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THERMAL PROPERTIES AND ESTIMATED GLYCEMIC INDEX OF DIFFERENT COMPOSITE FLOURS AND THEIR GLUTEN-FREE BREAD MAKING PERFORMANCES

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ABSTRACT
This study aimed to investigate thermal properties and estimated glycemic index of various gluten-free flours (rice, chestnut, chickpea and bean). Gluten-free bread was prepared by mixing chestnut, chickpea and bean flours with rice flour at different ratio (0:100, 25:75, 50:50) using straight dough bread making process. Bean flour reflected the lower estimated glycemic index as compared to the other gluten-free flours. The chestnut-rice flour (50:50) bread exhibited the highest texture firmness after 8 days of storage, followed by the rice flour bread. Chestnut-rice flour (25:75) bread was the swellest comparatively. Future studies may be focused on developing the sensorial characteristics of gluten-free breads prepared from chickpea and bean flours.
Keywords: Bean flour, chestnut flour, chickpea flour, gluten-free, estimated glycemic index

FARKLI UN BİLEŞİMLERİNİN TERMAL ÖZELLİKLERİ İLE GLİSEMİK İNDEKS DEĞERLERİNİN BELİRLENMESİ VE GLUTENSİZ EKMEK YAPIMINDAKİ PERFORMANSLARI

ÖZ
Çalışmanın amacı glutensiz pirinç, kestane, nohut ve fasulye unlarının termal özellikleri ile tahmini glisemik indeks değerlerinin belirlenmesidir. Bu kapsamda kestane, nohut ve fasulye unları pirinç unu ile farklı oranlarda (0/100, 25/75, 50/50) karıştırılarak glutensiz ekmekler üretilmiştir. Çalışmada direkt hamur metodu kullanılmıştır. Çalışma sonuçları, fasulye ununun glisemik indeks değerinin diğer unlara göre daha düşük olduğunu göstermiştir. Glutensiz ekmeklerin tekstür analizi sonuçları, 8 günlük depolamanın sonunda en sert ekmeklerin sırasıyla kestane-pirinç unundan (50:50) elde edilmiş ekmekler ve pirinç unundan elde edilmiş ekmekler olduğunu göstermiştir. Kestane-pirinç unundan (25:75) elde edilmiş ekmeklerin hacmi diğer glutensiz ekmeklerden daha yüksek bulunmuştur. Yapılacak yeni çalışmalarda, özellikle glutensiz nohut ve fasulye unlarından elde edilen ekmeklerin duyusal özelliklerinin geliştirilmesi üzerine odaklanılması uygun olacaktır.
Anahtar kelimeler: Fasulye unu, glisemik indeks, glutensiz, kestane unu, nohut unu

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INTRODUCTION

Celiac disease (CD) is known as a chronic enteropathy that is caused by the consumption of gluten protein; originated mostly from barley, rye, wheat, etc. (Moore et al., 2006). CD is described as an intolerance to the gliadin fraction of wheat and the prolamins of barley (hordeins), possibly oats (avidins) and rye (secalins) (Murray 1999; Fasano and Catassi 2001; Farrell and Kelly 2002; Fasano, 2005; Moore et al., 2007). Gluten consumption produces an inflammatory response to the CD patients that damages the villous structure of the small intestine (Shan et al., 2002; Moore et al., 2007).

Today, the only known remedy against CD is to take gluten-free diet throughout the lifespan (Gallagher et al., 2004; Lazaridou et al., 2007). Studies show that 11% of the consumers buy gluten-free products for health, 27 % for weight loss while 65 % consumers think that gluten-free products are healthy (Witczak et al., 2016). The number of CD patients are increasing day by day (Houben et al., 2012), therefore, gluten-free diet is necessary to prevent severe intestinal damage (Sciarini et al., 2008; Torbica et al., 2010). There are different reports of gluten-free formulations prepared from various flours including rice, corn, soybean, chestnut, cassava, and chickpea, starches together with enzymes, proteins, and hydrocolloids to mimic the viscoelastic structure of gluten (Gujral and Rosell 2004; Sanchez et al., 2004; Toufeili et al., 1994; Anton and Artfield, 2008; Demirkesen et al., 2010a; Torbica et al., 2010).

