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**Effect of cocoa bean shells granulometries
on qualitative properties of gluten-free bread during storage**

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1 **Abstract**

2 Cocoa bean shells (CBS) are by-products of cocoa processing. Their rich chemical composition makes them interesting
3 ingredients to improve the overall quality of gluten free bread. The addition of different dimensional fractions (F1, 1.00-
4 1.99 mm; F2, 0.50-0.99 mm; F3, 0.355-0.49 mm) of CBS to gluten-free bread was investigated in terms of volume,
5 moisture content, crumb grain, texture and colour during 3 days of storage. No significant differences in moisture content
6 were observed among fractions, but protein, fat and fibre content resulted influenced by the dimensional range. With
7 reference to water absorption indexes, F2 showed the highest water binding capacity (WBC) and water holding capacity
8 (WHC), having instead the lowest water solubility index (WSI), in comparison to the other fractions, and confirming a good
9 retention of the fibres' structure. Results on bread showed that CBS addition modified crumb grain leading to a lower
10 specific volume, especially for F2. The presence of CBS accelerated significantly the crumb staling process in comparison
11 to the control, resulting however F1 softer than control at the end of the storage period. Finally, colour was improved by the
12 presence of CBS becoming darker, with F3 showing the highest impact on L^* and a^* .

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14 **Keywords:** cocoa by-products, celiac disease, shelf-life, water absorption, structure, colour

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18 **Introduction**

19 World cocoa beans (*Theobroma cacao* L.) production results of 5.2 Mtons in 2018 [1] with Côte d'Ivoire (2.0 Mtons),
20 Ghana (0.9 Mtons), Indonesia (0.7 Mtons), Nigeria (0.3 MTons), Ecuador (0.3 Mtons), Cameroon (0.3 Mtons) and Brazil
21 (0.2 Mtons) as the main producers. Cocoa bean shells (CBS) are the outer portions of beans that encase the nibs and, since
22 they represent 12-20% of the cocoa seed, they result as a by-product produced in great quantity. Chemical composition of
23 CBS is very interesting as protein content ranges from 11 to 18 g/100g dm, fat content from 2 to 21 g/100g dm, fibre
24 content from 43 to 60.5 g/100g dm and polyphenols from 1 to 5.8 g/100g dm [2-4]. Several efforts were spent in valorising
25 this waste material, starting from the use in animal feed with the limit of theobromine content (2–3%) [5] to the use for the
26 production of extracts rich in fibres, polyphenols, antioxidants and pectins [6-9]. In addition, in the last years several studies
27 on application of CBS fibre as a functional ingredient in food products were published. Martínez-Cervera et al. [10] used
28 soluble cocoa dietary fibre as a fat replacer in chocolate muffins with some advantages such as higher moisture, more tender
29 and crumbly structure and a reduction of hardening during storage. Similarly, Collar et al. [11] investigated the effects of
30 soluble dietary fibre from cocoa shell wheat bread and suggested that dietary fibres from cocoa shell added up to 6% could
31 be used to design technologically feasible, sensory acceptable, and long-term stored fibre-enriched wheat bread. In the same
32 way, Grillo et al. [12] obtained dietary fibre powder from CBS and substituted oat and whole wheat with no significant
33 differences between CBS powder and other fibres during organoleptic test. Recently, Jozinović et al. [13] successfully
34 enriched extruded snacks with cocoa husks and reported a proportional increase in resistant starch, polyphenol content and
35 antioxidant activity with the addition of cocoa husks, while starch damage slightly decreased. A similar approach was
36 followed by Rojo-Poveda et al. [14] who successfully obtained new functional beverages with potential health benefits.
37 Kreibich et al. [15] described CBS structure from a microscopical point of view and observed thick-walled protective cells
38 providing somewhat rigid and flexible support properties. Interestingly, authors established that the shell has different layers
39 and vascular bundles distributed among the tissues: the first tissue (epidermis) layer is comprised of long cells (smaller
40 sizes) with thick walls and the second of long flattened parenchyma cells, closely surrounding the vascular bundles. It is
41 possible to visualize different formats (spiral – tracheid), sizes and numbers of vascular tissues (protoxylem and phloem) for
42 water and nutrient transportation. Starting from this morphological characteristic, a technological approach, similar to what
43 reported by de Sousa et al. [16], could be applied for separating CBS into different granulometric fractions that potentially
44 had different profiles in chemical composition and technological properties, as consequence. Rojo-Poveda et al. [14]

45 demonstrated that the extraction of polyphenols for obtaining a functional beverage from CBS was directly influenced by
46 the grinding degrees when using percolation techniques, whereas the maceration technique showed no dependence on the
47 particle size. The present paper represents the first attempt to use sieving of CBS as a preliminary method for modifying
48 functional properties of this kind of industrial waste. Thus, the aim of this work was to evaluate the effects of the addition of
49 different CBS granulometries on physico-chemical properties of a commercial gluten-free bread formulation during 3 days
50 of shelf-life.

