

## Research Article

# Chemical Composition and Antioxidant Properties of Five White Onion (*Allium cepa* L.) Landraces

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Five onion landraces belonging to *Bianca di Pompei* cv., cultivated in Campania region (Italy), were characterized for their main quality parameters. The onion landraces were harvested at the end of the growth cycle corresponding to the ripening time and harvest month, respectively: February, March, April, May, and June. The total content of volatile compounds as well as the sulfur-containing compounds in *Aprilatica* was significantly ( $p \leq 0.05$ ) higher than the other landraces investigated. The nutraceutical feature investigated through the total phenols, phenols profile, and antioxidant activity showed higher values for the samples harvested in spring months. High pungency values ranging from 9 to 14  $\mu\text{mol/g}$  FW were found in all onion landraces investigated as enzymatically (alliinase) produced pyruvate (EPY). The organic acids profile (malic, citric, succinic, pyruvic, oxalic, ascorbic, and tartaric acids) highlighted malic and citric acids in higher amounts in all landraces. Fructose, glucose, and sucrose were found as soluble sugars and fructose was the most abundant. Generally, the results highlighted the growth temperature influence on the investigated quality parameters.

## 1. Introduction

Onion (*Allium cepa* L.) is the most widely cultivated species of the genus *Allium* [1].

The plant portion commonly used is the bulb, which is utilized as a food ingredient to give flavour and aroma to a great variety of dishes.

Onions are an important source of several phytonutrients as flavonoids, fructooligosaccharides (FOS), and thiosulfonates and other sulfur compounds, recognized as important elements of the Mediterranean diet [2].

In fact, onions contain high levels of phenolic compounds, which have antioxidant properties besides beneficial effects against different degenerative pathologies (cardiovascular and neurological diseases, dysfunctions based on oxidative stress) [3].

Flavonoids are the major phenolics in onions, which can be classified to different subclasses (flavones, flavanones,

flavonols, isoflavones, flavanonols, flavanols, chalcones, and anthocyanins) on the basis of the degree of unsaturation and the degree of oxidation of the central ring. Flavonoids subclasses can be further differentiated on the basis of the number and nature of substituent groups attached to the rings [4].

Flavonols are the most abundant in onions, present as their glycosides, that is, quercetin and kaempferol [5, 6], in higher concentration (280–400 mg/kg) than other vegetables (i.e., 100 mg/kg in broccoli, 50 mg/kg in apple) [7]. Anthocyanins, belonging to anthocyanidins, are mainly present in red onions (250 mg/kg), besides having a composition rich in flavonols as yellow onions [7].

FOS represent another source of phytochemicals in onions bulbs. They are mainly inulin, kestose, nystose, and fructofuranosylnystose. The health benefits of these carbohydrates have been widely reported in the past years due to their prebiotic effect [8].

In onions, sulfur compounds are responsible for typical odour and flavour and are also active antimicrobial agents [9]; hence, onions may be used as natural preservatives to control microbial growth [10]. Furthermore, they have also protective effects against cardiovascular diseases.

The precursors of sulfur-containing compounds are S-alk(en)yl-L-cysteine sulfoxides (ACSOs, i.e., methiin, propiin, and isoalliin) which are hydrolysed by means of alliinase enzyme into pyruvate, ammonia, and a mixture of both volatile and nonvolatile sulfur compounds [11], after the breakage of the tissue caused by cutting, mastication, and cooking.

The concentration of pyruvate produced by alliinase activity allows assessing the pungency of onions [12, 13]. The major flavour compounds are generated by spontaneous reactions of the sulfenic acids. These latter undergo rearrangement to form a mixture of sulfur-containing compounds (S-compounds) including thiosulfinates, thiosulfonates, and mono-, di-, and trisulfides as well as specific compounds such as thiopropanal S-oxide, the lachrymatory or tear factor, all responsible for the typical flavour of onions [3].

The bioaccumulation of organosulfur compounds in onions depends on different factors but especially on the sulfur-based fertilization, the environment, and the genotype of the cultivars [14–16]. Also, other compounds such as organic acids and sugars can contribute to the sensory profile of onions. Hence, organic acids influence the acidity and pH of the onion juice in a larger or smaller degree; the soluble sugars influence the sweetness of onions and hence the acceptability of this vegetable by consumers. In fact, there is increasing interest in the role that some nonstructural carbohydrates play in the taste preference [17].

Onion (*Allium cepa* L.) is an *Allium* vegetable widely cultivated in Campania region (South Italy), in particular in the two contiguous areas of Nocerino-Sarnese and Stabiese-Vesuvius plains, where *Bianca di Pompei* cultivar is mostly present. This cultivar is actually composed of a set of locally named landraces, all sharing common shapes and colours (white-greenish flattened bulbs). The landraces differ mainly in the bulb harvest time that is a function of the end of the growth cycle and so of the ripening time; the latter ranges from February to June. In this way, the farmer can supply a fresh product for a long time on the market, avoiding problems and costs due to the preservation [18]. The aim of this paper was to characterize different onion landraces belonging to *Bianca di Pompei* cv in terms of main quality parameters for this crop (volatile compounds, organic acids, sugars, polyphenols, antioxidant activity, and pungency).

## 2. Materials and Methods

**2.1. Onion Samples.** Raw onions (*Allium cepa*, *Bianca di Pompei* cv) were supplied by factories located in the Salerno area (Campania region, Italy). White onion landraces were harvested at the end of the growth cycle and classified according to the harvest month: *Febbraese*, *Marzatica*, *Aprilatica*, *Maggiola*, and *Giugnese* reaped in February, March, April, May, and June, respectively. The production of the five white onion landraces was carried out using the conventional cultivation methods for this crop. The bulbs were stored

in the dark at 7°C for a maximum of 5 days before analysis.

