52^{Bbc}	358.73 ± 29.78^{ABde}	411.71 ± 39.06^{Ad}	67.42 ± 14.21^{e}
P1	3.50 ± 0.21^{a}	86.32 ± 0.53^{Aa}	82.32 ± 0.53^{Ba}	151.20 ± 7.51^{Be}	307.43 ± 77.70^{Ae}	409.77 ± 56.00^{Ad}	137.76 ± 29.03^{cd}
P2	2.10 ± 0.07^{f}	77.65 ± 1.01^{Af}	74.65 ± 1.01^{Be}	956.93 ± 120.88^{Ba}	1657.71 ± 110.28^{Aa}	1878.21 ± 195.57^{Aa}	460.64 ± 38.32^{a}

Mean ± standard deviation. Different lowercase letters in the same column indicate a significant difference ($p < 0.05$). Different capital letters in the same row indicate a significant difference ($p < 0.05$), for Pontuation and Hardness separately. C: bread control. S1: bread containing 0.01% sorbitol. S2: bread containing 0.05% sorbitol. T1: bread containing 1% trehalose. T2: bread containing 2% trehalose. A1: bread containing 0.0005% α-amylase. A2: bread containing 0.001% α-amylase. P1: bread containing 0.1% polysorbate 80 and 1% vegetable oil. P2: bread containing 0.2% polysorbate 80 and 1% vegetable oil.

In the study by Sciarini *et al.* (2012) it was reported that the addition of surfactant did not lead to an increase in specific volume, in fact, the addition of 1% sodium stearoyl lactate (SSL) decreased SV of gluten-free bread prepared from 45% rice flour, 45% cassava starch and 10% soy flour when compared to the control bread. The SV of bread in this case was found in the range of 1.71 to 1.99 $mL.g^{-1}$, volumes much smaller than those found in this study.

The bread with greater SV and the best score was the one added with 0.1% polysorbate 80. Among the food additives used in bakery, emulsifiers are an extremely important group because they are responsible for a host of benefits, ranging from ease of manipulation of the dough, to increases in volume and shelf life of the final products (Gandra *et al.*, 2008).

To assess the quality of bread the system of total score has been used, it assess the characteristics of bread (internal, external, aroma and flavor). According to Dutcosky (1996) and according to the parameters evaluated by the scoring (El-Dash, 1978) system, the bread that has a score of 81-100 can be classified as good quality bread, 61-80 regular, 31-60 bad and less than 30 points is of unacceptable quality. Thus, analyzing Table 2, after one hour baking, the control bread and the breads with 0.1% sorbitol and 0.2% polysorbate 80 can be classified as regular quality bread, while the others are classified as breads of good quality. However, when stored for 48 hours, only two breads showed good quality, the bread with 0.1% polysorbate 80 and the bread with 1% trehalose. The other formulations showed regular quality, due to having lower values of the evaluated items shown in the quality spreadsheet.

In the study by Soares Jr. *et al.* (2008) scores between 51.59 and 83.46 were reported for wheat breads made from roasted rice bran, with some classified as regular quality and only one as poor quality. In Table 2, we can see that there is a significant difference in hardness values (1, 24 and 48 h after baking) and hardness rate of bread. The lowest hardness (1 h) reported was 151.20 g for bread with 0.1% polysorbate 80, followed by 177.16 g for bread with 0.01% sorbitol. The initial crumb hardness and hardness rate were reduced with the incorporation of additives, with the exception of the bread with 0.2% polysorbate 80. The breads that had lower hardness rates were the breads added with 0.001% of α-amylase and the bread with 1% trehalose.

It was found that the bread with 0.0005% of α- amylase had a 43% reduction in crumb hardness compared to the control bread. This confirms what occurred in the study by Sciarini *et al*. (2012) which reported a decrease in 31% of hardness of the gluten-free breads prepared with 45% rice flour, 45% of tapioca starch, 10% soy flour and with the addition of 0.0006% of α-amylase, wherein the control bread (without the α- amylase) has a hardness of 249 g.

