Food Research International 54 (2013) 1400–1408

Contents lists available at ScienceDirect

Food Research International

journal homepage: www.elsevier.com/locate/foodres

Sensory test vs. electronic nose and/or image analysis of whole bread produced with old and modern wheat varieties adjuvanted by means of the mycorrhizal factor

Luisa Torri [a],*, Paola Migliorini [a], Giorgio Masoero [b]

[a] University of Gastronomic Sciences, Piazza Vittorio Emanuele 9, 12060 Bra, CN, Italy
[b] Accademia di Agricoltura di Torino, Via A. Doria 10, 10162 Torino, Italy

ARTICLE INFO

Article history:
Received 5 July 2013
Accepted 27 September 2013

Keywords:
Sensory analysis
Bread
Wheat old varieties
Electronic nose
Image analysis
Mycorrhizal factor

ABSTRACT

In order to promote local organic farming and healthy local products, the germplasm of common wheat (*Triticum aestivum* spp.) retrieved from old-varieties (G — Gentil Rosso, I — Inallettabile, S — Sieve) has been compared with that of the modern Blasco *Triticum*, treated with (Bm — Blasco mycorrhizal) or without (B — Blasco) Micosat F® mycorrhizal consortium, and with that of an ordinary reference flour (C — Control). A sensory test (18 attributes, 10 panelists) was compared with rapid analyses: electronic nose (e-nose, 10 sensors, 8 replicates) and/or image analysis (9 parameters, 3 replicates). The planned contrasts were able to establish the significance of the epoch and of the mycorrhizal factors. Chemometrics of the e-nose, image and concatenated scores was used to cluster the average groups. The reference groups (B and C) were clearly distinguished. The mycorrhizal factor has emerged as being a botanical modifier of the sensory properties of the bread: a modern wheat treated with the Micosat F® microbial consortium after breading was established as non-differentiable from the old Sieve variety and to be similar to the old Gentil Rosso and Inallettabile varieties. The rapid analyses forecast several traits: the raw average cross-validated r-square, calculated across the 18 attributes, was 0.69 for the e-nose and 0.56 for the imaging features. However the concatenated sets rose to 0.83 and only 4 traits were below a 2.0 threshold of the ratio–performance prediction (RPD) while 10 scores exceeded 2.5 RPD.

1. Introduction

The bread quality can refer to hygienic, sanitary, technological, nutritional, functional and sensorial properties. All of these features are affected by both the properties of the flour and baking process that is used (Kihlberg, Johansson, Kohler, & Risvik, 2004). In the 20th century, new varieties of wheat (*Triticum aestivum* L.) were selected mainly for agronomic and technological purposes on the basis of the improvement that could be reached in their productivity (Guarda, Silvano Padovan, & Delogu, 2004) and the potentially excellent bread-making properties of the old varieties were abandoned (Bordes, Branlard, Oury, Charmet, & Balfourier, 2008). After the Second World War, intensive plant breeding programs led to the complete replacement of landraces by modern semi- and high-yielding cultivars, correlating with a decrease in wheat genetic diversity (Ceccarelli, 2009).

The high yields of today's modern wheat cultivars require the use of mineral fertilizers and chemical herbicides and fungicides, all of which lead to both higher production costs and a greater risk of environmental pollution (Guarda et al., 2004). In order to harmonize the expectations of both producers and conscious consumers, it is essential to offer perceived high sensory quality products and incorporate environmentally friendly methods into the farming system used for the production of whole grain bread, which has been shown to have a protective function for human health (Adom, Sorrells, & Liu, 2003; Gasztonyi, Farkasa, Berkia, Petroczib, & Daood, 2011; Mader et al., 2007; Ward, Poutanen, & Gebruers, 2008). The use of microbial consortia with mycorrhizal factor represents environmentally friendly and sustainable methods. Mycorrhiza is a symbiotic association between a fungus and the root of a vascular plant. In a mycorrhizal association, the fungus colonizes the host plant's roots, either intercellularly (endomycorrhiza), as in arbuscular mycorrhizal fungi, or extracellularly (ectomycorrhiza). They are an important component of soil life and soil chemistry as they become part of the plant's rhizosphere. Mycorrhizae form a mutualistic relationship with the roots of most plant species. This mutualistic association provides the fungus with relatively constant and direct access to carbohydrates, such as glucose and sucrose. In return, the plant gains the benefits of the mycelium's higher absorptive capacity for water and mineral nutrients due to the comparatively large surface area of the mycelium/root ratio, thus improving the plant's mineral absorption capabilities (Bonfante & Genre, 2010; Maronek, Hendrix, & Kiernan, 2011; Strack, Fester, Hause, Schliemann, & Walter, 2003). Mycorrhizae are present in 92% of plant families that have been studied and

---

* Corresponding author. Tel.: +39 0172 458509; fax: +39 0172 458500.
E-mail address: l.torri@unisg.it (L. Torri).

0963-9969/$ – see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.foodres.2013.09.045

arbuscular mycorrhizae are found in 85% of all plant families; these occur in many crop species, for example in wheat (Wang & Qiu, 2006).

Arbuscular mycorrhizal fungi (AMF) are considered the most important soil organisms for agro-ecosystem sustainability, as they establish root symbioses with most crop plants and, acting as a living interface between the plant roots and soil, translocate mineral nutrients – mainly P, N, Zn, Ca and Cu – from the soil to the plants through the large extraradical mycelial network that spreads from the mycorrhizal roots to the surrounding environment (Giovannetti and Avio, 2002; Smith & Read, 2008).

The objective of this research was to compare the sensory properties of bread made with old vs. modern common wheat varieties under organic conditions. Moreover, the qualitative effect of the mycorrhizal factor – together with mycorrhizal helper bacteria (Kannan et al., 2011) – has been observed in the modern Blasco variety of wheat; using the Micosat F® microbial consortium (Di Cesare et al., 2012).

Several approaches based on sensory methods and instrumental techniques, have been reported in literature to evaluate the quality of bread. Sensory quality of bread was generally studied by means of descriptive analysis (Holtekjølen, Baevre, Rødbotten, Berg, & Knutsen, 2008; Škrbić & Filipčev, 2008; Škrbić et al., 2009). It was used also in combination with a consumer test (Heenan, Dufour, Hamid, Harvey, & Delahunty, 2008) or with mass spectrometry (Heenan et al., 2009; Jensen, Oestdal, Skibsted, Larsen, & Thybo, 2011), to reveal which attributes are correlated to the perception of bread freshness. However, sensory analysis is relatively expensive and very time-consuming (Lawless & Heymann, 2010). Attention has recently been focused on the development of rapid, cheap, non-invasive and non-destructive instrumental techniques based on the use of electronic nose devices, image analysis methods and texture analyzer machines. Several works have been aimed at investigating the potential correlation between the instrumental data and the sensory scores. The influence of the farming system, and the milling and baking techniques on the quality of whole pan bread has been evaluated by means of descriptive profiling and image analysis (Kihlberg et al., 2004). These two techniques were also applied to show the effect of flour quality, the production process (traditional French and industrially modified), the mixing and the proofing time on the quality of baguettes (Baardseth, Kvaal, Lea, Ellekjær, & Færgestad, 2000). An overview of the literature pertaining to the measurement of crumb appearance and mechanical bread properties (visual and physical texture) has been reported in literature (Scanlon & Zghal, 2001). Only a few works have been aimed at the study of the aroma of bread using an electronic nose (Piazza & Benedetti, 2008; Ponzoni et al., 2008).

