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Effect of chestnut flour supplementation on physico-chemical properties and volatiles in bread making

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ABSTRACT

In this study, wheat breads supplemented with different contents of chestnut flour (wheat/chestnut flour ratios: 100/0; 80/20; 50/50), were evaluated on the basis of physico-chemical properties (proximate composition, fatty acids, texture, colour, crumb grain, antioxidant capacity, volatile profile). Proximate composition, fatty acids, antioxidant capacity and volatiles for wheat and chestnut flours and their blends in the same proportion were also determined.

Antioxidant capacity increased with chestnut flour content in bread, and in accordance with flour values. A richer volatile profile was shown by bread supplemented with this type of flour as well as for flours. In particular, a marked increase was observed in furans, with their toasty and nutty notes, and phenolic compounds, with their woody and smoky notes.

A more heterogeneous crumb structure characterized 80/20 breads added of chestnut flour with larger and more asymmetrical cavities as compared to a finer and more homogeneous pore distribution of the other formulated breads. A lower volume, harder and darker crumb was also shown by bread formulated with 50/50 ratio of chestnut flour in comparison with the other formulations probably due to its higher fibre and sugar contents.

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1. Introduction

Bread is one of the most popular and wide spread baked products in the world and its quality depends on several physical (*i.e.* texture, volume, colour) and organoleptic characteristics (*e.g.* volatiles), which could be influenced by many factors, such as flour type and other ingredients, bread-making procedure, fermentation, cooking time and temperature. In the recent years, bread showed an increasing attention as a potential functional food based on its great diffusion and consumption. Thus, industries and researchers are involved in optimizing bread-making technology to improve the variety, quality, taste and availability of active compounds, adding such components with nutritional and functional properties (Balestra, Cocci, Pinnavaia, & Romani, 2011; Pasqualone et al., 2011) with the final aim to formulate a product with physiological effectiveness encountering consumers' acceptance in terms of appearance, taste and texture (Siró, Kápolna, Kápolna, & Lugasi, 2008). In this context, the utilization of flours derived from minor cereals, pseudocereals, and other non-traditional crops that could be included in bread formulation to obtain a healthier product with excellent sensorial properties was recently explored in the literature (Angioloni & Collar, 2012; de Escalada Pla, Rojas, & Gerschenson, 2013) and also reviewed (Sivam, Sun-Waterhouse, Quek, & Perera, 2010).

Among these flours, the use of chestnut flours was recently evaluated in terms of rheological properties of dough to establish the effect derived by additives and processing procedures (Moreira, Chenlo, & Torres, 2011; Moreira, Chenlo, Torres, & Prieto, 2010). In particular, chestnut flour dough performances were compared with those obtained in gluten and gluten free flour dough, pointing out that chestnut-based products could probably present problems of staling and crumbs firmness (Moreira, Chenlo, Torres, & Prieto, 2012). Chestnut flour utilization was also recently proposed for the production of gluten-free bread. Encouraging results were achieved if moderate levels of chestnut flour were added to rice flours (Demirkesen, Mert, Sumnu, & Sahin, 2010) also optimising content of emulsifier and baking conditions (Demirkesen, Sumnu, Sahin, & Uysal, 2011), while high level led to some deterioration in quality parameters (lower volume, harder texture and darker colour).

Chestnut fruits have a long history of reported health effects related to their composition (excellent energy source due to its high starch content), to the presence of nutritional effective compounds

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such as omega-3 fatty acids, vitamins E and C (De Vasconcelos, Bennet, Rosa, & Ferreira-Cardoso, 2010) and to the richness in antioxidant compounds as simple phenolics and more complex tannins (De Vasconcelos et al., 2010). Chestnuts represent a traditional product of European mountain areas from Italy, Spain, Portugal and France, where it has been considered in the past centuries as a staple food thanks to its dietary characteristics and quality. The Italian production of chestnut fruit was the first among European countries in 2010, with a value equal to 53,577 tons (FAO 2010), reaching also very good qualitative standards. Italian chestnut flour generally presents high-quality proteins with essential amino acids (~5.8%), low amount of fat (~3.7%), relatively high amount of dietary fibre (~10.8%) (IEO, 2012) and a rich volatile profile, recently characterised by Cirlini et al. (2012). It is traditionally employed for the production of typical bakery products such as bread and cakes.

Although its use is regaining interest among consumers due to its nutritional qualities and potential health benefits, to the authors' best knowledge, the evaluation of its performance in bread making in association with wheat flour is little or not explored in literature, yet. Thus, starting from flour composition, the scope of this work is to establish the feasibility of manufacturing bread supplemented with different contents of chestnut flour obtained from some Italian traditional cultivars from Parma province (located in Emilia Romagna Region). The influence of chestnut flour supplementation on physico-chemical properties (*i.e.* texture, colour, crumb grain characteristic, antioxidant capacity, and volatile profile) was thus evaluated with the final aim of valorising the traditional production chain.

2. Materials and methods

2.1. Chemical materials

Hexane, methanol, DPPH (2,2-diphenyl-1-pirylhydrazyl free radical) and Trolox ((±)-6-hydroxy-2,5,7,8-tetramethylchromane-2-carboxylic acid) were purchased from Sigma–Aldrich (St. Louis, USA), while potassium hydroxide was purchased from Carlo Erba Reagents (Milano, Italy).

2.2. Flour and bread formulation and making

Chestnut flour was obtained by mixing, with the same percentage, five cultivars (*Ampollana*, *Leccardina*, *Mondadì*, *Perticaccia* and *Luetta*) from Ceno valley (Parma, Italy). Flours were prepared starting from milled fruits dried at constant temperature (40 °C) for 30 days in a traditional drying kiln called "metato" and peeled (Cirlini et al., 2012). On the other hand, soft wheat flour type "0", as legally defined in the Italian Government Official Bulletin (2001), was purchased in a local market from a single lot.

Three types of bread samples were prepared in this study on the basis of different soft wheat/chestnut flour ratios: soft wheat 100 g/100 g (SW_{100}); chestnut 20 g/100 g of soft wheat flour replacement (SW_{80}/ChN_{20}); chestnut 50 g/100 g of soft wheat flour replacement (SW_{50}/ChN_{50}). The names reported into brackets will be used to refer to the different samples throughout the text.

