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Sensory evaluation of gluten-free breads assessed by a trained panel of celiac assessors

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Abstract

An increase in celiac consumers has caused an increasing interest of food companies in research and development of substitute products, which may exhibit higher sensory acceptability. Although specific gluten-free breads (GFBs) have been developed, little is known about the sensory profile of such products. The purpose of the present study was to apply the sensory profiling method to the six most consumed gluten-free breads in the Italian market in order to identify sensory descriptors that can best characterize these products. Sensory evaluation was combined with chemical and physical measurements.

Products were evaluated by ten trained celiac assessors who identified 17 descriptors for appearance, aroma, taste and texture. The most significant variables in discriminating among samples were the sensory descriptors Porosity, crust and crumb Color, Softness by hand and mouth, Cheese odor, Corn odor and Fermented odor, Sweet, Salty, Adhesive and Rubbery) and the instrumental parameters associated to crust and crumb color and texture. A good correlation between sensory and instrumental measurements was observed.

The identification of the most relevant sensory features of GFB could facilitate the setting up of new formulations of bakery products that interpret at best the hedonic dimension of this increasing target of consumers.

***Keywords**: gluten-free bread, sensory profiling, color, texture, food perception*

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Introduction

In the early 1980's celiac disease was considered a rare condition. At present, also as a result of improved screening tests, celiac disease is known to be one of the most frequent chronic diseases, showing a prevalence in the European population of 1:100 [1].

The increasing incidence of this pathology, and resulting increase in demand for gluten-free products, has caused a growing interest of food companies in research and development of a wide range of substitute products, which may exhibit higher sensory acceptability. Until a few decades ago, even finding some food products suitable for celiacs was a major concern. Once this problem has been overcome, research focused on identifying raw materials that are "technologically" similar to wheat flour, which is the main ingredient in the western diet.

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Gluten has unique viscoelastic properties, which supply wheat with a specific technological quality required for both bread making and pasta making. Gluten has also additional functional properties (*i.e.*, high water sorption capability, swelling, networking/structuring, emulsifying and foaming properties), which make it suitable for food industry [2]. Conversely, corn rice, tapioca and potato flours, which are allowed in a gluten-free diet, are not able to supply the same technological characteristics as gluten.

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Even though replacement of gluten is one of the major challenge for gluten-free products development, only recently research has been carried out on this topic. GFB requires polymeric substances that mimic the viscoelastic properties of gluten in bread dough. Formulation of GFBs mainly involves the incorporation of starch, pre-gelatinized starch, hydrocolloids, protein-based ingredients like dairy or soy proteins and pseudo-cereals [3, 4].

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Although replacement of wheat flour with alternative novel/functional ingredients may improve technological and nutritional aspects, it is important to ensure that GFBs have appropriate characteristics of appearance, aroma, taste and texture, which are key determinants for celiac consumers' sensory acceptability. Indeed, it is reported that dissatisfaction with both the availability and the hedonic dimension of gluten-free food items is determinant for non-compliance with gluten-free diet [5]. Despite this, literature studies that pay the same attention to sensory properties of GFB as compared to technological, nutritional and safety aspects are scanty.

Arendt and colleagues [6] indicated that most of the gluten-free bakery products on the market have very poor quality, particularly when compared to traditional wheat flour yeast bread, since they have reduced flavor and a crumbly and dry texture.

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Gujral et al. [7] indicated that problems related to the structure and volume of GFB crumb are observed when rice flour is used as wheat substitute.

Mezaize et al. [8] found that bread prepared with buckwheat flour had an increased specific volume, a softer texture, and color characteristics and gas-cell size distribution similar to traditional bread.

However, in the above-mentioned studies the sensory properties of GFB were determined by means of instrumental devices instead of a group of assessors.

Very few works recently [9-11] described the sensory attributes of GFB using a trained panel. Nevertheless, it is noteworthy that in these cases sensory evaluation was performed by non-celiac subjects, who are not the specific consumer target of gluten-free products.

The purpose of the present study was to apply the sensory profiling method [12] involving a panel of celiac subjects in order to identify the sensory attributes that best characterize the properties of appearance, aroma, taste and texture of the 6 leader GFBs on Italian market. Sensory evaluation was carried out along with physicochemical analyses in order to obtain a more complete product description.

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Materials and Methods

Materials

Six commercial GFB samples were considered for the study (Fig. 1).

Insert Fig. 1 about here

Samples were chosen on the basis of purchase data in Italy, which were used to get the brands with the highest market share. GFB ingredients and their nutritional values are reported in table 1. It is worth noting that, according to the European Council Directive 89/398 [13] relating to foodstuffs intended for particular nutritional uses, specific labeling requirements are to be adopted by producers/importers. In fact, when a product as referred to above is placed on the market for the first time the producer or the importer, shall notify the competent authority of the Member State where the product is being marketed by forwarding it a model of the label used for the product. An indication of the nutritional characteristics of the product must be provided on the label and the producer/importer shall demonstrate compliance of the product with its claimed nutritional purposes.

Insert table 1 about here

All the samples considered in the present study were sliced ready-to-consume breads, with the exception of sample A which required a final baking phase before consumption. However, information for use reported on the labels of each GFB suggested to heat the sample in order to obtain a better product from a sensory point of view. For this reason, samples (10 slices of each GFB) were heated at 180°C for 7 min (sample A), 4 min (samples, B, C, D E) and 3.5 min (sample F), according to the information for use reported on their labels. A laboratory ventilated electric oven (model S 98, Smeg, S.p.A., Italy), which had been pre-heated to 180°C was used.