A combined approach based on the application of a sensory descriptive method, the image analysis technique and the use of a commercial electronic nose has been proposed in this study, in order to evaluate the effects of old vs. modern organic common wheat varieties, adjuvanted by means of a mycorrhizal factor, on the properties of bread.

2. Materials and methods

2.1. Bread samples

Six wheat flour samples (type 2) were obtained from the stone milling of the following varieties of old and modern wheat varieties grown in an experimental field under organic agriculture management practices, using an inoculum of mycorrhizal Micosat F® (CCS-Aosta s.r.l., Italy: *www.micosat.it*): I — Inallettabile (old), G — Gentil Rosso (old), S — Sieve (old), B — Blasco (modern), Bm — Blasco with mycorrhiza (modern), C — Control (a mixture of commercially available flour; modern).

The same bread dough formula was adopted (5 kg of flour, 3 kg of water, 1.5 kg of liquid mother yeast, 25 g of malt extract, 100 g of salt) and the same bread making process was followed: first dough (flour, water, yeast) for 4–5 min; left to rest for 15–20 min; second dough (addition of malt, salt, water) for 3–4 min; left to rest at 27 °C for 45 min; forming (600 g size loaf); rising at 29 °C for 4 h; etching of the surface of the bread; baking in two consecutive steps (first step: 5 min at 240 °C; second step:45 min at 220 °C).

For the sensory and instrumental analysis the samples were made by a local bakery and delivered to the evaluation laboratory within 2 h of baking.

2.2. Descriptive sensory analysis

The sensory evaluation was carried out 3 h after baking by a panel of 10 well trained testers at the University of Gastronomic Sciences, Bra (Italy). The sensory panel was made up of 5 males and 5 females and the average age was 33 (range = 24–42). Each tester had a minimum of two years of experience in sensory evaluation, using descriptive analysis, on various kinds of food and beverages. Prior to the assessment, the panel was specifically trained on bread using the so called "ballot training method" (Lawless & Heymann, 2010). Four 2 h training sessions were conducted. In the first training session the panelists were provided with the entire range of the products and with a list containing 28 attributes and their definitions. The list has been fully described in detail in Heenan et al. (2008). After evaluating the samples, panelists indicated, through consensus, which of the words and definitions would be used in the study. In three subsequent sessions, the panelists were provided with standards by the panel leader and the list of words was refined. At the end of the training the panelists, through consensus, deleted 15 attributes pertaining to appearance (speckled, crust smoothness); to odor (dairy, floury, musty, malty); to flavor (bitter, buttery, oily, seedy; for oral-texture: coarse, fat); to after-flavor (bitter, sour, toasted), and added 4 new attributes (odor intensity, flavor intensity, elasticity and crumbliness as oral-texture properties), and split the porosity attribute into two (pore quantity, pore dimension). The panelists provided a definition of the six new attributes and indicated the sequence of all the attributes for the evaluation. The final list of 18 attributes, definitions and standards used for the final assessment of the samples is reported in Table 1.

The trained panel scored the perceived intensity for a given sample for each descriptor on a linear continuous scale. Verbal expressions, e.g. low intensity–high intensity, were indicated at the extreme ends, according to Holtekjølen et al. (2008). The panel scored samples by marking the scale on a computer with a cursor (mouse). The left side of the scale corresponded to the 0.0 value and the right side corresponded to the 10.0 value. A computerized system (FIZZ, Version 2.46A, Biosystèmes) was used to record the data. A half-loaf of bread (~200 g) of each sample, including the crust and the crumbs, was presented to the assessors during all the evaluation sessions on three-digit coded disposable plastic plates, in a completely randomized order. A 60-second interval was set between samples to reduce the likelihood of carryover. In addition, each assessor was provided with still water and unsalted crackers and asked to cleanse their palate between tastings. The samples were evaluated in individual sensory booths under white light at room temperature (23 ± 1 °C).

2.3. Electronic nose

The volatile compound emissions from the samples were monitored during storage by means of a commercial portable electronic nose (PEN 3 model, Win Muster Airsense Analytics GmbH, Schwerim, Germany). This nose consists of a sampling apparatus, a detector unit containing an array of sensors and pattern recognition software (Win Muster v.16) for data recording and elaboration. The sensor array system is composed of 10 metal oxide semiconductors (MOS) of different chemical compositions and thicknesses to provide selectivity towards volatile compound classes, as indicated by the instrument supplier: W1C (aromatic

Table 1
Sensory descriptors (No. = 18) evaluated in bread together with the respective definitions and references.

i	Attributes	Definition	Low intensity reference	High intensity reference
Appearance				
1	Crust darkness	Degree of color darkness in the crust, ranging from light brown to dark brown	Multigrain sandwich bread	Whole rye sandwich bread
2	Crumb darkness	Degree of color darkness in the crumbs, ranging from white to light brown	White sandwich bread	Multigrain sandwich bread
3	Pore quantity	Quantity of pores in the crumbs	Matera bread IGP	White sandwich bread
4	Pore dimension	Dimension of pores in the crumbs	1 mm^2	2 cm^2
Odor				
5	Odor intensity	Total odor intensity of the sample	–	–
6	Yeasty	Odor associated with aromatic exchange from yeast fermentation	Distilled water	Beer yeast powder
7	Grain	Aromatic impression of cereal derived products, usually rye, wheat, oats, cornmeal and barley	Distilled water	Oats, rye, barley, wheat grains (proportion 1:1) ground finely
8	Nutty	Aromatics associated with a blend of mixed nuts, e.g. walnuts, hazelnuts and pine nuts	Distilled water	Walnuts, hazelnuts and pine nuts (proportion 1:1) cut finely
9	Toasted	Odor impression of bread and crumb after baking/heating	Distilled water	White sandwich bread cut finely and toasted in a pan
Taste/Flavor				
10	Salty	Fundamental taste sensation elicited by means of sodium chloride	Distilled water	Marine salt diluted in distilled water (2 g/L)
11	Sweet	Fundamental taste sensation of which sucrose is typical	Distilled water	Refined sugar diluted in distilled water (5 g/L)
12	Sour	Fundamental taste sensation evoked by acids, e.g. citric acid	Distilled water	Citric acid diluted in distilled water (0.5 g/L)
13	Flavor intensity	Strength of the total flavor in the sample	–	–
Texture				
14	Elasticity	Ability of the bread to retain its shape after squeezing by hand	Rye sandwich bread	White sandwich bread
15	Crumbliness	Tendency of the bread crust to crumble	White sandwich bread	White crackers
16	Hardness	Force required to bite completely through a sample placed between the molars	White sandwich bread	Whole wheat stale bread
17	Adhesiveness	Force required to remove a sample completely from the palate, using the tongue during consumption	White sandwich bread	Whole rye sandwich bread
18	Moisture	Amount of moisture perceived on the surface of the product, when in contact with the oral cavity	White sandwich bread	Whole rye sandwich bread

compounds), W5S (broad-range compounds, polar compounds, nitrogen oxides and ozone), W3C (ammonia, aromatic compounds, aldehydes, ketones), W6S (hydrogen), W5C (alkanes, aromatic compounds, less polar compounds), W1S (methane, broad-range compounds), W1W (sulphur compounds, terpenes and sulphur organic compounds), W2S (alcohols, partially aromatic compounds, ketones), W2W (aromatic compounds, sulphur organic compounds) and W3S (methane). The sensor response is expressed as resistivity (Ohm). The MOS sensors rely on changes in conductivity induced by the adsorption of molecules in the gas phase and on subsequent surface reactions. They consist of a ceramic substrate coated with a metal oxide semiconducting film, and are heated by means of a wire resistor. Owing to the high operating temperatures (200–500 °C), the organic volatiles transferred to the surface of the sensors are combusted totally to carbon dioxide and water, and this leads to a change in the resistance. The use of a high temperature prevents water interference and encourages a rapid response and recovery times (Kohl, 1992). The detection limit of the hot sensors is in the 1 ppm range.

