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Sensory, Digestion and Texture Quality of Commercial Gluten-Free Bread: Impact of Broken Rice Flour Type

Running title: Broken Rice Commercial Gluten-Free Bread

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Abstract

This research investigated the effects of two varieties of broken rice (Khouzestan and Lenjan) from warm and dry regions, and two (Hashemi and Tarom) from mild and humid regions on different parameters including dough rheology, digestibility and quality (color, specific volume, textural properties and sensorial properties) of a commercial gluten-free bread. Furthermore, the rice varieties' hydration properties, gelatinization temperatures and starch-granule morphology were assessed. Significant differences were observed in the varieties' proximate composition and hydration properties from both climate zones. The granules' average size was 3.17-4.9 μm . The specific volume of the breads showed no correlation with either the damaged starch content or the amylose content, but had a significant negative correlation with hardness ($r = -0.923$, $P < 0.05$). The crumb hardness of bread was positively correlated with water-binding capacity and was affected by elastic modulus of dough. Results of predicted glycemic index were in accordance with total carbohydrates. Khouzestan received the highest score in sensory evaluation test. Based on the outcomes for bread-quality attributes, Khouzestan from the warm and dry region, which is a cheaper rice variety in Iran, was the most appropriate variety for gluten-free bread production. Moreover, it was determined that the rice varieties currently used in commercial manufacture of gluten-free bread do not necessarily yield the highest-quality bread.

Keywords: Rice, Gluten-free bread, Digestion, Texture

Practical applications

Gluten-free breads (GFBs) are generally used by Coeliac patients. In comparison to wheat bread, the quality of GFBs is lower. Rice is one of the main ingredients of GFBs' formulation, thence by determining the quality-related features of the rice, improvement in the final product could be achieved. In addition, by implementing the cheap and the broken rice variety, the price of the final product could be decreased and be more affordable for the patients.

1 Introduction

Bakery products, especially bread, form the basis of most people's diet (Kihlberg & Risvik, 2007). Bread is one of the world's most popular food product because of its relatively high nutritional value and unique sensory characteristics (texture, taste and flavor). However, a number of people suffer from coeliac disease (CD) (Matos Segura & Rosell, 2011) or other gluten sensitivities. Intolerance to gluten in coeliac patients is lifelong; lifetime adherence to a gluten-free diet is the only solution (Nicolae, Radu, & Belc, 2016).

Rice as a gluten-free cereal has a neutral flavor and white color, easy digestibility and low levels of sodium, and it has hypoallergenic properties (Matos & Rosell, 2013). These features lead rice flour to be a desirable ingredient for producing gluten-free bakery products. The physicochemical properties of rice flour are significantly influenced by the rice variety (Fabiola Cornejo & Cristina M. Rosell, 2015), protein (Marcoa & Rosell, 2008; Sun, Hou, & Zhang, 2008), lipid, moisture (Dautant, Simancas, Sandoval, & Müller, 2007) and amylose content (Varavinit, Shobsngob, Varanyanond, Chinachoti, & Naivikul, 2003). There is a general opinion that the amylose content of each variety is a primary factor influencing the bread quality in gluten-free breads, but it is not influential enough by itself to allow a prediction of bread quality (Han, Cho, Kang, & Koh, 2012). Several other factors, such as gelatinization properties and damaged starch content of flour, must be considered in producing high-quality, gluten-free rice bread. Some rice is damaged during refining into white rice; the damaged grains are discarded as "broken rice". Wastage for this reason is high in Iran. Furthermore, gluten-free products are prohibitively expensive for most coeliac patients. One of the best ways to surmount this issue is to use rice-processing wastage in gluten-free formulations, particularly gluten-free bread. Furthermore, information about the physicochemical properties of broken-rice varieties, and the effects of different cultivation conditions on the rice or the quality of gluten-free bread made from each variety is little. The rice varieties we chose were from two different climate zones; two varieties were more expensive than the others.