Bread samples were all produced using the following formulation expressed on a flour basis: flour (100 g), water (67 g/100 g of flour), sugar (7 g/100 g of flour), yeast (3.8 g/100 g of flour), sunflower oil (6.5 g/100 g of flour) and salt (2 g/100 g of flour), by means of a home bread-maker (Severin BM3986, Sundern, Germany). The following programme was employed: pre-heating, 22 min, 30 °C; stirring, 3 min; kneading, 18 min, 35 °C; rising, 45 min, 45 °C; smoothing, 1 min; rising, 25 min, 40 °C; smoothing, 1 min; rising, 50 min, 40 °C; baking, 65 min, 210 °C). Bread loaves were allowed to cool at room temperature for 2 h prior to analysis. Four loaves were produced for each bread-type.

2.3. Chemical analysis on flour and bread

2.3.1. Proximate composition

The moisture content of flours and breads was measured in triplicate according to AACC Approved Methods 44-15A (AACC, 2000). Protein content was determined both on flour and bread samples by Kjeldhal method: 1 g of ground sample was digested by DKL fully automatic digestion unit and distilled with UDK 139 semi-automatic distillation unit (Velp Scientifica, Monza-Brianza, Italy). Nitrogen value derived from titration was multiplied for the correction factor of 5.7, typical of flour mixtures (Mccarthy & Meredith, 1988). The same factor was used for bread samples.

Fat content was determined both on flour and bread samples utilizing a Soxhlet extractor (Velp Scientifica, Monza-Brianza, Italy). In particular, 5 g of ground samples were extracted using diethyl ether as solvent. Fatty acid profile was obtained by GC–MS analysis, after transesterification with a KOH/CH_3OH 5 mL/100 mL solution, as already reported by Dall'Asta, Falavigna, Galaverna, and Battilani (2012). Fatty acids were also reported according to their unsaturation degree, as saturated (SFA), monounsaturated (MUFA) and polyunsaturated (PUFA) fatty acids. One sample from each loaf for each bread type was analysed ($n = 4$) and each analysis was replicated twice.

2.3.2. DPPH free radical scavenging activity test

Flour extracts were prepared starting from 0.1 g of flour, added with 5 mL of a methanol/water (70:30 v/v) mixture, extracted on a stirrer at room temperature for 1 h and then filtered on paper filter. The extract was evaporated and dissolved with 1 mL of a methanol/water (70:30 v/v) mixture. For bread analysis, 5 g of sample were added with 100 mL of a methanol/water (70:30 v/v) mixture, homogenized with a blender, extracted on a stirrer at room temperature for 1 h and then filtered on paper filter. The extract was evaporate, dissolved with 2 mL of a methanol/water (70:30 v/v) mixture and centrifuged at 5040× *g* for 15 min at 4 °C.

Analyses were performed in triplicate on 200 μL of extract, mixed with 2.6 mL of methanol and 2 mL of DPPH. The absorbance of the solution was recorded at 517 nm by a Perkin Elmer UV–Visible spectrophotometer after an incubation time of 30 min at room temperature. Blank was prepared and analysed following the same procedure.

The radical scavenging activity was calculated as follows: $I\% = [(Abs_0 - Abs_1)/Abs_0]^*100$, were Abs_0 was the absorbance of the blank and Abs_1 was the absorbance of the sample. TEAC value (Trolox Equivalent Antioxidant Capacity; μmol Trolox eq./g of d.w) of samples was obtained from the calibration curve calculated measuring the absorbance at 517 nm of Trolox methanolic solutions at different concentrations. One sample from each loaf for each bread type was analysed ($n = 4$) and each analysis was replicated three times.

2.3.3. Volatile compound analysis

The volatile fractions of flour and bread samples were analysed using solid phase microextraction technique (HS-SPME) coupled with GC/MS. For each SPME analysis, 3 g of flour or 2 g of bread were placed in a 30 mL glass vial, adding 200 μL of a toluene aqueous solution (250 ml/L), in according with method utilized by Cirlini et al. (2012). Identification of volatiles was obtained both by comparing mass spectra recorded with library mass spectra (NBS75K, WILEY275) and by Kovats Indices calculation. One sample from each loaf for each bread type was analysed ($n = 4$) and each analysis was replicated three times.

2.4. Physical analysis on bread

2.4.1. Crumb grain characteristics and specific bulk volume

Crumb grain was evaluated by means of a digital image analysis system as reported previously (Chiavaro, Vittadini, Musci, Bianchi, & Curti, 2008), on two central slices (20 mm thickness) from each loaf.

Briefly, images were acquired with a Scanjet 8200 flatbed scanner (Hewlett-Packard, Palo Alto, CA, USA), with a resolution of 600 dots per inch (dpi) and converted from true colour to 256 level grey scale. Analysis were performed on 40 × 40 mm squares taken from the centre of the images after their calibration, standardization and optimization by means of appropriate filters carried out with an Image-Pro Plus 4.5 (Media Cybernetics Inc., USA) software. The same software was employed for data processing, enumerating the pores present in five pre-selected dimensional classes based on their area (class 1 = 0.0010–0.0039 mm^2; class 2 = 0.0040–0.049 mm^2; class 3 = 0.05–0.99 mm^2; class 4 = 1.00–1.99 mm^2; class 5 = > 2 mm^2) to obtain the number of pores of each class and the area occupied by each class (expressed as percentage of the total number of pores).

Bread specific bulk volume was determined according to AACC Approved Method 10-05 (AACC, 2000) procedure: specific volume of bread was expressed as the volume/weight ratio of finished bread (cm^3/g). The measurements were done after 20 min of bread cooling at 25 °C on each loaf for bread type.

2.4.2. Texture and colour determination

Instrumental evaluation was performed by means of texture profile analysis (TPA) using a TA.XT2 Texture Analyzer equipped with a 25 kg load cell (Stable Micro Systems, Godalming, U.K.) and a Texture Expert for Windows software (version 1.22) for data analysis. The load cell calibration was daily performed according to the TA.XT2 manual (Stable Micro Systems).