Eight grams of bread, including the crust and the crumbs, was cut into small cubes (~1 cm^3) and placed in 45 mL glass air tight vials, hermetically sealed with a PTFE/silicone septum and a screw cap (Limbo, Torri, Sinelli, Franzetti, & Casiraghi, 2010). The vials were equilibrated at 25 ± 1 °C for 24 h and analyzed at the same temperature under standardized conditions. The measurement device sucked the gaseous compounds from the headspace of the sample, through the sensor array, at 300 mL/min for 180 s. After sample analysis, the system was purged for 400 s at a flow rate of 600 mL/min with filtered air prior to the next sample injection in order to allow the re-establishment of the instrument base line. Six replications were performed for each type of bread, and the individual records were used for the statistical analyses.

2.4. Image analysis

Three loaves of each type of bread were sampled from each production batch and cut into two halves. One half-loaf of each sampled bread was analyzed (three replicates). Individual bread images were acquired by digitalization using a Hewlett-Packard ScanJet 8200 desktop scanner with Hewlett-Packard Scanning version 2.2.1 software (Hewlett-Packard, Cupertino, CA, USA). The scanner and the software operated on a Lenovo 3000 N200 notebook with a Pentium Dual Core processor (Lenovo, Morrisville, NC, USA). Each half-loaf was placed on the flat bed scanner in pre-standardized conditions (black cardboard box over the half-loaf was imposed to enhance contrast) (Russ, 2011). The image was acquired at a resolution of 200 dpi (78.74 dots per cm) full color, and saved as a TIFF file. The image files were analyzed using the Image Pro Plus 6.0 software (Media Cybernetics Inc., Silver Spring, MD, USA). The bread images were spatially calibrated using millimeters as the unit before feature extraction (Russ, 2011). In this work, several features were extracted from the images to describe the morphological characteristics of the full slice and the gas holes present in the paste (Abdullah, 2008). The total area of each entire slice was measured by means of an automatic procedure, provided by the software, based on the automatic selection of the bright objects on a dark background (Russ, 2011). Simple threshold “magic wand” and “fill–black color” options were applied in order to isolate the gas holes present in the bread. After this operation, the selected area was measured applying the same procedure mentioned above for the full slice. An automatic procedure was then used to count and classify the isolated dark object into three classes as a function of their area: class 1 — small size (S; $0.2 < \text{area} < 3.0\ mm^2$); class 2 — medium size (M; $3.0 < \text{area} < 10.0\ mm^2$); class 3 — large size (L; $>10.0\ mm^2$). Several geometric features were measured for each hole class: number of holes, percentage of holes belonging to the class compared to the total number of holes on the half-loaf surface, total area of the holes belonging to the class, percentage of the area occupied by the holes belonging to the class compared to the total area occupied by all the holes present on the half-loaf surface, average roundness of the holes, aspect of the holes, pore density, height/width ratio of the holes (Y/X) (Russ, 2011).

2.5. Statistical analysis

The study concerned three sets of variables, namely the predicted sensory set (S_i, $i = 1–18$, 10 panelists) and the two predictive sets: e-nose (E_k, $k = 1–450$, 10 MOS traces for 180 s reduced by 8, 8 replicates) and image set (I_j, $j = 1–9$, 3 replicates). The groups were characterized by a univariate linear model and by multivariate chemometric methods, that is, using the linear partial least squares (PLS) method (Barker & Rayens, 2003; Dardenne, Sinnaeve, & Baeten, 2000).

2.5.1. Univariate analysis

A univariate approach was adopted to investigate the 18 sensory variables, the sum of the 10 MOS resistivity Ω_i/Ω_0 values plus the integral and the 9 image scores. A generalised linear model of the SAS V. 9 software (SAS Institute, Cary, NC, USA) was used considering a single group factor with 6 levels (B, Bm, C, G, I, S) that featured each single score or analysis. The comparison of the mean values was carried out according to Fisher's test at $P \leq 0.05$ and $P \leq 0.01$ probability levels. The mycorrhizal effect was ascertained according the Bm vs. B contrast. The effect of the epoch was tested by examining the contrast GIS vs. BmBC. The mycorrhizal modern group, Bm, was also tested to check if it was similar to the old groups using a BmGIS/BC contrast, and a BmS/GI contrast pointed out some further differences.

2.5.2. Multivariate analyses

2.5.2.1. Matrix distance and average cluster analysis. The 3 sets of $[\underline{S}_{18}]$ — sensory, $[\underline{E}_{450}]$ — e-nose and $[\underline{I}_9]$ — image were elaborated in order to build 3 distance matrices. For this purpose, each couple of the 6 groups was fitted as a binary dummy variable in order to build the $\{\underline{S}, \underline{E}, \underline{I}\}$ distance matrices, which was composed of 15 couples of contrasts and a zero diagonal. The parameter retained to characterize the distances between the 6 groups in the matrix was the r-square obtained in cross-validation mode (R^2cv) pertinent to each of the 15 contrasts.

The z-score obtained by Fisher transformation according to Preacher (2002) was used to testify the differences in the R^2cv values. Chemometrics was performed by means of the WinISI II v1.04 software (Infrasoft International, ISI: State College, PA, USA). A cross-validation system was adopted to assess the optimal number of latent variables to be included in the PLS equations, while permitting one passage for elimination of the outliers ($t > 2$; $H > 10$, Fearn, 1997).

In order to connect the e-nose to the image information, a mixed matrix was built with the reduced $\{\underline{E}\}$ and $\{\underline{I}\}$ triangular matrices.

The distance matrices were then analyzed by means of Ward's Hierarchical Clustering Analysis (HCA), performed via StatBox software vs 6.5 (Grimmer Logiciel, Paris) in order to compare the relative average similarity patterns (Jobson, 1992). HCA performs agglomerative hierarchical clustering of objects on the basis of distance measures of dissimilarity. The hierarchy of clusters can be represented by a binary tree, called “dendrogram”. A final partition, i.e., the cluster assignment of each object, is obtained by cutting the tree at a specified level of the scree plot (Gardner & Bartlett, 1992).

2.5.2.2. Prediction of the sensory variables by e-nose, imaging and their concatenated sets. In order to evaluate the prediction ability of the instrumental analyses in estimating the sensory scores, the 3 sets described above were divided into predicted (sensory) and predictor (e-nose, image, e-nose & image sets); a series of quantitative calibrations was then performed by means of the linear PLS method. For this purpose, the records obtained from the e-nose in each analysis of the bread were fitted to the averages of the $[\underline{S}_{18}]$ sensory variables, for each of the 6 groups.

The same process was adopted for the image records, in order to predict the sensory scores.

Finally, in order to capitalize on the complementarily of the results, a further prediction of the sensory scores was obtained by concatenating the e-nose records – reduced to the average of the 10 MOS sensors plus their sum – with an image record that was randomly assigned from those carried out in the same group. The concatenated set [E & I] consisted of 20 predictor variables.