Gluten is responsible for the viscoelastic properties of dough, which is essential for maintenance of yeast-produced gas in bread structure (Ngemakwe, Le Roes-Hill, &

Jideani, 2015; Nicolae et al., 2016). Gluten-free dough has neither cohesiveness nor elastic properties; the resulting dough is more fluid than wheat dough, similar to cake batters in terms of viscosity and rheological properties. Consequently, the baked bread has poor color and crumbling texture, and shows other post-baking defects (Ngemakwe et al., 2015). It is important to define GFBs' sensory characteristics, including texture, appearance, taste, and aroma that are appealing to coeliac consumers, as to date most GFBs have exhibited poor quality and consumer acceptability, especially in comparison to traditional wheat flour yeast bread.

The glycemic index of bread is also an important aspect that should be considered in gluten-free products because of the relatively high prevalence of type I diabetes mellitus in coeliac patients (Murray, 2005; Smyth et al., 2008). Therefore, it is necessary for patients to maintain acceptable glycemic control while following a strict gluten-free diet.

This study is the first to compare the macronutrients of broken rice from two climate zones and investigate the dough rheology, sensory, digestibility and quality of bread made with these varieties with those of commercial gluten-free bread.

2 Materials and methods

2.1 Materials

Four varieties of broken rice were used: three (Hashemi, Khouzestan, Lenjan) grown in two regions of Iran with different climate zones and one (Tarom) used by a company making gluten-free products. Hashemi and Tarom, cultivated in mild and humid regions were more expensive than the other samples; Khouzestan and Lenjan, cultivated in warm and dry regions, were less expensive. All varieties were harvested between August and November 2015. The broken kernels were milled (Maadani Machinery Mill, Iran) with a 500 μm screen.

2.2 Flour characterization

The protein, damage starch, amylose, lipid, ash and moisture content of rice flours milled from each sample were analyzed following AACC (11th edition) methods, showing values

of 46-11/02, 76-30/02, 61-03/01, 30-25/01, 08-01/01 and 44-01/01, respectively. All the analyses were done in triplicate.

2.3 Flour hydration

The samples' water-holding capacity (WHC) and water-binding capacity (WBC) were evaluated according to the procedure described by Cornejo and Rosell (F. Cornejo & C. M. Rosell, 2015). WHC was determined by mixing 1.000 g $\pm$ 0.005 g of flour with 10 ml of distilled water and maintaining it at room temperature for 24 h, after which the supernatant was removed.

WBC was determined by mixing 1.000 g $\pm$ 0.005 g flour with 10 ml of distilled water and centrifuging the mixture at 2,000 $\times$ g for 10 min. WHC and WBC were expressed as grams of water retained per gram of solid. All the analyses were made in triplicate.

2.4 Gelatinization temperatures

Gelatinization was evaluated using differential scanning calorimetry (NETZSCH, 200 F 3 Maia, Germany). Flour samples (dry-weight basis) and distilled water were added to aluminum pans in a ratio of 1:3. The pans were hermetically sealed and placed at room temperature for 1h before analysis and heated from 20 to 120 °C at a rate of 10 °C/min. Indium was used for calibration and an empty pan was used as a reference. The transition temperatures (onset (T_o), peak (T_p) and conclusion (T_c)) and the enthalpy of gelatinization (ΔH) were measured. In addition, the gelatinization temperature range (I_g) and the peak height index (PHI) were calculated as shown in equations (1) and (2). PHI describes the relative shape of the endotherm. Low PHI values represent a less structured starch matrix (Correia & Beirão-da-Costa, 2012).

$$I_g = T_c - T_o \quad (1)$$

$$PHI = \Delta H / T_p - T_c \quad (2)$$

2.5 Scanning electron microscopy

To analyze the morphology of starch granules, alkaline extraction of starch (Souza, Sbardelotto, Ziegler, Marczak, & Tessaro, 2016) from rice varieties was performed first.

Then starch samples were affixed to aluminum stubs with double-sided carbon tape and coated with sputtered gold (KYKY, sbc-12 sputter coater, china). An acceleration potential of 26 kV was used in scanning electron microscopy (SEM) (KYKY, EM3200, China). The diameter of the starch granules was measured by averaging the largest dimension of 20 starch granules at 2500x magnification from three micrographs.