51 **Materials and Methods**

52 **Samples, breadmaking and storage**

53 A commercial (NT FOOD S.p.A., Altopascio, Lucca, Italy) gluten-free bread mixture with the following ingredients was
54 used: corn, rice cream soup, tapioca starch, sugar, vegetable fibres, salt, thickening agents: guar flour and
55 hydroxypropylmethylcellulose, flavourings. The proximate composition of the mixture was: moisture 9.7g/100g,
56 carbohydrate 85.7g/100g, fibres 1.9g/100g, protein 1.8g/100g, fat 0.9g/100g, salt 1.9g/100g. Ground CBS were kindly
57 donated by the company Ferri dal 1905, Castel Goffredo (MN), Italy.

58 To obtain the different particle sizes of CBS, it was classified in sieves with opening sizes 2.00 mm, 1.00 mm, 0.50 mm and
59 0.355 mm, respectively with an automatic sieve (Giuliani Tecnologie, Torino, Italy). The obtained fractions were named F1
60 (1.00 to 1.99 mm), F2 (0.50 to 0.99 mm) and F3 (0.355 to 0.49 mm).

61 Bread samples were prepared with the following recipe: gluten free mixture (250.0 g), water (225.0 g), sunflower oil (12.5
62 g), compressed yeast (6.25 g) (control, C). CBS from F1, F2 and F3 fractions were then added by substituting the mixture at
63 4 g/100g in order to achieve a total final fibre content equal to 3 g/100g for the claim “source of fibre” [17]. The amount of
64 water was determined on the basis of preliminary tests until obtaining a workable dough for all the formulations. The same
65 amount of water was added to all the recipes in order to evaluate the unique effect of the different CBS added fractions.

66 A domestic bread machine (Moulinex, Groupe Seb Italia S.p.A., Milano, Italy) was used for breadmaking, with the rapid
67 program: stirring + kneading + rising, 80 min; baking, 55min at 210 °C, obtaining sandwich bread loaves with mean weight
68 of about 400 g. Cooking losses after baking were measured and ranged from 22.1 ± 1.2 g/100g for C and F1 samples to
69 about 24.5 ± 1.3 g/100g for F2 and F3 breads. The cooked breads were cooled at room temperature, packaged in alcohol-
70 sprayed sealed air-tight plastic bags and stored in a 25 °C temperature-controlled chamber in the dark (ISCO 9000, Milan,
71 Italy). Samples were analysed at 0, 1 and 3 days of shelf life. Three loaves were used for the characterization of the breads
72 at each storage time analysing 9 loaves for each bread type.

73 **Proximate composition and water absorption tests of flour and CBS**

74 Moisture, protein, lipid and ash of cocoa bean shell fractions were determined using standard procedures [18]. Moisture was
75 determined in oven at 105 °C for 24 h. Crude fat content was determined using an automatized Soxhlet extractor (SER
76 148/3 VELP SCIENTIFICA, Usmate Velate, Italy) using diethylether. Total ash was determined after mineralization at 550
77 °C for 5 h + 5 h. Total nitrogen was determined with a Kjeldahl system (DKL heating digestor and UDK 139 semiautomatic
78 distillation unit, VELP SCIENTIFICA).

79 Water absorption tests on CBS fractions were determined as described by Sarangapani et al. [19], as average of three
80 replicates. Water binding capacity (WBC) and water holding capacity (WHC) were expressed as grams of water retained
81 per gram of solid. Water absorption index (WAI) and water solubility index (WSI) were expressed as grams of gel per
82 grams of sample (db) and grams of dried supernatant per grams of sample (db), respectively.

83

84 **Physical analyses**

85 Specific volume of breads was determined in triplicate according to the AACC Approved Method 10-05.01 [20] and
86 expressed as the volume/weight ratio of cooked bread (mL/g).

87 Crumb grain was evaluated by means of a digital image analysis system, as reported previously [21]. Images of three central
88 slices (20 mm thickness) of each loaf were acquired with a scanner (Hewlett Packard, Palo Alto, CA, USA) at 600 dots per
89 inch (dpi) taking squares (40x40 mm) from the centre of the images after calibration, standardization and optimization by
90 means of Image-Pro Plus 4.5 (Media Cybernetics Inc., USA) software. The number of pores (expressed as percentage of the
91 total number) was obtained according to pre-selected dimensional classes based on their area. Selected classes were: class-1:
92 0.01-0.099 mm²; class-2: 0.1-0.199 mm²; class.3: 0.2-0.99 mm²; class-4: > 1 mm².

