

1	Developing gluten free bakery improvers by hydrothermal treatment of rice and corn
2	flours
3	
4	Hayat Bourekoua ^{a,b} , Leila Benatallah ^b , Mohammed Nasreddine Zidoune ^b , Cristina M. Rosell ^a
5	^a Institute of Agrochemistry and Food Technology (IATA/CSIC), Avenida Agustin Escardino
6	7, Paterna, 46980 Valencia, Spain (crosell@iata.csic.es).
7	^b Institute of Nutrition, Food and Agrofood Technology (I.N.A.T.A.A.). University of Brother
8	Mentouri, Constantine. Algeria (bourekoua.h@hotmail.fr).
9	Abbreviations
10	RFBS Rice /Faba Bean Semolina
11	CFBS Corn /Faba Bean Semolina
12	H/W ratio Height/Width ratio
13	Abstract

The impact of hydrothermal treatment of rice and corn flours on their potential as gluten free 14 bakery improvers was tested. Hydrothermal treatment of flours was carried out by 15 suspending flours in water on the basis of 5/1 (w/w) and heated until 65°C. Corn or rice 16 semolina supplemented by faba bean semolina in ratio of 2/1 (w/w) were used for obtaining 17 protein enriched gluten free breads, where improvers functionality was tested. Two central 18 composite designs involving water hydration levels (X_1, X'_1) and the level of hydrothermally 19 treated rice or corn flours (X2, X'2) were used. Instrumental analyses of breads (specific 20 volume, moisture content, crumb texture and height/width ratio) were carried out to assess the 21 impact of experimental factors. Results showed that hydrothermal treatment of rice or corn 22 flours affected in different extent the bread properties, increasing the specific volume of 23 breads and H/W ratio, and decreasing the hardness and chewiness of both types of breads. 24

The optimum formulation for rice/faba bean bread contained 7.59 g/100 g treated rice flour and 96.66 g/100 g water, and for corn/faba bean bread the optimum included 4.73 g/100 g treated corn and 78.81g/100 g water. Optimized breads were found acceptable according to color and texture structure.

29 Key Words: breadmaking, gluten free, hydrothermal treatment, rice, corn.

30

31 1. Introduction

Gluten is the main structure-forming protein in flour, because resulting dough presents high 32 elastic characteristics and it contributes to the appearance and crumb structure of many baked 33 products (Gallagher et al., 2004). Nevertheless, when pathologies require the removal of 34 gluten from the diet, the replacement of gluten presents a major technological challenge. In 35 recent years there has been extensive research for the development of gluten free bread, 36 involving diverse approaches, like the use of different starches (maize, potato, cassava or 37 rice), dairy products, gums and hydrocolloids, emulsifiers, other non-gluten proteins and 38 prebiotics, as alternatives to gluten, to improve the structure, mouth feel, acceptability and 39 shelf-life of gluten free bakery products (Matos & Rosell, 2012). Other gluten free ingredients 40 such as leguminous seeds, buckwheat, potato and sorghum flours (Arendt & Dal Bello, 2008; 41 42 Matos & Rosell, 2015) have been also investigated.

Nevertheless, some autochthonous products offer great opportunities to develop high quality gluten free products, like breads. Studies on celiac disease in Algeria report that adherence to gluten free diet is poor and not easy for concerned population, with the subsequent negative effects on nutritional and health status of patients (Boukezzoula & Zidoune, 2014). One way to improve that status is to find or to develop gluten free products resembling the traditional products on the Algerian market, like *Khobz eddar*. This is the traditional bread made from durum wheat semolina and characterized by a crispy crust and a soft crumb.

Pre-gelatinized starches obtained by heating in the presence of water, are used widely for their 50 51 technological properties such as solubility in hot or cold water, high viscosity and smooth texture and can be used in food processing whenever thickening is required (Lai, 2001). For 52 instance, in cereal porridge, gelatinized flour was used as ingredient to improve texture 53 (Chiang & Johnson, 1976). Despite the extended use in soups and creams, scarce information 54 has been reported on bakery products. Tsai et al. (2012) confirmed that the addition of rice 55 porridge improved the quality of baked bread promoting soft crumb texture. Even later on, 56 Hesso et al. (2014) proposed the use of pregelatinized starches for delaying the staling of 57 cakes. Despite the previous use of those starches as antistaling, there is limited information 58 59 about the use of those starches as structuring agents, which are required to obtain gluten free 60 bread.

The aim of this study was to evaluate the potential of hydrothermal treatment of rice and corn flour to obtain pregelatinized slurries for manufacturing gluten free bread based on rice or corn semolina supplemented with faba bean semolina. The study was carried out following a RSM (Response surface methodology) to define the levels of water and pregelatinized slurries (rice or corn), based on a traditional Algerian breadmaking process.

66 2. Materials and methods

67 2.1. Materials

Rice (*Oryza sativa*) (Basmati, Thaïland) was grinded using a stone mill grinder until
obtaining semolina (200-500 µm particle size) or flour (lower than 200 µm particle size).
Rice semolina and flour had 10.30% moisture, 0.04% ash, 0.21% lipid and 10.73% protein
content. Corn (*Zea mays*) semolina (>300 µm particle size) or flour (<300 µm particle size)
were provided by DACSA (Valencia, Spain), containing 13.69% moisture, 0.33% ash and
7.50% protein. Faba bean semolina (*Vicia faba*) (10% moisture, 0.08 % ash, 1.08% lipid,

30.01% protein) was obtained after grinding the hulled bean seeds purchased from AlamirCompany (Albehera, Egypt).