**2.2. Chemicals.** Analytical grade chemicals, methanol, dichloromethane, trichloroacetic acid, acetic acid, acetonitrile, sodium hydroxide, Folin-Ciocalteu reagent, gallic acid, 2,2-diphenyl-1-picrylhydrazyl (DPPH), dinitrophenyl hydrazine (DNPH), quercetin, kaempferol, 2-octanol, sodium sulfate, and ferulic and chlorogenic acids, were purchased from Sigma-Aldrich (St. Louis, MO, USA).

**2.3. Extraction of Volatile Compounds.** One hundred grams of edible part of onion samples was homogenized in an Ultra-Turrax blender (T25, IKA Werke, Staufen, Germany) at room temperature. The slurry was transferred into the flask with 300 mL of distilled water and 1  $\mu$ L of 2-octanol, as internal standard, and sonicated at 50 Hz for 30 minutes. Ice was added to the ultrasonic bath (Sonica 22000 MH, Soltec, Italy) to avoid matrix heating phenomena. The slurry was subjected to steam distillation by a vertical steam distillation unit. The flask containing the homogenized onion was heated for 3 h and the condensed vapour was extracted with fresh dichloromethane 3 times by separatory funnel. The organic phases were collected, dehydrated with anhydrous Na<sub>2</sub>SO<sub>4</sub>, filtered through Whatman filter paper, and finally concentrated using a Kuderna-Danish device.

**2.4. GC-MS Analysis.** The volatile compounds were determined by GC-MS (Trace MS plus, Thermo Finnigan, USA) and by GC-FID (HP 6890, Agilent), both equipped with a capillary column (SUPELCO WAX 10; 60 m, 0.25 mm, and 0.25  $\mu$ m, Supelco, USA). Chromatographic separation was performed on 2  $\mu$ L of sample, using helium as carrying gas at constant flow of 1 mL/min. The temperature program was as follows: 3 min at 40°C, first ramp at 2°C/min to 150°C, second ramp at 4°C/min to 220°C, and 10 min at 220°C. The GC injector was held at 40°C for 3 min, and the column temperature program increased up to 150°C at 2°C/min, from 150°C to 220°C at 4°C/min (held for 10 min). Detector and transfer line were held at 250°C. The identification of volatile compounds was carried out by injection of commercial standards (Sigma-Aldrich, Milano, Italy), by spectra comparison using the NIST and Wiley libraries, and by comparison of their retention indices to reference data from the literature. The quantitative analyses were done assuming the response factors equal to the 2-octanol, used as internal standard [19].

**2.5. Polyphenols.** 150 mL of methanol was added to 50 g of fresh onion tissue. The resulting mixture was homogenized and stirred for 30 minutes; homogenates were held for 15 min in an ultrasonic bath (Fungilab Ultrasound, Barcelona, Spain) and the extract was separated from the residue by centrifugation at 1900  $\times$ g. Extraction was repeated increasing the stirring time to 60 min and to 90 min. The combined methanol : water extracts were filtered through a Whatman No. 2 filter paper and were evaporated at 40°C (rotary evaporator IKA RV-8, Staufen, Germany) to remove methanol. The extracts

were redissolved in 10 mL of methanol [5] and were used for the determination of polyphenols and antioxidant activity.

Total phenols were estimated by Folin-Ciocalteu colourimetric assay [20] and results were expressed as mg equivalent gallic acid (GAE)/g dry weight. The absorbance of solutions was read at 765 nm using a UV-Vis spectrophotometer (Lambda Bio 40; PerkinElmer, Waltham, MA, USA), after 2 hours of incubation in the dark.

The quali-quantitative profile was determined by HPLC (1100, Agilent, Waldbronn, Germany) according to Cinquanta et al. (2015). The polyphenol extracts were filtered through 0.45  $\mu\text{m}$  filter syringe and directly injected into Agilent/HPI100 system (CA, USA). The phenolic compounds were separated on Supelco Ascentis RP-Amide Columns  $C_{18}$  (150  $\times$  4.6 mm; 5  $\mu\text{m}$ ) at a flow rate of 1.2 mL/min. The mobile phase used was (A) water/acetic acid (99:1, v/v) and (B) acetonitrile/acetic acid (99:1 v/v), with a gradient as follows: 0 min, 100% A; 6.5 min, 85% A and 15% B; 8.0 min, 80% A and 20% B; 12 min, 75% A and 25% B; 16 min, 70% A and 30% B; 25 min, 60% A and 40% B; 40 min, 60% A and 40% B. The eluates were detected at 280 and 350 nm. The concentration of identified phenols was calculated with external standards method.

**2.6. Antioxidant Activity.** Antioxidant activity was measured by 1,1-diphenyl-2-picrylhydrazyl (DPPH) solution ( $6 \cdot 10^{-5}$  M in methanol) and the free radical scavenging activity was expressed as the  $EC_{50}$  value: the volume ( $\mu\text{L}$ ) required to reduce 50% of the initial DPPH radical activity. Total antioxidant activity of pomegranate juice was determined by the 1,1-diphenyl-2-picrylhydrazyl (DPPH) method [22]. Onion extracts in different concentrations were mixed with  $6 \cdot 10^{-5}$  M methanol solution of DPPH radical. The mixture was shaken vigorously and left to stand for 30 min in the dark at room temperature. After the reaction was allowed to take place in the dark for 30 min, the absorbance at 517 nm was recorded to determine the concentration of remaining DPPH.