The prediction capacity of the calibrated models was then evaluated for these quantitative sensory traits with the ratio performance deviation (RPD) (Williams, 1987; Williams & Sobering, 1996), a capacity parameter, defined as the relationship between the standard deviation of the chemical method (SD reference) and the standard error in cross-validation (SECV). When the RPD values were ≥ 2.5, the relevant calibration models were considered to be suitable for routine use for single case discrimination purposes, but an $RPD \geq 2.0$ showed significant differences between groups.

3. Results and discussion

3.1. Bread properties

The analysis of the sensory profiles of the six groups of bread reported in Table 2 indicated an average r-square level of 0.30. The univariate analysis results, although reporting significant differences between the wheat varieties for all but one of the sensory attributes (moisture), did not seem satisfactory. For this reason, an analysis of the contrasts was conducted by pooling specific groups in contrast with other groups, in order to improve the power of the statistics for particular effects, i.e. old varieties vs. modern, or to assess whether the mycorrhizal had an old or modern aspect.

The G–I–S old varieties compared to the modern Bm–B–C, exhibited significant positive contrast values for crust darkness, pore quantity, grain odor, flavor intensity and significant negative contrasts for yeasty and nutty odor, and adhesiveness. These results indicated higher quality traits for the old varieties than the modern ones. It has in fact been reported (Laureati, Giussani, & Pagliarini, 2012; Morais, Cruz, Faria, & Bolini, 2013) that crust color and porosity are positively correlated to consumers' preferences while yeasty aroma, flavor and adhesiveness are negatively correlated to the overall liking expressed by consumers'.

The authors have hypothesized that the mycorrhizal factor can provide an “*old look*” to a modern genetic tool and in this way the Bm–G–I–S vs. B–C contrast was calculated. The results have demonstrated that the mycorrhizal factor modified the Bm group from the B and C reference groups and induced several significant differences in appearance (higher pore quantity), odor intensity (less nutty and toasted but more pronounced yeasty aroma), taste and flavor intensity (increased, except sweetness), and texture scores (improved elasticity and crumbliness with less adhesiveness). In particular, the high positive contrast observed for the salty taste perception, could be important for the production of bread considering the EC directive which has the aim of reducing the salt concentration in bread (WHO, 2007).

As far as the old wheat products, including the mycorrhizal group are concerned, three significant contrasts were positive for Bm–S while six were positive for G–I.

The responses of the ten e-nose sensors (Table 3) showed more variance than the sensory panel (R^2 avg. 0.50). The results of the

Table 2
Univariate analysis. LSMean scores of the $[\underline{S_{18}}]$-sensory attributes and contrasts. (Means within a row with different letters are significantly different; Fisher's test, $\alpha \leq 0.05$).

	$[\underline{S_{18}}]$				LSMean groups						Contrasts[1] (%)		
i	Sensory attributes		R^2	SE	G	I	S	Bm	B	C	GIS/BmBC	BmGIS/BC	BmS/GI
1	*Appearance*	Crust darkness	0.23	1.99	5.71a	6.5a	5.57a	3.42b	6.34a	4.96ab	**21**	−6	**−26**
2		Crumb darkness	0.08	2.17	3.53b	5.44a	4.69ab	4.45ab	4.14ab	4.87ab	1	0	2
3		Pore quantity	0.34	1.92	7.83a	4.75b	4.19b	4.62b	3.94b	4.36b	**30**	**29**	**−30**
4		Pore dimension	0.24	2.25	7.23a	6.89ab	4.5bc	5.13bc	6.26ab	4.08c	20	15	**−32**
5	*Odor*	Odor intensity	0.43	1.99	6.01a	5.96a	2.04b	2.26b	4.46a	5.37a	16	−17	**−64**
6		Yeasty	0.49	1.82	3.81c	2.78c	2.38c	7.02a	3.00c	5.74b	**−43**	−9	**43**
7		Grain	0.12	2.26	5.84ab	5.63ab	6.28a	4.55ab	4.27b	4.31b	**35**	**30**	−6
8		Nutty	0.42	2.08	2.68c	4.75b	2.77c	3.93bc	7.04a	6.55a	**−42**	**−48**	−10
9		Toasted	0.20	2.27	4.51ab	5.81ab	3.53c	4.26bc	6.45a	6.1ab	−18	**−28**	−25
10	*Taste/flavor*	Salty	0.15	2.31	6.04a	6.52a	5.27ab	5.95a	4.64ab	3.78b	24	**41**	−11
11		Sweet	0.19	2.20	3.59b	6.04a	4.52ab	4.08b	6.36a	5.44ab	−11	**−23**	−11
12		Sour	0.31	1.99	5.56a	4.77a	1.48b	4.15a	4.44a	3.91a	−6	−4	**−45**
13		Flavor intensity	0.46	2.00	4.13b	6.50a	8.13a	6.73a	3.03b	4.49b	**32**	**69**	**40**
14	*Texture*	Elasticity	0.35	2.01	3.69bc	3.11bc	4.72b	6.64a	4.36b	2.16c	−12	**39**	**67**
15		Crumbliness	0.46	1.73	2.28c	5.80a	3.29bc	5.90a	4.08b	2.22c	−7	**37**	14
16		Hardness	0.31	2.28	5.98a	6.59a	2.11c	4.64ab	3.96bc	4.3ab	14	17	**−46**
17		Adhesiveness	0.46	1.65	3.05b	2.61b	2.22b	2.90b	5.82a	5.56a	**−45**	**−53**	−10
18		Moisture	0.07	2.07	5.28	5.19	6.04	4.52	5.42	6.19	2	−9	1

[1] The significant contrast values are in bold ($\alpha \leq 0.05$).

univariate statistical analysis highlighted the ability of the e-nose to differentiate the groups between whole wheat bread samples. Our findings are in agreement with those of Sapirstein, Siddhu, and Aliani (2012), who reported that e-nose was capable of differentiating between bread volatiles whose composition varied due to differences in flour or bran type. Similarly, Botre and Gharpure (2006) demonstrated the ability of e-nose to cluster bread odor data according to the freshness of the bread.

Mycorrhizal factor significantly differentiated the two Blasco (B and Bm) groups for 6 sensors out of 11. In the planned contrasts, the differences in B vs. Bm amplified the divergence between the modern and old varieties; the G–I–S vs. Bm–B–C contrast was in fact significant for 8 out of 11 cases and the Bm–G–I–S vs. B–C contrast for 9 out of 11. As far as the old and "*old look*" varieties are concerned, the e-nose differentiated the G–I group from the Bm–S groups in 7 cases out of 11.

Elaboration of the image scores (Table 4) showed a higher level of fitting (R^2 avg. 0.70), but the Sieve group was diminished to a great extent from the others for 5 out of 9 image traits. Because of the abnormal minus-variance in the S group, the contrasts of the old vs. modern (G–I–S vs. Bm–B–C) varieties enhanced 5 negative traits out of 9 while the Bm–G–I–S vs. B–C contrast showed 5 negative and 1 positive cases. The distinct behavior of Sieve suggested a higher variability between the old varieties than between the modern ones, thus confirming that the largest ranges of variation were found in landraces and old cultivars rather than in more recent varieties (Bordes et al., 2008). The mycorrhizal factor applied to Blasco wheat increased the pores in the bread in number, in percentage of the area and in the Y/X ratio. Taking into account that porosity was positively correlated with consumers' preferences (Laureati et al., 2012), the observed increase in the pore attributes has revealed a positive effect of the mycorrhizal factor on Blasco appearance.