Instant dry yeast (Saf-instant) was donated by Lessafre Iberica (Valladolid, Spain). Egg
powder was purchased from EPSA company (Valencia, Spain). Salt and sunflower oil were
acquired in local market.

79 **2.2. Hydrothermal treatment**

80 2.2.1. Preparation of treated rice and treated corn

Hydrothermally treated rice or corn was prepared according to TangZhong method (Yvonne, 2007): the treatment was done by suspending flours (rice or corn) in cold distilled water ($21\pm$ 1°C) at the ratio of 1/5 (w/w). Slurries kept thoroughly stirred were heated until the inner temperature reaches 65°C; it took about 6-7 min for corn and 8-9 min for rice to reach that temperature. After cooling down at room temperature for 1 h, slurries were kept for 24 h at 4°C and used as bread ingredients to improve the breadmaking.

87 2.2.2. Pasting properties

Pasting properties of hydrothermally treated slurries were determined after 1 hour of cooling 88 at room temperature and after 24 hours storage at 4 °C. Previously prepared slurries (30 g) 89 were weighed into the aluminum canisters and analyzed using a rapid visco analyser (RVA) 90 (RVA 4500, Perten Instruments SA, Stockholm, Sweden) by following ICC standard method 91 No 162 (ICC, 1996). Peak viscosity, through, breakdown, final viscosity, setback (difference 92 between final viscosity and peak viscosity) and onset temperature were recorded by 93 Thermocline software (Perten Instruments SA, Stockholm, Sweden). Three replicates were 94 carried out per sample. 95

96 2.3. Experimental Design

Two central composite designs with 2 factors each were employed to determine the effect of water (X_1 for RFBS, X'_1 for CFBS) and the amount of hydrothermally treated slurries (X_2 : 99 treated rice for RFBS, X'₂: treated corn for CFBS) on rheological and technological properties
100 of breads followed by optimisation of the process using surface response methodology.

Factorial section was a 2^2 test; the star section included four tests. Five replicates centerpoints were also added, for a total of $2^2+2^2+5=13$ runs for each type of bread (Table 1).

For each response variable, model selection appeared to be quadratic; the significance level was set at 0.05. Responses of each variable were subjected to statistical analysis in order to define the optima points for breadmaking recipe using the desirability function approach (DFA). The desirability function approach is a multi-criteria optimization method useful to find the best compromise between several responses. D = $(d1 \times d2 \times d3.....\times dn)^{1/n}$ where *di* are the desirability indices for each response (*di* = 0 least desirable; *di* = 1 most desirable) and *n* is the number of responses in the measure (Simurina et al., 2012).

110 **2.4. Bread making process**

Rice /faba bean semolina (RFBS) and corn /faba bean semolina (CFBS), in a ratio of 2/1 111 (w/w) cereal/faba bean semolina, were the two gluten free recipes studied, aiming to offer a 112 better nutritional balance in amino-acids (Benatallah et al., 2012; Storck et al., 2013). The 113 hydration range applied in the experimental design was determined for each formula by 114 preliminary experiments (55 to 110% for rice breads and 51 to 105% for corn breads). The 115 levels of hydrothermally treated slurries were fixed up to 14.75% or 13.8%, for rice or corn 116 breads, respectively. Hydration and hydrothermally treated slurries percentages are expressed 117 based on the semolinas blend basis. 118

The basic bread recipe consisted of: 666 g of rice or corn semolina, 333 g of faba bean semolina, 20 g of salt, 20 g of instant dry yeast, 20 g powder egg, 200 ml of sunflower oil and the amount of water defined in the experimental design (Table 1). The making procedure involved manual premixing of dry ingredients, with exception of powder egg and then water was added. When hydrothermally treated slurries were added, the corresponding amount of rice or corn semolina and water were replaced. It must be kept in mind that slurries contained flour/water at the ratio of 1/5 (w/w). The mixture was left to rest for 10 min, then the powder egg and the rest of water were added and manually mixed for 10 min and sunflower oil was added and mixed for additional 5 min. The resulting dough was divided in lumps (100 g) and put into mold and proofed for 45 min at 37 °C in a fermentation cabinet (Salva Industrial S.A., Lezo, Guipuzcoa, Spain).The baking tests were carried out at 165°C for 25 min into an electric oven (Salva Industrial S.A., Lezo, Guipuzcoa, Spain).

131 **2.5. Bread quality evaluation**

Bread characterization after one hour post-baking consisted of specific volume, crumb textureprofile analysis, height/width ratio and bread moisture.

Volume was determined by the rapeseed displacement method according to the AACC Approved Method 10.05. (AACC, 2000); specific volume (cm^3/g) of an individual loaf was calculated by dividing volume by weight. Moisture content was calculated based on ICC 110/1 method (ICC, 1996).

Height/width ratio was measured by capturing the image of the central slice with an HPScanjet G 3110 scanner in the presence of scale.

Texture profile analysis (TPA) was evaluated on the breadcrumb using a texture analyzer (TA-XT plus, StableMicro Systems Ltd., Godalming, UK), using two bread slices of 1-cmthickness, which underwent a double compression test up to 50% strain (penetration of its original height) at a cross head speed of 1 mm/s and a 30 s gap between compressions, with a cylindrical stainless steel probe P/25 (25 mm diameter) (Rosell et al., 2009). The parameters recorded were hardness, cohesiveness, springiness and chewiness.