The antioxidant activity was expressed according to Albanese et al. [23] as percentage inhibition of DPPH and then calculated according to the following equation:

$$\% \text{ inhibition of DPPH} = \left[ \frac{(A_{C(0)} - A_{S(t)})}{A_{C(0)}} \right] * 100, \quad (1)$$

where  $A_{C(0)}$  is the absorbance of the control at  $t = 0$  min and  $A_{S(t)}$  is the absorbance of sample at  $t = 30$  min. The free radical scavenging activity determined by DPPH was expressed as the  $EC_{50}$  value: mg of the extract per mL required to reduce 50% of the initial DPPH radical activity.

**2.7. Pungency Analysis.** Pungency of onions was determined as enzymatically (alliinase) produced pyruvate (EPY) by colourimetric analysis according to Schwimmer and Weston (1961) with slight modifications. Onion landraces were sliced in half longitudinally: 50 g was homogenized by Ultra-Turrax blender (T25, IKA Werke, Staufen, Germany) with 50 mL of distilled water for the determination of total pyruvate alliinase produced, whereas 50 g of onions was pretreated with 50 mL of 5% trichloroacetic acid solution to inactivate

the alliinase in order to quantify pyruvate basal level. Both mixtures were left at room temperature for 15 min and filtered with Whatman filter paper (grade 1) and 10 mL of the filtrate was diluted ten times with bidistilled water. One millilitre of sample was placed in a reaction tube with 1 mL of 2,4-dinitrophenyl hydrazine (DNPH) solution (0.0125% DNPH in 2 M HCl) and 1 mL of bidistilled water. Reaction tube was vortexed and placed in a water bath at 37°C for 10 minutes. After the incubation time, 5 mL of 0.6 M NaOH was added to the tube and allowed to stand for 5 min. The DNP hydrazine derivative of pyruvate was measured using PerkinElmer Lambda 25 UV-Vis spectrometer at 420 nm. Enzymatically (alliinase) produced pyruvate (EPY) in each sample was calculated from the difference of total and basal concentration of pyruvate. A blank sample was prepared with 2 mL of water and 1 mL of DNPH; standards were prepared replacing onion sample with 1 mL of sodium pyruvate solution, ranging from 20 to 100  $\mu\text{M}$ .

**2.8. Sugars Analysis.** Sugars were determined by HPLC (Hewlett Packard, mod. 79852, USA) [21]. The HPLC system was equipped with 4.6  $\times$  250 mm (60  $\text{\AA}$ , 4  $\mu\text{m}$ ) carbohydrate-cartridge column (Waters, USA). The mobile phase was an acetonitrile-water solution (75:25), with a flow rate of 1.2 mL/min and a column temperature of 60°C. Peaks were detected by a refractive index detector (Hewlett Packard, mod. 100, USA) and concentrations were calculated with external standards method.

**2.9. Organic Acids Analysis.** 1 g of fresh onion was added to distilled water up to 10 mL and homogenized in an Ultra-Turrax blender (T25, IKA Werke, Staufen, Germany) for 2 min. The samples were centrifuged at 4000 rpm for 10 min and filtered through 0.45  $\mu\text{m}$  syringe cellulose filter (Millipore, USA) before ion exchange chromatography analysis. The apparatus (Dionex Corp., USA) was equipped with an ED 500 electrochemical detector, Ionpac AS11 column (250  $\times$  4 mm), and Ionpac AS11 Guard (50  $\times$  4 mm). The elution phase at 0.5 mL/min was bidistilled water (E1) and NaOH 100 mM (E2) for a total running time of 25 min, using the following gradient: from 93% E1 at time 0 to 65% E1 at 20 min and then to 93% E1 in 5 min [24]. The organic acids were identified by the overlapping of their retention time with those of commercial standard acids prepared from 1 g/L stock solution and diluted to the required concentration before use. A calibration curve of organic acid standards was obtained and used for quantitative analysis. Acquisition and integration of chromatograms were performed with Peaknet G4GIT0 (Dionex Corp.) software.

**2.10. Statistical Analysis.** Analysis of parameters investigated was carried out on five different samples belonging to each onion group. Results were reported as the mean and standard deviation. The analysis of variance (ANOVA) was applied to the data. The least significant differences were obtained using an LSD test ( $p \leq 0.05$ ). Statistical analysis was performed using SPSS version 13.0 for Windows (SPSS, Inc., Chicago, IL, USA).



### 3. Results and Discussion

A total of 22 compounds (Table 1) were identified in the volatile fraction of onion samples belonging to the following chemical classes: sulfur-containing compounds (S-compounds), aldehydes, and ketones.

S-compounds were the main volatile compounds of the onion profile [11] which in our study ranged from a minimum of 207.87 mg/Kg in *Febbrarese* landrace to a maximum of 1459.40 mg/Kg in *Aprilatica* one, corresponding to 63% and 74% of total volatile compounds, respectively. Nine out of fifteen S-compounds were di- and trisulfides and were the most numerous. These results were in agreement with Lanzotti [25] who proved that di- and trisulfides were the main compounds in the volatile fraction of the onion isolated by steam distillation. Furthermore, the volatile S-compounds of *Bianca di Pompei* onion landraces were the typical molecules of the volatile fraction characterizing the onion's pungent flavour [26–29]. Other compounds, aldehydes and ketones, were found in a smaller amount ranging between 17–21% and 3–7% of the total volatile compounds, respectively, in landrace samples.