The hardness of the crumb of all breads increased with storage time regardless of the treatment as can be seen in Figure 1. The aging of the bread is detected by changes in texture, besides taste and the aroma. In fact, the processes that cause aging begin during cooling, even before the starch has solidified enough to cut the product. In general, during storage, the crumb becomes dry, crumbly and harder, and the crust is soft and coriaceous (Cauvain and Young, 2009).

As previously reported, the breads with 0.2%

Table 3. Crumb and crust moisture, and hydration capacity of the crumb of gluten free breads

Bread	Crumb Moisture (%)		ML (%)	Crust Moisture (%)		MG (%)	HCC		Loss of HCC (%)
	1hs	48hs		1hs	48hs		1h	48h	
C	52.58 ± 0.21^{Ade}	51.63 ± 0.26^{Bbc}	1.81	25.83 ± 0.19^{Bf}	32.87 ± 0.50^{Ac}	27.26	3.32 ± 0.14^{Aab}	3.30 ± 0.06^{Aa}	0.60
S1	52.99 ± 0.33^{Acd}	51.53 ± 0.06^{Bbc}	2.76	31.47 ± 0.42^{Abc}	31.91 ± 0.26^{Ad}	1.40	3.35 ± 0.10^{Aa}	2.77 ± 0.02^{Bb}	17.31
S2	53.07 ± 0.20^{Abcd}	51.98 ± 0.22^{Bb}	2.05	33.20 ± 0.37^{Ba}	36.89 ± 0.37^{Aa}	11.11	3.32 ± 0.10^{Aab}	2.63 ± 0.03^{Bc}	20.78
T1	53.25 ± 0.19^{Abc}	51.70 ± 0.18^{Bbc}	2.91	32.17 ± 0.35^{Ab}	32.05 ± 0.16^{Ad}	0.00	3.20 ± 0.02^{Ab}	2.73 ± 0.15^{Bbc}	14.69
T2	52.36 ± 0.09^{Ae}	51.48 ± 0.14^{Bc}	1.68	30.85 ± 0.73^{Bc}	34.75 ± 0.09^{Ab}	12.64	3.04 ± 0.11^{Ac}	2.65 ± 0.05^{Bbc}	12.83
A1	53.49 ± 0.09^{Abc}	51.74 ± 0.08^{Bbc}	3.27	28.20 ± 0.08^{Be}	34.21 ± 0.17^{Ab}	21.31	2.80 ± 0.06^{Ad}	2.12 ± 0.05^{Bc}	24.29
A2	53.12 ± 0.07^{Abcd}	51.68 ± 0.07^{Bbc}	2.71	30.81 ± 0.21^{Bc}	34.59 ± 0.14^{Ab}	12.27	2.53 ± 0.03^{Ae}	2.01 ± 0.07^{Bc}	20.55
P1	53.64 ± 0.81^{Ab}	52.85 ± 0.42^{Aa}	1.47	28.86 ± 0.58^{Be}	30.40 ± 0.38^{Ae}	5.34	2.95 ± 0.06^{Abc}	2.74 ± 0.05^{Bbc}	7.12
P2	54.69 ± 0.34^{Aa}	52.50 ± 0.59^{Ba}	4.00	29.91 ± 0.39^{Bd}	32.14 ± 0.85^{Ad}	7.46	2.75 ± 0.07^{Ad}	2.66 ± 0.08^{Abc}	3.27

Mean ± standard deviation. Different lowercase letters in the same column indicate a significant difference ($p < 0.05$). Different capital letters in the same row indicate a significant difference ($p < 0.05$), for Crumb Moisture, Crust Moisture and HCC separately. C: bread control. S1: bread containing 0.01% sorbitol. S2: bread containing 0.05% sorbitol. T1: bread containing 1% trehalose. T2: bread containing 2% trehalose. A1: bread containing 0.0005% α-amylase. A2: bread containing 0.001% α-amylase. P1: bread containing 0.1% polysorbate 80 and 1% vegetable oil. P2: bread containing 0.2% polysorbate 80 and 1% vegetable oil. ML: moisture loss. MG: moisture gain. HCC: hydration capacity of the crumb.