3.2. Comparison of techniques

A marked similarity between the 6 groups emerged from the sensory, e-nose and image analyses data elaboration. The distance matrices {S}, {E} and {I} reported in Table 5, which were based on the 15 couple comparisons, in fact, enhanced the average R^2cv by 0.65, 0.81 and 0.79, respectively, and these values were more elevated than the previous 0.30, 0.50 and 0.70 values one, which, however, were averages of the different univariate models.

The mycorrhizal effect on the modern Blasco wheat appeared very high and was highlighted in the distance matrices with R^2cv 0.66 for sensory, 0.88 for e-nose and 0.85 for images.

When the 6 groups were clustered on the basis of the Euclidean distances according to their R^2cv coefficients (Fig. 1: I-IV), a good degree of symmetry emerged between the sets; the reference flour couple, composed of the C — Control and the non mycorrhizal B — Blasco (cluster 1; C–B) was in fact always distinguished from the three old varieties and from the mycorrhizal Bm — Blasco. In the sensory cluster, Bm was closer to the S — Sieve than to the couple formed by G — Gentil_Rosso and I — Inallettabile. This pattern was confirmed in the

Table 3
Univariate analysis. LSMean scores of the 10 MOS sensors of the $[\underline{E}]$ e-nose and of the integral and contrasts. (Means within a row with different letters are significantly different; Fisher's test, $\alpha \leq 0.05$).

$[\underline{E}]$ — e-nose			LSMean groups						Contrasts[1] (%)		
Sensors	R^2	SE	G	I	S	Bm	B	C	GIS/BmBC	BmGIS/BC	BmS/GI
W1C	0.23	0.34	4.82a	4.33b	4.56ab	4.81a	4.52ab	4.53ab	−1	2	2
W5S	0.66	128	939a	922ab	807b	677c	509d	557cd	**53**	**57**	**−20**
W3C	0.44	0.35	5.23a	4.86b	4.73b	4.87b	4.43bc	4.37c	**9**	**12**	**−5**
W6S	0.63	1.02	20.9c	22.5b	22.4b	22.3b	24.3a	24.6a	**−8**	**−10**	3
W5C	0.63	0.34	4.85a	4.62ab	4.24b	4.31b	3.79c	3.69c	**16**	**20**	**−10**
W1S	0.57	23.80	112c	126bc	146.b	145b	179a	183a	**−24**	**−27**	**22**
W1W	0.63	4.95	49.0a	46.7a	44.6ab	40.9b	32.5c	35.0c	**29**	**34**	**−11**
W2S	0.72	18.0	89.8c	91.0c	119b	122b	152a	160a	**−31**	**−32**	**34**
W2W	0.16	6.08	58.9b	59.8ab	62.3ab	59.5ab	63.3ab	66.1a	−4	**−7**	3
W3S	0.42	0.40	20.4bc	21.1a	20.3bc	20.2bc	20.2c	20.2bc	**2**	**1**	**−2**
Integral	0.37	171	1306a	1304a	1237ab	1102b	994b	1059b	**22**	**20**	**−10**

[1] The significant contrast values are in bold ($\alpha \leq 0.05$).

Table 4
Univariate analysis. LSMean scores of the $[I_9]$ — image traits and contrasts. (Means within a row with different letters are significantly different; Fisher's test, $\alpha \leq 0.05$).

	$[I_9]$ — image set			LSMean groups						Contrasts[1] (%)		
j	Variables	R^2	SE	G	I	S	Bm	B	C	GIS/BmBC	BmGIS/BC	BmS/GI
1	Slice total area	0.85	657	9657b	10416ab	7418c	11497a	10972ab	10030b	**−18**	**−13**	−7
2	Slice pore number	0.74	77.81	520.3b	531.6b	361.3c	588ab	683.3a	666.3a	**−14**	0	−12
3	Pore total area	0.80	247	2396a	2611a	1506b	2546.a	2454a	2759a	**−12**	**−12**	**−23**
4	Large pore area (%)	0.59	5.60	52.86ab	54.86a	43.63b	50.26ab	38.63c	48.4ab	1	**−13**	**−19**
5	Large pore number (%)	0.58	1.48	10.400a	10.460a	8.366ab	9.133a	6.233b	8.733ab	6	**−14**	**−24**
6	Medium pore roundness	0.69	0.46	4.200ab	3.833b	3.666b	4.933a	5.233a	4.333a	**−13**	−1	**4**
7	Small pore aspect	0.63	0.10	2.266a	2.166a	1.933b	2.200a	2.2a	2.233a	−2	−1	**−8**
8	Y/X	0.83	0.05	0.433b	0.533a	0.300c	0.500a	0.466b	0.566a	**−13**	**−15**	**−23**
9	Pore density	0.61	0.63	5.333b	5.100b	4.900b	5.133b	6.233ab	6.633a	2	**13**	−7

[1] The significant contrast values are in bold ($\alpha \leq 0.05$).

e-nose and in the e-nose with image concatenated patterns. S — Sieve instead in the image cluster showed a marked originality.

On the whole, the multivariate cluster analysis highlighted similitude among the 6 groups, and the reference C bread in particular was similar to the B — Blasco non mycorrhizal bread.

The present results on the mycorrhizal factor have shown that some putative secondary constituents could have been modified (Ceccarelli et al., 2010; Strack et al., 2003). For this reason a specific study was underway. Giovannetti et al. (2012) have shown that the symbiosis in tomato plants positively affected the growth and mineral nutrient content and enhanced the nutritional and functional value of tomato fruit through modifications of the secondary metabolism of the plant, which led to increased levels of lycopene; moreover, such changes did not result in the production of mutagenic compounds, since the tomato extracts induced no in vitro genotoxic effects. Similar antioxidant fortification results in tomato have been obtained by Ordookhani and Zare (2011).

3.3. Prediction of the sensory variables

As far as the prediction is concerned (Table 6), e-nose was more efficient than the image analysis except for the texture traits, where the image analysis was better (avg. RPD 1.95 vs. 1.80). Considering all of the 18 attributes, the e-nose prediction exceeded the RPD 2.0 threshold in 8 cases, with a maximum for the pore dimension (3.2) followed by adhesiveness, and then nutty and salty, while the image analysis exceeded RPD 2.0 in 3 cases, with a maximum (3.0) for the moisture trait. When considering the representation of the sensory traits in the whole objects provided by the two instruments, the raw average cross-validated r-square, calculated across the 18 attributes, was 0.69 for the e-nose and 0.56 for the image analysis features, but the similarity provided by the concatenated sets rose to 0.83, with consistent improvements in prediction of the odor, taste, flavor and texture: of the 18 sensory variables, only 4 were below 2.0 and 10 resulted over 2.5 RPD. The high prediction for several sensory traits obtained from the e-nose and image analysis instruments confirmed the fact that an a-priori difference assessed by the rapid instruments, could be transformed in a a-posteriori verification of the differentiation of the groups. The relationships between the mechanical parameters and the odor release responses that were captured by e-nose which have emerged in this work, have confirmed the results of the Piazza and Benedetti (2008) study performed on low moisture bakery products by means of the acoustic–mechanical technique combined with e-nose.

Table 5
Distance matrices {S} — sensory, {E} — e.nose, {I} — image and concatenated {E & I} of the six groups on the basis of the PLS multivariate. R-squares in cross-validation (R^2cv) values over the diagonal and contrasts[1] below the diagonal.