93 The crumb moisture content (g/100g) within the bread loaves was evaluated in triplicate at each shelf-life time for each
94 bread type following the AACC standard method, 44-15.02 [20].

95 Texture analysis was performed on crust and crumb using a TA.XT2 Texture Analyzer equipped with a 25 kg load cell
96 (Stable Micro Systems, Godalming, UK) and Texture Expert for Windows software (version1.22) for data analysis on each
97 loaf. Crust hardness was measured by means of a puncture test using a 3 mm diameter stainless steel probe and a test speed
98 of 2 mm/s. Maximum peak force (N) was measured from the penetration curve and taken as crust hardness. Measurements
99 were taken on five preselected points of the crust.

100 Crumb evaluation was carried out on ten cubes of 20×20×20 mm extracted from two central slices of the samples. A TPA

101 test was performed with a 35 mm diameter cylindrical aluminium probe by means of a double compression with a speed of
102 2mm/s up to the 40% of the original sample height. The textural parameters considered were hardness (maximum peak
103 force of the first compression cycle, N), cohesiveness (ratio of positive force area during the second compression to that
104 during the first compression area, dimensionless), resilience (area during the withdrawal of the penetration, divided by the
105 area of the first penetration, dimensionless), and chewiness (product of hardness x cohesiveness x springiness, N) [22].

106 Colour was determined on ten pre-selected locations of the crust and crumb of each bread loaf. The analyses were
107 performed using a Minolta Colorimeter (CM 2600d, Minolta Co., Osaka, Japan) equipped with a standard illuminant D65
108 and a 10° position of the standard observer. The instrument was calibrated before each analysis with white and black
109 standard tiles. L^* (lightness), a^* (redness) and b^* (yellowness) were quantified on each sample using the Spectramagic
110 software (Ver. 3.6).

111 **Statistical analysis**

112 Means and standard deviations calculated with SPSS (Version 25.0, SPSS Inc., Chicago, USA) statistical software were
113 used to perform one way (ANOVA) to evaluate the effect of CBS addition at a significance level of 0.05 ($p < 0.05$). A
114 Tukey-Kramer post-hoc test at a 95% confidence level was also applied using the same software to verify the differences
115 among groups.

116 **Results and Discussion**

117 **Chemical and technological analyses of CBS fractions**

118 Moisture content of CBS (Table 1) was in the range 4.79-5.01 g/100 g with no significant differences among the considered
119 fractions. Observed moisture content was quite lower compared to those reported by Martínez et al. [2] but the composition
120 of CBS is reported to be quite variable and it depends, among other factors, on its origin and mainly on the process to which
121 it has been subjected. The highest ash content (Table 1) was observed for F3 while the lowest for F2 with F1 as intermediate
122 value: in general, obtained data are in the range previously reported for this cocoa co-product [2]. Regarding protein
123 content, no differences were observed between F2 and F3 while F1 presented a significantly lower value. Obtained data are
124 in the range already published in the scientific literature [3]. Moreover, the fat content resulted significantly different among
125 the three CBS size classes, being the lowest for F1 and the highest for F3, as also reported by other authors [3].

126 Finally, fibre content of F1 resulted the highest one with no differences between F2 and F3; the observed results confirm the
127 feasibility of fibre enrichment by means of grinding and sieving [23]. Starting with the work of Knuckles et al. [24]
128 separation of different fractions with different chemical composition and technological properties, as consequence, was

129 successfully achieved for cocoa bean shell.

130 Hydration properties of the fractions are reported in Table 1. WBC and WHC data presented a similar trend with the highest
131 values for F2 probably due to the higher fibre structure retention as WHC and WBC are strictly related to them [25]. The
132 same authors, working on coffee by-products, reported also an inverse relation with the granulometry of the material but in
133 the present work, this relationship was not observed. Technological functionality was in the range published by Lecumberri
134 et al. [26] for cocoa shell and according to their observations, it could be confirmed that all fractions presented only few
135 amounts of soluble fibres.