Chestnut flour is used in gluten-free bread formulations due to its functional benefits (Demirkesen et al., 2010a). Additionally, chestnut flour is a source of dietary fiber, starch, sugar, fat, high quality proteins with essential amino acids, vitamin B groups, vitamin E, magnesium, potassium and phosphorus (Sacchetti et al., 2004; Chenlo et al., 2007; Demirkesen et al., 2010a). As most of the gluten-free bread formulations lacks these constituents, chestnut flour is a beneficial alternative (Moroni et al., 2009; Demirkesen et al., 2010a).

Chickpea flour may also be used in gluten-free formulations; thanks to its functional properties. Miñarro et al. (2012) found better physicochemical characteristics among the gluten-free bread formulations tested. They also stated that chickpea flour could be used as soya protein substitute in gluten-free bread formulations. Giuberti et al. (2015) successfully introduced bean flour into gluten-free spaghetti formulation. Kohajdová et al. (2013) investigated the influence of bean flour on specific properties and rheological characteristics of wheat dough. They reported that the addition of legume flours to bread formulation provides high levels of bioactive compounds; thus nutritional and functional properties to the final product (Angioloni et al., 2012; Rizzello et al., 2014).

A fundamental insight for the development of novel gluten-free functional formulations lies in the effects of individual ingredients on internal bread characteristics. Therefore, the present study was designed to use different flours such as chestnut, chickpea, bean and rice flours in the gluten-free bread making. The chemical characterizations of gluten-free flours were also carried out. Moreover, the estimated glycemic index of both the gluten-free breads and flours were determined during this study.

MATERIALS AND METHODS

Materials

Bean (pre-cooked, dried, white), chestnut, chickpea and rice flours were purchased from Naturelka (Aydın, Turkey). The instant (Dr. Oetker, Turkey) natural dough yeast (*S. cerevisiae*), sugar, margarine, salt and baking powder were bought from local markets. Xanthan gum and emulsifier DATEM (diacetyl tartaric acid esters of monoglycerides) were purchased from Tito (İstanbul/Turkey).

Hexane, sulphuric acid, boric acid, hydrochloric acid, methylene blue, methylene red, sodium hydroxide, potassium sulfate and copper sulfate were purchased from Sigma (St-Louis, USA). All the chemicals used were of analytical grade.

Methods

Chemical analyses

Moisture and ash content of gluten-free flours were determined according to the methods (925.10 and 923.3) of AOAC (1990). The total fat content was determined using Soxhlet method (AOAC, 1990). Nitrogen content was determined according to the Kjeldahl method and converted to protein with a factor of 6.25. All experiments in the study were carried out in duplicate.

Determination of thermal properties

The gelatinization properties of gluten-free flours were determined using differential scanning calorimetry (Perkin-Elmer DSC 6000) according to the method of Chung et al. (2008a). The samples were kept at -20 ºC for 1 min, then heated to 250 °C (with 20 °C/min). Thermograms were analysed in terms of glass transition temperatures and enthalpy values.

Determination of the estimated glycemic index

Estimated glycemic index of gluten-free flours and breads were determined according to the method of Chung et al. (2008a). 0.1 g of gluten-free flour was incubated with porcine pancreatic α-amylase (10 mg) and amyloglucosidase in 4 mL of 0.1 mol L^{-1} sodium maleate buffer (pH 6.0) in a shaking water bath that was adjusted at 37 ºC (0.5–16 h). After incubation, ethanol (95 %) was added and the sample was centrifuged at 2000 rpm for 10 min. Glucose content of supernatant was measured using glucose oxidase–peroxidase (GOPOD) kit.