Sensory evaluation, water activity, moisture content and the rheological properties of the GFBs were performed strictly within 1 h after sample heating. All the other physicochemical parameters were evaluated within the same day.

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All the GFBs and the relevant breads used as reference standards for the formal sensory sessions were purchased and analyzed after the same storage time (within 7 days) from the production date, in order to minimize possible differences due to the staling process.

Physicochemical evaluation

Crumb moisture content of GFBs was determined before and after the heating phase by oven-drying samples for 5 h at 105°C, and expressed as g_{water} /100g_{sample} [14]. Water activity (a_w) of the crumb was measured by a dew point hygrometer (AquaLab®, Decagon Devices Inc., Washington).

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For titratable acidity and pH evaluation, GFB was diluted with water (10 g of crumb in 100 mL) and homogenized for 30 min. pH was measured with a model PHM62 pHmeter (Radiometer, De Mori S.p.A., Milan, Italy). Titratable acidity was determined by recording the volume of 0.1 N NaOH required to increase the pH to 8.5 [15].

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Color was evaluated using a Cr 210 Chromameter (Minolta Ldt, Milano), which provided the Hunter L^*, a^*, and b^* colorimetric coordinates. Color parameters range from $L^* = 0$ (black) to $L^* = 100$ (white), - a^* (greenness) to + a^* (redness), and - b^* (blueness) to + b^* (yellowness). The chromameter was calibrated with a white standard. Yellow Index (YI) was calculated according to Pasqualone et al. [16] by the following equations:

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$$\text{Yellow Index} = b^*/L^* \times 142{,}86 \qquad (1)$$

The colorimetric parameters (L_o^*, a_o^*, and b_o^*) of a commercial loaf made with wheat flour by Mulino Bianco, Barilla S.p.A were taken as a reference to evaluate ΔE according to the equation:

$$\Delta E = ((a^*-a_o^*)^2 + (b^*-b_o^*)^2 + (L^*-L_o^*)^2)^{1/2} \qquad (2).$$

The colorimetric parameters for the reference product were as follows: $a_o^* = 14.05$, $b_o^* = 32.80$, $L_o^* = 59.90$ (crust); $a_o^* = -1.13$, $b_o^* = 19.29$ $L_o^* = 81.72$ (crumb).

Firmness of the GFB samples was measured on a Texture Analyzer (Stable Micro Systems, Vienna Court, England) equipped with Texture exponent software, according to Raffo and colleagues [17]. From the central part of each GFB cylindrical discs were cut. Compression of discs was performed under the following conditions: cross-head speed 15mm/min; plunger diameter: 160 mm; load cell: 15g; deformation: 40%. The linear section of the stress-strain plot was used to determine the Young modulus by the following equation:

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$$E = \text{stress/strain} = (F/A)/(\Delta L/L) \qquad (3)$$

where E = Young' s modulus; F/A =force/surface of the plunger, or stress; ΔL/L = relative displacement or strain.

All the instrumental analyses were performed on three replicates, the same used for the formal sensory evaluation

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Sensory evaluation

The sensory profiling method [12] was applied to all the GFB samples. Ten celiac panellists (5 females and 5 males, aged between 20 and 33) recruited amongst the students and staff of the University of Milan, took part in the experiment.

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The training phase consisted of ten 1-h sessions over a period of 2 months to develop terminology to describe the key sensory attributes of GFB samples [18]. First, panellists were asked to write down terms describing appearance, aroma, taste, flavor and texture sensations that, in their opinion, represented at best GFB samples. As training progressed, descriptive terms were defined through panel discussion and redundant terms were excluded by panel consensus. Panel discussions also determined the order of appearance for each term and the reference standard used to anchor the scale endpoint label.

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Overall, 22 sensory attributes covering appearance, aroma, taste, flavor and texture were generated. The list of sensory attributes, with their relevant definition and reference standard is reported in table 2.

Insert table 2 about here

During the formal evaluation, panellists evaluated in triplicate three GFB samples per session over the course of 2 days of evaluation. Assessors received 1 full slice of bread and were asked to rate the intensity of each sensory attribute using a continuous, unstructured 100 mm line scale anchored at both extremes (“not at all intense” on the left and “very intense” on the right).

Samples were served in plastic plates coded with 3-digit numbers and evaluated in individual booths under white light at room temperature. In order to balance the effects of serving order and carry-over, presentation orders were produced according to a Latin square and were systematically varied over the tests [19]. The sessions were held at the sensory laboratory of DISTAM (University of Milan) designed according to ISO guidelines [20].

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Data analysis

One-way Analysis of Variance (ANOVA) was performed on physicochemical data considering *Samples* (6) as factors and instrumental parameters as dependent variables.

Sensory data were analyzed by means of 3-way ANOVA considering *Samples* (6), *Judges* (10), *Replications* (3) and their relevant 2-way interactions as factors, and sensory attributes as dependent variables.

When the ANOVA showed a significant effect ($p<0.05$), the Least Significant Difference (LSD) was applied as a multiple comparison test using the statistical software program STATGRAPHIS PLUS version 5.0 (Manugest KS Inc., Rockville, USA).

Principal Component Analysis (PCA) was performed on sensory and instrumental data using The Unscrambler® version 9.7 (CAMO PROCESS AS, Norway). The variables were standardized (1/sdev) and full cross validation was applied.

Results and Discussion

Instrumental analyses

Table 3 shows the physicochemical characterization of the 6 GFB samples.