Variables	Groups	Bm	B	G	I	C	S	Mean ± SD
{S} — Sensory	Bm	0	0.66	0.84	0.85	0.68	0.64	0.65 ± 0.16
6 groups	B	abc	0	0.65	0.56	0.20	0.82	
*	G	a	abc	0	0.47	0.60	0.79	
10 panelists	I	a	bcd	cd	0	0.57	0.67	
*	C	abc	d	bcd	bcd	0	0.68	
18 scores	S	abc	ab	ab	abc	abc	0	
{E} — E-nose	Bm	0	0.88	0.92	0.96	0.93	0.64	0.81 ± 0.25
6 groups	B	bc	0	0.97	0.97	0.07	0.74	
*	G	b	a	0	0.53	0.97	0.73	
8 replicates	I	ab	ab	d	0	0.98	0.92	
*	C	ab	e	a	a	0	0.96	
225 scores (1800/8)	S	d	cd	cd	b	ab	0	
{I} — Image	Bm	0	0.85	0.78	0.58	0.92	0.78	0.79 ± 0.14
6 groups	B	abcd	0	0.75	0.84	0.74	0.82	
*	G	cde	cdef	0	0.42	0.87	0.95	
8 replicates	I	ef	abcd	f	0	0.69	0.92	
*	C	ab	cdef	abcd	def	0	0.91	
9 scores	S	cde	bcd	a	ab	abc	0	
Concatenated	Bm	0	0.88	0.92	0.96	0.93	0.64	0.82 ± 0.15
{E & I}	B	abc	0	0.75	0.84	0.74	0.82	
6 groups	G	abc	de	0	0.42	0.87	0.95	
*	I	a	bcd	e	0	0.69	0.92	
8 replicates	C	ab	de	bcd	de	0	0.91	
*	S	de	cd	ab	abc	abc	0	
20 scores (11 E-nose + 9 Image)								

[1] Below diagonal contrasts for $(R^2cv)^{-5}$ of Bm|S groups; $a > b > c > d > e$: ($\alpha \leq$ P-value < 0.05); test: z-score by means of Fisher's transformation according to Preacher (2002).

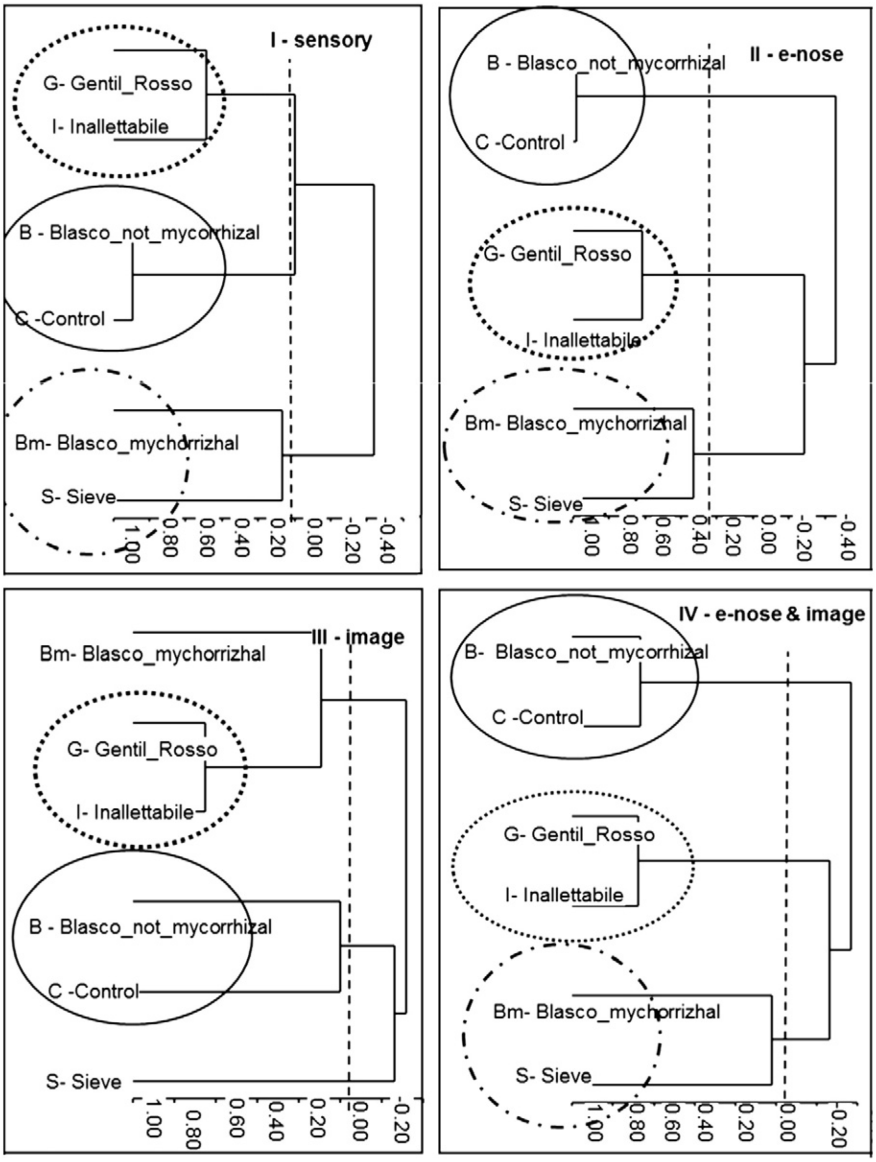

Fig. 1. Ward's hierarchical clusters of the similarity between the six groups according to the intergroup distances in the distance matrices {S} — sensory (I), {E} e-nose (II), {I} image analysis (III) and concatenated {EI} e-nose-image (IV). Abscises: Euclidean distance.

Table 6
Prediction of the [$\underline{S_{18}}$]-sensory variables on the basis of the e-nose measurements of the bread, the image analysis variables, and a concatenation of the two sets.

				E-nose			Image analysis			Concatenated		
	Attributes	Mean	SD	R2cv	RPD	RPD_mean	R^{2}cv	RPD	RPD_mean	R^{2}cv	RPD	RPD_mean
Appearance	Crust darkness	5.26	1.03	0.66	1.7		0.47	1.3		0.68	1.8	
	Crumb darkness	4.51	0.59	0.76	2.1		0.60	1.6		0.69	1.8	
	Pore quantity	5.02	1.29	0.75	2.0		0.55	1.5		0.73	1.9	
	Pore dimension	5.61	1.26	0.9	3.2	2.25	0.21	1.1	1.37	0.75	2.0	1.88
Odor	Odor intensity	4.21	1.83	0.65	1.7		0.80	2.2		0.83	2.5	
	Yeasty	4.27	1.81	0.47	1.4		0.32	1.2		0.76	2.1	
	Grain	5.31	0.77	0.7	1.8		0.78	2.0		0.98	6.9	
	Nutty	4.07	1.42	0.87	2.8		0.62	1.5		0.93	3.7	
	Toasted	4.83	0.95	0.74	2.0	1.94	0.62	1.5	1.69	0.89	3.0	3.62
Taste/flavor	Salty	5.56	0.92	0.85	2.6		0.61	1.5		0.96	4.9	
	Sweet	4.59	0.87	0.74	2.0		0.07	1.0		0.85	2.6	
	Sour	4.64	0.63	0.59	1.6		0.55	1.5		0.82	2.4	
	Flavor intensity	5.91	1.49	0.62	1.6	1.95	0.41	1.2	1.31	0.50	1.4	2.82
Texture	Elasticity	4.23	1.53	0.58	1.6		0.71	1.9		0.95	4.7	
	Crumbliness	3.93	1.61	0.66	1.7		0.56	1.5		0.82	2.4	
	Hardness	4.71	1.56	0.58	1.6		0.65	1.6		0.88	2.9	
	Adhesiveness	3.18	1.12	0.86	2.7		0.72	1.8		0.88	2.9	
	Moisture	5.45	0.58	0.45	1.4	1.8	0.90	3.0	1.95	0.96	4.7	3.53

SD: standard deviation; SECV: standard error in cross-validation; R^{2}cv: R-square in cross-validation; RPD: ratio-performance deviation.