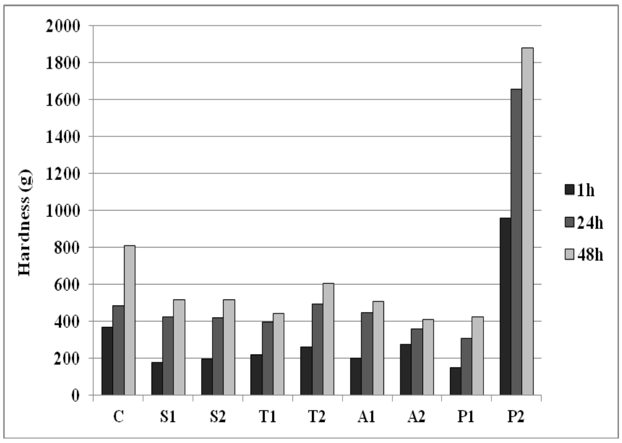

Figure 1. Hardening of the crumb during storage

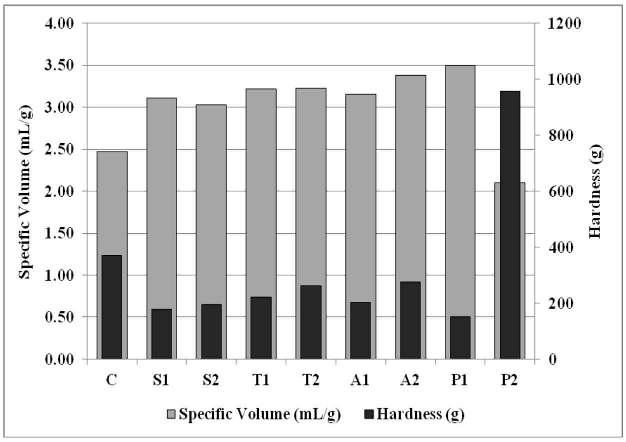

Figure 2. Relationship between specific volume and hardness of breads

polysorbate 80 have the lowest SV and higher initial hardness, differing significantly from the others, which led to a higher hardness rate. This can be explained according to the second theory of Moore *et al.* (2006) and Mezaize *et al*. (2009), which indicate that there is a strong positive correlation between SV and hardness of breads, justified by the greater compression of gas cells in bread with lower SV, that cause increased resistance to deformation of these breads resulting in higher crumb hardness. Figure 2 demonstrates this characteristic of the breads.

According to Table 3, crumb moisture values were found (1h after baking) between 52.36 and 54.69%, and moisture from the crust (1h after baking) between 25.83 and 33.20%. In the breads made with the control formulation, mean crumb moisture values of 52.58% and crust moisture of 25.83% were obtained. Similar values were found in the study by Ronda and Ross (2011), where the crumb moisture ranged from 53.25 to 53.76% and the crust was between 28.70 and 31.84% for gluten-free breads made from rice flour.

It can be seen that there was a reduction in the moisture of the crumb and increase in the crust moisture after 48 hours of storage of the bread. According to Pyler (1988), the aging process is accompanied by the migration of moisture from the center of the bread to the outer regions. Breads P1, T2 and C were those with a smaller reduction in crumb moisture, and the bread added with 0.1% polysorbate 80 showing the smallest loss. For bread crust moisture, the breads that had lower moisture gain were T1, S1 and P1. Thus, these breads were those with the slowest water migration rate, which is of interest for a better conservation of the product.

During aging of bread several changes occur in the properties associated with hardening of the crumb, including an increase in moisture crust, starch crystallinity, opacity and firmness, loss of taste and also a reduction in the crumb moisture, of soluble starch and the hydration capacity of the crumb

(D'Appolonia and Morad, 1981; Martin *et al.*, 1991).

Observing Table 3, which presents the hydration capacity of gluten-free bread crumbs (1 to 48 h after baking), it can be seen that the results showed agreement with the literature, as there was a reduction of up to 24.29% in hydration capacity of the crumbs after 48h of storage. The breads that showed greater preservation of crumb hydration capacity (CHC) after 48h bread were control (C) and the bread added with polysorbate 80 (P1 and P2).