Insert table 3 about here

Samples were significantly different for all the instrumental parameters considered.

Young's modulus was evaluated to compare the crumb texture of GFBs, which is linked to the cellular structure including two macroscopic phases, namely the gas cells and cell walls solids [21].

Concerning texture, sample A was found to be the hardest sample, being characterized by the highest Young's modulus value, which was similar to that of a wheat flour bread in which staling is begun [21]. Samples E and D, which is the leader product in the Italian market, were found to have an intermediate Young's modulus value, which is in good agreement with that of conventional fresh wheat flour bread [21]. This was an indication of a good choice of gluten substitutes, namely polymeric substances that mimic the viscoelastic properties of gluten in bread dough. Samples B, C and F were found to be the softest GFB samples.

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As expected, samples with a higher moisture content and a_w (B and C) had the softest texture; on the contrary, sample A, which was characterized by the hardest texture, was found to have a significantly lower moisture content and a_w than the other GFB samples.

Color measurements were carried out on both GFB crumb and crust. ΔE values, which represent the difference in term of color between each GFB sample and a reference sample (white wheat bread), evidenced that sample C was the most different in terms of color from the reference sample, being characterized by a high ΔE value for both crumb and crust and the absence of yellow color (extremely low YI crumb value). This result should be expected since sample C was produced mainly with rice flour and starch. On the contrary, all other samples were produced mainly using corn starch. Among GFBs containing corn, sample A, which had the lowest protein content, had a remarkably high ΔE value for the crust, depending on low a* and b* values and high L* value. Opposite to this sample, samples which were characterized by higher protein content, especially sample E, had a lower ΔE value due to lower L* value and higher a* and b* values for the crust. This behavior is indicative of crust browning due to the Maillard reaction between reducing sugars and amino acids. The darkening of the crust colour is desirable as gluten free breads tend to have a lighter crust color than white wheaten breads [22].

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Sensory evaluation

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Results provided by judges during replicates for each descriptor and for each sample, were analyzed by means of 3-way ANOVA. This procedure enabled us to investigate whether significant differences could be observed for single factors

(*Judges*, *Samples*, *Replicates*) and for interaction factors (*Judges by Replicates*, *Judges by Samples*, *Samples by Replicates*) for all the sensory descriptors considered.

The interaction *Judges by Replicates* refers to how reliable the judges were during replicates (*i.e.* degree of training), *Judges by Samples* refers to judges agreement in the use of the scale during the evaluation, *Samples by Replicates* refers to the invariability of GFB samples characteristics during replicates.

ANOVA results showed that all the sensory descriptors considered significantly (p<0.001) discriminated the GFB samples (table 4).

Insert table 4 about here

The factors *Judges* and *Judges by Samples* were also significant (p<0.001) for all the sensory descriptors, thus indicating some inconsistencies among panellists. Inconsistencies among panellists are common in sensory evaluation and may be ascribed to a different use of the scale. One reason of this inconsistency may be explained by the fact that the tasting group was composed by subjects who were following a gluten-free diet from birth and subjects who were diagnosed celiacs during childhood or adulthood and, therefore, had a memory of the sensory characteristics of traditional wheat bread which may have had an influence on their judgment. Indeed, it has been shown that experiences with sensory stimuli (for instance a flavor), learned during the first period of life, are highly influential on hedonic perception of food later in life [23,24]. Therefore, it is reasonable to hypothesize that consumers, who were diagnosed celiacs at birth, have a sensory and hedonic perception of food, which differs from that of consumers, who were diagnosed celiacs during adulthood. As compared to the former, the latter, due to their "memory" of the sensory properties of wheat-based bakery food, are likely to be more difficult with gluten-free products. As a result, they should be considered as a distinct category of celiac consumers, which may not be easily satisfied, when setting up gluten-free food formulations.

Despite this inconsistency among judges, *Replicates* and *Samples by Replicates* were not significantly different for most of the sensory descriptors as well as the interaction *Judges by Replicates* that show a good panel reliability for all the attributes with the exception of Fermented aroma.

Mean ratings and multiple comparison test results for each sensory descriptor and for each GFB sample are reported in table 5.

Table 5

As can be observed, GFBs B and E were found to have the most uniform crumb porosity, whereas GFB A, resulted to have a less homogeneous crumb porosity. Low homogeneity of the pore size in sample A, which is almost a "protein-free" bread, likely comes from a very low protein content in the formulation. Difficulty in managing GFB of very high starch content and extremely low protein content is not surprising, since these components impact the dough interfacial properties and rheology, affecting its gas retention capability [25]. Opposite to sample A, uniform crumb porosity in sample E could be linked to a high protein content, whereas in sample F it is indicative of a good balance in the hydrocolloids used as gluten substitutes [26].

Crumb and crust color was determined in comparison with a market-leader traditional loaf bread (Mulino Bianco, Barilla S.p.A., Italy), which served as a reference standard as made for the instrumental evaluation. GFBs presented large differences in terms of color, with sample A, which had the lowest protein content, resulting the most different from the reference and sample E, which had the highest protein content, the most similar. Sample C showed an intermediate behavior. These results are only partially in agreement with instrumental measurements; one possible explanation of this inconsistency may be found in the poor uniformity of porosity of samples that may have had an impact on color evaluation.