Heenan et al. (2009), using a proton transfer reaction mass spectrometry (PTR-MS) technique, identified 33 mass-ions, out of 160, and was able to distinguish specialty from commercial breads. In terms of validated r^2, the PTR-MS features of 12 sensory attributes (out of 18) was 0.74, a higher value than the e-nose performance (0.69) but lower than the 0.84 obtained for the concatenated e-nose and image rapid analyses in the present work.

Carson and Sun (2001) studied the mechanical analysis of several commercial breads and correlated the viscoelastic portion (50 mN) to creep deformation ($r^2 = 0.77$) and sensory springiness ($r^2 = 0.82$), which appear comparable with the non-mechanical features attained by the e-nose with adhesiveness (R^2cv = 0.86) and by the image analysis with elasticity (R^2cv = 0.71) (Table 6).

Ponzoni et al. (2008) examined bread baking aroma detection by means of a low-cost e-nose based on a resistance to period converter readout system, which was suitable to handle a wide range of resistance values (from kΩ to tens of GΩ) with a high accuracy (<1%). The applications of e-nose to detect the key aromas of different stages of the bread baking process showed the ability of the proposed e-nose to distinguish these volatiles in an ordered manner and to reflect the different baking steps that they represent.

The present paper capitalizes on the mutual relationships between the groups of sensory properties and the instrumental features. Gao, Tan, Shatadal, and Heymann (1999) focused on the expanded-food sensory-texture properties of an extruded corn puff through image analysis. The features derived from the saturation band for hardness-related sensory properties resulted in the best r^2 values. Brosnan and Sun (2004) wrote a review on how computer vision has been successfully adopted for the quality analysis of meat, and fish, pizza, cheese, and bread.

4. Conclusions

This work has shown two original features: crop production and methodology pathway. It has been shown along the crop production axis that the mycorrhizal factor is a botanical modifier of bread sensory properties: a modern productive wheat treated with the Micosat F® microbial consortium promoted a modified breading process which led to a product that could not be differentiated from bread obtained from the old variety and which is similar to the old Gentil Rosso and Inallettabile varieties. E-nose and image rapid analyses, supported by heuristic chemometrics, have shown accuracy in the clustering of many conglomerate groups along the methodology pathway. Furthermore, the e-nose examination predicted several sensory scores. The image analysis was somewhat erratic, but when concatenated with the e-nose the average coefficient of similarity with the sensory traits rose to 0.83, with consistent improvements in the prediction of the odor, taste, flavor and texture: of the 18 sensory variables, only 4 were below 2.0, and 10 were over 2.5 RPD, thus showing a sign of good predictability. This work has confirmed the usefulness of the development of rapid, cheap, non-invasive and non-destructive instrumental techniques, based on the use of electronic-nose devices and/or image analysis methods has proved the positive correlation between the instrumental data and the sensory scores. Nevertheless from our results it emerges that these instruments cannot replace completely the sensory analysis, and further studies on the hedonic scale would be necessary in order to conduct a liking test aimed to evaluate the acceptability of the bread samples by consumers.

References

Abdullah, M. Z. (2008). Quality evaluation of bakery products. In D. W. Sun (Ed.), *Computer Vision Technology for Food Quality Evaluation* (pp. 481–522). London: Academic Press.

Adom, K. K., Sorrells, M. E., & Liu, R. H. (2003). Phytochemical profiles and antioxidant activity of wheat varieties. *Journal of Agricultural and Food Chemistry*, *51*, 7825–7834.

Baardseth, P., Kvaal, K., Lea, P., Ellekjær, M. R., & Færgestad, E. M. (2000). The effects of bread making process and wheat quality on French baguettes. *Journal of Cereal Science*, *32*, 73–87.

Barker, M., & Rayens, W. (2003). Partial least squares for discrimination. *Journal of Chemometrics*, *17*, 166–173.

Bonfante, P., & Genre, A. (2010). Mechanisms underlying beneficial plant–fungus interactions in mycorrhizal symbiosis. *Nature Communications*, *1*, 1–11.

Bordes, J., Branlard, G., Oury, F. X., Charmet, G., & Balfourier, F. (2008). Agronomic characteristics, grain quality and flour rheology of 372 bread wheats in a worldwide core collection. *Journal of Cereal Science*, *48*, 569–579.

Botre, B., & Gharpure, D. (2006). Analysis of volatile bread aroma for evaluation of bread freshness using an electronic nose (e-nose). *Materials and Manufacturing Processes*, *21*, 279–283.

Brosnan, T., & Sun, D. W. (2004). Improving quality inspection of food products by computer vision — A review. *Journal of Food Engineering*, *61*, 3–16.

Carson, L., & Sun, X. S. (2001). Creep-recovery of bread and correlation to sensory measurements of textural attributes. *Cereal Chemistry*, *78*, 101–104.

Ceccarelli, N. (2009). Evolution, plant breeding and biodiversity. *Journal of Agriculture and Environment for International Development*, *103*, 131–145.

Ceccarelli, N., Curadi, M., Martelloni, L., Sbrana, C., Picciarelli, P., & Giovannetti, M. (2010). Mycorrhizal colonization impacts on phenolic content and antioxidant properties of artichoke leaves and flower heads two years after field transplant. *Plant and Soil*, *335*, 311–323.

Dardenne, P., Sinnaeve, G., & Baeten, V. (2000). Multivariate calibration and chemometrics for near infrared spectroscopy: Which method? *Journal of Near Infrared Spectroscopy*, *8*, 229–237.

Di Cesare, L. F., Migliori, C., Ferrari, V., Parisi, M., Campanelli, G., Candido, V., et al. (2012). Effects of irrigation–fertilization and irrigation–mycorrhization on the alimentary and nutraceutical properties of tomatoes. In T. Shui Lee (Ed.), *Irrigation Systems and Practices in Challenging Environments* (pp. 207–232). Shanghai: InTech.

Fearn, T. (1997). Qualitative analysis I. *NIR news*, *8*, 4–5.

Gao, X., Tan, J., Shatadal, P., & Heymann, H. (1999). Evaluating expanded-food sensory properties by image analysis. *Journal of texture studies*, *30*, 291–304.

Gardner, J. W., & Bartlett, P. N. (1992). Pattern recognition in odour sensing. In J. W. Gardner, & P. N. Bartlett (Eds.), *Sensors and Sensory Systems for an Electronic Nose* (pp. 161–179). Boston: Kluwer Academic Publishers.

Gasztonyi, M. N., Farkasa, R. T., Berkia, M., Petroczib, I. M., & Daood, H. G. (2011). Content of phenols in wheat as affected by varietal and agricultural factors. *Journal of Food Composition and Analysis*, *24*, 785–789.

Giovannetti, M., & Avio, L. (2002). Biotechnology of arbuscular mycorrhizas. In G. G. Khachatourians, & D. K. Arora (Eds.), *Applied mycology and biotechnology. Vol. 2 Agriculture and Food Production* (pp. 275–310). Amsterdam: Elsevier.

