1	Prunus persica by-products: a source of minerals, phenols and volatile compounds
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3	Samira MAATALLAH <sup>1*</sup> , Samia DABBOU <sup>2*</sup> , Antonella CASTAGNA <sup>3</sup> , Monia GUIZANI <sup>1,2</sup> , Hichem HAJLAOUI <sup>1</sup> ,
4	Anna Maria RANIERI <sup>3,4</sup> , Guido FLAMINI <sup>4,5</sup>
5	
6	<sup>1</sup> Regional Centre of Agricultural Research (CRRA) PB 357, Sidi Bouzid 9100, Tunisia <sup>2</sup> Dentistry Faculty,
7	University of Monastir, Avicenne Street, 5019 Monastir, Tunisia <sup>3</sup> Department of Agriculture, Food and
8	Environment, University of Pisa, via del Borghetto 80, 56124 Pisa, Italy, <sup>4</sup> Interdepartmental Research Center
9	Nutrafood "Nutraceuticals and Food for Health", University of Pisa, via del Borghetto 80, 56124 Pisa, Italy,
10	<sup>5</sup> Dipartimento di Farmacia, University of Pisa, via Bonanno 6, 56126 Pisa, Italy
11	
12	*These two authors contributed equally to this work
13	
14	Correspondence:
15	Dr. Samia DABBOU, <u>e-mail: dabbou_samia@yahoo.fr</u>
16	Dentistry Faculty, University of Monastir, Avicenne Street, 5019 Monastir, Tunisia.
17	<b>Phone</b> : +216 73 46 08 32; <b>Fax</b> : +216 73 46 11 50
18	
19	ORCID numbers
20	Samira MAATALLAH: https://orcid.org/0000-0002-4508-2260
21	Samia DABBOU: <u>https://orcid.org/0000-0001-6463-9717</u>
22	Antonella CASTAGNA: https://orcid.org/0000-0001-6481-4570
23	Monia GUIZANI: <u>https://orcid.org/0000-0001-5580-7328</u>
24	Hichem HAJLAOUI: https://orcid.org/0000-0001-9478-0105
25	Anna Maria RANIERI: https://orcid.org/0000-0002-5939-656X
26	Guido FLAMINI: http://orcid.org/0000-0003-2418-9349
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## 31 Abstract

Large amounts of peach (*Prunus persica*) leaves and stems are by-products deriving from peach tree cultivation and canned industries. This work aimed to evaluate mineral nutrients, phenolic and volatile profile and antioxidant activities from the by-products of five peach cultivars (Early Maycrest, Sweet Cap, O'Henry, Flordastar and Rubirich).

36 Minerals showed significant variations with respect to peach by-product. N showed higher contents in peach leaves 37 among macronutrients, while Mn showed higher contents among micronutrients. Stems had high levels of Ca and 38 traces of micronutrient levels. The HPLC-DAD phenols analysis showed twelve compounds identified 39 (neochlorogenic and chlorogenic acids, catechin and epicatechin, gallic, caffeic, syringic, ferulic and coumaric 40 acids, quercetin-3-rutinoside, quercetin-3-galactoside, cyanidin-3-glucoside, cyanidin-3-galactoside). Flavonols showed the highest values (quercetin-3-galactoside with 70.79 - 232.16 mg 100g<sup>-1</sup> DW, quercetin-3-rutinoside with 41 42 63.88 - 206.37 mg 100g<sup>-1</sup> DW), while the least content was observed for anthocyanins. Cultivar had a significantly 43 (P < 0.05) impact on phenolic compounds. Comparing by-products, stems showed higher levels of phenols. The GC-MS volatile compounds analysis revealed 43 compounds in different percentages and occurrences, depending 44 45 on the cultivar and the by-product. Benzaldehyde was detected as the major volatile leaf component (70-95%), 46 whereas myrcene (18-21%) and terpinolene (18-26%) were found to be the most important compounds in stems. 47 Methanolic extracts of mature leaves were characterized by lower antioxidant capacity. Finally, peach by-products 48 could represent a natural source of minerals, volatiles and phenolic compounds with high antioxidant activities 49 having a great potential use in food products as natural flavouring agents and as nutraceutical supplements and 50 pharmaceutical and cosmetic molecules.

51 Keywords: *Prunus persica*, Agricultural by-products, Mineral nutrients, Phenol profile, Volatile compounds,
 52 Antioxidant activity.

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# 61 **1. Introduction**

Interest for bioactive compounds of natural origin with a high antioxidant capacity has increased considerably in the past two decades, mostly due to their potential for the treatment and prevention of cancer, cardiovascular, chronic and neurodegenerative diseases (Joana Gil-Chávez et al., 2013). Phenolic compounds are ubiquitous in vegetation, constitute an important part of the human diet, pharmaceutical, and cosmetic industry and have aroused much interest due to their antioxidant properties (Kuppusamy et al., 2016) and health-promoting properties (Shashank and Pandey, 2013).