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All the GFB samples were found to be characterized by different intensities of aroma and flavor. This result suggests that considerable variation can occur with different bread formulation due to varying ingredients used during production, thus highlighting the essentiality of sensory evaluation for these attributes. Accordingly to the ingredients reported on label, GFB C (produced mainly with rice starch and flour) was the sample with the lowest intensity of corn aroma and flavor and the highest intensity of Yeast aroma and flavor, whereas, on the contrary, GFBs A, D and E were characterized by a high level of corn aroma and flavor since they were mainly produced with this kind of starch. GFBs B and F lie in the middle with intermediate mean values of corn aroma and flavor. Only GFB B was found to be characterized by cheese flavor; this was probably due to the presence of calcium propionate added as preservative.

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Taste was also found to discriminate the samples. In general, all the GFB samples were characterized by a low intensity of sweet and salty taste, with the exception of GFB B which was perceived as the sweetest sample.

Concerning texture properties, sensory results confirmed instrumental measurements. Sample A, characterized by the highest value of Young's modulus, was also perceived as the hardest both by hands and by mouth. On the contrary, GFBs B and C were found to be the softest samples both by instrumental and sensory assessment. GFBs E, D and F were characterized by intermediate values of softness and Young's Modulus.

Finally, from table 5 it can be observed that softness perceived by hands and mouth had an opposite trend as compared to the sensory descriptor Rubbery: the softest samples were found to be the less Rubbery and viceversa. Softness perception was linked to the lipid content of the samples. Indeed, samples B and C, which were characterized by the highest fat content, were perceived as the softest both in mouth and by hands.

Multivariate analysis of sensory and physicochemical data

Since ANOVA results indicated that the mean scores for each GFB given by the panel for each attribute could be assumed satisfactory estimates of the sensory profile of samples, sensory data were averaged across assessors and submitted to PCA along with instrumental measurements.

At this stage, according to correlation loadings plot, redundant variables or variables with less than 50% explained variance were left out from the analysis. Thus, 5 sensory and 4 instrumental variables were removed from the analysis. The scores and loadings plot based on the remaining 22 variables are shown in Fig. 2 and 3, respectively.

Insert Fig. 2 and 3 about here

A multidimensional space based on sensory and instrumental data was obtained. The variance explained by the first two principal components was 75%.

In Fig. 2 (PC1 vs PC2) the GFB samples appear to be well separated. Moving left to right along the first component (explained variance 43%), GFB A is separated from the rest of the products. The second component (explained variance 32%) distinguishes the samples D, E and F from samples A, B and C. Furthermore, from Fig. 2 it can be observed that the 3 replicates performed for each GFB sample are very close to each other thus indicating a good reliability of the assessment.

In Fig. 3, texture parameters were located along PC1, with the instrumental measurement Young's modulus and the sensory descriptors Rubbery, Adhesive on the left hand panes negatively correlated to water content expressed by Aw and Moisture and the sensory parameters Soft (by hands and mouth) on the right hand panes. Similar results were found by Gallagher et al. (2003) [22] who evidenced that both crust and crumb hardness was reduced with increasing water addition.

The instrumental (ΔE for crust and crumb) and sensory (crust and crumb color) parameters representing the difference in color from the reference standard (wheat bread) are located in the upper left hand panes and are negatively correlated to crumb Yellow Index (YMc). The sensory descriptors related to taste (Salty) and aroma (Corn and Fermented aroma) are located in the lower left hand panes opposite to Sweet taste and Cheese Aroma.

Comparison of the scores and loadings plots (Fig. 2 and 3) showed that sample A on the left of the first component of Fig. 2 is completely separated from the other samples since it received high scores for the attributes Rubbery and Adhesive and, accordingly, was also characterized by a higher Young's modulus value. Sample A was also associated to high values of crust and crumb color indicating that it was judged as not similar to the standard loaf wheat bread. Sample A is differentiated from samples D, E and F also for the poor homogeneous porosity, probably due to low values of softness and the poor water content.

Sample C was mainly described by the instrumental color parameters being associated to high colorimetric values, poor corn aroma and being opposed to YIcrumb it resulted to be characterized by a low intensity of yellow color.

Sample B, the only one to be added with calcium propionate as preservative, was characterized by cheese aroma and it was perceived as more sweet, soft during chewing and by hands, having a more homogeneous porosity and to be more similar to the wheat bread (reference standard).

Samples D, E and F were close to each other in the negative axis of PC2 and were mainly associated to an intense yellow color (high YIcrumb value) and high intensity of Corn and fermented aroma probably due to the presence of corn flour and starch as main ingredients.

According to our study, Wronkowska et al. (2008) [11] evidenced that some of the sensory attributes that better contributed to the characterization of GFBs produced with different amount of buckwheat flour were connected with bread appearance (colour and porosity), odour (yeast odour) and texture (adhesiveness and rubberiness).

Finally, it is noteworthy, that sample D was the GFB with the highest market share. This could be seen as an indirect measure of celiac consumer degree of liking since this sample is the most consumed on Italian celiac gluten-free bread market. This suggests that GFB should be characterized by sensory attributes related to presence of corn as ingredient.

Conclusion

The present study identified the sensory properties of GFBs, which are representative of the Italian market by means of a panel of celiac consumers. Some inconsistencies among celiac assessors were observed probably depending on the different period of life during which the disease occurred (from birth onward). In this respect, the influence of the time of appearance of the celiac disease on food perception deserves further investigation.

Beside the description of GFBs from a sensory point of view, characterization of their appearance and texture evaluated by instrumental devices was also achieved. A good correlation between sensory and instrumental measurements was observed.

The results of this study are a contribution to further investigate the underestimated sensory aspects associated to food products designed for celiac consumers diet. Indeed, the identification of the most relevant sensory features of GFB will facilitate the setting up of new formulations of bakery products that interpret at best the needs and attitudes of this increasing and neglected target of consumers.