TPA test was carried out using a cylindrical aluminium probe (35 mm diameter) and a crosshead speed of 60 mm/min to compress a crumb samples to 50% of their original height. Measurements were carried out on three slices (20 mm thickness) taken from the centre of each loaf for bread type. Two crumb samples (20 × 20 × 20 mm) were extracted from the centre of each slice.

The textural parameters considered were hardness (HD, peak force of the first compression cycle, in N), cohesiveness (CO, ratio of positive force area during the second compression to that during the first compression area, dimensionless), springiness (ratio of the time duration of force input during the second compression to that during the first compression, dimensionless), chewiness (CH, hardness × cohesiveness × springiness, in N) (Bourne, 1978).

Colour was determined on crust and crumb using a Minolta Colorimeter (CM 2600d, Konica Minolta Sensing, Osaka, Japan) equipped with a standard illuminant D65 and a Spectramagic software (Version 3.6) for data analysis. The instrument was calibrated before each analysis with white and black standard tiles. L^* (lightness, black = 0, white = 100), a^* (redness >0, greenness <0), b^* (yellowness, $b^* > 0$, blue <0), C (chroma, 0 at the centre of the colour sphere) and Hue° (Hue angle, red = 0°, yellow = 90°, 180° = green, 270° = blue) were quantified on each sample using a 10° position of the standard observer.

Crust colour was determined on nine pre-selected locations on the crust of each loaf, while crumb colour was determined on three points on the three central slices of each sample for bread type.

2.5. Statistical analysis

Means and standard deviations (SD) of data were calculated with SPSS (Version 19.0, SPSS Inc., Chicago, IL) statistical software. SPSS was used to perform one-way analysis of variance (ANOVA) and the least significant difference (LSD) test at a 95% confidence level ($p < 0.05$) was further used to identify differences among breads.

3. Results and discussion

3.1. Chemical analysis of flours

The chemical compositions of chestnut and wheat flours were determined and expressed as percentages on the dry matter. The proximate composition of flours was as follow: water ranged from 10.5 to 4.1 g/100 g, protein from 13.2 to 5.8 g/100 g and fat from 1.4 to 4.7 g/100 g, from SW_{100} to ChN_{100}, respectively. Flour mixtures presented proximate intermediate composition, related to chestnut and wheat flour proportion. Results showed that the moisture and protein content of chestnut flour are lower than wheat flour, as expected: the latter, indeed, was chosen for bread making because of its high protein content, useful to create the characteristic gluten structure. The fat content of chestnut flour (ChN_{100}) is higher compared to wheat (SW_{100}), but with high quality on account of the higher percentage of unsaturated fatty acids, as already observed by Borges, Carvalho, Correia, and Silva (2007). Among fatty acids, the most representative in chestnuts are linoleic, oleic and palmitic, those generally contributed for more than 85% of the total fatty acid content, according to previous studies (De Vasconcelos et al., 2010). In particular, for the chestnut flour used in this work, this amount was higher (Table 1), representing about the 95 % of total fatty acid fraction, mainly due to the higher level of oleic acid found in the considered flour in comparison to data usually reported in the literature (De Vasconcelos et al., 2010). On the contrary, it was observed that for wheat flour the more representative fatty acid was linoleic with a percentage of about 54.2 g/100 g (Table 1).

Antioxidant activity was tested in flours utilizing a method extensively used in the literature (Neri, Dimitri, & Sacchetti, 2010) and expressed as TEAC value. None antioxidant capacity was detected for wheat flour samples, while chestnut flour presented values (about 0.6 μmol Trolox eq./g of d.w.) similar to watermelon and leek ones (Pellegrini et al., 2003). It is demonstrated that non-

Table 1
Main fatty acid composition of flour and bread samples.

(g/100 g fat)[a]	Flour				Bread		
	SW_{100}	SW_{80}/ChN_{20}	SW_{50}/ChN_{50}	ChN_{100}	SW_{100}	SW_{80}/ChN_{20}	SW_{50}/ChN_{50}
Palmitic acid	18.3 ± 0.5	20.7 ± 1.6	19.8 ± 0.3	19.4 ± 0.2	13.9 ± 0.4	16.3 ± 3.2	16.9 ± 0.9
Stearic acid	1.1 ± 0.0	0.9 ± 0.1	0.7 ± 0.2	1.0 ± 0.0	1.9 ± 0.1	1.5 ± 0.2	1.4 ± 0.1
Oleic acid	26.7 ± 0.4	31.3 ± 0.7	40.8 ± 0.1	44.2 ± 0.3	29.3 ± 0.1	29.6 ± 1.3	32.2 ± 0.5
Linoleic acid	52.2 ± 0.1	46.0 ± 0.2	37.0 ± 0.4	33.6 ± 0.1	54.2 ± 0.2	51.8 ± 1.3	48.8 ± 0.3
Linolenic acid	1.6 ± 0.2	1.2 ± 0.6	1.8 ± 0.2	1.8 ± 0.2	0.7 ± 0.1	0.8 ± 0.3	0.9 ± 0.1
SFA	19.4 ± 0.3	21.5 ± 0.2	20.4 ± 0.2	20.4 ± 0.3	15.8 ± 0.3	17.8 ± 0.5	18.2 ± 0.2
MUFA	26.7 ± 0.4	31.3 ± 0.2	40.8 ± 0.4	44.2 ± 0.3	29.3 ± 0.5	29.6 ± 0.7	32.2 ± 0.3
PUFA	53.9 ± 0.4	47.2 ± 0.5	38.8 ± 0.5	35.4 ± 0.6	54.9 ± 0.5	52.6 ± 0.3	49.6 ± 0.7

[a] $n = 4$, sample size = 2 for each bread type. Abbreviations: SW, soft wheat flour; ChN, chestnut flour; SFA, saturated MUFA, monounsaturated, PUFA polyunsaturated fatty acids.

enzymatic antioxidant activity measurable by DPPH test of fresh chestnut fruit is mainly due to phenolic compound and ascorbic acid content (You et al., 2012), but it could decrease upon technological treatments on account of the elimination of skins. The phenolic compound levels in chestnut integuments is, indeed, higher than in the fruit (De Vasconcelos et al., 2010).