Hydrolysis index (HI) was determined by dividing the area under the curve of the samples by the area obtained for white bread. The estimated glycemic index (eGI) was calculated using the equation described by Goñi et al. (1997) as given in equation 1:

$$eGI = 39.71 + 0.549\ (HI) \qquad \text{(eq. 1)}$$

Gluten-free bread making

In the first step, chestnut, chickpea and bean flours were mixed with rice flour at different ratio (0:100, 25:75, 50:50, respectively). The remaining dry ingredients (instant yeast, salt, sugar, xanthan gum, and emulsifier) were added to the formulation. After the addition of melted margarine, water was added, and mixed for 2 minutes using a hand mixer (Fakir, Stuttgart/Germany). The water content of gluten-free formulations was determined according to the results of preliminary experiments for each formulation. The dough was fermented at 30 °C for 40 min. After fermentation, the dough samples were baked at 220 °C for 30 minutes in a domestic oven (Arçelik, Turkey). Dough samples containing only rice flour were used as controls. The produced gluten-free pan breads were stored in a refrigerator at 4 °C totally for 8 days. The breads were produced in duplicate.

Determination of specific volume of gluten-free breads

The specific volume of gluten-free breads was determined using the rapeseed displacement method (AACC, 1995).

Texture analysis

The firmness of gluten-free breads during storage period (at 4 °C for 8 days) was tested according to the standard method (AACC, 2000, 74-09.01) using Stable Micro Systems Texture Analyzer, equipped with a 25 kg load cell and a 3.5 cm aluminium cylindrical probe. Freshly prepared gluten-free breads (day 0) and stored gluten-free breads (day 4 and 8) were placed on the texture analyzer's platform. The breads were compressed with a round compression probe. The peak force of the first penetration was termed as firmness.

Sensory evaluation

The produced gluten-free breads were evaluated by 8 panellists for flavor, odor, color, texture and general acceptability. A 9-point hedonic scale quality analysis was used (Annett et al., 2008).

Statistical analysis

Statistical evaluation was performed using SPSS 15 statistical software (SPSS Inc., Chicago, USA). The results were evaluated by one-way ANOVA using Duncan's multiple range test to check the significant difference among the analyzed parameters. The level of significance was set at $p<0.05$.

RESULTS AND DISCUSSION

The chemical compositions of the gluten-free flours are presented in Table 1. The moisture contents of the flours were found in the range of 7.95 and 9.67%. Protein contents of the bean and chickpea flours were similar and higher than the others as expected. The chemical compositions of gluten-free flours were found as similar to previous reports. Chestnut flour has 3.80 % fat, 4.61 % protein, 1.99 % ash and 10.79 % moisture content (Demirkesen et al., 2010), whereas the rice flour has 1.26 % fat, 7.27 % protein and 0.63 % ash content (Ji et al., 2007). Additionally, Debnath et al. (2003) has reported that chickpea flour contains 4.7 % fat, 26.7 % protein, 9.9 % moisture and 3.3% ash contents, whereas Dzuide and Hardy (1996) found 2.2 % fat, 21.2 % protein, 2.1 % ash and 4.7 % moisture contents in bean flour.

Table 1. Chemical composition of gluten-free flours

	Fat content (%)	Protein content (%)	Ash content (%)	Moisture content (%)
Chestnut flour	2.83 ± 0.02^{b}	7.44 ± 0.31^{a}	2.87 ± 0.05^{c}	7.95 ± 0.07^{a}
Chickpea flour	5.02 ± 0.07^{c}	22.09 ± 0.15^{c}	2.05 ± 0.00^{b}	9.67 ± 1.14^{a}
Bean flour	2.07 ± 0.02^{a}	22.75 ± 0.00^{c}	3.92 ± 0.00^{d}	8.95 ± 0.38^{a}
Rice flour	2.22 ± 0.06^{a}	8.97 ± 0.15^{b}	0.94 ± 0.01^{a}	9.37 ± 0.05^{a}

± Standard error of the mean. Lowercase superscript letters in the same column indicate the significant differences among gluten-free flours ($P<0.05$).

Thermal properties of gluten-free flours are presented in Table 2. Glass transition temperature was the highest in rice flour (52.84 °C) while the least was found in bean flour (47.93 °C). Singh et al. (2000) has found the glass transition temperature of different rice flours ranged from 209.3 to 221.8 °C. However, Cham and Suwannaporn (2010) reported that the glass transition temperature of heat-moisture treated rice flour was 58 °C. According to Khamthong and Lumdubwong (2012), heat-moisture treatment led to a decrease in glass transition temperature. This situation was attributed to an increase in the free volume of polymers that caused a decrease in the glass transition temperature of the samples (Chung et al., 2004). The gluten-free flours analysed throughout the current study were moisture-treated before DSC analysis. Therefore, the low glass transition temperatures can be attributed to the observed condition.