The color of the crumb samples was measured at three different locations by using a Minolta colorimeter (Chroma meter CR-400/410, Konica Minolta, Tokyo, Japan) after standardization with a white calibration plate ($L^{*}=96.9$, $a^{*}=-0.04$, $b^{*}=1.84$). The color was recorded using 149 CIE- $L^*a^*b^*$ uniform color space, where L^* indicates lightness, a^* indicates hue on a green 150 (-) to red (+) axis, and b^* indicates hue on a blue (-) to yellow (+) axis (Matos & Rosell, 151 2013). Data from three slices per sample were averaged.

High resolution images (600 ppi) of the bread slices 10 mm thick were captured by HP Scanjet G 3110 scanner. Crumb cell analysis of breads was performed by Image J software according to Gonzales-Barron & Butler (2006) in the optimized breads. Number of cells, average cells area, and average diameter were calculated. Values were the mean of four replicates.

157 **2.6. Statistical analysis**

158 Multiple regression analysis was performed to fit second order model to dependent variables by using Minitab Release 16 (Minitab Inc., State College PA, USA). The models were used to 159 determine responses surfaces in Statistica 10 (Stat soft, France). One way analysis of variance 160 (ANOVA) was applied to compare the effect of water $(X1, X^1)$ and treated flours $(X2, X^2)$ 161 on the dependent variables (Y, Y). A coefficient of determination (R^2) was computed and the 162 adequacy of models was tested by separating the residual sum of squares into pure error and 163 lack-of-fit. Optimization was performed with Minitab Release 16 (Minitab Inc., State College 164 PA, USA). 165

166 **3. Results and discussion**

167 **3.1. Pasting performance of hydrothermally treated slurries**

Rice and corn flours were submitted to hydrothermal treatment to modify their functionality looking for obtaining bakery improvers. After hydrothermally treatment, rice and corn flours were stored for one and 24 hours and the pasting profile of those treated flours were compared with the native ones to check the impact of the thermal treatment on starch gelatinization (Figure 1 and 2). Significant differences were observed among the pasting profiles of the flours before and after treatment. The RVA profile confirmed that hydrothermal treatment

partially gelatinized the starch, increasing the initial viscosity of the flours (1800 cP and 2000 174 175 cP for rice and corn treated flours, respectively) and decreasing the viscosity during heating. The high initial viscosity for the pre-gelatinized rice and corn was attributed to the disruption 176 of the molecular order within the starch granules during the treatment, resulting in the loss of 177 granule integrity and destruction of starch crystallinity (Marti et al., 2013). Besides the 178 reduction in the maximum viscosity during heating, a shift in the onset pasting temperature 179 180 was observed due to higher temperature was required to gelatinize the remaining intact starch granules. Nevertheless, differences were observed depending on the flours source, rice 181 (Figure 1) or corn (Figure 2), since botanical origin is responsible of morphological, thermal 182 183 and rheological properties of starches (reviewed by Sing et al., 2003). Rice flour after treatment showed a significant reduction in the maximum viscosity during heating, but the 184 trend changed during cooling, whose viscosity was higher than in the native flour. During 185 186 cooling rapid reorganization of the amylose chains is produced increasing the viscosity of the flour (Rosell et al., 2007). It seems that the structure disorder promoted by the thermal 187 treatment favored the amylose released, which was rapidly enabled to reorganize during 188 cooling. It must be stressed that the degree of granule disintegration in rice is greatly 189 dependent on the amount of amylose, particularly low amylose starch granules are weak and 190 fragile whereas high amylose rice starch is strong and rigid (Sandhya & Bhattacharya, 1995), 191 leading to higher viscosities during heating and cooling (Klug Tavares et al., 2010). The 192 storage of the thermally treated rice flour during 24 hours induced a reduction in the viscosity 193 obtained during heating and cooling. In opposition, thermal treatment of corn flour induced a 194 decrease in the viscosity during the heating and cooling stages compared to the native flour. 195 Moreover, the storage of the treated flour resulted in an increase of the viscosity along the 196 temperature gradient likely due to the progress of starch retrogradation during the post-197 treatment storage. It must be remarked that the staling kinetics is dependent on the starch 198

source, thus differences must be ascribed to the nature of rice or corn starches (Jane et al.,1999).

The two profiles revealed that the treated rice achieved higher final viscosity (about 9000 cP to 12000 cP) than those of corn (6000-7000 cP) at the end of RVA measurement. Therefore, depending on the extend of the hydrothermal treatment would be possible to obtain different degree of gelatinization and in consequence different level of viscosity, which would be useful for obtaining gluten free bread.

206 **3.2.** Improving effect on *khobz eddar* gluten free breads quality

207 **3.2.1. Model fitting**

208 The analysis of variance induced by the hydration level and the amount of treated flours on the quality parameters of rice/faba bean semolina and corn/faba bean semolina breads is 209 shown in Tables 2 and 3. The statistical analysis indicated that the fitting models were 210 adequate because they gave satisfactory values of the R^2 (higher for corn breads) for all the 211 responses and model significance. Results showed that the lack-of-fit test was significant for 212 the specific volume, hardness, chewiness and moisture content for experimental design 1 213 (Table 2), whereas it was significant for the specific volume, hardness, cohesiveness and 214 chewiness for experimental design 2 (Table 3). This can be attributed to the very large 215 experimental region covered in which all appropriate functions of independent variables were 216 not included. However, when large amounts of data were included in the analysis, a model 217 with a significant lack of fit could still be used (Phatcharee et al., 2014). In the case of gluten 218 free breads this model was really convenient due to their large variability derived from their 219 complex formulations (Matos & Rosell, 2011), and it is always more convenient to extend the 220 range of analysis than to carry out the experimental design within narrow limits to give more 221 robust information. 222

223 **3.2.2.** Gluten free bread quality characteristics

The effect of different concentrations of water and treated rice or corn on the response surface graphs for specific volume and height/width ratio of RFBS bread and CFBS is shown in Figures 3 and 4, respectively. For RFBS breads, the specific volume varied from 1.50 to 2.35 cm^{3}/g , showing a positive effect of increasing water content and treated rice, although in the later a quadratic effect was observed with a maximum value of 2.35 cm³/g against 1.70 cm³/g for the bread without treated flour.