However, this study did not take into consideration the hedonic aspects associated to celiac consumer perception, thus future perspectives should be addressed to understanding which sensory characteristics of GFB are key determinants for food acceptance of celiac consumers.

Acknowledgment

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Figure caption

Fig. 1 GFBs considered in the experiment

Fig. 2 Principal components product scores plot of GFB samples (Letters from A to F refer to GFB samples, numbers from 1 to 3 refer to replicates)

Fig. 3 Principal components attribute loadings plot from of GFB samples

Table 1 – GFB samples with relevant ingredients and nutritional value reported on label (d.w.=dry weight; f.w.=fresh weight; n.r. not reported)

GFB	Ingredients	Nutritional value (% d.w.)					
		Moisture (% f.w.)	Lipids	Proteins	Carb.	Minerals	Fibers
A	Corn starch, water, yeast, extravirgin olive oil (3%), fibers, dextrose, thickening agents: guar gum and hydroxypropyl methyl cellulose, salt, sugar, acidifying agent: sodium diacetate, emulsifying agents: mono- and diglycerides, aroma	38.9	5.4	2.3	81.0	3.9	7.4
B	Water, corn starch, milk powder, rice flour, sunflower oil, sugar, palm oil, margarine, yeast, thickening agents: modified cellulose and guar gum, salt, rice starch, vinegar, glucose syrup, emulsifying agents: soy lecithin and mono and diglycerides of fatty acids, calcium propionate.	43.5	14.0	3.5	81.1	n.r.	n.r.
C	Water, rice flour, rice starch, margarine, tapioca starch, dextrose, yeast, locust bean flour, potato flakes, corn starch, salt, emulsifying agents: mono- and digliceride, thickening agents: hydroxypropyl methyl cellulose, beetroot fiber; sorbitol, acidifying agent: tartaric acid	47.2	11.4	4.2	77.6	1.9	4.9
D	Water, corn starch, rice flour, vegetable oil, thickening agents: guar flour and hydroxypropyl methyl cellulose, lupin proteins, yeast, salt, fibers, aroma, emulsifying agents: mono- and diacetyl tartaric acid esters of mono- and diglycerides of fatty acids.	45.1	9.1	5.3	72.3	2.4	10.9
E	Water, corn starch, rice flour, vegetable oil, thickening agents: guar flour and hydroxypropyl methyl cellulose, soy bean isolate, fiber, yeast, salt, aroma, tartaric acid, emulsifying agents: mono- and diacetyl tartaric acid esters of mono- and diglycerides of fatty acids, calcium, niacin, thiamin, iron	42.0	9.1	6.0	66.4	n.r.	n.r.
F	Water, corn starch, rice starch, rice flour, sunflower oil, thickening agents: locust bean gum, hydroxypropylmethylcellulose, guar gum; dextrose, rice proteins, yeast, salt, sorbitol, sugar, psyllium fiber, acidifying agent: tartaric acid	40.0	8.3	4.3	80.7	2.2	4.5

Table 2 – Sensory attributes, definitions, and endpoint labels used for the evaluation of the 6 samples of GFB.

Sensory attributes	Definition	Reference standard
Appearance		
Porosity	The extend of perforation of the bread surface, this encompassing the holes, cracks allowing the permeation of air	Loaf bread Pancarré Mulino Bianco (Barilla S.p.A.)
Crumb color	Difference of crumb color between the sample and the reference standard	Loaf bread Pancarré Mulino Bianco (Barilla S.p.A.,)
Crust color	Difference of crust color between the sample and the reference standard	Loaf bread Pancarré Mulino Bianco (Barilla S.p.A.)
Touch (by hands)		
Soft	Force necessary to compress the slice on a flat surface with one finger obtaining a deformation of about 50%	Gluten-free white bread DS Food Dr. Schär S.r.l.)
Aroma		
Corn	Characteristic aroma of corn perceived by olfaction	Sweet corncob (Prestopronti Ghisetti S.r.l.)
Yeast	Characteristic aroma of yeast fermentation perceived by olfaction	Yeast Paneangeli (Cameo S.p.A.)
Cheese	Characteristic aroma of melted cheese perceived by olfaction	Sliced melted cheese Sottilette Fidel (Esselunga S.p.A.)
Fermented	Characteristic aroma of yoghurt perceived by olfaction	Full-fat yoghurt Centrale del Latte Milano (Granarolo S.p.A.)
Taste		
Sweet	Fundamental taste sensation of which sucrose is typical	Sweet gluten-free bread Bon Matin (Dr. Schär S.r.l.)
Salty	Fundamental taste sensations elicited by sodium chloride	Gluten-free crackers Bi-Aglut (Heinz S.p.A.)
Flavor		
Corn	Characteristic aroma of corn perceived by taste and olfaction during swallowing	Sweet corncob (Prestopronti Ghisetti s.r.l.)
Yeast	Characteristic aroma of yeast fermentation perceived by taste and olfaction during swallowing	Yeast Paneangeli (Cameo S.p.A.)
Cheese	Characteristic aroma of melted cheese perceived by taste and olfaction during swallowing	Sliced melted cheese Sottilette Fidel (Esselunga S.p.A.)
Fermented	Characteristic aroma of yoghurt perceived by taste and olfaction during swallowing	Full-fat yoghurt Centrale del Latte Milano (Granarolo S.p.A.)
Texture		
Adhesive	Force required to remove sample completely from the palate, using the tongue during consumption	Gluten-free bread Duo (Dr. Schär S.r.l.)
Rubbery	Persistent density perceived during chewing: time required to crumble and swallow a bite of bread	Gluten-free bread Duo (Dr. Schär S.r.l.)
Soft	Force required to press a sample with the teeth	Gluten-free white bread DS Food Dr. Schär S.r.l.)