136 WAI and WSI can be used to indicate the behaviour of the starches contained in the three fractions [19]: F1 gave the highest
137 values while on the contrary F3 the lowest ones. Low WAI values are generally linked to high level of starch damage and
138 interactions with non-starch components and cell wall matrices [19, 27]. Actually, F3 samples represent the littlest fraction
139 with probably the highest impact of grinding on structure but with the highest content of starch that was subjected to
140 damages during the whole process of cocoa beans. This hypothesis is confirmed by the high WSI value of F3 fraction
141 probably due to the depolymerization of starch, which leads to the production of low molecular weight fragments of
142 amylose and amylopectin [19]. Thus, the decrease in the values of water interaction indexes could be explained by the fact
143 that some of the molecules of the solubilized material, in particular amylose molecules, may interact each other generating a
144 modified structure that impedes water absorption [28].

145 **Crumb grain characteristics and specific bulk volume of bread**

146 Specific volumes of tested breads are reported in Figure 1: at time 0 significant differences were observed among the cocoa
147 bean shell fractions. Particularly, F2 presented a significantly lower value compared to F1 and F3 probably due to its high
148 WHC and WBC values (Table 1) that are generally related to low volume in bread [29]. Moreover, F2 samples presented
149 both a higher cooking loss (24.8 ± 0.6 %) and a higher percentage of holes belonging to the greatest class (Figure 2)
150 compared to the other samples. Results are in agreement with Coda et al. [30] that reported a volume variation depending on
151 wheat bran particle size. Similarly, Gómez et al. [31], studying the effect of fibre size on the quality of fibre-enriched cakes,
152 reported a relation between specific volume, regardless the origin of fibres, and dimensions of them. During storage,
153 specific volume of C samples remained constant with no significant variations. Otherwise, all CBS-enriched breads
154 presented a significant volume reduction (Figure 1), also showing a significant increase in the percentage of holes belonging
155 to the greatest class and a decrease in the lowest one, as consequence, with a significant variation in the total number of
156 holes only for F1 samples (data not shown). During storage F1, F2 and F3 breads showed a little weight loss (data not

157 shown) ranging from 1 to 2 % that, together with water migration from crumb to crust, could have caused the reduction of
158 wall thickness and the increase of holes' dimensions, as consequence. On the contrary, C samples did not show any weight
159 loss during storage.

160 The presence of fibres contained in cocoa bean shells caused a significant reduction of total number of holes (about 350
161 holes/16cm²) compared to C samples (about 600 holes/16cm²) with a higher presence of holes with large dimensions
162 (Figure 2). Bigger holes are more sensible to collapse and this fact could explain the volume reduction observed for F1, F2
163 and F3 samples (Figure 1). Probably, cocoa shell fibres interfered with thickening agents (guar flour and
164 hydroxypropylmethylcellulose) of the gluten free mixture used for the preparation of breads as a competitor for water when
165 water is limited [32].

166 Crumb moisture content (Figure 3) of enriched samples were significantly lower than C ones at all storage times confirming
167 the higher evaporation caused by a coarser crumb grain characteristic observed for F1, F2 and F3 samples (Figure 2). As
168 stated above, weight loss after cooking was significantly higher for F2 and F3 samples while no statistical differences were
169 observed for others. During storage, no samples showed significant difference in crumb water content thanks to packaging
170 and to the short-term shelf life.

171 Crust colorimetric data (Table 2) showed significant differences due to the addition of cocoa bean shell. Particularly, crust
172 L* values increased and a* decreased thanks to the cocoa bean fibre probably due to the lower moisture content (Figure 3)
173 that generally leads to lighter bread crust [33]. On the contrary, the impact of cocoa bean shell on bread crumb colour was
174 evident: addition of CBS caused a significant reduction of L* and simultaneously a significant increase of a* and b* (Table
175 2) with darker crumb in accordance with Kārklīņa et al. [34], due to the natural colour of cocoa bean shells. F3 showed the
176 greatest impact on L* and a* values probably due to the very low dimensions of particles. In general, colorimetric changes
177 caused by cocoa bean shells' addition can be positively considered as gluten free breads are generally reported to present
178 pale colour that is not attractive for consumers [35].

179 Bread crumb and crust hardness (Table 3) at time 0 days were not significantly affected by cocoa bean shell addition.
180 During storage, F2 samples showed the highest hardness increase at time 3 with a value two folds higher compared to the
181 control. This could be due to the presence of largest holes that generally tend to collapse (Figure 3) and to the high WHC
182 value (Table 1) of this fraction with a probably strong competition for water with starches and thickening agents [36]. On
183 the contrary, F1 gave the lowest hardness value at time 3 with significant lower values also compared to C, demonstrating
184 an antistaling effect probably due to the high WAI value (Table 1) of this fraction related to a high water retention capacity

185 which could help to prevent the retrogradation of amylopectin [37]. Cohesiveness (Table 3) decreased for all samples with
186 increasing storage time, as expected [38]. All samples added with cocoa bean shell fractions exhibited lower values at day 1
187 compared to C in accordance with Rocha Parra et al. [39] probably in relation with crumb moisture content that resulted
188 lower in F samples.