The specific volume of CFBS bread ranged from 1.40 to 2.90 cm³/g (Figure 4), 230 indicating that a higher specific volume can be obtained with corn in comparison to rice. The 231 response surface (Figure 4) again showed a positive effect of water hydration and treated flour 232 233 on specific volume, although the water effect was less pronounced than that was observed in rice bread. Many authors confirmed the dependence of gluten free bread volume with water 234 amount and indicated also its dependence on the type of raw material (Marco & Rosell, 2008; 235 Schoenlechner et al., 2010). A quadratic effect of the amount of treated corn rice was 236 observed, with a maximum when 5 g of treated corn were present. Therefore, hydrothermally 237 treated slurries, which have partially gelatinized starch, improved the specific volume of 238 gluten free rice or corn based *khobz eddar*. Presumably, the higher initial viscosity induced by 239 treated flours favor the entrapment of air bubbles in the dough structure, and it is even enough 240 241 to hold the gas pressure during expansion at the early stage of baking (Shibata et al., 2011). In order to achieve a good bread volume, the dough should have enough strength to develop and 242 maintain the cells gas and gelatinized starch must be able to withstand the rapid expansion of 243 cells during the initial phase of the cooking (Pongjaruvat et al., 2014). 244

Response surface of height/width ratio for RFBS indicated that it varied from 0.61 to 0.82 with the most significant effect observed with the amount of water. A significant different response was observed for CFBS breads, in which a quadratic effect was observed for the water hydration and the amount of treated corn flour. For CFBS the H/W ratio ranged from 0.63 to 0.81 obtaining the optimum at 75-105 ml of water and 3.0-13.8 g of treated corn flour. Taking into account that breads were baked in molds, the H/W ratio gave information about the oven rise and thus the holding ability of the dough structure during baking.

Hardness is one of the most important quality characteristic of bread texture, 252 with consumers desiring soft and flexible crumbs and low hardness (Hager & Arendt, 2013). 253 The response surface for RFBS breads showed a quadratic effect of both factor, with a 254 minimum hardness located in the interval 7 g-10 g of treated rice and 85 ml-98 ml of water 255 per 100 g semolina blends (Figure 3). The same response, although less pronounced, was 256 obtained with the CFBS breads (Figure 4) and lower hardness was obtained within the range 257 258 1.8 g-13.8 g of treated corn and 76 ml-110 ml of water. Shibata et al. (2011) also reported higher dough expansion and softer texture when added gelatinized rice flour or rice porridge 259 to wheat bread. 260

Regarding chewiness (Figures 3 and 4), different effect was observed in the rice and 261 corn breads. Water content and treated rice amount promoted quadratic negative effects on the 262 chewiness of rice breads, with minimum values when containing 5 g-12 g of treated rice and 263 80 ml-100 ml of water per 100 g semolina blends, indicating easy chewing of the breads. For 264 CFBS bread chewiness, a deep decrease was observed when increasing water, and a smoother 265 266 decrease was induced by the addition of treated corn. Therefore, the use of rice or corn semolina as ingredients obligated to adapt recipes and even the addition of pregelatinized 267 flours confers different characteristics. 268

The opposite effect was induced by water hydration and treated flour amount on springiness in rice and corn based breads, being in both cases a quadratic effect. It must be remarked that springiness is associated to fresh, aerated and elastic product, thus high springiness values are recommended (Matos & Rosell, 2012). Values obtained in rice and corn breads fall within the springiness values (0.76 to 1.00) found for gluten free breads (Matos & Rosell, 2012). Cohesiveness reflects the ability of a material to be deformed before breaking down; in breads high cohesiveness is desirable indicating the bolus formation during mastication without crumbling. Water content and the amount of treated flour induced a quadratic negative effect on the rice and corn bread cohesiveness; with the exception of the positive effect promoted by treated corn flour on CFBS bread (Figures 3 and 4).

Generally, the hydrothermally treated cereal flours improved the quality of gluten free bread, 279 280 because the partial gelatinization of starch increased the dough consistency and likely the entrapment of gas during mixing and baking. Similarly, Tsai et al. (2012) reported that the 281 addition of rice porridge improved the texture of wheat bread, which was ascribed to the 282 283 gelatinization of rice starch granules. In addition, results showed the fundamental role of water hydration in the quality of the resulting gluten free breads, and in particular these gluten 284 free khobz eddar breads, based on rice or corn semolina. The moisture content of the breads 285 286 varied from 30 to 45 %, which agree with previous results in commercial gluten free breads (Matos & Rosell, 2012). Gluten free breads usually show high moisture content due to the 287 importance of hydrating the dough. In fact, the response surface obtained with RFBS and 288 CFBS breads showed the increase moisture content of breads when increasing water 289 hydration. For CFBS bread the moisture content ranged from 30 to 45 % and presented a 290 291 linear rise with increasing water and it was independent on the quantity of treated corn.

292 **3.2.3.** Optima gluten free breads

From the results obtained in the experimental design, optimized recipes for rice and corn based gluten free breads were developed with the aim to maximize specific volume, H/W ratio and minimize hardness, chewiness and moisture content. Figure 5 showed the crosssection of the bread slices compared to the references ones without treated flours.