The overall content of volatile compounds seems to be influenced by growth cycle and harvest month which is a function of temperature. Metabolite compositions in onions are strongly affected by climate conditions, in particular by air temperature [30]. In fact, *Febbrarese* and *Marzatica* landraces grown and harvested in colder winter months showed lower concentrations of total volatile compounds than those found in *Aprilatica*, *Maggiaiola*, and *Giugnese*, which were characterized by a milder crop temperature. These three onion landraces showed a concentration of volatile compounds almost five times higher than that in *Febbrarese* and *Marzatica*. Likewise, aldehydes, ketones, and S-compounds increased from *Febbrarese* to *Giugnese*; in particular, *Giugnese* showed the highest amounts equal to 395.28 and 133.46 mg/kg for aldehydes and ketones, respectively, while S-compounds content was maximum in *Aprilatica* (1459.40 mg/kg).

For all the investigated onion landraces, quantitative analysis showed that di- and trisulfides, such as *cis*- and *trans*-methyl-1-propenyl disulfide, methyl-2-propenyl disulfide, dipropyl disulfide, *cis*- and *trans*-propenyl propyl disulfide, methyl propyl trisulfide, and dipropyl trisulfide, were the largest part, whose sum represents about 60% of S-compounds.

Di- and trisulfides increased in landraces harvested in spring months (*Aprilatica*, *Maggiaiola*, and *Giugnese*) in comparison to those detected in *Febbrarese* and *Marzatica* landraces harvested in colder winter months.

According to Lanzotti (2006), di- and trisulfides were formed by the degradation of thiosulfinates whose biosynthetic pathway came from the condensation reactions of alk(en)yl-sulfenic acids (e.g., Z,E-propanethial S-oxide or lachrymator factor). The amount of alk(en)yl-sulfenic acids (the lachrymator factor) was closely linked to the concentration of the aroma precursors: S-alkenyl cysteine sulfoxides (ACSOs). The higher content of di- and trisulfides than other volatile compounds in onion samples cultivated during warmer spring months was probably due to an increase of

S-alkenyl cysteine sulfoxides (ACSOs), according to Randle and Coolong [31] who proved that growth temperature affected ACSOs concentration of *Granex 33* onion. In particular, they recorded that ACSO concentrations from bulbs grown at 15.6°C were roughly a third of those grown at 32.2°C. Among di- and trisulfide compounds, methyl propyl trisulfide was the most abundant compound for all onion samples investigated, followed by *trans*-methyl-1-propenyl disulfide. A similar result has been documented by Arnault et al. [32] who reported that S-compounds such as thiosulfonates, propyl-containing disulfides and trisulfides, propenyl-containing disulfides and trisulfides, and thiophene derivatives mainly contributed to the volatile compounds of onion.

About the aldehydes class, four compounds were detected in the volatile pattern of onion samples except for 2-methyl-2-pentenal which was not detected in *Febbrarese* landrace. Aldehydes content was different ( $p \leq 0.05$ ) for the five onion landraces investigated and it increased in landraces harvested from February (*Febbrarese*) to June (*Giugnese*). Among the aldehydes, furfuraldehyde was the most abundant in all samples, and its highest content ( $p \leq 0.05$ ) was found in *Aprilatica* landrace. Propionaldehyde and 2-methyl-2-pentenal contents were different ( $p \leq 0.05$ ) in landraces samples: the highest values were in *Aprilatica* landrace, followed by *Maggiaiola* and *Giugnese*. These aldehydes, together with di- and trisulfide compounds, arose from the lachrymator factor [28, 33] as well as the pungency and the overall flavour of onion.

The other volatile compounds were two ketones and furfuryl alcohol (Table 1). The 1,2-cyclopentanedione concentration differed at harvest time: in spring months, *Aprilatica*, *Maggiaiola*, and *Giugnese* landraces had a higher content ( $p \leq 0.05$ ) than those yielded in winter (*Febbrarese* and *Marzatica*). The butyrolactone compound was detected only in landraces harvested in spring months (*Aprilatica*, *Maggiaiola*, and *Giugnese*).

Total phenols, quantified in the onion samples, were reported in Table 2. The concentration ranged from a minimum of 4.75 in *Febbrarese* to a maximum of 5.31 in *Giugnese*. The results were in accordance with Santas et al. (2008) and Prakash et al. (2007) who evaluated white Spanish and red, violet, and green onion varieties, respectively.

The onion landraces were also studied for their specific phenols composition (Figure 1 and Table 2). For all landraces, the most abundant phenol was gallic acid, whose quantity changes from 55.66 to 64.90  $\mu\text{g/g dw}$  in *Febbrarese* and *Giugnese*, respectively. Among identified phenols, quercetin has an important role from a nutritional point of view. Quercetin is the aglycone form of several other flavonoid glycosides such as rutin and quercitrin that are found in citrus fruits, buckwheat, and onions [35]. Functional benefits of quercetin include anti-inflammatory activity, antihistamine effect, allergy medication, and anticancer and antiviral activities. It has also been claimed to regulate blood pressure in hypertensive subjects [36].

In our samples, this flavonol was found to be between 6.98 and 8.14  $\mu\text{g/g}$ ; higher amounts were detected by Prakash et al. (2007) who studied quercetin in four (red, white, violet, and green) onion varieties.

TABLE 1: Volatile compounds (mg/kg dw; mean  $\pm$  SD) detected in *Bianca di Pompei* onion landraces.