Table 3 – Mean values of physicochemical analysis for the 6 GFB samples. Values marked with different letters by row were significantly different (p<0.05). *** indicates a significant difference for p<0.001

Parameters	F values	Samples					
		A	B	C	D	E	F
pH	1277.29***	5.05 [d]	5.55 [f]	3.98 [a]	4.75 [b]	5.10 [e]	4.81 [c]
Acidiy (mL NaOH 0.1 N)	582.36***	2.6 [a]	3.5 [c]	3.7 [e]	4.5 [f]	3.6 [d]	3.0 [b]
Young modulus (MPa)	203.40***	0.075 [c]	0.015 [a]	0.015 [a]	0.024 [b]	0.027 [b]	0.016 [a]
Aw	474.52***	0.84 [a]	0.94 [e]	0.93 [d]	0.90 [b]	0.92 [c]	0.92 [d]
Moisture $g_{H20}/100g_{sample}$	459.78***	28.2 [a]	39.2 [d]	41.5 [e]	35.3 [b]	38.8 [d]	37.4 [c]
a* crust	4621.35***	4.59 [b]	12.47 [e]	3.15 [a]	13.04 [f]	9.37 [d]	8.40 [c]
b* crust	8337.54***	1.91 [b]	10.07 [c]	-2.35 [a]	11.46 [d]	20.89 [e]	21.23 [f]
L* crust	12266.30***	85.3 [e]	78.9 [d]	98.4 [f]	69.5 [c]	54.3 [a]	60.8 [b]
ΔE crust	20164.60***	41 [e]	19 [c]	53 [f]	23 [d]	14 [b]	13 [a]
a* crumb	348.29***	-0.09 [a]	-0.11 [a]	0.80 [b]	1.32 [c]	1.24 [c]	1.52 [d]
b* crumb	10303.30***	1.56 [b]	1.79 [b]	-5.14 [a]	8.98 [c]	24.09 [e]	17.05 [d]
L* crumb	3432.45***	98.1 [c]	98.1 [c]	99.4 [d]	94.2 [b]	76.7 [a]	77.2 [a]
ΔE crumb	2705.95***	24 [d]	24 [d]	30 [e]	16 [c]	7 [b]	5 [a]
YI crumb	13537.40***	2.29 [b]	2.61 [b]	-7.39 [a]	13.63 [c]	44.88 [e]	31.57 [d]

Table 4 – Effect of samples (6), judges (10) and replicates (3) on the 17 sensory descriptors considered (*** p<0.001; ** p<0.01; * p<0.05; n.s. not significant)

Sensory descriptors	Samples	Judges	Replicates	J x S	S x R	J x R
Appearance						
Porosity	162.19 ***	3.88 ***	1.21 n.s.	4.28 ***	2.24 *	0.55 n.s.
Crumb color	278.18 ***	6.20 ***	3.13 *	4.18 ***	1.36 n.s.	0.74 n.s.
Crust color	240.01 ***	4.03 ***	3.62 *	4.13 ***	2.13 *	1.15 n.s.
Touch (by hands)						
Soft	419.86 ***	8.36 ***	2.98 n.s.	3.04 ***	0.81 n.s.	0.74 n.s.
Aroma						
Corn	156.08 ***	8.49 ***	0.03 n.s.	3.72 ***	0.63 n.s.	0.71 n.s.
Yeast	158.62 ***	4.55 ***	1.56 n.s.	3.63 ***	1.20 n.s.	1.03 n.s.
Cheese	2608.10 ***	2.88 **	2.18 n.s.	2.88 ***	2.18 *	1.00 n.s.
Fermented	77.53 ***	8.11 ***	3.76 *	3.06 ***	0.49 n.s.	1.80 *
Taste						
Sweet	115.43 ***	10.16 ***	2.73 n.s.	4.11 ***	0.96 n.s.	0.89 n.s.
Salty	105.27 ***	30.43 ***	0.54 n.s.	4.34 ***	1.09 n.s.	1.54 n.s.
Flavor						
Corn	304.31 ***	6.69 ***	1.01 n.s.	4.00 ***	0.78 n.s.	1.20 n.s.
Yeast	147.06 ***	3.59 ***	0.28 n.s.	2.76 ***	1.94 n.s.	0.44 n.s.
Cheese	2348.52 ***	3.43 **	1.12 n.s.	3.43 ***	1.12 n.s.	1.00 n.s.
Fermented	71.18 ***	10.48 ***	0.83 n.s.	2.45 ***	1.45 n.s.	0.56 n.s.
Texture						
Adhesive	32.53 ***	14.67 ***	0.73 n.s.	2.30 ***	1.24 n.s.	0.82 n.s.
Rubbery	174.66 ***	15.42 ***	0.11 n.s.	5.86 ***	1.03 n.s.	0.77 n.s.
Soft	647.86 ***	10.50 ***	2.24 n.s.	2.59 ***	0.79 n.s.	1.32 n.s.

Table 5 – Mean ratings for the 17 sensory descriptors of the 6 GFB samples. Values marked with different letters along the same row were significantly different ($p < 0.05$).