The selection of improver content and water concentration was fundamental for obtaining the two gluten free breads with the best quality characteristics and they were obtained by

applying the desirability function. The optima concentrations were 7.59 g/100 g (semolina 299 300 blends basis) of rice improver and 96.66 g water/100 g semolina blends for the rice formula, with a desirability value of 0.747. At these concentrations of improver and water, maximum 301 specific volume and best textural parameters were obtained compared to the bread without 302 improver. For corn based bread, 4.73 g of corn improver and 78.81 g of water (based on 303 semolina blend) were obtained with a desirability value of 0.627. These concentrations of 304 305 corn improver and water allowed obtaining a bread with maximum specific volume and minimum hardness and chewiness comparing bread without improver. 306

The specific volume of the two gluten-free breads, textural properties (hardness, chewiness, 307 308 springiness and cohesiveness), H/W ratio and moisture content, besides the color and crumb image analysis of the selected gluten free breads are described in table 4. Optimization of 309 gluten free breads based on rice or corn gave different quality characteristics. Specific volume 310 of corn bread was higher than that of rice bread, which also led to softer crumbs when using 311 corn semolina. The estimated values of specific volume in optimally formulated bread were 312 2.24 cm³/g for RFBS bread and 2.60 cm³/g for CFBS bread. H/W ratio for RFBS is 0.79 and 313 for CFBS is 0.85. Bread containing treated corn presented higher specific volume and H/W 314 ratio than bread containing treated rice, which might be related to the pasting properties of the 315 316 improvers derived from rice or corn flour. Both breads presented the best textural parameters. For RFBS bread, it was obtained lower hardness and chewiness (566 g, 217) than its 317 counterpart without treated rice (2604 g, 1314). For CFBS optimized bread, (435 g, 298) 318 hardness and chewiness improved compared to the reference bread (495 g hardness and 330 319 chewiness). 320

The color of the breads was related to the color of the corresponding semolina, being in the case of CFBS higher b^* due to the yellowish color derived from corn semolina. No differences were observed in lightness (L^*). Lightness of ingredients plays an important role in bakery products due to consumer preferences. In fact, numerous efforts have been devoted to lighten the color of the grains and grains products (Metzger, 2003). The hue green (- a^*) varied from 0.55 for RFBS to 1.74 for CFBS.

Image crumb analysis revealed that CFBS breads contained higher number of small holes,
whereas rice based bread contained bigger holes. In both cases, breads exhibited an aerated
crumb structure.

330 4. Conclusion

Gluten-free breads based on rice or corn semolina were successfully developed following the 331 Algerian (khobz eddar) traditional breadmaking, but applying needed changes for making 332 333 gluten free products. A recipe based on rice and faba bean semolina or corn and faba bean semolina was used adding hydrothermally treated rice or corn flours as bakery improvers. A 334 experimental design, used to optimize the level of water hydration and the amount of 335 336 improver, revealed the importance of those factors in the quality characteristics of gluten free breads and allowed determining the optima levels for improving specific volume, H/W ratio 337 and textural parameters. The optimum gluten free rice/faba bean bread was produced by 338 incorporating 7.59 g of treated rice and 96.66 g of water (based on 100 g semolina blends), 339 whereas the optimum gluten free corn/faba bean bread was produced by incorporating 4.73 g 340 of treated corn and 78.81 g of water (based on 100 g semolina blends). According to the 341 results of specific volume, texture parameters, color and crumb structure, it can be concluded 342 that optimized rice or corn breads were obtained using hydrothermally treated slurries as 343 344 bakery improvers, leading to gluten free *khobz eddar* breads for celiac patients.

345 Acknowledgements

Authors acknowledge the financial support of Spanish Scientific Research Council (CSIC), the Spanish Ministry of Economy and Sustainability (Project AGL2014-52928-C2-1-R), the European Regional Development Fund (FEDER) and the Generalitat Valenciana (Project

- Prometeo 2012/064). H. Bourekoua acknowledges the financial support of Institute of
 Nutrition, Food and Agrofood Technology (I.N.A.T.A.A.).
- 351

352 **REFERENCES**

- AACC. (2000). American Association of Cereal Chemists. Approved methods of the AACC
 (10th ed.). St Paul, USA: American Association of Cereal Chemists.
- Arendt, E.K., & Dal Bello, F. (2008). Gluten-free cereal products and beverages. Academic
 press. A volume in *Food Science and Technology*. (1st ed.). 443 pages. London:
 Academic Press.
- Benatallah, L., Zidoune, M.N., & Michon, C. (2012). Optimization of HPMC and Water
 Addition for a Gluten-Free Formula with Rice and Field Bean Based on Rheological
 Properties of Doughs, *International Review of Chemical Engineering*, *4*, 474-481.
- Boukezoula, F., & Zidoune, M.N. (2014). Gluten-free diet adherence and its consequences
 on the nutritional and health status of 100 celiac patients in Tébessa. Algeria. *Médecine des maladies Métaboliques. Elsevier, 8,* 440-444.
- Chiang, B.Y., & Johnson, J.A. (1976). Gelatinization of starch in extruded products. *Cereal Chemistry*, 54, 436-443.
- Gallagher, E., Gormleya, T.R., & Arendt. E.K. (2004). Recent advances in the formulation of
 gluten free cereal- based products. *Journal of Food Science and Technology*, *15*, 143-152.
- Gonzales-Barron, U., & Butler, F. (2006). A comparison of seven thresholding techniques
 with the k-means clustering algorithm for measurement of bread-crumb features by digital
- image analysis. *Journal of Food Engineering*, 74, 268-278.
- Hager, A.S., & Arendt, E.K. (2013). Influence of hydroxypropylmethylcellulose (HPMC),
 xanthan gum and their combination on loaf specific volume, crumb hardness and crumb