Number	RT	Volatile compound	Febrarese	Marzatica	Aprilatica	Maggiola	Giugnese
<i>Aldehydes</i>							
1	6.7	Propionaldehyde	5.30 $\pm$ 0.10 <sup>a</sup>	12.65 $\pm$ 0.26 <sup>b</sup>	72.39 $\pm$ 1.51 <sup>c</sup>	48.37 $\pm$ 1.01 <sup>d</sup>	132.51 $\pm$ 2.77 <sup>e</sup>
2	23.4	2-Methyl-2-pentenal	n.d.	3.79 $\pm$ 0.08 <sup>a</sup>	49.65 $\pm$ 1.04 <sup>b</sup>	65.43 $\pm$ 1.37 <sup>c</sup>	38.82 $\pm$ 0.81 <sup>d</sup>
3	44.1	Furfuraldehyde	39.38 $\pm$ 1.36 <sup>a</sup>	82.25 $\pm$ 2.84 <sup>b</sup>	166.21 $\pm$ 5.73 <sup>c</sup>	97.26 $\pm$ 3.35 <sup>b</sup>	135.11 $\pm$ 4.66 <sup>d</sup>
4	49.9	5-Methyl-2-furfuraldehyde	35.68 $\pm$ 0.87 <sup>a</sup>	13.48 $\pm$ 0.33 <sup>b</sup>	68.76 $\pm$ 1.67 <sup>c</sup>	105.06 $\pm$ 2.56 <sup>d</sup>	88.84 $\pm$ 2.16 <sup>e</sup>
		<i>Total</i>	80.36 $\pm$ 1.67 <sup>a</sup>	112.16 $\pm$ 2.33 <sup>b</sup>	357.02 $\pm$ 7.42 <sup>c</sup>	316.16 $\pm$ 6.58 <sup>d</sup>	395.28 $\pm$ 8.23 <sup>e</sup>
<i>Sulfur-containing compounds</i>							
5	7.3	1-Propanethiol	n.d.	5.42 $\pm$ 0.11 <sup>a</sup>	64.04 $\pm$ 1.34 <sup>b</sup>	127.95 $\pm$ 2.67 <sup>c</sup>	94.69 $\pm$ 1.98 <sup>d</sup>
6	9.7	Propylene sulfide	26.01 $\pm$ 0.84 <sup>a</sup>	27.83 $\pm$ 1.49 <sup>a</sup>	59.83 $\pm$ 1.94 <sup>c</sup>	29.02 $\pm$ 0.94 <sup>a</sup>	52.08 $\pm$ 1.69 <sup>d</sup>
7	17.1	Dimethyl sulfide	n.d.	22.35 $\pm$ 0.47 <sup>a</sup>	18.00 $\pm$ 0.38 <sup>b</sup>	53.87 $\pm$ 1.13 <sup>c</sup>	16.88 $\pm$ 0.35 <sup>b</sup>
8	25.8	Methyl propyl disulfide	10.04 $\pm$ 0.27 <sup>a</sup>	25.51 $\pm$ 0.69 <sup>b</sup>	5.21 $\pm$ 0.14 <sup>c</sup>	84.34 $\pm$ 2.28 <sup>d</sup>	11.59 $\pm$ 0.31 <sup>a</sup>
9	29.3	<i>cis</i> -Methyl-1-propenyl disulfide	3.67 $\pm$ 0.15 <sup>a</sup>	4.01 $\pm$ 0.17 <sup>a</sup>	23.95 $\pm$ 0.99 <sup>b</sup>	99.70 $\pm$ 4.11 <sup>c</sup>	54.23 $\pm$ 2.23 <sup>d</sup>
10	29.5	5-Methyl-1,3-thiazole	n.d.	n.d.	21.76 $\pm$ 0.45 <sup>a</sup>	20.02 $\pm$ 0.42 <sup>a</sup>	17.75 $\pm$ 0.37 <sup>a</sup>
11	31.9	<i>trans</i> -Methyl-1-propenyl disulfide	34.98 $\pm$ 0.72 <sup>ac</sup>	30.96 $\pm$ 0.67 <sup>a</sup>	232.11 $\pm$ 3.47 <sup>b</sup>	138.67 $\pm$ 0.87 <sup>c</sup>	181.23 $\pm$ 2.76 <sup>d</sup>
12	32.9	3,4-Dimethyl thiophene	n.d.	n.d.	184.18 $\pm$ 3.85 <sup>a</sup>	40.46 $\pm$ 2.90 <sup>b</sup>	127.86 $\pm$ 3.79 <sup>c</sup>
13	34.3	Methyl-2-propenyl disulfide	4.16 $\pm$ 0.17 <sup>a</sup>	7.97 $\pm$ 0.33 <sup>a</sup>	7.62 $\pm$ 0.32 <sup>a</sup>	16.44 $\pm$ 0.69 <sup>b</sup>	102.86 $\pm$ 4.31 <sup>c</sup>
14	37.5	Dipropyl disulfide	33.42 $\pm$ 0.78 <sup>a</sup>	22.92 $\pm$ 0.51 <sup>b</sup>	40.23 $\pm$ 0.90 <sup>c</sup>	40.46 $\pm$ 0.90 <sup>c</sup>	65.46 $\pm$ 1.46 <sup>d</sup>
15	39.8	1,2,4-Trithiolane	3.51 $\pm$ 0.09 <sup>a</sup>	4.87 $\pm$ 0.13 <sup>a</sup>	160.66 $\pm$ 6.12 <sup>b</sup>	116.808 $\pm$ 3.1 <sup>c</sup>	54.74 $\pm$ 1.44 <sup>d</sup>
16	41.3	<i>trans</i> -Propenyl propyl disulfide	16.63 $\pm$ 0.67 <sup>a</sup>	30.31 $\pm$ 1.23 <sup>b</sup>	94.20 $\pm$ 3.82 <sup>c</sup>	79.91 $\pm$ 3.24 <sup>d</sup>	58.25 $\pm$ 2.36 <sup>e</sup>
17	42.9	<i>cis</i> -Propenyl propyl disulfide	n.d.	n.d.	112.60 $\pm$ 2.35 <sup>a</sup>	21.32 $\pm$ 0.45 <sup>b</sup>	33.42 $\pm$ 0.70 <sup>b</sup>
18	45.6	Methyl propyl trisulfide	38.86 $\pm$ 1.01 <sup>a</sup>	46.03 $\pm$ 0.77 <sup>a</sup>	276.67 $\pm$ 7.67 <sup>b</sup>	379.52 $\pm$ 10.52 <sup>c</sup>	226.48 $\pm$ 6.28 <sup>d</sup>
19	55.9	Dipropyl trisulfide	36.58 $\pm$ 0.89 <sup>a</sup>	37.54 $\pm$ 0.86 <sup>a</sup>	158.34 $\pm$ 3.63 <sup>b</sup>	37.01 $\pm$ 0.85 <sup>a</sup>	96.68 $\pm$ 2.22 <sup>c</sup>
		<i>Total</i>	207.87 $\pm$ 4.61 <sup>a</sup>	265.72 $\pm$ 5.89 <sup>a</sup>	1459.40 $\pm$ 32.42 <sup>b</sup>	1285.48 $\pm$ 28.53 <sup>c</sup>	1194.19 $\pm$ 26.51 <sup>c</sup>
<i>Ketones</i>							
20	51.1	1,2-Cyclopentanedione	12.15 $\pm$ 0.34 <sup>a</sup>	12.90 $\pm$ 0.37 <sup>a</sup>	59.43 $\pm$ 1.69 <sup>b</sup>	72.06 $\pm$ 2.05 <sup>c</sup>	69.21 $\pm$ 1.97 <sup>c</sup>
21	53.7	Butyrolactone	n.d.	n.d.	47.56 $\pm$ 0.99 <sup>a</sup>	59.98 $\pm$ 1.25 <sup>a</sup>	64.25 $\pm$ 1.34 <sup>a</sup>
		<i>Total</i>	12.15 $\pm$ 0.34 <sup>a</sup>	12.90 $\pm$ 0.37 <sup>a</sup>	106.99 $\pm$ 4.09 <sup>b</sup>	132.04 $\pm$ 5.04 <sup>c</sup>	133.46 $\pm$ 5.10 <sup>c</sup>
<i>Other</i>							
22	54.5	Furfuryl alcohol	34.90 $\pm$ 0.73 <sup>a</sup>	18.34 $\pm$ 0.38 <sup>b</sup>	43.97 $\pm$ 0.92 <sup>c</sup>	64.81 $\pm$ 1.35 <sup>d</sup>	156.85 $\pm$ 3.28 <sup>e</sup>
		<i>Total volatile compounds</i>	335.20 $\pm$ 9.77 <sup>a</sup>	409.08 $\pm$ 11.90 <sup>a</sup>	1967.39 $\pm$ 43.70 <sup>b</sup>	1798.45 $\pm$ 39.93 <sup>c</sup>	1874.48 $\pm$ 41.75 <sup>ac</sup>