189 Regarding resilience data at time 0 (Table 3), F2 bread showed a significantly lowest value compared to the others probably
190 due to the lowest volume and to the presence of a highest percentage of holes belonging to the greatest class. Characteristics
191 of F2 breads could be probably related to water hydration properties of this CBS fraction. In general, CBS enriched breads
192 presented significantly lowest resilience values compared to C at all times probably due to the lowest total number of holes
193 per area and to the highest presence of holes with great dimensions. Collar et al. [11] already reported significant reduction
194 of resilience in breads with soluble dietary fiber preparations obtained from cocoa shell.

195 Finally, chewiness values of C samples were in almost all cases significantly higher compared to the enriched ones:
196 chewiness is a secondary texture parameter that is associated with difficulty in chewing the sample and forming a bolus
197 before swallowing. These results could be related to the greater moisture retained by C samples (Figure 3), making them
198 denser to chew [10].

199 **Conclusions**

200 This study attempted to evaluate the effect of the addition of three different dimensional fractions from cocoa bean shells on
201 gluten free bread during 3 days of storage. Different proximate composition and water absorption properties observed for
202 the different fractions of cocoa bean shell were reflected also in bread characteristics. The fibre deriving from CBS
203 fractions, probably, negatively influenced the crumb grain, with coarser structure, as well as specific volume and moisture
204 content, with significantly lower values. On the other hand, the CBS supplementation led to a pleasant darker colour, with
205 F3 showing the more consistent improvement. Moreover, textural attributes (hardness) confirmed the possibility to add CBS
206 F1 fraction with limited impact on product quality also during storage.

207 Overall, it was evidenced that cocoa bean shell fractions represent a promising functional ingredient for the development of
208 improved gluten free bread formulations. Nevertheless, further studies would be necessary to improve the characteristics of
209 final product also evaluating sensorial attributes as well as water dynamic during shelf life.

210 **Compliance with ethical standards**

211 **Conflict of interest** The authors declare that they have no conflict of interest.

212 **Compliance with ethics requirements** This article does not contain any studies with human or animal subjects.

213

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315 **Captions for figures**

316 **Figure 1:** Specific volume of tested breads. Error bars represent +/- 1 standard deviation, (n = 3,
317 sample size = 3 for each bread type). Different capital letters indicate significant differences ($p < 0.05$)
318 among different times for the same bread while different lowercase letters indicate significant
319 differences ($p < 0.05$) among the four types of bread at the same storage time.

320 **Figure 2:** Number of holes, as percentage of the total number, for the selected dimensional classes at
321 time 0 day. Error bars represent +/- 1 standard deviation, (n = 9, sample size = 3 for each bread type).
322 Bars of histograms with the same letters for each class are not significantly different ($p < 0.05$).

323 **Figure 3:** Crumb moisture content for tested breads during storage. Error bars represent +/- 1 standard
324 deviation, (n = 9, sample size = 3 for each bread type). Different capital letters indicate significant
325 differences ($p < 0.05$) among different times for the same bread while different lowercase letters
326 indicate significant differences ($p < 0.05$) among the four types of bread at the same storage time.

327

328 **Table 1.** Water binding capacity (WBC), water absorption index (WAI), water holding capacity (WHC), water solubility index (WSI) and proximate
329 composition of cocoa bean shell fractions^a

	moisture	ash	protein	fat	fibre	WBC	WAI	WHC	WSI
F1	4.9±0.3a	6.0±0.1b	15.2±0.4b	6.9±0.6c	63.7±0.6a	2.89±0.16b	5.52±0.16a	3.22±0.18ab	0.124±0.062a
F2	4.7±0.2a	5.7±0.1c	17.3±0.3a	14.6±1.6b	59.4±0.6b	3.75±0.13a	4.38±0.23b	3.50±0.03a	0.023±0.004b
F3	5.1±0.2a	6.7±0.1a	18.2±0.6a	21.8±1.9a	57.2±0.7b	2.95±0.20b	3.98±0.18c	3.08±0.11b	0.150±0.073a

330

331 ^a n=3. Means in columns followed by different letter differed significantly ($p < 0.05$).