2.6 Dynamic oscillatory test

To identify the doughs' viscous and elastic features, dynamic oscillatory tests were performed. Dynamic tests on dough (all ingredients without yeast) were carried out with an MCR 301 rheometer (Anton Paar, Austria) using serrated parallel plate geometry (40-mm diameter). The gap was adjusted to 1-mm. Petroleum jelly (Vaseline) was used to cover the exposed sample surfaces. Before evaluation, the dough was set to relax for 5 min. First, a strain sweep test (0.1-200 %) was performed at 25 °C with a constant frequency of 1 Hz to identify the linear viscoelastic region (LVR). Based on the results, 0.1 % constant strain was used in a frequency sweep test at 25°C with a frequency range of 0.1-100-Hz. The dynamic rheological properties of the samples were measured by the storage modulus (G' [Pa]) (elastic modulus), loss modulus (G'' [Pa]) (viscous modulus) and $\tan \delta$ (G''/ G') for different frequency values (Hz) (C. M. Mancebo, San Miguel, Martínez, & Gómez, 2015).

2.7 Preparation of gluten-free rice bread

Gluten-free breads were prepared according to the formula for a popular gluten free bread manufactured by an Iranian company. All gluten-free formulations contained rice flour, potato flour, corn starch, instant yeast, sunflower oil, salt, sugar, water and stabilizers. Rice flour constituted 50 % of the dry ingredients by weight and water to the weight of 80 % of the dry ingredients' weight was added. All dry ingredients were mixed in a spiral mixer (Diosna, Germany) for 5 min. Then oil and water were added gently and mixed for 5 min at 1 rpmf. After mixing, 240 g of each dough was put into pans (25×11×5.55 cm) and fermented for 20 min at 35 °C and 85% RH. Finally, the baking process was carried out at 185°C for 20 min. Breads were kept for 2 h at room temperature before evaluation.

2.8 Crumb-color measurement

The crumb color was determined using a SP64 Portable Sphere Spectrophotometer (X.Rite, USA) according to the standard ASTM E308 (ASTM E308-15). L^* (0 =black, 100 = white), a^* (+value = red, -value = green), and b^* (+value = yellow, -value =blue) values were recorded from three different areas of the bread crumb.

2.9 Measurement of specific volume and crumb textural properties

The breads' volume was determined using the rapeseed displacement method. Specific volume (cm^3/g) was calculated as the ratio of the volume (cm^3) to the weight of the bread (g) (Fabiola Cornejo & Cristina M. Rosell, 2015).

Texture measurements were performed by a TA.XT Plus texture analyzer (Stable Micro Systems Ltd., Surrey, UK) according to the AACC (2000) Approved Method 74-09 (AACC International. Approved Methods of Analysis) with a slight modification. A slice of the crumb measuring $20 \times 20 \times 25$ mm was compressed to 40 % of its original height at a crosshead speed of 100 mm/min with a 36 mm cylinder probe using a 5 kg load cell (Kittisuban, Ritthiruangdej, & Suphantharika, 2014). The analysis was carried out at 20 ± 3 °C on bread slices. The resulting TPA curves were used to measure hardness, cohesiveness, springiness, resilience and chewiness of the crumb. The average of six replicates was reported.

2.10 In vitro starch digestibility and predicted glycemic index

In vitro starch digestibility was performed to mimic the hydrolysis reactions in the human intestine as described by Brennan and Tudorica (Brennan & Tudorica, 2008). It involves simulated mastication, a proteolytic stage followed by incubation for mimicking the belly, and restriction of pancreatic α -amylase by dialysis tubings for mimicking intestine. Dialysis tubings, chemicals and enzymes were obtained from Sigma-Aldrich, Arklow, Ireland. Triplicate samples of homogenized gluten-free and wheat breadcrumb (4 ± 0.001 g)