Since the flour used in this study was obtained by traditional drying and milling, the integument content in the final product was higher than that usually found in industrially milled chestnut flours, although lower than values commonly found in fresh whole fruits. In fact, Neri et al. (2010) found considerable TEAC values ranged between 3.02 and 3.11 μmol Trolox eq./g of d.w measuring the antioxidant activity of three different chestnut fruit varieties.

The volatile profile of both chestnut and wheat flours as well as of the mixtures was investigated by HS-SPME/GC–MS analysis. A total of 49 chromatographic peaks was detected and identified in this study. Since almost all the compounds considered in this paper have been already identified in a previous study (Cirlini et al., 2012), RI values from the literature were not reported.

Comparing the different flour samples, a great difference between wheat (SW_{100}) and chestnut (ChN_{100}) can be noticed: the volatile profile of the former showed only 10 gas-chromatographic signals, while the latter is characterised by 44 peaks. The most abundant compound in ChN_{100} flour was hexanal, followed by octanal and nonanal, while 1-hexanol mainly characterised SW_{100} flour.

In general, chestnut flour was found to be richer in volatiles than wheat flour, as clearly shown in Fig. 1A where the main volatile groups found in the considered flours are reported. As an example, in chestnut flour some terpenes are present, such as limonene and β-ocimene, while in wheat flour no terpene was detected. The aromatic profile found for ChN_{100} is mainly due to the effect of the traditional drying and milling process applied to chestnut fruit, which are already rich in volatiles (Cirlini et al., 2012). As an example, the significant level of phenolic compounds found in ChN_{100} should be ascribed to the pyrolitical degradation of lignin occurring upon drying.

The classes of volatiles those mainly concur to compose the aromatic profile of wheat flour (SW_{100}) are alcohols (73%), furans (12%) and aldehydes (11%). On the contrary, it is possible to observe that the compounds representing ChN_{100} are aldehydes (65%), alcohols (15%) and terpenes (7.6%). Aldehydes probably derive from the drying process to which chestnuts are subjected to obtain flour, as reported by Morini and Maga (1995): these authors found aldehydes in boiled and roasted chestnuts demonstrating that heating leads to lipid peroxidation with following degradation of fatty acid to aldehydes and ketones. Among phenolic compounds, guaiacol was the most abundant (0.23 μg/g): it is probably generated by the thermal degradation of lignin that occurs during drying treatment in kilns, where fruits are placed on a rack located on a flame (Cirlini et al., 2012).

Furan occurrence was observed in both flours, with a higher number and amount of these compounds in chestnut. In the latter, a relevant content of furfural and 5-methylfurfural was found, probably deriving from the drying treatment of chestnut fruit before milling; these compounds are responsible for a characteristic aroma of bread and caramel to the product (Cirlini et al., 2012).

The volatile fraction of samples with different soft wheat/chestnut flour ratios (SW_{80}/ChN_{20} and SW_{50}/ChN_{50}) presented all the compounds already detected in wheat and chestnut flour, but, obviously, the volatiles deriving only from chestnut resulted diluted.

3.2. Chemical analysis of breads

Regarding the chemical composition of breads, fat (3.4–4.2 g/100 g) and protein (10.9–12.3 g/100 g), as well as moisture content

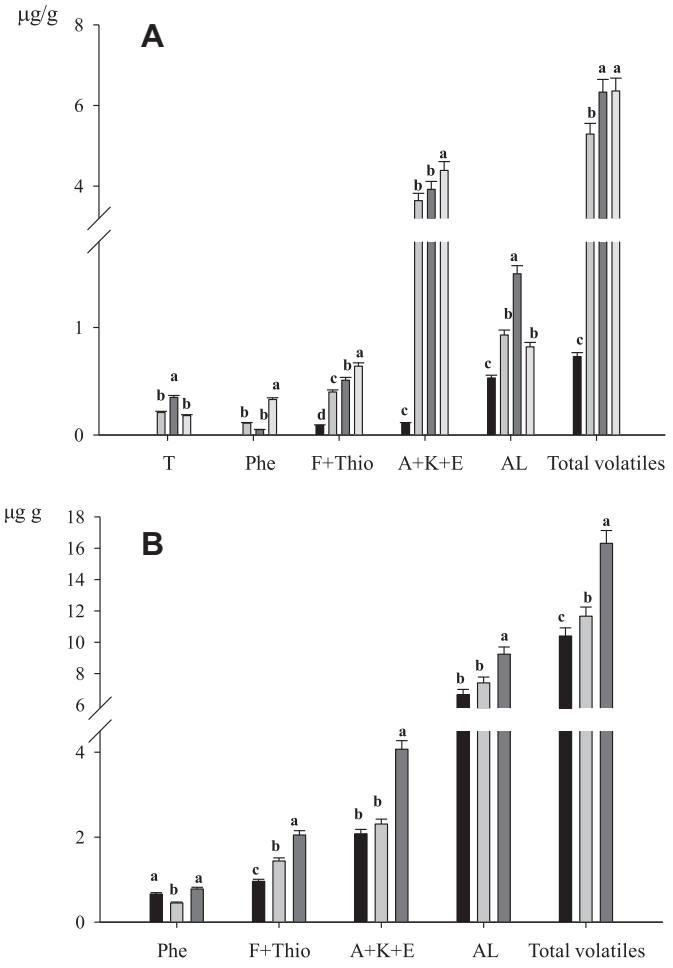

Fig. 1. Main classes of volatile compounds detected in flours (A) and breads (B) expressed as μg/g fresh weight for SW_{100} (black histogram), SW_{80}/ChN_{20} (grey histogram), SW_{50}/ChN_{50} (dark grey histogram) and ChN_{100} (light grey histogram) (SW, soft wheat; ChN, chestnut; T: terpenes; Phe: phenolic compounds; F + Thio: furans and thioazoles; A + K + E: aldehydes, ketons and esters; AL: alcohols). Bars of histograms with the same letters are not significantly different ($p \leq 0.05$, $n = 4$ for each bread type).