Table 2. Thermal properties of gluten-free flours

	Tg (°C)	ΔH (J/g)
Chestnut flour	51.16 ± 0.94^{b}	166.61 ± 2.23^{a}
Chickpea flour	48.21 ± 0.01^{a}	206.85 ± 1.57^{c}
Bean flour	47.93 ± 0.15^{a}	190.93 ± 0.37^{b}
Rice flour	52.84 ± 0.77^{b}	225.09 ± 1.77^{d}

± Standard error of the mean. Lowercase superscript letters in the same column indicate the significant differences among gluten-free flours ($P<0.05$).

The enthalpy values of gluten-free flours ranged from 166.61 (chestnut) to 225.09 J/g (rice). Correia and Beirão-da-Costa (2012) stated that the enthalpy values of chestnut flours belonging to two different cultivars were 24.8 and 27.6 J/g. However higher ΔH values were reported by Attanasio et al. (2004) for chestnut flour, where they connected the high enthalpy values to the low water content of flours. Singh et al. (2000) found the ΔH value of different rice flours between 178.1 and 320.8 mj/mg. Kaur and Singh (2005) stated that gelatinization enthalpy of chickpea flours from different cultivars was between 3.5 and 4.9 (J/g). Chung et al. (2008a)

reported that gelatinization enthalpy of pea, lentil and chickpea flours were around 3.0-5.1 (J/g).
Table 3 summarizes the existing research about the estimated glycemic index value of gluten-free flours and breads. The estimated glycemic index value of gluten-free flours ranged from 47.26 (bean flour) to 52.08 (rice flour), while the glycemic index value of gluten-free breads was between 56.67 (50:50 CPF/RF) and 70.42 (25:75 BF/RF). Higher estimated glycemic index values of gluten-free breads, when compared to gluten-free flours, may be attributed to the incorporation of sugar into gluten-free bread formulations. Considering gluten-free flours, lower glycemic index values were reported by Chung et al. (2008a) for pea (44.4-48.9), lentil (41.4-41.5) and chickpea (48.9-56.1) flours. Kim and White (2012) reported the glycemic index values of different oat flours between 61.1-66.7.

Table 3. Estimated glycemic index of gluten-free flours and breads

Gluten free flours	Estimated glycemic index
Chestnut flour	49.55±1.12^{ab}
Chickpea flour	49.06±0.90^{ab}
Bean flour	47.26±0.42^{a}
Rice flour	52.08±0.25^{b}
Gluten free breads	
50:50 CF/RF	56.84±0.19^{c}
25:75 CF/RF	58.35±0.43^{c}
50:50 CPF/RF	56.67±0.21^{c}
25:75 CPF/RF	56.90±0.21^{c}
50:50 BF/RF	64.90±1.90^{d}
25:75 BF/RF	70.42±1.27^{e}
100 RF	58.42±1.10^{c}

± Standard error of the mean. Lowercase superscript letters in the same column indicate the significant differences among gluten-free flours and breads ($P < 0.05$). CF: Chestnut flour, RF: Rice flour, CPF: Chickpea flour, BF: Bean flour.

The results demonstrated that bean flour showed lower estimated glycemic index when compared to other gluten-free flours. Similarly, different studies highlighted that the beans can reduce glycemic and insulinemic responses and thus they have been considered as low glycemic index foods (Reyes-Moreno and Paredes-Lopez, 1993; Foster-Powell and Brand Miller, 1995; Chung et al., 2008b). Different studies shows that the incorporation of different legume flours to gluten-free cake formulation based on rice flour, reduces both the hydrolysis index and predicted glycemic index (Arocha Gularte et al., 2012). According to Osorio-Díaz et al. (2008), hydrolysis index and predicted glycemic index can be decreased by the addition of legume flour to pasta formulation. However, considering the gluten-free formulations tested, a general trend regarding the estimated glycemic index by the addition of different gluten-free flours could not be observed.