“as eaten” were mixed with 20 mL sodium-potassium phosphate buffer (pH 6.9). The pH was adjusted to 1.5 using concentrated hydrochloric acid (12 N), which is appropriate for porcine pancreatic pepsin activity. Samples were incubated for 30 min at 37 °C with 5 mL pepsin solution (EC 3.4.23.1; porcine gastric mucosa, 115 U/mL). The pH was then adjusted to 6.9 with NaOH (12N), and α -amylase solution (EC 3.2.1.1; porcine pancreatic; 110 U/mL in TRIS HCl buffer) was added. Finally, samples were brought to 50 mL with a sodium-potassium phosphate buffer and the mixtures were transferred into dialysis tubings (molecular weight cut-off 11331 Da). Each tube was transferred into a beaker containing 450 mL potassium phosphate buffer and incubated in a shaking incubator (5 h at 37 °C, 100 rpm). An aliquot of dialysate was withdrawn every 30 min for quantification of reducing sugar content, and replaced each time with the equal amount of sodium potassium phosphate buffer. The amount of reducing sugars in the withdrawn dialysates (DNS) was measured using the 3,5 dinitrosalicylic acid method. Thereafter, 200 mL dialysate was boiled for 10 min together with 200 mL 3,5-dinitrosalicylic acid (DNS) reagent. Absorbance was measured at 546 nm. A standard curve using maltose (Sigma-Aldrich, Ireland) was prepared. The DNS solution was prepared by mixing solution A (10 g of 3,5 dinitrosalicylic acid powder in 200 mL 2 N NaOH) and solution B (300 g potassium sodium tartrate tetrahydrate in 500 mL distilled water) and adjusting the volume to 1 L with distilled water. For quantification of reducing sugars released (% RSR), hydrolysis index (HI) (area under the curve (AUC) from 0 to 180 min as a percentage of the corresponding area of the reference white wheat bread) and the predicted GI, the following equations were used:

$$\% RSR = A_{sample} \times 500 \times 0.95 / A_{maltose} \times carbohydrate \times 100$$

$$A_{sample} = \text{sample absorbance at 546 nm}$$

$$A_{maltose} = \text{absorbance of solution containing maltose (1 mg/ml)}$$

$$Carbohydrate = \text{mg starch and sugars contained in 4 g sample}$$

$$HI = AUC(0-180\text{min})_{sample} / AUC(0-180\text{min})_{wheatbread} \times 100$$

$$GI_{predicted} = 0.862HI + 8.189$$

The amount of carbohydrates available in 4 g of bread was calculated based on the following formula: $[100 - (\text{moisture} + \text{fat} + \text{protein} + \text{ash})]$.

2.11 Sensory evaluation

Sensory analysis of the breads was carried out by 20 semi-trained panelists, both male and female, to evaluate color, texture, appearance, taste, odor and overall acceptability in a five-point hedonic scale (1=dislike extremely, 3= neither like nor dislike, 5= like extremely). The samples were coded and evaluated at room temperature.

2.12 Statistical analysis

The results were expressed as mean values. Significance differences in treatment means were identified by one-way analysis of variance (ANOVA) followed by Duncan's multiple range tests at $p \leq 0.05$ (IBMSPPSS v.21, Armonk, NY). The sensory data were analyzed using independent samples t-test.

3 Results and discussion

3.1 Proximate analysis, WHC and WBC

Significant differences were seen in the proximate analysis and hydration properties of different rice flours (Table 1). Lenjan had the highest content of damaged starch, followed by Hashemi, Khouzestan and Tarom

[Table 1 here]

The apparent amylose contents (AACs) of the rice flours were intermediate (A. A. Wani et al., 2012), and varied between 21.38 and 25.72 %. In contrast, the fat content ranged from 0.74 to 2.96 %. Khouzestan had the highest protein, ash and moisture content, while Hashemi showed the lowest AAC, protein and moisture content. Lenjan exhibited the highest AAC and the lowest WBC; this was in agreement with the trend reported by Gani et al (Gani, Wani, Masoodi, & Salim, 2013). However, Hashemi, with the lowest AAC, did not follow the trend; this was likely due to other factors such as particle size, molecular

structure and hydrophilic parts of proteins and carbohydrates (Gani et al., 2013; I. A. Wani, Sogi, Wani, & Gill, 2013). WBC and WHC are important factors for making gluten-free breads. Low WBC is a factor in making a fresh bread with suitable volume and firmness (Han et al., 2012). High WHC could retard staling by interfering in starch retrogradation in gluten-free bread (Sciarini, Ribotta, León, & Pérez, 2010). Tarom had the highest WHC. Overall, varieties from warm and dry regions had more AAC, ash, fat and protein than varieties from mild and humid regions.

3.2 Starch-granule morphology

SEM images of rice starch granules are presented in fig. 1. The granules' microstructure was mainly polyhedral, with an average granule size of 3.17-4.9 μm .