332 Hydration properties are expressed as g/g while proximate data are expressed as g/100g

333

334 **Table 2.** Crumb and crust colorimetric parameters for analysed breads. ^a

	<i>day</i>	<i>L*</i>	<i>a*</i>	<i>b*</i>	<i>L*</i>	<i>a*</i>	<i>b*</i>
		<i>Crust</i>			<i>Crumb</i>		
<i>C</i>	0	78.77±0.01 B	1.84±0.01 aA	13.01±0.03 aB	86.99±2.42 aA	0.128±0.035 cB	6.59±0.13 aA
	1	83.83±1.27 bA	1.98±0.31 aA	15.97±0.99 aA	81.12±2.18 aB	0.440±0.121 dA	6.97±0.43 cA
	3	83.26±0.38 bA	0.42±0.10 bB	7.92±0.36 cC	83.26±0.38 aAB	0.307±0.135 dA	6.70±0.78 cA
<i>F3</i>	0	84.04±0.88 aA	2.03±0.16 aA	12.67±0.71 aA	71.96±1.56 cA	2.207±0.251 aA	9.27±0.46 bA
	1	84.75±2.11 bA	1.61±0.28 aB	11.46±0.89 bA	73.82±2.18 bA	2.038±0.301 aAB	8.80±0.66 bB
	3	83.12±1.49 bA	1.84±0.22 aB	11.76±1.08 bB	75.42±2.18 bA	1.866±0.316 aB	10.21±0.99 aA
<i>F2</i>	0	84.23±2.10 aA	1.37±0.38 bA	14.09±1.30 aA	81.08±1.92 bA	1.367±0.38 bA	14.33±1.28 aA
	1	86.76±0.69 abA	0.84±0.16 aA	10.89±0.54 bB	76.42±2.19 abB	1.050±0.286 cB	8.46±0.48 bB
	3	84.36±1.72 bA	1.63±0.37 bA	15.17±1.16 aA	81.84±2.97 aA	1.321±0.2001 bA	11.08±0.94 aAB
<i>F1</i>	0	86.26±2.33 aA	1.19±0.13 bA	10.54±0.88 bA	73.09±1.85 cA	1.398±0.155 bAB	8.88±1.25 bB
	1	88.02±1.37 aA	0.80±0.19 bB	11.58±1.05 bA	74.75±2.11 bA	1.609±0.284 bA	11.45±0.89 aA
	3	87.85±1.37 aA	0.82±0.19 bAB	10.88±1.05 aA	73.73±5.01 bA	0.907±0.270 cB	8.03±0.88 bB

335 ^a n=10. Means in column followed by different capital letters significantly differ ($p < 0.05$) among different times for the same bread. Means followed by different
336 lowercase letters significantly differ ($p < 0.05$) among the four types of bread at the same storage time.

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Table 3. Crumb textural profile analysis (TPA) and crust penetration parameters for the four analysed breads during storage.^a

<i>crumb</i>						<i>crust</i>
	<i>day</i>	<i>Hardness (N)</i>	<i>Cohesiveness</i>	<i>Resilience</i>	<i>Chewiness (N)</i>	<i>Hardness (N)</i>
<i>C</i>	0	0.53±0.06 aB	0.77±0.10 aA	100.10±0.32 aA	0.43±0.09 aB	1.92±0.11 aA
	1	0.76±0.12 bB	0.84±0.09 aA	102.67±3.02 aB	0.90±0.22 aA	1.16±0.15 bB
	3	1.57±0.48 bA	0.64±0.08 aB	103.16±3.87 aC	1.10±0.42 abA	2.22±0.46 bA
<i>F3</i>	0	0.52±0.10 aB	0.78±1.07 aA	98.68±0.02 aA	0.40±0.19 aB	1.83±0.49 aC
	1	1.21±0.24 aAB	0.58±0.02 aB	97.05±0.71 bA	0.63±0.15 bA	2.39±0.51 aB
	3	1.43±0.34 bA	0.55±0.06 bB	91.53±3.99 bA	0.72±0.16 bA	3.30±0.31 aA
<i>F2</i>	0	0.61±0.21 aC	0.74±0.06 aA	81.55±15.48 bB	0.35±0.10 aB	1.84±0.43 aA
	1	1.31±0.29 aB	0.60±0.04 bB	98.00±0.53 bA	0.71±0.16 bB	1.52±0.31 bB
	3	2.93±0.47 aA	0.58±0.09 aB	97.06±1.14 bA	1.64±0.25 aA	1.81±0.19 bA
<i>F1</i>	0	0.57±0.16 aB	0.72±0.02 aA	98.77±0.42 aA	0.43±0.11 aA	2.18±0.60 aAB
	1	0.84±0.07 bA	0.60±0.04 bB	97.55±0.51 bAB	0.51±0.06 bA	2.61±0.27 aA
	3	0.95±0.24 cA	0.57±0.08 aB	92.62±4.46 bB	0.49±0.13 bA	1.91±0.26 bB