- 373 grain characteristics of gluten free breads based on rice, maize, teff and buckwheat. *Food*374 *Hydrocolloids*, *32*, 195 -203.
- Hesso, N., Loisel, C., Chevallier, S., & Le-Bail, A. (2014). Impact of pregelatinized starches
 on the texture and staling of conventional and degassed pound cake. *Food Bioprocess Technology*, 7, 2923-2930.
- Hsi-Mei Lai. (2001). Effects of hydrothermal treatment on the physicochemical properties of
 pregelatinized rice flour, *Food Chemistry*, 72, 455-463.
- ICC. (1996). Standard methods of the International Association for Cereal Science and
 Technology, Vienna. Austria.
- Jane, J., Chen, Y.Y., Lee, L.F., McPherson, A.E., Wong, K.S., Radosavljevic, M. &
- Kasemsuwan, T. (1999). Effects of amylopectin branch chain length and amylose content
 on the gelatinization and pasting properties of starch. *Cereal Chemistry*, *76*, 629–637.
- 385 Klug Tavares, A. C., Zanatta, E., Zavareze, E. R., Helbig, E., & Guerra Dias, A. R. (2010).
- 386 The effects of acid and oxidative modification on the expansion properties of rice flours
- with varying levels of amylose. *LWT-Food Science and Technology*, *43*, 1213-1219.
- 388 Marti, A, Caramanico, R., Bottega, G., & Pagani, M.A. (2013). Cooking behavior of rice
- pasta: Effect of thermal treatments and extrusion conditions. *LWT Food Science and Technology*, 54, 229-235.
- Marco, C., & Rosell, C.M. (2008). Breadmaking performance of protein enriched gluten free
 breads. *European Food Research and Technology*, 227, 1205-1213.
- Matos, M.E., & Rosell, C.M. (2011). Chemical composition and starch digestibility of
 different gluten free breads. *Plant Food for Human Nutrition*, 66, 224-230.
- Matos, M.E., & Rosell, C.M. (2012). Relationship between instrumental parameters and
 sensory characteristics in gluten free breads. *European Food Research and Technology*,
 235, 107-117.

- Matos, M.E., & Rosell, C.M. (2013). Quality Indicators of Rice-Based Gluten-Free Bread Like Products: Relationships Between Dough Rheology and Quality Characteristics. *Food Bioprocess Technology*, 6, 2331–2341.
- Matos, M.E., & Rosell, C.M. (2015). A review: understanding gluten free dough for reaching
 breads with physical quality and nutritional balance. *Journal of the Science of Food and Agriculture*, 95, 653–661.
- 404 Metzger, L.E. (2003). Bleached grain and grain products and methods of preparation. U.S.
 405 454 patent 0,082.280 A1.
- Phatcharee, K., S. (2014). Optimization 406 Pitiporn, R., & Manop, of hydroxypropylmethylcellulose, yeast b-glucan, andwhey protein levels based on physical 407 properties of gluten free ricebread using response surface methodology. Food Science and 408 Technology, 57, 738-748. 409
- 410 Pongjaruvat, W., Methacanon, P., Seetapan, N., Fuongfuchat, A., & Gamonpilas, C. (2014).
- Influence of pregelatinised tapioca starch and transglutaminase on dough rheology and
 quality of gluten free jasmine rice breads. *Food Hydrocolloids*, *36*, 143-150.
- 413 Rosell, C.M., Santos, E., Sanz-Penella, J.M., & Haros, M. (2009). Wholemeal wheat bread:
- A comparison of different breadmaking processes and fungal phytase addition. *Journal of Cereal Science*, 50,272-277.
- Rosell, C.M., Collar, C., & Haros, M. (2007). Assessment of hydrocolloid effects on the
 thermo-mechanical properties of wheat using the Mixolab. *Food Hydrocolloids*, *21*, 452418 462.
- 419 Sandhya Rani, M. R., & Bhattacharya, K. R. (1995). Microscopy of rice starch granules
 420 during cooking. *Starch-Stärke*, 46,334-337.

- Singh, N., Singh, J., Kaur, L., Sodhi, N. S., & Gill, B. S. (2003). Morphological, thermal and
 rheological properties of starches from different botanical sources. *Food Chemistry*, *81*(2),
 219-231.
- Schoenlechner, R., Mandala, I., Kiskini, A., Kostaropoulos, A., & Berghofer, E. (2010).
 Effect of water, albumen and fat on the quality of gluten-free bread containing
 amaranth. *International Journal of Food Science and Technology*, 45, 661-669.
- Shibata, M., Sugiyama, J., Tsai, C. L., Tsuta, M., Fujita, K., Kokama, M., & Araki, T.
 (2011). Evaluation of viscoelastic properties and air-bubble structure of bread containing
 gelatinized rice. *Procedia Food Science*, *1*, 563-567.
- 430 Šimurina, O,D., Ikonić, B,B., Jevtić-Mučibabić, R.C., Belović, M.M., Koprivica, G.B., &
- Mišljenović, N.M. (2012). Application of response surface methodology in the
 development of specialty bread with sugar beet molasses, flax seed and vital wheat gluten. *Food and Feed Research*, *39*, 11-21.
- 434 Storck, R., da Rosa Zavareze, E., Gularte, M.A., Elias, M.C., Rosell, C.M., & Guerra Dias,
- 435 A.R. (2013). Protein enrichment and its effects on gluten free bread characteristics. *LWT* -
- 436 *Food Science and Technology.* 53, 346–354.
- 437 Tsai, C.L., Sugiyama, J., Shibata, M., Kokawa, M., Fujita, K., Tsuta, M., Nabetani, H., &
- 438 Araki, T. (2012). Change in the texture and viscoelastic properties of bread containing rice
- 439 porridge during storage. *Bioscience Biotechnology Biochemistry*, 76, 331-335.
- 440 Yvonne, C. (2007). 65°C Bread doctor. Orange culture Ltd.
- 441