The sorbitol, trehalose and α-amylase showed no positive effect on this characteristic of the crumb, a minor loss of CHC was only found with the addition of polysorbate 80. Trehalose, the α- amylase and polysorbate 80 were influential in the initial CHC, all caused a reduction compared to control bread, probably due to interaction of the additive with the damaged starch granules. According to Martin *et al.* (1991), during baking, monoglycerides and fats interact with the starch molecules and reduce their swelling. Because the starch granules swell less, the solubility of the starch molecules is lower and the hydration capacity of the bread crumb is lower too.

Conclusion

The use of added components influenced the technological characteristics of gluten free breads. Improvements were observed in specific volume, hardness and score with the use of sorbitol, trehalose, α- amylase and lower concentrations of polysorbate 80 used.

The bread that got the best features was that added with 0.1% polysorbate 80 and reduction of vegetable oil, presenting a great potential to expand the shelf life of gluten-free breads, due to low hardness and hardening rate, besides showing little variation in moisture values in the crumb and the crust, and in the crumb hydration capacity.

References

AACC (American Association Of Cereal Chemists) 2000. Approved Methods of the AACC.10th edition. American Association of Cereal Chemists, St. Paul, MN.

ACELBRA (Associação dos Celíacos do Brasil). Disponível em: <*https://www.acelbra.org.br*>. Accessed on: April, 2013.

Ahlborn, G. J., Pike, O. A., Hendrix, S. B., Hess, W. M. and Huber, C. S. 2005. Sensory, mechanical, and microscopic evaluation of staling in low-protein and gluten-free breads. Cereal Chemistry 82(3): 328-335.

Aquarone, E., Lima, U. A., Borzani, W. and Schmidell, W. 2001. Biotecnologia Industrial. São Paulo: Editora Blucher, p.183-207.

Brasil. Resolução – ANVISA – RDC n° 263 de 22 de setembro de 2005. Regulamento Técnico Para Produtos de Cereais, Amidos, Farinhas e Farelos. Disponível em: <*http://www. abima. com. br/dload /13_46_resol_263_ 05_leg_alim_nac.pdf*>. Accessed on: june, 2012.

Capriles, V. D. and Arêas, J. A. G. 2011. Avanços na produção de pães sem glúten: aspectos tecnológicos e nutricionais. Boletim CEPPA 29(1): 129-136.

Cauvain, S. P. and Young, L. S. 2009. Tecnologia da panificação. Barueri, SP. Manole. 2ed. p.418.

D'Appolonia, B. L. and Morad, M. M. 1981. Bread Staling. Cereal Chemistry 58(3): 186-190.

Demirkesen, I., Mert, B., Sumnu, G. and Sahin, S. 2010. Rheological properties of gluten-free bread formulations. Journal of Food Engineering 96: 295-303.

Dutcosky, S. D. 1996. Análise Sensorial dos Alimentos. Curitiba: Champagnat, p.123.

El-Dash, A. A. 1978. Standardized mixing and fermentation procedure for experimental baking test. Cereal Chemistry 55(4): 436-446.

FAO. 1989. Wheatless bread. January.

Feighery, C. F. 1999. Coeliac disease. British Medical Journal, London, 319: 236-239.

Figueira, F. S. 2010. Produção de pão sem glúten enriquecido com Spirulina platensis. Dissertação (Mestrado em Engenharia e Ciência de Alimentos). Rio Grande, Universidade Federal do Rio Grande, p.108.

Gallagher, E., Gormley, T. R. and Arendt, E. K. 2004. Recent advances in the formulation of gluten-free cereal-based products. Food Science and Technology 15: 143-152.

Gandra, K. M., Del Bianchi, M., Godoy, V. P., Queiroz, F. P. C. And Steel, C. J. 2008. Aplicação de lipase e monoglicerídeo em pão de forma enriquecido com fibras. Ciência e Tecnologia de Alimentos 28(1): 182-192.

Gujral, H. S., Haros, M. and Rosell, C. M. 2003. Starch hydrolyzing enzymes for retarding the staling of rice bread. Cereal Chemistry 80(6): 750-754.

Gutkoski, L. C., Brehm, C. M., Santos, E. And Mezzomo, N. 2005. Efeito de ingredientes na qualidade da massa de pão de forma congelada não fermentada durante o armazenamento. Ciência e Tecnologia de Alimentos 25(3): 460-467.