[Figure 1 here]

Khouzestan starch had the smallest granules, at an average of 3.17 μm ; Hashemi starch had the largest granules, at an average of 4.9 μm . Lenjan and Tarom had average starch-granule sizes of 3.35 μm and 3.68 μm respectively. Khouzestan and Lenjan cultivated in warm and dry regions had smaller granules than Hashemi and Tarom cultivated in mild and humid regions. Starch-granule size is an important factor affecting the composition, gelatinization, pasting properties, swelling, solubility and crystallinity of starch (A. A. Wani et al., 2012).

3.3 Gelatinization parameters of rice flour

The flours' gelatinization temperatures ranged from 70.7 to 83.4 $^{\circ}\text{C}$ (Fig. 2). There were no significant differences ($p>0.05$) in the samples' peak temperature (T_p), gelatinization enthalpy (ΔH) or peak height index (PHI); this was consistent with the results obtained by SEM (Fig. 1). The studied rice varieties' starch granules were similar in shape, and the range of average granule sizes was small, which may explain the similarity in gelatinization temperatures. Gelatinization enthalpy may be attributed to the enthalpy of amylose-lipid complex formation during heating (Juliano, 1998), and can reflect the loss of

molecular order (double-helical) (N. Singh, Kaur, Sandhu, Kaur, & Nishinari, 2006). Low ΔH values represent lower stability of crystals in starch (Chiotelli & Le Meste, 2002). Gelatinization temperatures were high but similar to those reported by other authors in studies of rice flour and starch (Ahmed, Ramaswamy, Ayad, & Alli, 2008; Fabiola Cornejo & Cristina M. Rosell, 2015; N. Singh et al., 2006). A high degree of crystallinity can be a reason for high gelatinization temperatures in rice flour (N. Singh et al., 2006). The differences seen in T_o and I_g among the rice cultivars examined in this study might be due to differing amounts of longer-chain amylopectins (Yamin, Lee, Pollak, & White, 1999).

[Figure 2 here]

[Table 2 here] Tarom showed the highest T_o and T_p , while Lenjan showed the lowest T_o , T_p , ΔH and PHI; that could be attributed to with each variety's damaged starch content (Table 1). Intact starch granules (less starch damage) need more energy to be gelatinized (Asmeda, Noorlaila, & Norziah, 2016). Higher contents of damaged starch accelerate the water absorption and swelling that result in lower gelatinization temperatures (Asmeda et al., 2016). The results did not show a significant correlation between amylose content and gelatinization parameters; this is in agreement with the results of Cornejo and Rosell (F. Cornejo & C. M. Rosell, 2015).

3.4 Dough rheology

The viscoelastic behavior of the dough samples was studied using oscillation frequency sweep experiment. At all frequencies tested, the storage modulus (G') was higher than the loss modulus (G'') in all dough samples, which represents the prevalence of elastic features to the viscous character of the dough. Hence $\tan \delta$, ratio of the lost energy to the stored energy per cycle, was lower than 1 for all dough samples (Fig. 3). A solid elastic-like behavior was seen due to a slight rise in storage and loss modulus with increase in frequency (Lazaridou, Duta, Papageorgiou, Belc, & Biliaderis, 2007). $\tan \delta$ rose by increasing frequency, which shows the transition of solid-like to liquid-like behavior in samples at higher frequency ranges. Tarom dough had the lowest $\tan \delta$ than all the other doughs, this represents the stiffness of this sample in comparison to the others.

[Figure 3 here]

These results were in relation with the samples' damaged starch content and WBC (Table 1). Tarom, with the highest WBC, possessed the highest G' and G'' and the lowest damaged starch content. In contrast, Lenjan, with the highest damaged starch content and lowest WBC, showed the lowest G' and G'' along with Khouzestan. The differences in dough rheology could be related to the internal structure of the starch in each flour, the more damaged starch leading to enhanced water binding and reduced elastic modulus of flour and dough (C. Mancebo, Merino, Martínez, & Gómez, 2015). There was not any relation between protein content and rheological properties of the dough within samples that could be due to moderate effect of protein on water absorption of dough (Dexter, Preston, Martin, & Gander, 1994; Tara, Bains, & Finney, 1972). The varieties from mild and humid region had higher storage modulus than the ones from warm and dry region due to having greater amount of AAC and WBC.