442 FIGURE CAPTIONS

- **Figure 1.** RVA profiles of rice flour, hydrothermally treated rice flour after 1 h and after 24 h
- 444 storage. Legend: (----): temperature profile; (----) rice flour; (---) treated rice 1 h; (...)
- treated rice 24 h.

- 446 Figure 2. RVA profiles of corn flour, treated corn after 1h and pre-gelatinized corn after 24 h.
- 447 Legend: (----): temperature profile; (----) rice flour; (---) treated rice 1 h; (...) treated rice
 448 24 h.
- 449 Figure 3. Responses surfaces of experimental design for rice/faba bean semolina (RFBS)
 450 bread.
- 451 Figure 4. Responses surfaces of experimental design for corn/faba bean semolina (CFBS)
 452 bread.
- 453 Figure 5. Cross section of central bread slice obtained with the optima recipes for rice and
- 454 corn breads. A.Control rice based bread; B. Optimum rice based bread; C. Control corn based
- 455 bread; **D**. Optimum corn based bread.

456	Table 1. Factors, levels and code values used in the two Central Composite Designs (CCD1,
457	CCD2) for rice bread and corn breads, respectively.

	Code	values			Real va	alues
-	Hydration	Treated flour	CC	D 1	CC	D2
Run	(ml) X1	(g) X2	Hydration X1	Treated rice	Hydration X`1	Treated corn
			(ml)	X2 (g)	(ml)	X`2 (g)
1	1.41421	0	110	7.375	105	6.9
2	-1.41421	0	55	7.375	51	6.9
3	0	1.41421	82.5	14.75	78	13.8
4	-1	1	63.055	12.589	58.908	11.779
5	1	-1	101.945	2.160	97.092	2.021
6	1	1	101.945	12.589	97.092	11.779
7	-1	-1	63.055	2.160	58.908	2.021
8	0	-1.41421	82.5	0	78	0
9	0	0	82.5	7.375	78	6.9
10	0	0	82.5	7.375	78	6.9
11	0	0	82.5	7.375	78	6.9
12	0	0	82.5	7.375	78	6.9
13	0	0	82.5	7.375	78	6.9

Table 2. Analysis of variance (ANOVA) test tracing parameters for the response surfaces formula of rice/faba bean breads.

	Sequential sum of squares						
Source	Specific volume (cm ³ /g)	Texture parameters			H/W ratio Moisture content (%		
	(0111 (8)	Hardness (g)	Springiness	Cohesiveness	Chewiness (g)		
Lack of fit	0.040*	0.019*	0.072	0.804	0.000*	0.597	0.020*
Pure error	0.017954	400591	0.013233	0.028575	529	0.001887	3.032
F	7.52 ^{*NS}	11.69 ^{*NS}	5.21 ^{*NS}	0.33 ^{*NS}	3078.51 ^{*NS}	$0.70^{*\mathrm{NS}}$	11.35 ^{*NS}
R-Sq (%)	71.67	67.67	67.05	81.33	71.39	86.20	78.50

463 H/W: Height/Width ration; F: variance Ficher—Snedecor; *NS: not significant (P>0.05); *: Significant at $P \leq 0.05$.

Table 3. Analysis of variance test tracing parameters response surfaces formula corn/faba bean.

Sequential sum of squares						
Specific volume (cm ³ /g)				H/W ratio M	loisture content (%)	
	Hardness (g)	Springiness	Cohesiveness	Chewiness (g)		
0.008*	0.000*	0.061	0.449*	0.010*	0.173	0.182
0.02507	98.78	0.000106	0.001803	308.8	0.001073	1.251
19.26 * ^{NS}	23.2 ^{*NS}	5.81 ^{*NS}	1.09 ^{*NS}	16.94 ^{*NS}	2.79 ^{*NS}	2.69 *NS
74.90	96.38	71.82	91.78	89.37	75.47	97.41
	(cm ³ /g) 0.008* 0.02507 19.26 ^{*NS}	(cm ³ /g)Hardness (g) $0.008*$ $0.000*$ 0.02507 98.78 19.26 *NS 23.2 *NS	Specific volume (cm ³ /g) Texture Hardness (g) Springiness $0.008*$ $0.000*$ 0.061 0.02507 98.78 0.000106 19.26^{*NS} 23.2^{*NS} 5.81^{*NS}	Specific volume (cm ³ /g) Texture parameters Hardness (g) Springiness Cohesiveness 0.008* 0.000^* 0.001 0.449^* 0.02507 98.78 98.78 0.00106 19.26 *NS 23.2 *NS 5.81 1.09^* NS	Specific volume (cm ³ /g) Texture parameters Hardness (g) Springiness Cohesiveness Chewiness (g) $0.008*$ $\overline{0.000*}$ 0.061 $0.449*$ $0.010*$ 0.02507 98.78 0.000106 0.001803 308.8 19.26^{*NS} 23.2^{*NS} 5.81^{*NS} 1.09^{*NS} 16.94^{*NS}	Specific volume (cm ³ /g) Texture parameters H/W ratio M Hardness (g) Springiness Cohesiveness Chewiness (g) 0.173 0.008* $\overline{0.000^*}$ $\overline{0.061}$ $\overline{0.449^*}$ $\overline{0.010^*}$ $\overline{0.173}$ 0.02507 98.78 0.000106 0.001803 308.8 0.001073 19.26 *NS 23.2 *NS 5.81^{*NS} 1.09 *NS 16.94 *NS 2.79 *NS

468 H/W: Height/Width ration; F: Ficher—Snedecor variance; *NS: not significant (P>0.05); *: Significant at $P \leq 0.05$.