Giovannetti, M., Avio, L., Barale, R., Ceccarelli, N., Cristofani, R., Iezzi, A., et al. (2012). Nutraceutical value and safety of tomato fruits produced by mycorrhizal plants. *British Journal of Nutrition*, *107*, 242–251.

Guarda, G., Silvano Padovan, S., & Delogu, G. (2004). Grain yield, nitrogen-use efficiency and baking quality of old and modern Italian bread-wheat cultivars grown at different nitrogen levels. *European Journal of Agronomy*, *21*, 181–192.

Heenan, S. P., Dufour, J. P., Hamid, N., Harvey, W., & Delahunty, C. M. (2008). The sensory quality of fresh bread: descriptive attributes and consumer perceptions. *Food Research International*, *41*, 989–997.

Heenan, S. P., Dufour, J. P., Hamid, N., Harvey, W., Harvey, W., & Delahunty, C. M. (2009). Characterisation of fresh bead flavour: relationship between sensory characteristics and volatile composition. *Food Chemistry*, *116*, 249–257.

Holtekjølen, A. K., Baevre, A.B., Rødbotten, M., Berg, H., & Knutsen, S. H. (2008). Antioxidant properties and sensory profiles of breads containing barley flour. *Food Chemistry*, *110*, 414–442.

Jensen, S., Oestdal, H., Skibsted, L. H., Larsen, E., & Thybo, A. K. (2011). Chemical changes in wheat pan bread during storage and how it affects the sensory perception of aroma, flavour and taste. *Journal of Cereal Science*, *53*, 259–268.

Jobson, J.D. (1992). *Applied Multivariate Data Analysis. Volume II: Categorical and Multivariate Methods*. New York: Springer-Verlag.

Kannan, V. R., Suganya, S., King Solomon, E., Balasubramanian, V., Ramesh, N., & Rajesh, P. (2011). Analysis of interaction between arbuscular mycorrhizal fungi and their helper bacteria by MILPA model. *Research in Plant Biology*, *1*, 48–62.

Kihlberg, I., Johansson, L., Kohler, A., & Risvik, E. (2004). Sensory qualities of whole wheat pan bread—Influence of farming system, milling and baking technique. *Journal of Cereal Science*, *39*, 67–84.

Kohl, D. (1992). Fundamentals and recent developments of homogeneous semiconducting sensors. In J. W. Gardner, & P. N. Bartlett (Eds.), *Sensors and Sensory Systems for an Electronic Nose* (pp. 53–76). Boston: Kluwer Academic Publishers.

Laureati, M., Giussani, B., & Pagliarini, E. (2012). Sensory and hedonic perception of gluten-free bread: Comparison between celiac and non-celiac subjects. *Food Research International*, *46*, 326–333.

Lawless, H. T., & Heymann, H. (2010). *Sensory Evaluation of Foods: Principles and Practices* (2nd ed.)New York: Chapman & Hall.

Limbo, S., Torri, L., Sinelli, N., Franzetti, L., & Casiraghi, E. (2010). Evaluation and predictive modeling of shelf life of minced beef stored in high-oxygen modified atmosphere packaging at different temperatures. *Meat Science*, *84*, 129–136.

Mader, P., Hahn, D., Dubois, D., Gunst, L., Alfoldi, T., Bergmann, H., et al. (2006). Wheat quality in organic and conventional farming: Results of a 21 year field experiment. *Journal of Science of Food and Agriculture*, *87*, 1826–1835.

Maronek, D.M., Hendrix, J. W., & Kiernan, J. (2011). Mycorrhizal fungi and their importance in horticultural crop production. In J. Janick (Ed.), *Horticultural Reviews*, *vol. 3*. Hoboken: Wiley & Sons (pp.).

Morais, E. C., Cruz, A. G., Faria, J. A. F., & Bolini, H. M.A. (2013). Prebiotic gluten-free bread: Sensory profiling and drivers of liking. *LWT — Food Science and Technology*, http://dx.doi.org/10.1016/j.lwt.2013.07.014.

Ordookhani, K., & Zare, M. (2011). Effect of *Pseudomonas*, *Azotobacter* and arbuscular Mycorrhiza fungi on lycopene, antioxidant activity and total soluble solid in tomato (*Lycopersicon Esculentum* F1 Hybrid, Delba). *Advances in Environmental Biology*, *5*, 1290–1294.

Piazza, L., & Benedetti, S. (2008). Study of structure and flavour release relationships in low moisture bakery products by means of the acoustic–mechanical combined technique and the electronic nose. *Journal of Cereal Science*, *48*, 413–419.

Ponzoni, A., Depari, A., Falasconi, M., Comini, E., Flammini, A., Marioli, D., et al. (2008). Bread baking aromas detection by low-cost electronic nose. *Sensors and Actuators B: Chemical*, *130*, 100–104.

Preacher, K. J. (2002). Calculation for the test of the difference between two independent correlation coefficients [Computer software]. (Available from). http://quantpsy.org (Accessed 09/05/2013)

Russ, J. C. (2011). *The Image Processing Handbook* (6th ed.)Boca Raton: CRC Press (Chapters 1, 5, 7,10, 11, and 12).

Sapirstein, H. D., Siddhu, S., & Aliani, M. (2012). Discrimination of volatiles of refined and whole wheat bread containing red and white wheat bran using an electronic nose. *Journal of Food Science*, *77*, S399–S406.

Scanlon, M. G., & Zghal, M. C. (2001). Bread properties and crumb structure. *Food Research International*, *34*, 841–864.

Škrbić, B., & Filipčev, B. (2008). Nutritional and sensory evaluation of wheat breads supplemented with oleic-rich sunflower seed. *Food Chemistry*, *108*, 119–129.

Škrbić, B., Milovac, S., Dodig, D., & Filipčev, B. (2009). Effects of hull-less barley flour and flakes on bread nutritional composition and sensory properties. *Food Chemistry*, *115*, 982–988.

Smith, S. E., & Read, D. J. (2008). *Mycorrhizal Symbiosis* (3rd ed.)New York: Academic Press.

Strack, D., Fester, T., Hause, B., Schliemann, W., & Walter, M. H. (2003). Arbuscular mycorrhiza: Biological, chemical and molecular aspects. *Journal of Chemical Ecology*, *29*, 1955–1979.

Wang, B., & Qiu, Y. L. (2006). Phylogenetic distribution and evolution of mycorrhizas in land plants. *Mycorrhiza*, *16*, 299–363.

Ward, J. L., Poutanen, K., & Gebruers, K. (2008). The HEALTHGRAIN Cereal Diversity Screen: concept, results, and prospects. *Journal of Agricultural and Food Chemistry*, *56*, 9699–9709.

WHO (World Health Organization) (2007). Reducing salt intake in populations: Report of a WHO forum and technical meeting. Paris, France, 5–7 October 2006. (Available from http://www.who.int/dietphysicalactivity/reducingsalt/en/index1.html. Accessed 09/09/2013)

Williams, P. C. (1987). Variables affecting near-infrared reflectance spectroscopic analysis. In P. C. Williams, & K. H. Norris (Eds.), *Near-infrared Technology in the Agricultural and Food Industries* (pp. 143–167). St. Paul: American Association of Cereal Chemists.

Williams, P. C., & Sobering, D. (1996). How do we do it: A brief summary of the methods we use in developing near infrared calibrations. In A.M. C. Davies, & P. C. Williams (Eds.), *Near Infrared Spectroscopy: The Future Waves* (pp. 185–188). Chichester: NIR Publications.