Kelly, C. P., Feighery, C., Gallagher, R. B. and Weir, D. G. 2004. The diagnosis and treatment of gluten-sensitive enteropathy. Advanced Internal Medicine 35: 341-364.

Kotze, S. L. M. 2006. Doença celíaca. Jornal Brasileiro de Gastroenterologia 6(1): 23-34.

Martin, M. L., Zeleznack, K. J. and Hoseney, R. C. 1991. A mechanism of bread firming. I.Role of starch swelling. Cereal Chemistry 68(5): 498-503.

Mezaize, S., Chevallier, S., Le Bail, A. and De Lamballerie, M. 2009. Optimization of gluten-free formulations for french-style breads. Journal of Food Science 74(3): 140-146.

Moore, M. M., Heinbockel, M., Dockery, P., Ulmer, H. M. and Arendt, E. K. Network formation in gluten-free bread with application of transglutaminase. Cereal Chemistry 83(1): 28-36.

Moreira, M. R. 2007. Elaboração de pré-mistura para pão sem glúten para celíacos. Dissertação (Mestrado em Ciência e Tecnologia dos Alimentos). Universidade Federal de Santa Maria, Santa Maria, p.102.

Neumann, H. and Bruemmer, J.M. 1997. Investigations with the production of gluten free bread and roll specialties. Getreide Mehl und Brot 51: 50-55.

Nunes, M. H. B., Moore, M. M., Ryan, L. A. M. and Arendt, E. K. 2009. Impact of emulsifiers on the quality and rheological properties of gluten-free breads and batters. European Food Research and Technology 228: 633-642.

Onyango, C., Unbehend, G.and Lindhauer, M. G. 2009. Effect of cellulose-derivatives and emulsifiers on creep-recovery and crumb properties of gluten-free bread prepared from sorghum and gelatinised cassava starch. Food Research International 428): 949–955.

Pyler, E. J. 1988. Baking Science and Technology.Vol.II, Sosland Publishing Company.

Renzetti, S., Bello, F. D. and Arendt, E. K. 2008. Microstruture, fundamental rheology and baking characteristics of batters and breads from different gluten-free flours treated with a microbial transglutaminase. Journal of Cereal Science 48(1): 33-45.

Rodrigo, L. 2006. Celiac disease. World Journal of Gastroenterology, 12: 6585-6593.

Ronda, F. And Roos, Y. H. 2011. Staling of fresh and frozen gluten-free bread. Journal of Cereal Science 53: 340-346.

Sciarini, L. S., Ribotta, P. D., Léon, A. E. And Pérez, G. T. 2012. Incorporation of several additives into gluten free breads: effect on dough properties and bread quality. Journal of Food Engineering 111: 590-597.

Sdepanian, V. L., Morais, M. B. and Fagundes-Neto, U. 2001. Doença celíaca: avaliação da obediência à dieta isenta de glúten e do conhecimento da doença pelos pacientes cadastrados na Associação dos Celíacos do Brasil (ACELBRA). Arquivos de Gastroenterologia São Paulo 38(4): 232-239.

Shin, M., Gang, D. and Song, J. 2010. Effects of protein and transglutaminase on the preparation of gluten-free rice bread. Food Science Biotechnology 19(4): 951-956.

Soares Jr., M. S., Bassinello, P. Z., Lacerda, D. B. C. L., Koakuzu, S. N., Gebin, P. F. C., Junqueira, T. L. And Gomes, V. A. 2008. Características físicas e tecnológicas de pães elaborados com farelo de arroz torrado. Semina: Ciências Agrárias 29(4): 815-828.

Storck, C. R., Pereira, J. M., Pereira, G. W., Rodrigues, A. O., Gularte, M. A. And Dias, A. R. G. 2009. Características tecnológicas de pães elaborados com farinha de arroz e transglutaminase. Brazilian Journal of Food Technology 12: 71-77.

Zhou, J. C., Peng, Y. F. And Xu, N. 2007. Effect of trehalose on fresh bread and bread staling. Cereal Foods World 52(6): 313-316.