Λ	7	1
4	1	Ŧ

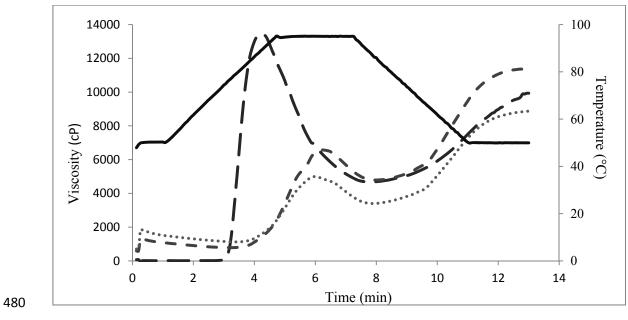
472	Table 4.	Characterisics	of optima	gluten	free rice of	or corn bread.
-----	----------	----------------	-----------	--------	--------------	----------------

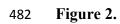
473			

Parameters	Rice/faba bean Bread	Corn/faba bean Bread	
Specific volume (cm ³ /g)	2.24 ±0.56	2.60 ±0.32	
Hardness (g)	566 ± 5	435 ±5	
Springiness	0.977 ± 0.011	0.918 ± 0.010	
Cohesiveness	0.766 ± 0.010	0.752 ± 0.040	
Chewiness	217 ±6	298 ±4	
Height/Width ratio	0.79 ± 0.13	0.85 ± 0.01	
Moisture content (%)	37.80 ± 0.69	36.78 ± 0.30	
L^*	61 ±1	61 ±1	
<i>a</i> *	-0.55 ±0.38	-1.74 ± 0.04	
<i>b</i> *	17.6 ±1.5	27.8 ± 0.1	
Image crumb analysis			
Number of holes	286 ±2	382 ± 0.1	
Total area (cm ²)	1.20 ± 0.1	1.44 ± 0.02	
Diameter (cm)	0.03 ± 0.02	0.02 ± 0.01	

Values are means±standard deviation.







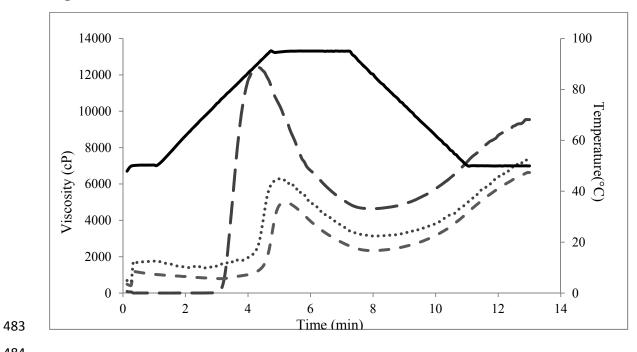
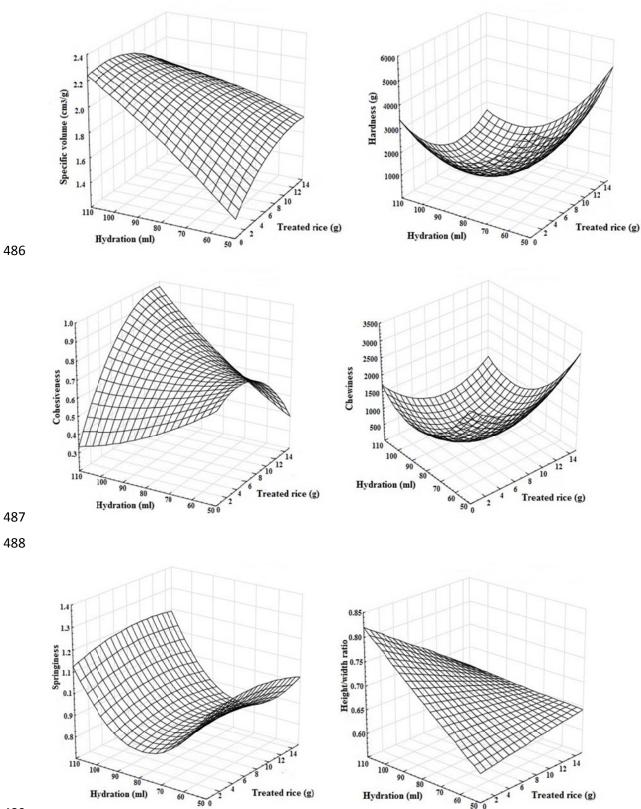


Figure 3.



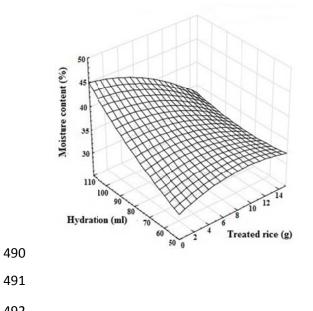




Figure 4.