The specific volumes of gluten-free breads were between 1.43 (50:50 CF/RF) and 2.12 (25:75 CF/RF) mL/g (Figure 1). The specific volume of the breads increased as rice flour content was increased from 50 to 75 %. Higher loaf specific volume may be due to the massive holes in breadcrumb as an earlier report shows (Sanchez et al., 2002). McCarthy et al. (2005) reported the specific volume of gluten-free bread based on rice flour and potato starch as 3.03 mL/g. Gallagher et al. (2003) reported the specific volume of wheat-starch-based gluten-free bread as 2.57 mL/g. Demirkesen et al. (2010b) stated that the specific volume of different gluten-free breads based on rice flour was lower than 2.5 g/cm^3. Lower bread specific volume was reported by Hager and Arendt (2013) for rice (1.93 mL/g), teff (1.47 mL/g) and buckwheat (1.77 mL/g) containing gluten-free breads. Additionally, Schober et al. (2005) reported the specific volume of gluten-free sorghum breads between 1.77-1.84 mL/g, whereas López et al. (2004) reported specific volumes of breads prepared with corn and cassava starch and rice flour as 2.53, 2.04 and 1.92 cm³/g, respectively.

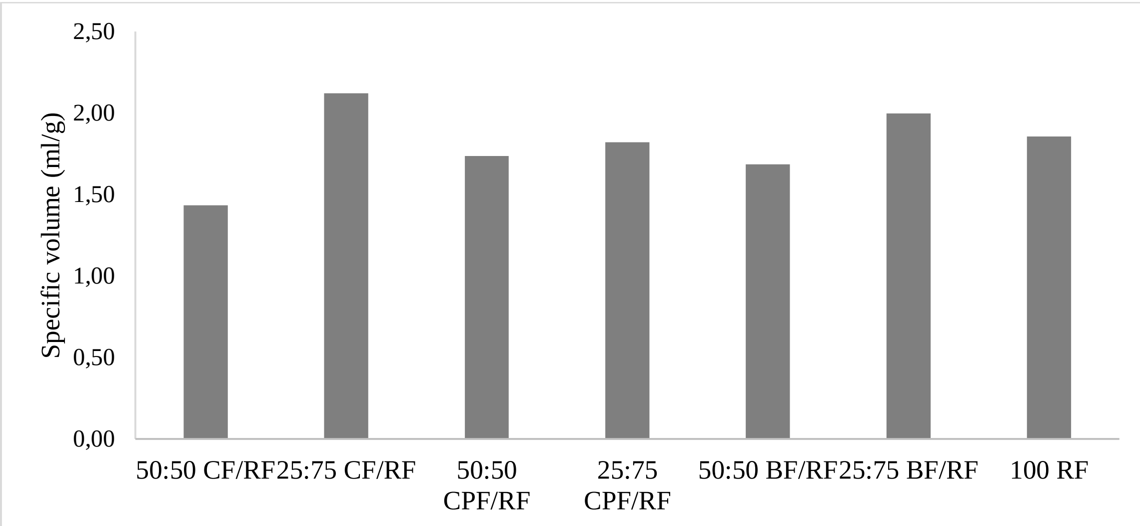

Figure 1. Specific volume (mL/g) of gluten-free breads
CF: Chestnut flour, RF: Rice flour, CPF: Chickpea flour, BF: Bean flour.

Crumb firmness is defined as the peak force of the first compression of the product (Moore et al., 2007). The crumb firmness of all-gluten-free breads increased upon storage (Table 4). Breads supplemented with chestnut-rice flour (50:50) exhibited the highest firmness at the end of 8 days, which was followed by rice flour bread. Additionally, crumb firmness increased as the amount of rice flour increased considering day 0 and day 4. At the end of the storage period, the significant softer crumb was found for chickpea-rice flour (50:50) bread when compared to the other formulations.