Sensory descriptors	Samples					
	A	B	C	D	E	F
Appearance						
Porosity	3.53 [a]	7.43 [e]	5.50 [c]	6.53 [d]	7.43 [e]	4.13 [b]
Crumb color	7.43 [e]	3.70 [b]	4.83 [c]	5.27 [d]	2.20 [a]	4.67 [c]
Crust color	7.77 [d]	3.97 [b]	3.83 [b]	4.43 [c]	2.43 [a]	4.33 [c]
Touch (by hands)						
Soft	1.20 [a]	8.17 [d]	7.97 [d]	6.33 [bc]	6.30 [b]	6.67 [c]
Aroma						
Corn	6.20 [c]	4.07 [b]	1.97 [a]	6.60 [c]	6.47 [c]	3.90 [b]
Yeast	4.50 [d]	1.83 [a]	6.93 [f]	2.93 [b]	3.63 [c]	5.13 [e]
Cheese	1.00 [a]	8.20 [b]	1.00 [a]	1.00 [a]	1.00 [a]	1.00 [a]
Fermented	3.83 [c]	2.03 [a]	2.50 [b]	4.33 [d]	4.77 [e]	3.97 [c]
Taste						
Sweet	1.53 [a]	6.00 [e]	3.00 [b]	3.63 [c]	4.30 [d]	3.20 [b]
Salty	4.93 [d]	1.80 [a]	4.43 [bc]	4.90 [d]	4.63 [cd]	4.30 [b]
Flavor						
Corn	6.03 [c]	3.97 [b]	1.47 [a]	6.70 [d]	6.43 [d]	3.93 [b]
Yeast	4.53 [c]	1.77 [a]	6.87 [d]	3.03 [b]	3.33 [b]	4.63 [c]
Cheese	1.00 [a]	8.00 [b]	1.00 [a]	1.00 [a]	1.00 [a]	1.00 [a]
Fermented	3.83 [c]	1.70 [a]	3.10 [b]	4.37 [d]	4.37 [d]	3.63 [c]
Texture						
Adhesive	5.60 [d]	4.03 [a]	5.07 [c]	5.53 [d]	6.10 [e]	4.63 [b]
Rubbery	7.47 [f]	4.50 [b]	3.40 [a]	5.60 [c]	5.93 [d]	6.53 [e]
Soft	1.33 [a]	8.07 [d]	8.13 [d]	6.27 [b]	6.87 [c]	6.33 [b]