3.5 Crumb color

Table 3 summarizes the L^* , a^* and b^* values for all the breads' crumb. The color of the crumb is an important feature of GFBs. The L^* value indicates the lightness of the crumb, and many studies consider it the most important color parameter (Nunes, Ryan, & Arendt, 2009; Sabanis & Tzia, 2011). In the current study, the L^* value ranged from 70 to 76 in bread samples; this was consistent with previous reports (Fabiola Cornejo & Cristina M. Rosell, 2015; Matos & Rosell, 2012, 2013). Among the rice-bread samples, Khouzestan resulted in a darker crumb color, which could be considered more desirable because it causes the bread to more closely resemble wheat breads than other GFBs, which tend to be obviously lighter (Gallagher, Kunkel, Gormley, & Arendt, 2003). Actually, in our study, varieties from warm and dry regions (Khouzestan, Lenjan) were darker than those from mild and humid regions (Hashemi, Tarom). The a^* values were negative, which indicates the lack of red hue for the crumbs. The b^* scale showed a positive value (yellow hue) for all the evaluated samples. Lenjan showed a significantly higher b^* value than did the other samples.

[Table 3 here]

3.6 Specific volume and textural properties

Table 4 gives the specific volume and textural properties of the GFBs in this study.

Specific volume is one of the important features of breads, and is a key parameter in evaluating bread quality. Tarom (the variety from mild and humid region used by the company manufacturing the commercial gluten-free bread and used in this study) had the lowest specific volume and the highest G', in contrast to the Khouzestan cultivated in the warm and dry region. In fact, dough systems with excessive elasticity lead to limited gas-cell expansion during proofing. (Lazaridou et al., 2007) No correlation was found between amylose content and specific volume of the bread, as was also reported by Cornejo et al (Fabiola Cornejo & Cristina M. Rosell, 2015).

[Table 4 here]

As seen for Tarom, the specific volume of the breads were depended on WBC; this agrees with previous research (Han et al., 2012). Although, a negative correlation between the specific loaf volume and damaged starch content has also been reported (Araki et al., 2009), this correlation was not evident in our breads with the commercial formula. This could be due to the effects of other parameters such as the storage modulus of the dough and WBC of the flours.

Table 4 gives the crumb texture properties. Crumb hardness ranged from 2.20 N to 4.14 N, softer than commercial GFBs as reported by Matos and Rosell (Matos & Rosell, 2012). Crumb hardness had a negative correlation with specific volume ($r = -0.923$, $P < 0.05$) and positive correlation with WBC ($r = 0.958$, $P < 0.05$); this was most obvious for the Tarom and Khouzestan varieties. Tarom had significant differences with the other varieties in almost all the textural properties. There was no significant difference in springiness among the samples, which indicates the freshness and elasticity of the bread. Cohesiveness represents the extent to which the material deforms before it ruptures. The cohesiveness of the Tarom variety was significantly higher than that of the other breads. Breads with low cohesiveness are more susceptible to fracturing or crumbling, which makes them less

desirable (Matos & Rosell, 2012). Breads with low chewiness are broken easily in the mouth. Tarom had the highest chewiness and Khouzestan had the lowest. As previously reported by Matos and Rosell (Matos & Rosell, 2012), hardness and chewiness showed similar trends. Among the breads, Tarom showed the highest resilience, which can be taken to represent the ability to recover after compression. Amylose molecules play an important role in the formation of bread's crumb structure (Kang, Sohn, Yoon, Lee, & Ko, 2015), but no correlation was found in this study between textural properties and amylose content. This could be due to other factors influencing the structure, such as interactions among proteins, lipids and carbohydrates and their synergic effects.

3.7 Predicted glycemic index

A dialysis system was used to investigate the in vitro digestion of rice-bread samples as eaten. The HI and GI indices of the rice-bread samples were predicted as in vitro in this study. Previous studies have confirmed that there is a correlation between the rate of starch uptake in vivo, as judged from the postprandial glucose response, and the rate of in vitro amylolysis when applying the dialysis system (Björck, Granfeldt, Liljeberg, Tovar, & Asp, 1994; Goñi, Garcia-Alonso, & Saura-Calixto, 1997). As expected, the control wheat bread had a much higher reducing sugars release than did the rice-bread samples.