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341 ^a n=5 for crust and n=10 for crumb. Means in column followed by different capital letters significantly differ ($p < 0.05$)
342 among different times for the same bread. Means followed by different lowercase letters significantly differ ($p < 0.05$)
343 among the four types of bread at the same storage time

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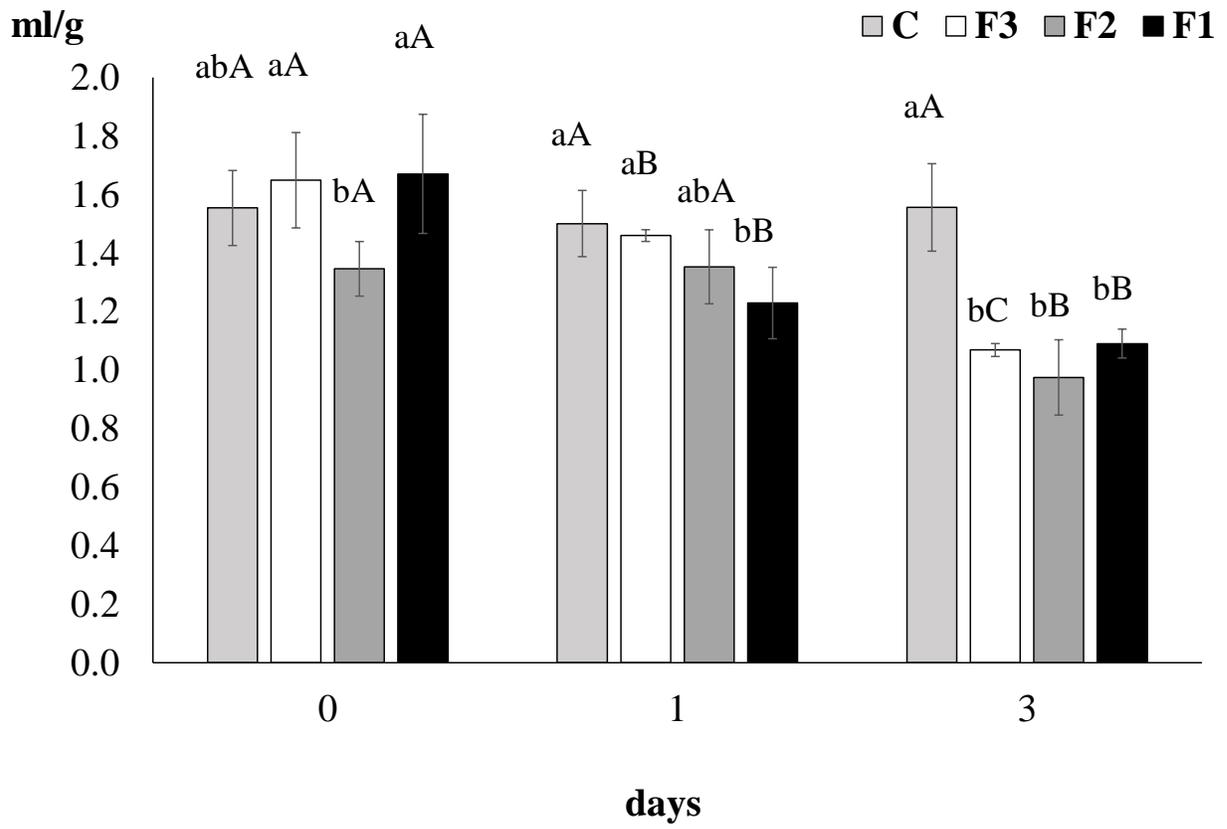
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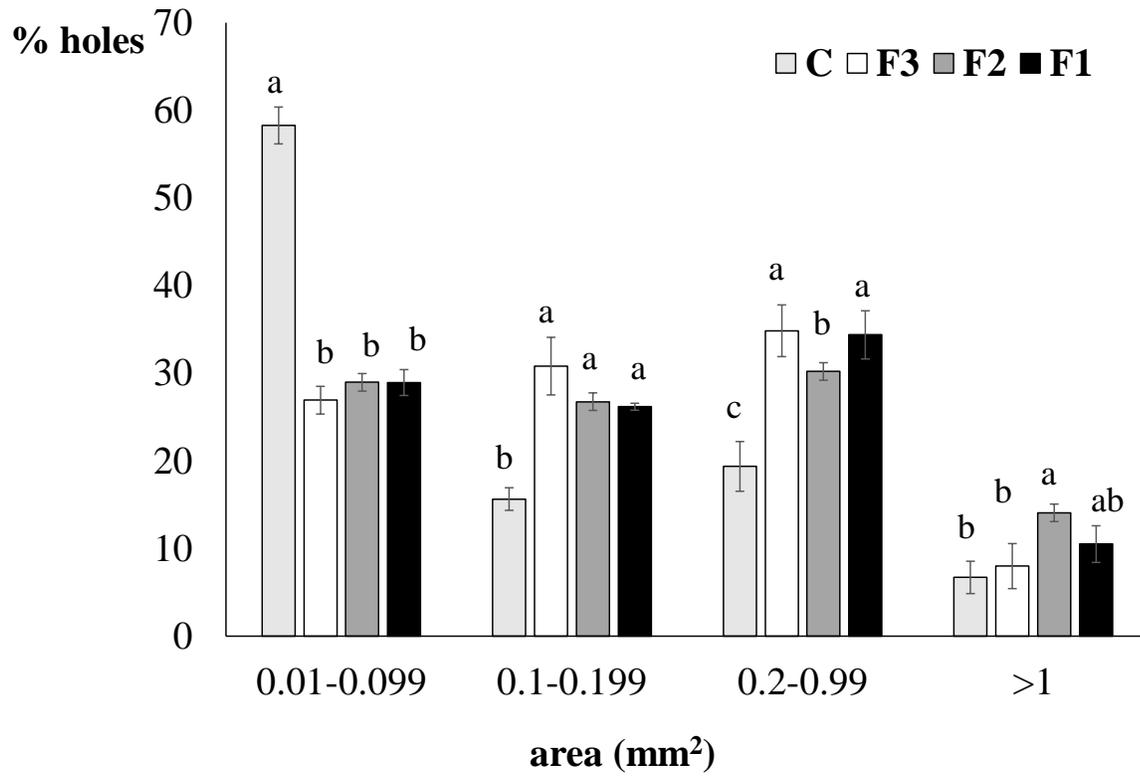
349 **Figure 1.**