(34.9–35.3 g/100 g), were similar in all the considered samples: this fact was probably due to the addition of other ingredients during bread-making and to the baking process itself. In the same way, the addition of seed oil also affected fatty acid profiles, concurring to the alignment of fatty acid percentages in all products.

Antioxidant capacity of bread samples were 0.73, 1.00 and 1.04 μmol Trolox eq./g of d.w for SW_{100}, Sw_{80}/ChN_{20} and Sw_{50}/ChN_{50}, respectively. Generally, the antioxidant activity of breads resulted higher than that of starting flours: this can be ascribed to Maillard or non-enzymatic browning reaction products, such as pyrroles and furans, which concur to improve this value in particular on bread crust (Kitts, Chen, & Jing, 2012). According to our data, breads containing chestnut flour showed a significantly higher TEAC values compared to SW_{100} samples, supporting the possible exploitation of chestnut flour for the formulation of functional bread. In particular, the possible flour enrichment with very low amount of chestnut integuments may potentially lead to an increase in the antioxidant capacity of the final products.

Volatile profile of bread is characterized by well-defined fractions, depending on the type of product, while fermentation and baking mainly influence the volatile compound formation. In particular, Bianchi Careri, Chiavaro, Musci and Vittadini (2008)

reported that fermentation directly affected crumb aromatic profile, while crust flavour mainly depends on baking treatment. The main volatile groups occurring in the breads considered in this study are reported in Fig. 1B.

The molecules that contribute to the volatile fraction of the different breads were alcohols, followed by aldehydes, ketones and furans. A total of 38 gas-chromatographic signals have been detected and identified in bread samples, most of them already present in chestnut flours. Some differences can be observed comparing the aromatic profiles of flours and breads: bread flavour is more complex as influenced by several factors, such as ingredients, fermentation and thermal treatment (Martinèz-Anaya, 1996). In particular, some volatiles formed during the fermentation process (Rehman, Peterson, & Piggot, 2006) are present only in the volatile fraction of breads, such 3-methyl-1-butanol, 3-hydroxy-2-butanone and ethyl octanoate, those give alcoholic, waxy and pungent characteristic odour to the product.

Although wheat flour was very poor in volatile compounds compared to chestnut flour, the SW_{100} bread volatile profile was qualitatively comparable to those obtained for SW_{80}/ChN_{20} and SW_{50}/ChN_{50} breads, on account of the common rising and baking process. In particular, most of the typical chestnut flour volatiles were due to Maillard-type reaction occurring upon fruit drying in kilns; the same compounds were also generated during the baking process, on account of the thermal degradation occurring in dough. Nonetheless, SW_{50}/ChN_{50} showed the highest amount of total volatiles, followed by SW_{80}/ChN_{20}, clearly demonstrating that the use of chestnut flour in the formulation of bread actually affects the organoleptic properties of the final product.

Several compounds significantly differentiate breads containing chestnut flour from that containing only wheat flour. As an example, furans such as furfural, 5-methylfurfural and 2-furfuryl alcohol strongly increased from SW_{100} to SW_{50}/ChN_{50}. This behaviour can be explained considering the different composition of chestnut and wheat flours: the high level of reducing sugars occurring in the former may favour caramelization processes leading to the formation of furfural-based compounds. Similarly, guaiacol and 4-ethyl-guaiacol, which are likely formed during the chestnut smoking process in kiln, can be found only in chestnut containing breads.

3.3. Physical analysis on bread

Characteristic images of the central slice as well as of the area considered for the crumb grain for each type of breads prepared in this study are reported in Fig.2 for SW_{100} (A), SW_{80}/ChN_{20} (B) and SW_{50}/ChN_{50} (C) samples.

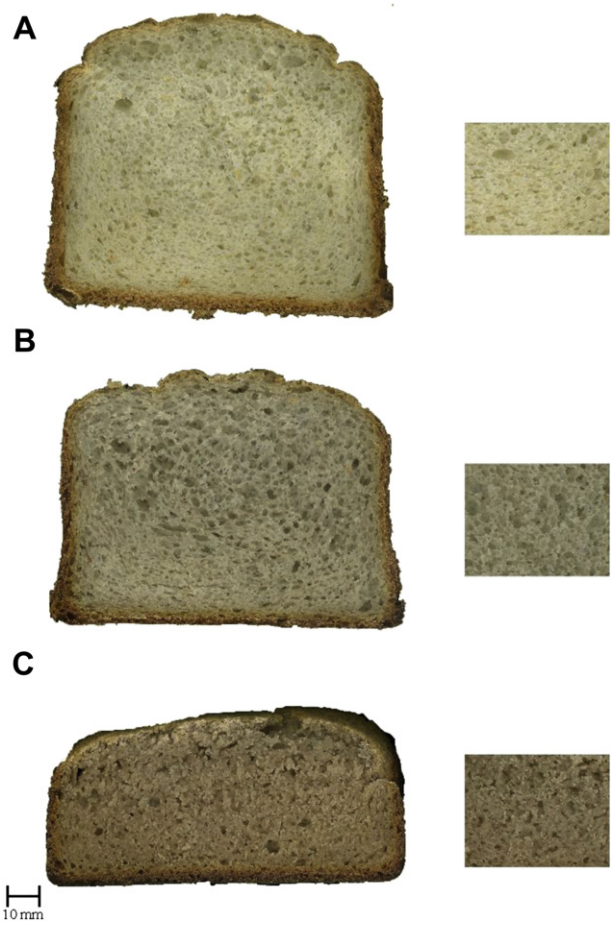

Fig. 2. Crumb slice images and relative area used for image analysis for SW_{100} (A), SW_{80}/ChN_{20} (B) and SW_{50}/ChN_{50} (C).