Different letters (a, b, c, etc.) correspond to significant differences ( $p \leq 0.05$ ) among onion landraces.

TABLE 2: Phenols (mg/g dw; mean  $\pm$  SD), antioxidant activity (EC<sub>50</sub> mg extract/mL; mean  $\pm$  SD), and total phenols (mg GAE/g dw) of *Bianca di Pompei* onion landraces.

	<i>Febbrarese</i>	<i>Marzatica</i>	<i>Aprilatica</i>	<i>Maggiaiola</i>	<i>Giugnese</i>
<i>Phenols (mg/g dw)</i>					
Gallic acid	55.66 $\pm$ 2.30 <sup>a</sup>	59.56 $\pm$ 1.10 <sup>ab</sup>	61.23 $\pm$ 2.50 <sup>b</sup>	61.94 $\pm$ 1.91 <sup>b</sup>	64.90 $\pm$ 1.22 <sup>b</sup>
Ferulic acid	1.52 $\pm$ 0.20 <sup>a</sup>	1.62 $\pm$ 0.25 <sup>a</sup>	1.67 $\pm$ 0.41 <sup>a</sup>	1.69 $\pm$ 0.19 <sup>a</sup>	1.77 $\pm$ 0.30 <sup>a</sup>
Quercetin	6.98 $\pm$ 0.42 <sup>a</sup>	7.47 $\pm$ 0.30 <sup>b</sup>	7.68 $\pm$ 0.28 <sup>b</sup>	7.77 $\pm$ 0.30 <sup>b</sup>	8.14 $\pm$ 0.20 <sup>b</sup>
Kaempferol	1.62 $\pm$ 0.33 <sup>a</sup>	1.73 $\pm$ 0.27 <sup>a</sup>	1.78 $\pm$ 0.15 <sup>a</sup>	1.80 $\pm$ 0.21 <sup>a</sup>	1.89 $\pm$ 0.32 <sup>a</sup>
Chlorogenic acid	0.84 $\pm$ 0.06 <sup>a</sup>	0.90 $\pm$ 0.02 <sup>a</sup>	0.92 $\pm$ 0.08 <sup>a</sup>	0.93 $\pm$ 0.04 <sup>a</sup>	0.98 $\pm$ 0.07 <sup>a</sup>
<i>Antioxidant activity</i>					
EC <sub>50</sub> (mg extract/mL)	18.80 $\pm$ 1.0 <sup>a</sup>	18.50 $\pm$ 0.50 <sup>a</sup>	20.90 $\pm$ 0.60 <sup>b</sup>	20.25 $\pm$ 0.40 <sup>b</sup>	21.27 $\pm$ 0.8 <sup>b</sup>
<i>Total phenols (mg GAE/g dw)</i>	4.75 $\pm$ 0.24 <sup>a</sup>	4.90 $\pm$ 0.10 <sup>a</sup>	5.14 $\pm$ 0.35 <sup>a</sup>	5.06 $\pm$ 0.28 <sup>a</sup>	5.31 $\pm$ 0.30 <sup>a</sup>

Different letters (a, b) correspond to significant differences ( $p \leq 0.05$ ) among onion landraces.