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352 **Figure. 2.**



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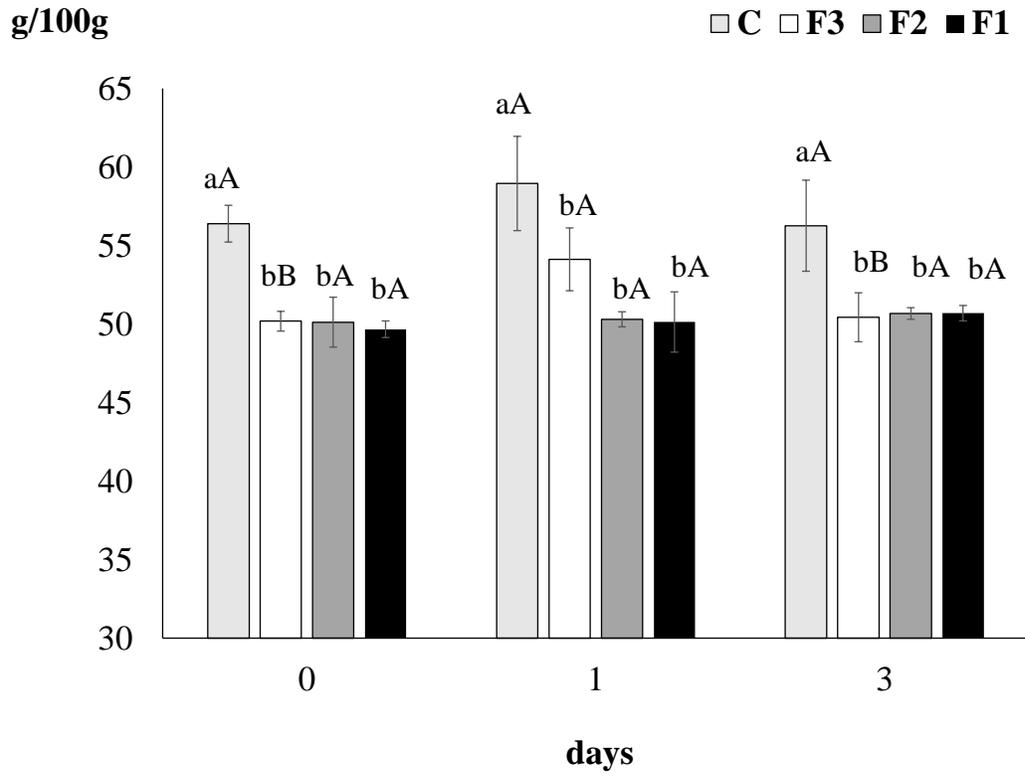
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357 **Figure. 3.**

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