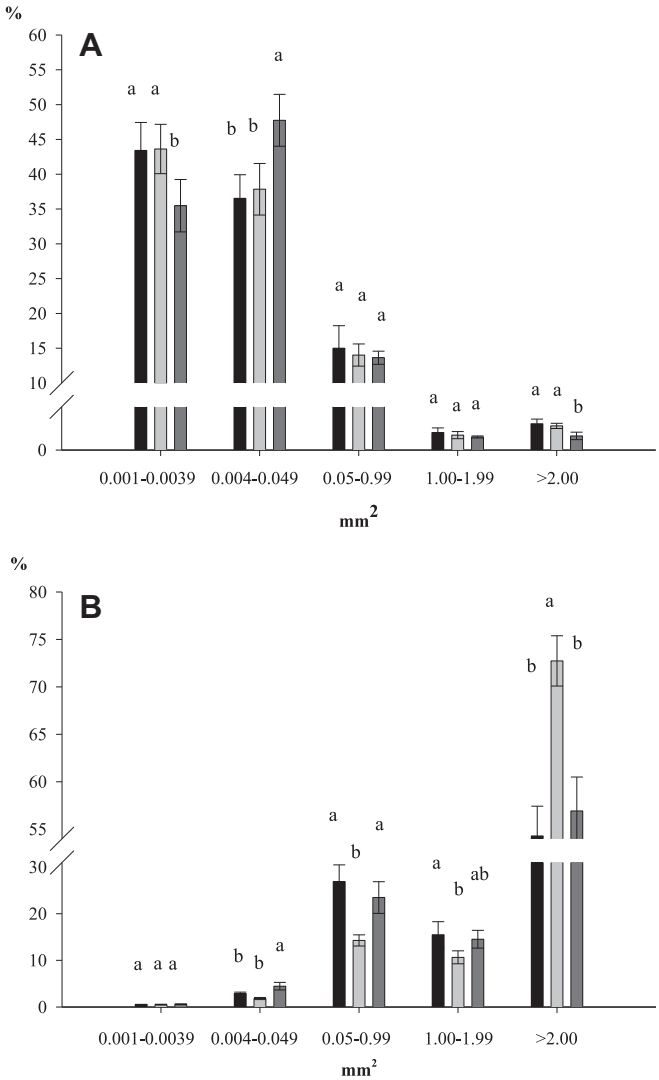

Fig. 3. Number of pores as percentage of total number of pores (A) and area as percentage of total pore area (B) for the selected five-dimensional classes for SW_{100} (black histogram), SW_{80}/ChN_{20} (grey histogram) and SW_{50}/ChN_{50} (dark grey histogram) breads (SW, soft wheat; ChN, chestnut). Bars of histograms with the same letters are not significantly different ($p \leq 0.05$, $n = 4$, sample size = 2 for each bread type).

Table 2
Colour (L^*, a^*, b^*) and texture profile analysis parameters of breads analysed.

	Crust			Crumb		
	SW_{100}	SW_{80}/ChN_{20}	SW_{50}/ChN_{50}	SW_{100}	SW_{80}/ChN_{20}	SW_{50}/ChN_{50}
Colour[a]						
L^*	54.1 ± 4.1 a	48.1 ± 4.0 b	47.9 ± 9.4 b	66.4 ± 2.9 a	57.4 ± 4.0 b	49.4 ± 3.3 c
a^*	8.5 ± 1.7 a	8.8 ± 1.4 a	8.2 ± 2.6 a	1.0 ± 0.2 c	1.9 ± 0.4 b	4.9 ± 0.9 a
b^*	16.4 ± 2.4 a	16.6 ± 6.6 a	18.3 ± 7.1 a	11.5 ± 1.0 a	9.2 ± 2.0 b	11.5 ± 3.3 a
C^*	18.5 ± 2.5 a	19.0 ± 6.3 a	19.8 ± 6.3 a	11.5 ± 1.0 a	9.4 ± 1.9 b	12.5 ± 3.3 a
H°	62.7 ± 4.6 a	59.8 ± 5.8 a	66.9 ± 6.3 a	84.9 ± 1.2 a	78.0 ± 1.8 b	66.4 ± 3.5 c
Texture[b]						
Hardness (*N*)	–	–	–	3.0 ± 0.4 a	2.0 ± 0.4 b	3.3 ± 0.4 a
Cohesiveness	–	–	–	0.52 ± 0.01 c	0.54 ± 0.01 b	0.58 ± 0.02 a
Chewiness (*N*)	–	–	–	1.8 ± 0.2 b	1.1 ± 0.2 b	2.7 ± 0.4 a

Same letters within each row do not significantly differ ($p < 0.05$). Abbreviations: SW, soft wheat flour; ChN, chestnut flour.
[a] $n = 3$, sample size = 3 for each type of bread.
[b] $n = 3$, sample size = 2 for each type of bread.

A more heterogeneous crumb structure characterized SW_{80}/ChN_{20} product as compared to SW_{100} and SW_{50}/ChN_{50} (Fig. 2). Crumb of SW_{80}/ChN_{20} bread presented larger and more asymmetrical cavities as compared to a finer and more homogeneous pore distribution of SW_{100} and SW_{50}/ChN_{50}. This was confirmed by data reported in Fig. 3 where pore distribution among selected classes and area occupied by each class, both expressed in percentages, were shown. Comparing the two breads formulated with different contents of chestnut flour, SW_{80}/ChN_{20} exhibited significantly lower area percentages occupied by pores from 0.004 to 1.99 mm^2 and higher of larger pores (>2 mm^2). On the contrary, SW_{50}/ChN_{50} showed higher number of small pores (0.004–0.049 mm^2). In addition, the loaves of SW_{100} and SW_{80}/ChN_{20} were characterized by similar values of specific volume (2.8 ± 0.4 cm^3/g and 2.5 ± 0.2 cm^3/g, respectively) whereas SW_{50}/ChN_{50} presented the lowest one (1.3 ± 0.2 cm^3/g). A decrease of volume for gluten free formulated bread prepared with chestnut flour was previously observed by Demirkesen et al. (2010) and related to the high fibre content of this type of flour. Fibre was reported to reduce bread volume due to their interaction with gluten that led to a decrease of the gas retention capacity and to restrict expansion of the gas cells (Gómez, Ronda, Blanco, Caballero, & Apesteguía, 2003) producing pores with low mean cell area (Angioloni & Collar, 2012). Thus, SW_{50}/ChN_{50} bread could not probably entrap gas bubbles leading to a dense crumb structure. Chestnut flour also showed the highest speed of weakening of the protein network in comparison with other gluten free flours (Moreira et al., 2012)