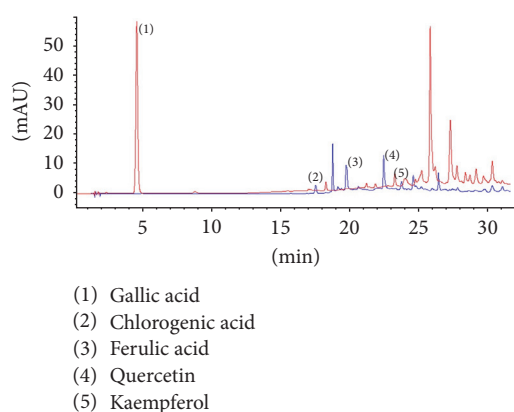


FIGURE 1: Chromatograms of phenols in onion samples at 280 and 350 nm (red and blue lines, resp.).

All the other phenols increased in landraces harvested in spring months. Our results highlighted the notion that the quantity of phenolic pool may change not only with the cultivar as reported in the literature [6, 34] but also with the growth stage and environmental conditions.

The antioxidant activity varied from 19.00 to 21.27 mg of extract/mL (Table 2). Our results were in agreement with previous EC<sub>50</sub> data on white onions [6] and consistent with quali-quantitative phenols previously described. In fact, the highest values were found for *Aprilatica*, *Maggiaiola*, and *Giugnese* without significant differences ( $p \leq 0.05$ ) among them.

It is generally accepted that there is a high correlation between levels of enzymatically produced pyruvate (EPY) present in onions and the perception of pungency [12]. The investigation of this parameter was important to estimate the potential flavour and to define the aroma characteristics of various onion cvs. Classification of onions according to pungency was proposed as follows: low, 0–3  $\mu$ mol of pyruvic acid/g; moderate, 3–7  $\mu$ mol of pyruvic acid/g; and high, higher than 7  $\mu$ mol of pyruvic acid/g [37].

Herein, all onion landraces had high pungency values expressed by EPY amount, ranging from 9 to 14  $\mu$ mol/g FW.

Significant differences ( $p \leq 0.05$ ) were found among the five landraces with the highest pungency level in *Aprilatica* landrace.

In order to define the sensory profile of onion, the sugars content was investigated. Three soluble sugars, sucrose, glucose, and fructose, were detected in the five onion landraces (Table 3). Other water soluble sugars (oligosaccharides of fructose, named fructans) were found by Davis et al. (2007) in the characterization of different onion cvs cultivated in the United Kingdom.

The total content of soluble sugars in *Bianca di Pompei* landraces changed from 3 to 5 g/100 g FW according to the ranges reported in the literature for other onion cultivars [38–42]. No statistical difference ( $p \leq 0.05$ ) was found among the onion landraces as regards fructose, while significant differences ( $p \leq 0.05$ ) were found for glucose, sucrose, and the total sugar content. Anyway, these differences do not influence the sweetness of the onion because the pungency values are higher than 4  $\mu$ mol/g FW, a medium-low level that would allow appreciating these differences during taste, as reported by Crowther et al. [43].

Seven organic acids were identified and quantified in onion samples (Table 3). The results were in accordance with those found in *Recas* cv onions [40], in four onion Greek cvs [39], and in six onion Spanish cvs [41] with fumaric and glutamic acids too. The main acids that contribute mostly to edible flesh acidity are malic (78.94–57.61 mg/100 g fw) and citric (63.32–19.70 mg/100 g fw) acids. Similar results were reported by Caruso et al. [44].

Malic acid was the most abundant organic acid for all onion samples except for *Febbrarese* landrace, where citric acid was the abundant organic acid. Significant differences ( $p \leq 0.05$ ) were found among all landraces for organic acids amount, probably due to a different activation level of multiple metabolic pathways, among which the Cycle of Krebs is the main one. Regarding ascorbic acid, it contributes to the nutritional value of the onion with respect to the other acids; the highest amount was found in *Aprilatica* landraces, followed by *Maggiaiola* and *Giugnese*, suggesting that the samples harvested in spring months had higher vit. C than those harvested in winter. Furthermore, ascorbic acid concentration in *Aprilatica*, *Maggiaiola*, and *Giugnese*

TABLE 3: Organic acids (mg/100 g fw; mean  $\pm$  SD), sugars (g/100 g fw; mean  $\pm$  SD) content, and pungency ( $\mu$ mol/g fresh weight) of *Bianca di Pompei* onion landraces.