Furthermore, one of the most important and overlooked diet constituents today is represented by minerals (Kuppusamy et al., 2016). People have increased interest to incorporate high nutrient levels with sufficient amounts of essential minerals into the regular diet (Alagić et al., 2018), due to their role as structural components of organs (e.g. bones and teeth) and their involvement in many physiological and metabolic processes (Soetan et al., 2010).

72 Besides, plants emit into the environment many types of volatile organic compounds (VOC), which behave as info 73 chemicals in biotic communication (Blée, 2002; Niinemets et al., 2013) and in abiotic stress acclimation. VOCs 74 represent also a method for plants to face stress and reduce the negative effects (Loreto and Schnitzler, 2010). 75 Furthermore, many authors have proven the important role of VOC in food industry to improve shelf-life and safety 76 of fresh or minimally processed fruits (Lanciotti et al., 2004) as well as in industrially produced food, which are 77 often subjected to aroma losses during processing and storage (Tylewicz et al., 2017). Indeed, investigating plant by-78 products, is one of the main topics of research and innovation nowadays, in the hope to reduce environmental 79 damage and provide new extracts of high purity. These enriched fractions and isolated compounds, assessed for their 80 safety and efficacy, should be incorporated in food and pharma formulations and products.

81 Prunus spp. is a member of the plant family Rosaceae that is widely distributed in most countries of the world. 82 During the last two decades, the Tunisian areas cultivated with *Prunus* showed a significant upward trend, with 83 more than 1.3 million hectares and an annual production of 123000.000 tonnes in 2016 (FAOstat, 2018). In fact, 84 peach cultivation occupies an important place in the sector of fruit trees in Tunisia; this culture has been expanding 85 since the eighties due to the introduction of new varieties and it has increased in areas equipped with modern 86 irrigation techniques. Therefore, production and exports have significantly increased in recent years (Dabbou et al., 87 2016). Trees producing nectarines, flat peach, white peach and yellow peach are characterized by different leaf traits 88 (Dai et al., 2017). Leaves and stems are considered as a by-product deriving from peach tree cultivation and canned 89 industries. Large amounts of these by-products are principally generated during harvesting of the peach fruit and 90 following leaf fall in winter. Winter fall is responsible for the most abundant accumulation of these by-product, 91 though this production has been never statistically quantified. Besides nutritional and nutraceutical value of the fruit, 92 also peach by-products as peel and remnant pulp have a hypoglycemic and hypotriglyceridemic effect (Rodríguez-

93 gonzález et al., 2018) whereas leaves showed pharmaceutical potential and are traditionally used as antihelmentic,

94 laxative and sedative (Kazan et al., 2014). A decoction of the leaves is used as a bath to treat heat rash, skin disease

95 and circulatory troubles (Perry and Metzger, 1980).

96 The full characterization of bioactive compounds obtained from natural matrices is a major challenge for researchers 97 in the food, pharmaceutical, and cosmetic industry (Karabegovic et al., 2014). The huge amount of food related 98 materials represents a potential reservoir of functional substances, whose recovery can create additional income and 99 contribute to reduce waste.

However, data on mineral, volatiles and nutraceutical composition of agricultural by-products of *P. persica* crops are scarce. Therefore, the present investigation is aimed to evaluate phenolic profile and mineral and volatile composition of leaves and stems from five peach cultivars grown in Tunisia under the same agricultural, geographical and climatic conditions.

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## 105 2. Material and methods

# 106 2.1. Plant material

107 Peach agricultural by-products (young and mature leaves and stems) were collected during the two summer seasons 108 2013-2014 from the experimental orchard of the Regional Centre of Agricultural Research Farm in the region of 109 Sidi Bouzid, Center-West of Tunisia (35°2'0"N, 9°30'0"E; at 313 m a.s.l.). The peach orchard is located in a semi-110 arid bioclimatic region, with a mean annual rainfall of 251.8 mm (concentrated mainly from autumn to spring), a 111 mean annual temperature varying from 12.5°C to 25.3°C and an average evapotranspiration (ETc.) of 1634.9 mm. 112 The soil horizons present a silt-clay-loam texture. Peach agricultural by-products of five different cultivars of 113 Prunus persica L. [Batsch], Early Maycrest, Sweet Cap, O'Henry, Flordastar and Rubirich grafted on the Germen 114 wild rootstock, were studied. Trees were planted in 2005 at a spacing of 4 m x 6 m. Trees were irrigated by a 115 network drop-by-drop with two pipes per row (4 L h<sup>-1</sup>). During the 2-year experimental period the three cultivars 116 were similarly fertilized with nitric acid, magnesium and potassium. Young leaves were a few days old and mature 117 leaves were >1-year-old. Three replicates were made for each peach by-products. Plant materials were hand 118 harvested, washed with tap water, lyophilised and stored at -20°C until analysis.