Textural properties of the three bread samples are reported in Table 2 and confirm these hypotheses. Crumb hardness of SW_{80}/ChN_{20} was significantly lower than SW_{100} and SW_{50}/ChN_{50} that presented similar values of this property, despite the differences in the specific volume. This is probably in relation with the high protein and/or gluten content of the soft wheat flour employed in this study: as a consequence, the formed dough was characterised by a strong cellular network being capable to show a high retention capacity after an optimal gas production (Dhingra & Jood, 2004), as also shown by data reported in Fig. 3. 100% wheat bread formulated with increasing content of protein in flour (from 9.5 to 13.5%) was previously found to exhibit an increase of crumb firmness (Salehifar, Ardebili, & Azizi, 2010). On the other hand, Demirkesen et al. (2010) found an increase of crumb hardness when chestnut flour was added in gluten free bread, showing also a relation with fibre amount. Gómez et al. (2003) reported a bread firmness increase related to addition of fibre, probably due to the thickening of the walls surrounding the air bubbles in the crumb. According to our results, mixing wheat and chestnut flours in a 80:20 proportion seemed to realize a good technological compromise, producing a gluten network that can entrap bubbles and, simultaneously, favouring starch gelatinization. This was also demonstrated by cohesiveness values of the crumb that were found to increase by increasing chestnut flour percentage from 0 to 50 g/100 g (Table 2): since cohesiveness relates to the ability of a material to stick to itself (Bourne, 1978), the higher amount of starch and sugars contained in chestnut flour caused a delay in gelatinization and, thus, stabilized the amorphous state of granules (Demirkesen et al., 2010). Chewiness (CH) was also significantly higher for SW_{50}/ChN_{50} due to the highest hardness (H) and cohesiveness (CO) values.

Colour parameters of bread crust and crumb are reported in Table 2. Regarding crumb colour, significant differences were found for all the considered samples: SW_{50}/ChN_{50} presented the lowest lightness (lower L^*) while SW_{100} exhibited the highest and SW_{80}/ChN_{20} had an intermediate value. The original colour of the chestnut flour had a darkening effect on flour mixture and on crumb bread, as consequence. Similar results were obtained for a^* but in this case, SW_{50}/ChN_{50} presented the highest redness (a^* and H° values) while SW_{100} the lowest and SW_{80}/ChN_{20} exhibited intermediate values. The addition of chestnut flour shifted the colour of the bread towards more red tones, as appreciable from central slice images (Fig. 2). Finally, for yellowness parameter (b^*) only SW_{80}/ChN_{20} was significantly different from the others with a lower value of b^*, indicating a less yellow appearance as shown in Fig. 2.

Colour analysis of the crust indicated that both breads with the addition of chestnut flour (SW_{80}/ChN_{20} and SW_{50}/ChN_{50}) had a darker crust (significantly higher L^*): this can be ascribed to the darkening effects of chestnut flour itself as well as to Maillard and caramelization browning due its high sugar content, as previously hypothesized by Demirkesen et al. (2010). No significant differences were found for redness (a^*, H°) and yellowness (b^*).

4. Conclusions

These findings show that breads formulated with incremental content of chestnut flour could give a different response by nutritional and qualitative points of view. For this reason, the identification of the exact content of chestnut flour required to obtaining the best compromise between all the above cited properties is very important for the formulation of a product characterised by a good sensory and nutritional balance.

By a nutritional point of view, a "functional bread" could be formulated with a high content of chestnut flour (ratio 50/50), to obtain a higher antioxidant capacity in the final product. Considering the volatiles, chestnut flour containing breads showed a higher amount of volatiles compared to wheat bread; the

characteristic flavour that furans, with their toasty and nutty notes, and phenolic compounds, with their woody and smoky notes, confer to the product, could be considered as an "added value" referred to the consumer preferences. Regarding physical properties, bread with 20 g/100 g of chestnut flour showed a heterogeneous crumb structure, lower hardness and cohesiveness as well a less dark colour than SW_{50}/ChN_{50}; on account of these properties, this formulation seems to be suitable to obtain a product that could encounter consumer satisfaction.

Starting from these data, further studies will be performed to find the best formulation that can realize a good compromise between nutritional and qualitative properties (taking also into account wheat flour composition), and to evaluate staling of bread containing different percentages of chestnut flour, furnishing useful information for the valorisation and implementation of traditional bread manufacture.

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References

AACC, American Association of Cereal Chemists. (2000). *Approved methods of the AACC* (10th ed.). St. Paul, USA: Am Assoc Cereal Chem.

Angioloni, A., & Collar, C. (2012). Suitability of oat, millet and sorghum in bread-making. *Food and Bioprocess Technology*. http://dx.doi.org/10.1007/s11947-012-0786-9. on line from January 31th 2012.

Balestra, F., Cocci, E., Pinnavaia, G. G., & Romani, S. (2011). Evaluation of antioxidant, rheological and sensorial properties of wheat flour dough and bread containing ginger powder. *LWT – Food Science and Technology, 44*, 700–705.

Bianchi, F., Careri, M., Chiavaro, E., Musci, M., & Vittadini, E. (2008). Gas chromatographic-mass spectrometric characterisation of the Italian protected designation of origin "Altamura" bread volatile profile. *Food Chemistry, 101*, 787–793.

Borges, O. P., Carvalho, J. S., Correia, P. R., & Silva, A. P. (2007). Lipid and fatty acid profiles of *Castanea sativa* Mill. Chestnut of 17 native Portuguese cultivars. *Journal of Food Composition and Analysis, 20*, 80–89.

Bourne, M. C. (1978). Texture profile analysis. *Food Technology, 32*, 62–66.