[Figure 4 here]

In fact, the rice-bread samples showed a slower rate of starch digestion compared to the control wheat bread. After 300 min of in vitro digestion, the percentage of reducing sugars released in all rice-bread samples was significantly lower than in the control wheat bread. In terms of HI and $GI_{\text{predicted}}$, there was no significant difference between Lenjan and Khouzestan breads, which showed the lowest values (Figure 4, Table 5). Results were in accordance with total carbohydrates: Hashemi, with the highest level of carbohydrates, presented the highest $GI_{\text{predicted}}$. Furthermore, a substantial negative correlation was observed between $GI_{\text{predicted}}$ and AAC ($r = -0.972$, $P < 0.05$), as had been reported before (J. Singh, Dartois, & Kaur, 2010). Hashemi, with the lowest AAC, had the highest $GI_{\text{predicted}}$. The results of $GI_{\text{predicted}}$ were associated with starch-granule size; this was in contrast with

previous studies (Capriles, Coelho, Guerra-Matias, & Arêas, 2008; Tester, Qi, & Karkalas, 2006). This could be due to unexpectedly large effects of other parameters, such as AAC and carbohydrate content. Overall, the warm and dry varieties had lower $GI_{\text{predicted}}$ due to the discussed parameters in this section.

[Table 5 here]

3.8 Sensory evaluation

According to sensory results, the GFBs differed significantly ($p < 0.05$) in crumb appearance and color (Table 6). Tarom showed the least acceptable appearance and color score. Khouzestan received the highest scores for odor, taste and texture. The scores that participants gave to texture were associated with the specific volume and hardness of the crumbs. In overall acceptability, Khouzestan received the highest score: 4.27 out of 5. The results of the color evaluation demonstrate that consumers prefer darker colors, which could be determined from the crumb-color results.

[Table 6 here]

4 Conclusions

In summary, AAC, WBC and damaged-starch content were the crucial factors in determining bread quality. In our study, WBC was a key parameter in determining the dough rheology, specific volume and crumb hardness. AAC was associated with $GI_{\text{predicted}}$. The content of damaged-starch can alter the gelatinization temperatures and dough rheology. Among the varieties being analyzed, Khouzestan had the highest specific volume and sensorial properties, and possessed a low predicted glycemic index. Furthermore, it represented the lowest hardness. Hence, it was chosen as the best variety for making gluten-free bread. Rice varieties (Khouzestan and Lenjan) cultivated in warm and dry regions with cheaper prices could be used instead of varieties (Hashemi and Tarom) being cultivated in mild and humid regions with higher prices. Moreover, varieties being used commercially are not necessarily the best ones for bread making, as shown by the results

for Tarom. In fact, could potentially increase both profit and product quality by using specific cheaper varieties.

5 Ethical Statements

Conflict of Interest: The authors declare that they do not have any conflict of interest.

Ethical Review: This study was approved by the Institutional Review Board of Shahid Beheshti University of Medical Sciences.

Informed Consent: Written informed consent was obtained from all study participants.

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Fig. 1 Scanning electron micrographs of starches from different rice cultivars: a) Taroni; b) Khouzestan; c) Lenjan; d) Hashemi

Fig. 2 Thermograms of different rice flours

Fig. 3 Evaluation of dynamic moduli (G' , G'') and $\tan \delta$ of dough samples at 25 oC. Filled symbols: G' , open symbols G''

Fig. 4 Reducing sugars released from 4 g of samples during hydrolysis

Table 1. Proximate composition and hydration properties of rice-flour varieties

Characteristics	Hashemi	Tarom	Khouzestan	Lenjan
AAC (g/100 g)	21.38±0.31d	23.57±0.55c	24.80±0.27b	25.72±0.4a
Damaged starch (g/100 g)	14.83±0.79b	12.03±0.27d	13.46±0.34c	17.45±0.26a
Ash (g/100 g)	0.79±0.01c	0.61±0.13d	2.16±0.05a	1.45±0.08b
Fat (g/100 g)	1.42±0.32b	0.74±0.04c	2.36±0.36a	2.96±0.43a
Protein (g/100 g)	8.91±0.05d	9.49±0.07c	11.7±0.05a	10.21±0.2b
Moisture content (g/100 g)	8.39±0.12c	8.93±0.24b	9.71±0.26a	8.72±0.13bc
WHC (g/g)	1.30±0.06b	1.52±0.05a	1.49±0.01a	1.23±0.04b
WBC (g/g)	1.00±0.06b	1.21±0.01a	0.98±0.01b	0.98±0.03b