	<i>Febbrarese</i>	<i>Marzatica</i>	<i>Aprilatica</i>	<i>Maggiaiola</i>	<i>Giugnese</i>
<i>Organic acids</i>					
Malic acid	60.43 $\pm$ 6.53 <sup>a</sup>	66.33 $\pm$ 6.61 <sup>a</sup>	61.85 $\pm$ 4.50 <sup>a</sup>	57.61 $\pm$ 3.34 <sup>a</sup>	78.94 $\pm$ 2.10 <sup>b</sup>
Citric acid	63.32 $\pm$ 5.90 <sup>a</sup>	57.05 $\pm$ 3.24 <sup>a</sup>	54.55 $\pm$ 5.43 <sup>a</sup>	34.46 $\pm$ 5.38 <sup>b</sup>	19.70 $\pm$ 3.95 <sup>c</sup>
Tartaric acid	13.93 $\pm$ 2.75 <sup>a</sup>	6.10 $\pm$ 1.21 <sup>b</sup>	16.15 $\pm$ 1.25 <sup>a</sup>	25.88 $\pm$ 4.57 <sup>c</sup>	12.02 $\pm$ 2.85 <sup>a</sup>
Oxalic acid	30.43 $\pm$ 2.97 <sup>a</sup>	19.93 $\pm$ 1.51 <sup>b</sup>	13.60 $\pm$ 1.41 <sup>c</sup>	12.21 $\pm$ 1.90 <sup>c</sup>	10.68 $\pm$ 2.92 <sup>c</sup>
Ascorbic acid	4.63 $\pm$ 0.85 <sup>a</sup>	4.85 $\pm$ 0.15 <sup>a</sup>	21.65 $\pm$ 2.80 <sup>b</sup>	17.69 $\pm$ 1.96 <sup>c</sup>	14.80 $\pm$ 1.73 <sup>c</sup>
Succinic acid	11.55 $\pm$ 1.04 <sup>a</sup>	9.28 $\pm$ 2.72 <sup>a</sup>	19.40 $\pm$ 0.89 <sup>b</sup>	23.35 $\pm$ 2.21 <sup>c</sup>	14.34 $\pm$ 0.82 <sup>ad</sup>
Pyruvic acid	1.07 $\pm$ 0.44 <sup>a</sup>	1.18 $\pm$ 0.66 <sup>a</sup>	0.35 $\pm$ 0.09 <sup>a</sup>	0.76 $\pm$ 0.21 <sup>a</sup>	0.50 $\pm$ 0.10 <sup>a</sup>
<i>Total</i>	185.36 $\pm$ 52.83 <sup>a</sup>	164.72 $\pm$ 30.07 <sup>a</sup>	187.55 $\pm$ 55.79 <sup>a</sup>	171.96 $\pm$ 52.16 <sup>a</sup>	150.98 $\pm$ 21.79 <sup>a</sup>
<i>Pungency (EPY)</i>	9.31 $\pm$ 0.97 <sup>a</sup>	10.01 $\pm$ 0.31 <sup>a</sup>	14.34 $\pm$ 0.13 <sup>b</sup>	13.00 $\pm$ 0.84 <sup>c</sup>	12.53 $\pm$ 0.27 <sup>c</sup>
<i>Soluble sugars</i>					
Fructose	1.74 $\pm$ 0.42 <sup>a</sup>	1.66 $\pm$ 0.62 <sup>a</sup>	2.11 $\pm$ 0.23 <sup>a</sup>	2.00 $\pm$ 0.60 <sup>a</sup>	2.26 $\pm$ 0.70 <sup>a</sup>
Glucose	1.09 $\pm$ 0.18 <sup>a</sup>	0.88 $\pm$ 0.43 <sup>a</sup>	1.97 $\pm$ 0.09 <sup>b</sup>	1.79 $\pm$ 0.54 <sup>b</sup>	2.00 $\pm$ 0.27 <sup>b</sup>
Sucrose	0.90 $\pm$ 0.11 <sup>a</sup>	0.81 $\pm$ 0.21 <sup>a</sup>	1.76 $\pm$ 0.36 <sup>b</sup>	1.04 $\pm$ 0.27 <sup>ac</sup>	1.21 $\pm$ 0.24 <sup>c</sup>
<i>Total</i>	3.73 $\pm$ 0.64 <sup>ac</sup>	3.35 $\pm$ 1.16 <sup>a</sup>	5.83 $\pm$ 0.67 <sup>b</sup>	4.84 $\pm$ 1.32 <sup>bc</sup>	5.46 $\pm$ 1.09 <sup>b</sup>

Different letters (a, b, c, etc.) correspond to significant differences ( $p \leq 0.05$ ) among onion landraces.

ranged from 14.80 to 21.65 mg/100 g fw, higher than that reported for other onion cultivars (range: 1.2–6 mg/100 g fw) [41].

#### 4. Conclusions

In our study, we highlighted the differences, of the most important quality parameters, existing among five onion landraces belonging to “*Bianca di Pompei*” cv. In regard to the investigation of onion volatile compounds, the volatile fraction was clearly dominated by sulfur compounds. These compounds seemed to be influenced by harvest month and so from by temperature of growth; in fact, the landraces *Aprilatica*, *Maggiaiola*, and *Giugnese* grown during the milder time period showed higher amount of sulfur compounds than those registered in *Febbrarese* and *Marzatica* cultivated and harvested in winter months. Similarly, total phenols, phenols profile, and antioxidant activity increased in onion landraces harvested in spring months. The pungency of all landraces was found to be high according to pungency classification of the onion. Also, for this parameter, the highest values were found for onion samples grown during mild months. Thus, even if the genetic factors as reported in the literature are predominant for the expression of quality and sensory parameters, this study highlights the notion that also the external factors, such as the period of growth, influence some of the sensory attributes such as aroma and taste.

#### Competing Interests

The authors declare that there are no competing interests regarding the publication of this paper.

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