Chiavaro, E., Vittadini, E., Musci, M., Bianchi, F., & Curti, E. (2008). Shelf-life stability of artisanally and industrially produced durum wheat sourdough bread ("Altamura" bread). *LWT – Food Science and Technology, 41*, 58–70.

Cirlini, M., Dall'Asta, C., Silvanini, A., Beghè, D., Fabbri, A., Galaverna, G., et al. (2012). Volatile fingerprinting of chestnut flours from traditional Emilia Romagna (Italy) cultivars. *Food Chemistry, 134*, 662–668.

Dall'Asta, C., Falavigna, C., Galaverna, G., & Battilani, P. (2012). Role of maize hybrids and their chemical composition in fusarium infection and fumonisin production. *Journal of Agricultural and Food Chemistry, 60*, 3800–3808.

De Vasconcelos, M., Bennett, R., Rosa, E., & Ferreira-Cardoso, J. (2010). Composition of European chestnut (*Castanea sativa* Mill.) and association with health effects: fresh and processed products. *Journal of the Science of Food and Agriculture, 90*, 1578–1589.

Demirkesen, I., Mert, B., Sumnu, G., & Sahin, S. (2010). Utilization of chestnut flour in gluten-free bread formulations. *Journal of Food Engineering, 101*, 329–336.

Demirkesen, I., Sumnu, G., Sahin, S., & Uysal, N. (2011). Optimisation of formulations and infrared-microwave combination baking conditions of chestnut-rice breads. *International Journal of Food Science and Technology, 46*, 1809–1815.

Dhingra, S., & Jood, S. (2004). Effect of flour blending on functional, baking and organoleptic characteristics of bread. *International Journal of Food Science and Technology, 39*, 213–222.

de Escalada Pla, M., Rojas, A. M., & Gerschenson, L. M. (2013). Effect of butternut (*Cucurbita moschata* Duchesne ex Poiret) fibres on bread making, quality and staling. *Food and Bioprocess Technology, 6*, 828–838.

FAO. (2010). *Food and agriculture organization of the United Nations, Faostat*. http://faostat.fao.org/site/291/default.aspx. Accessed 25.11.12.

Gómez, M., Ronda, F., Blanco, C. A., Caballero, P. A., & Apesteguía, A. (2003). Effect of dietary fibre on dough rheology and bread quality. *European Food Research and Technology, 216*, 51–56.

IEO. (1998). *European institute of oncology, food composition database for epidemiological studies in Italy*. http://www.ieo.it/bda2008/uk/index.aspx. Accessed 25.11.12.

Italian Government Official Bulletin. (2001). Regolamento per la revisione della normativa sulla produzione e commercializzazione di sfarinati e paste alimentari. English text was not available. *Italian Government Official Bulletin, 117*, 6–12.

Kitts, D. D., Chen, X. M., & Jing, H. (2012). Demonstration of antioxidant and anti-inflammatory bioactivities from sugar-amino acid Maillard reaction products. *Journal of Agricultural and Food Chemistry, 60*, 6718–6727.

Martinèz-Anaya, M. A. (1996). Enzymes and bread flavour. *Journal of Agricultural and Food Chemistry, 44*, 2469–2480.

Mccarthy, M. A., & Meredith, F. I. (1988). Nutrient data on chestnuts consumed in the United states. *Economic Botany, 42*, 29–36.

Moreira, R., Chenlo, F., & Torres, M. D. (2011). Effect of sodium chloride, sucrose and chestnut starch on rheological properties of chestnut flour doughs. *Food Hydrocolloids, 25*, 1041–1050.

Moreira, R., Chenlo, F., Torres, M. D., & Prieto, D. M. (2010). Influence of the particle size on the rheological behaviour of chestnut flour doughs. *Journal of Food Engineering, 100*, 270–277.

Moreira, R., Chenlo, F., Torres, M. D., & Prieto, D. M. (2012). Technological assessment of chestnut flour doughs regarding to doughs from other commercial flours and formulations. *Food and Bioprocess Technology, 5*, 2301–2310.

Morini, G., & Maga, J. A. (1995). Volatile compounds in roasted and bovile Chinese chestnut (*Castanea molissima*). *LWT - Food Science and Technology, 28*, 638–640.

Neri, L., Dimitri, G., & Sacchetti, G. (2010). Chemical composition and antioxidant activity of cured chestnut from three sweet chestnut (*Castanea sativa* Mill.) ecotypes from Italy. *Journal of Food Composition and Analysis, 23*, 23–29.

Pasqualone, A., Piergiovanni, A. R., Caponio, F., Paradiso, V. M., Summo, C., & Simeone, R. (2011). Evaluation of the technological characteristics and bread-making quality of alternative wheat cereals in comparison with common and durum wheat. *Food Science and Technology International, 17*, 135–142.

Pellegrini, N., Serafini, M., Colombi, C., Del Rio, D., Salvatore, S., Bianchi, M., et al. (2003). Total antioxidant capacity of plant foods, beverages and oils consumed in Italy assessed by three different *in vitro* assay. *Journal of Nutrition, 133*, 2812–2819.

Rehman, S., Peterson, A., & Piggott, J. R. (2006). Flavour in sourdough breads: a review. *Trends in Food Science and Technology, 17*, 557–566.

Salehifar, M., Ardebili, M. S., & Azizi, M. H. (2010). Effect of wheat flour protein variations on sensory attributes, texture and staling of Taftoon bread. *Ciência e Tecnologia de Alimentos, 30*, 833–837.

Siró, I., Kápolna, E., Kápolna, B., & Lugasi, A. (2008). Functional food. Product development, marketing and consumer acceptance – a review. *Appetite, 51*, 456–467.

Sivam, A. S., Sun-Waterhouse, D., Quek, S. Y., & Perera, C. O. (2010). Properties of bread dough with added fiber polysaccharides and phenolic antioxidants: a review. *Journal of Food Science, 75*, R163–R174.

You, Y., Jiang, Y., Sun, J., Liu, H., Song, L., & Duan, X. (2012). Effects of short-term anoxia treatment on browning of fresh-cut Chinese water chestnut in relation to antioxidant activity. *Food Chemistry, 132*, 1191–1196.