AAC: Apparent Amylose Content

Values with different letters within a row are significantly different ($P < 0.05$), ($n = 3$)

Table 2. Gelatinization parameters of flours from different rice varieties

Variety	T _o (°C)	T _p (°C)	T _c (°C)	ΔH (J/g)	I _g (°C)	PHI
Hashemi	71.9±0.4ab	77.5±0.7a	83.4±0.9a	2.54±0.6a	11.5±0.5a	0.44±0.08a
Tarom	72.5±0.6a	77.6±0.6a	83.2±1a	2.21±0.68a	10.7±0.4ab	0.43±0.13a
Khouzestan	71.0±0.5bc	76.3±1.2a	81.0±0.7b	2.45±0.31a	10.0±0.2c	0.46±0.01a
Lenjan	70.7±0.8c	76.2±0.6a	81.7±0.9ab	1.79±0.2a	11.0±0.1ab	0.32±0.05a

Values with different letters in the same column are significantly different ($P < 0.05$), (n=3)

Table 3. Crumb-color parameters

Variety	L*	a*	b*
Hashemi	72.53	-0.97	14.56
Tarom	76.32	-0.74	13.38
Khouzestan	70.26	-0.96	14.16
Lenjan	70.46	-0.65	16.37

Table 4. Quality parameters of gluten-free breads

Rice varieties	Hashemi	Tarom	Khouzestan	Lenjan
Specific volume, cm ³ /g	1.95±0.06a	1.80±0.03b	2.02±0.06a	1.92±0.04a
Hardness (N)	2.93±0.2b	4.14±0.08a	2.20±0.04d	2.57±0.14c
Springiness	0.98±0.04a	0.97±0.02a	0.99±0.02a	0.97±0.00a
Cohesiveness	0.70±0.01b	0.75±0.01a	0.71±0.01b	0.68±0.02b
Chewiness (N)	2.05±0.10b	3.11±0.09a	1.57±0.06c	1.73±0.15c
Resilience	0.44±0.01b	0.49±0.01a	0.44±0.00b	0.42±0.01c

Values with different letters within a row are significantly different ($P < 0.05$), (n = 3)

Table 5. Hydrolysis index (HI) and predicted glycemic index (GI_{predicted}) of rice-bread samples

Bread rice samples	HI(%)	GI _{predicted}
White-flour bread	-	100 ^a
Tarom rice bread	84.6 ± 1.6 ^b	81.14 ± 1.3 ^c
Hashemi rice bread	94.57 ± 1.7 ^a	89.7 ± 1.4 ^b
Khouzestan rice bread	68 ± 2.8 ^c	66.8 ± 2.4 ^d
Lenjan rice bread	65.1 ± 2.3 ^c	64.2 ± 2.0 ^d

Values with different letters in the same column are significantly different ($P < 0.05$), (n =3)

Table 6. Sensory analysis of gluten-free breads

Sensorial Parameters	Hashemi	Tarom	Khouzestan	Lenjan
Appearance	4.36±0.80a	2.81±1.40b	4±1.34ab	4.36±0.80a
Color	4.09±0.70a	3±1.18b	4.36±1.02a	4.45±0.65a
Odor	3.36±1.36a	3.54±1.21a	3.72±1.10a	3.27±1.48a
Taste	3.18±1.07a	3.54±0.93a	3.90±0.70a	3.27±1.19a
Texture	4±1.26a	3.9±1.04a	4.45±0.52a	3.81±1.32a
Overall	3.72±0.90a	3.54±0.82a	4.27±0.64a	3.81±1.07a

Evaluation was made on a five-point hedonic scale from 1 (dislike extremely) to 5 (like extremely). Values with different letters within a row are significantly different ($P < 0.05$)