Table 4. Firmness of gluten-free breads during storage period

Gluten-free bread	Firmness (N)		
	Day 0	Day 4	Day 8
50:50 CF/RF	5.18^{a,x}	11.02^{a,x}	31.82^{c,y}
25:75 CF/RF	6.83^{a,x}	16.28^{ab,x}	20.06^{abc,x}
50:50 CPF/RF	5.44^{a,x}	12.25^{a,x}	14.30^{a,x}
25:75 CPF/RF	7.88^{a,x}	21.53^{b,x}	22.94^{abc,x}
50:50 BF/RF	7.59^{a,x}	13.06^{a,xy}	19.54^{ab,y}
25:75 BF/RF	7.73^{a,x}	16.16^{ab,x}	29.81^{bc,y}
100 RF	7.73^{a,x}	16.92^{ab,xy}	30.66^{bc,y}

Uppercase letters indicate the differences in the same column, while "x-y" indicate the differences between storage periods (P <0.05). CF: Chestnut flour, RF: Rice flour, CPF: Chickpea flour, BF: Bean flour.

Sensory analyses of gluten-free breads were carried out by 8 panellists. All gluten-free formulations received scores higher than 5 regarding overall acceptability. However, the panellists gave the highest score for overall acceptability to the gluten-free formulation based on chestnut-rice flour (25:75) formulation (Table 5).

Table 5. Sensory analysis of gluten-free breads

	Flavour	Odour	Colour	Texture	Overall acceptability
50:50 CF/RF	6.19±0.51^{c}	6.94±0.35^{b}	6.63±0.29^{a}	6.63±0.37^{bc}	6.25±0.49^{ab}
25:75 CF/RF	6.44±0.26^{c}	6.88±0.23^{b}	7.13±0.26^{a}	7.06±0.24^{c}	6.75±0.30^{b}
50:50 CPF/RF	5.19±0.46^{abc}	5.81±0.39^{ab}	6.94±0.31^{a}	5.94±0.32^{ab}	5.81±0.40^{ab}
25:75 CPF/RF	4.88±0.34^{ab}	5.81±0.46^{ab}	6.50±0.34^{a}	5.50±0.39^{a}	5.50±0.32^{a}
50:50 BF/RF	4.44±0.35^{a}	5.25±1.25^{a}	6.75±0.36^{a}	5.25±0.37^{a}	5.19±0.28^{a}
25:75 BF/RF	5.63±0.29^{abc}	6.06±0.32^{ab}	7.00±0.36^{a}	6.63±0.28^{bc}	5.88±0.30^{ab}
100 RF	6.06±0.48^{bc}	6.00±0.44^{ab}	7.56±0.32^{a}	7.06±0.38^{c}	6.38±0.43^{ab}

Hedonic scale ratings: 9 = extremely like and 1 = extremely dislike

± Standard error of the mean. Lowercase superscript letters in the same column indicate the significant differences among gluten-free bread samples (*P* <0.05). CF: Chestnut flour, RF: Rice flour, CPF: Chickpea flour, BF: Bean flour.

It must also be highlighted that the gluten-free breads based on chestnut, chickpea, bean and rice flours were unfamiliar for nonceliac panellists, since the bread characteristics related with their expectations were mainly based on products made from wheat flour (Haque and Morris 1994). The low sensory scores may comparatively be attributed to this phenomenon (McCarthy et al., 2005), since people with no CD are usually unfamiliar with the organoleptic properties of gluten-free products.

CONCLUSION

The formulation and production of gluten-free bread with a high quality presents a challenge since gluten is responsible for basic crucial dough-forming properties of the flour. Different flours including chestnut, chickpea, bean and rice flours were used within the present study to produce gluten-free-breads. Additionally, estimated glycemic index and thermal properties of the flours were also analyzed. Rice flour provided the highest enthalpy (ΔH: 225.09 J/g) while chestnut flour (166.61 J/g) showed the least. Glass transition temperature of gluten-free flours was observed between 47.93 and 52.84 °C. The gluten-free breads showed significant differences in estimated glycemic indices ranged from 56.67 (50:50 CPF/RF) to 70.42 (25:75 BF/RF). The crumb firmness of all-gluten free breads increased in general when stored, however, it was higher in chestnut and rice flour (50:50). The specific volume of gluten-free breads increased with the increase in rice flour content (50 to 75 %) around 1.43 mL/g. In terms of overall acceptability, gluten-free breads based on chestnut-rice flour (25:75) formulation were mostly accepted by the panellists.

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