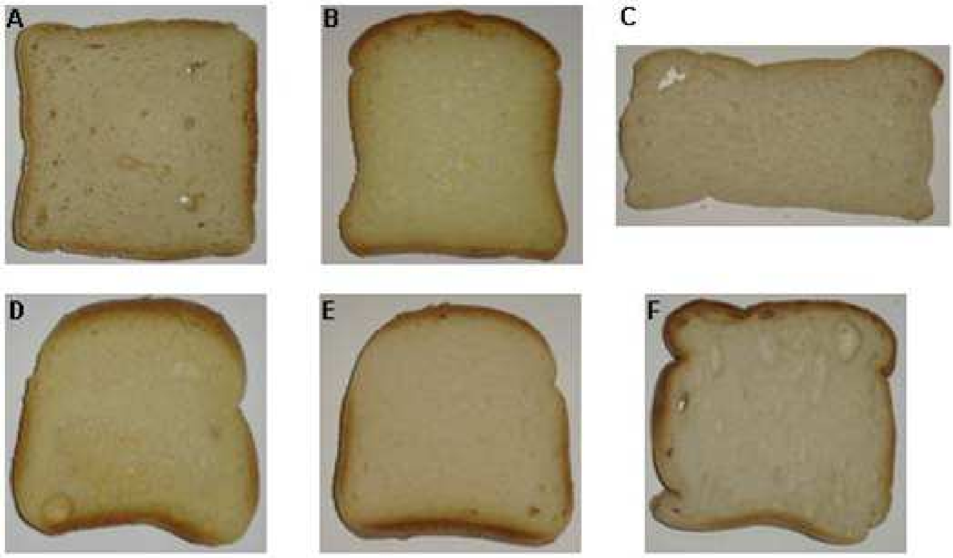

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Fig. 2

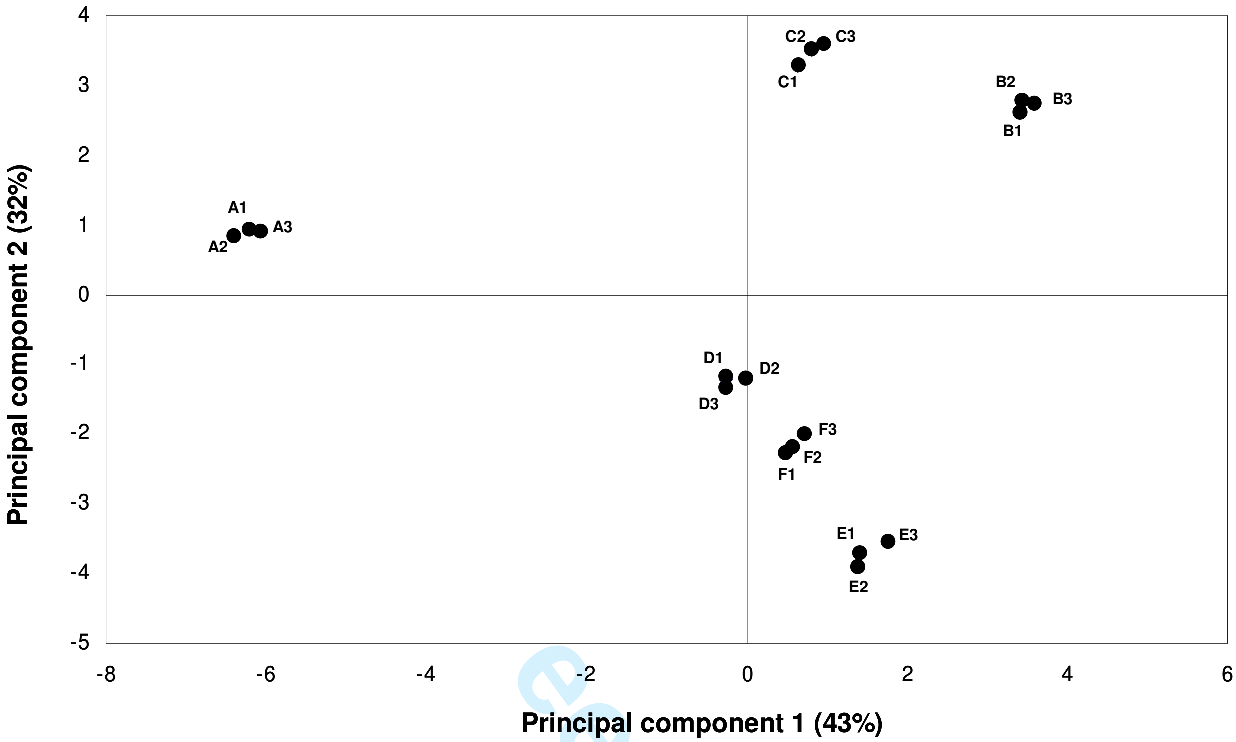
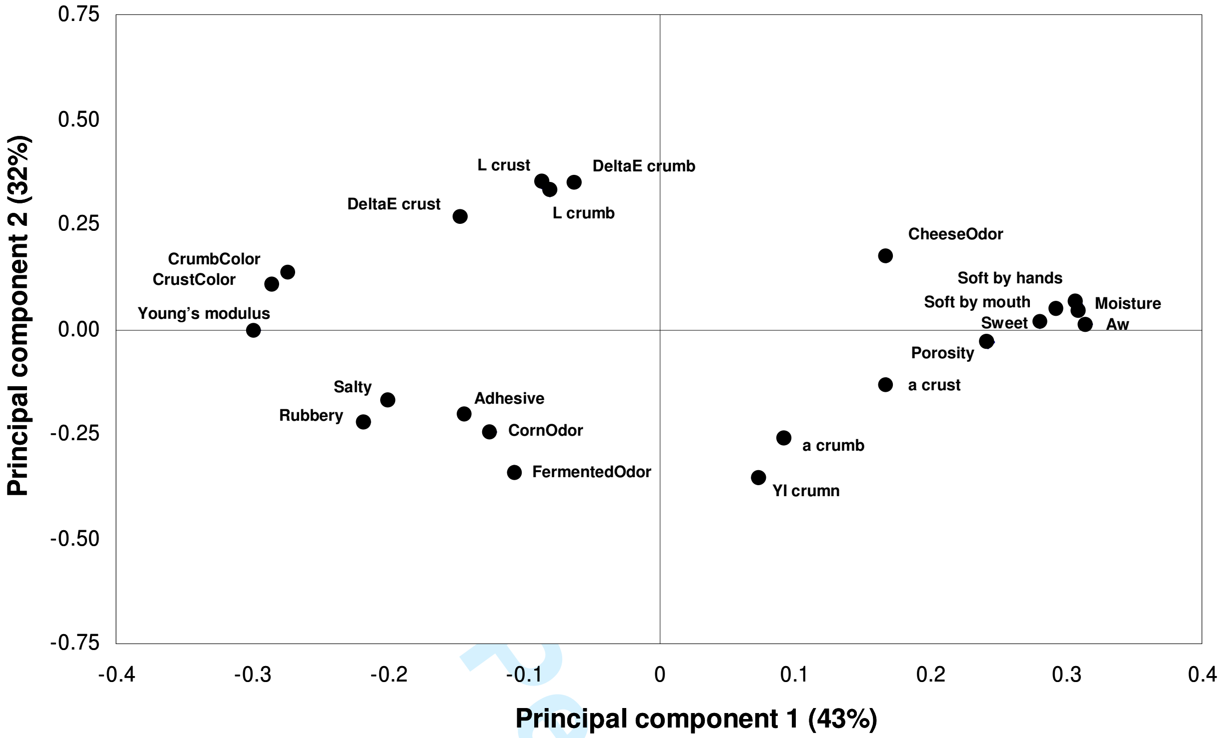

A1
A2
A3
B1
B2
B3
C1
C2
C3
D1
D2
D3
E1
E2
E3
F1
F2
F3
Principal component 1 (43%)
Principal component 2 (32%)
-8
-6
-4
-2
0
2
4
6
4
3
2
1
0
-1
-2
-3
-4
-5

Fig. 3


Principal component 2 (32%)
Principal component 1 (43%)
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0.50
0.25
0.00
-0.25
-0.50
-0.75
-0.4
-0.3
-0.2
-0.1
0
0.1
0.2
0.3
0.4
L crust
DeltaE crumb
DeltaE crust
L crumb
CrumbColor
CrustColor
Young's modulus
Salty
Rubbery
Adhesive
CornOdor
FermentedOdor
CheeseOdor
Soft by hands
Soft by mouth
Moisture
Sweet
Aw
Porosity
a crust
a crumb
Yl crumn