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- 2.2. Analytical methods
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### 2.2.1. Macro and microelements quantification

121 Cations was determined by atomic absorption spectrophotometry (PerkineElmer Analyst 100, Waltham, MA, USA) 122 as reported previously (Ranieri et al., 2005) with slight modifications. Briefly, the acidic digestion of dried samples 123 was carried out by dissolving 500 mg of dry matter in 9 mL flame-heated concentrated HNO<sub>3</sub>. H<sub>2</sub>O<sub>2</sub> drops were 124 added until complete clarification of samples, and final volumes were adjusted to 25 mL by milliQ water. The 125 concentrations of Ca, Mn, Na, Cu, K, Mg and Zn were successively measured using PerkineElmer INTENSITRON 126 lamps. Nitrogen was measured by Kjeldahl procedure after acidic digestion in H<sub>2</sub>SO<sub>4</sub>. Results were expressed as mg 127 g<sup>-1</sup> DW.

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## 2.2.2. Spectrophotometric analysis

130 The phenolic extract was obtained from the homogenation of 0.5 g of lyophilised peach leaves and methanol (80%) 131 and their centrifugation at 5,000 x g for 10 min at 4 °C (Dabbou et al., 2016); these steps was repeated twice. The 132 collected extracts were stored at -20°C until analysis.

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### 2.2.2.1. Total phenols and o-diphenols

134 The total phenols and o-diphenols content of peach leaves were determined calorimetrically using the Folin-135 Ciocalteu method (Montedoro et al., 1992). The results were expressed as mg hydroxytyrosol 100 g<sup>-1</sup> DW.

136 2.2.2.2. Total flavonoids

137 To determine the total flavonoid content of leaf extract, the aluminum chloride colorimetry method was used 138 (Zhishen et al., 1999). In brief, the methanolic extract sample was diluted with dH<sub>2</sub>O. Then, the diluted extract was 139 mixed with NaNO<sub>2</sub> (5%), AlCl<sub>3</sub> (10%) and NaOH (1M) solutions and dH<sub>2</sub>O and the absorbance of the obtained 140 solution was measured at 510 nm. The results were expressed as mg catechin 100 g<sup>-1</sup>DW.

- 141 2.2.2.3. Total flavonols

142 Total flavonols was analyzed spectrophotometrically at 360 nm using the colorimetric method described previously 143 (Romani et al., 1996). Diluted leaf extracts were added to ethanol (10%), 0.1% HCl in 95% ethanol and 2% HCl.

144 The mixed solution was allowed to stand for 15 min and the absorbance was read at 360 nm against the blank. The

total flavonols content was expressed as mg quercetin  $100 \text{ g}^{-1} \text{ DW}$ . 145

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2.2.2.4. In vitro antioxidant analysis

Antioxidant potential of the peach leaves extracts was studied using 2,2-azino-bis-3-ethylbenzothiazoline-6sulfonicacid (ABTS) radical scavenging assay (Re et al., 1999; Rice-Evans et al., 1996) and the reducing power assay (Oyaizu, 1986). The results were expressed as the effective concentration (EC50) value (mg 100g<sup>-1</sup> DW) (Dabbou et al., 2015).

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## 2.2.3. Individual phenolic compounds analyses

152 Individual phenols were determined from the lyophilised leaves and stems (0.25 g) homogenised, for 2 min on ice, 153 in an Ultra-Turrax T25 (IKA Labortechnik, Janke & Kunkel, Staufen, Germany), with 5 mL of 80% methanol 154 aqueous solution, BHT (0.1% w/v) to prevent oxidation. The homogenates were then centrifuged (16000 rpm, 15 155 min, 4°C). The procedure was repeated twice and the resulted supernatants were evaporated to dryness at 35°C 156 under vacuum through a rotary evaporator. The supernatant was recovered in 1 mL of MilliQ water and filtered 157 through 0.45 µm filter. Analysis of phenolic compounds was performed following the method of Tomás-Barberán et 158 al. (2001) using a Spectra System P4000 HPLC, equipped with a UV 6000 LP photodiode array detector (Thermo 159 Fisher Scientific, Waltham, MA) using a Phenomenex Prodigy LC-18 RP column (5 µm particle size, 250 x 4.6 mm, 160 Phenomenex Italia, Castel Maggiore, Italy). Concentrations were expressed as mg 100g<sup>-1</sup> dry weight (DW).

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## 2.2.4. Volatile compound analyses

Supelco (Bellefonte, PA) SPME devices coated with polydimethylsiloxane (PDMS, 100 µm) were used to sample the headspace of dry plant material inserted into a 15 mL glass vial and allowed to equilibrate for 30 min. After the equilibration time, the fiber was exposed to the headspace for 35 min at room temperature. Once sampling was finished, the fiber was withdrawn into the needle and transported to the injection port of the GC–MS system. All the SPME sampling and desorption conditions were identical for all the samples. Moreover, blanks were performed before each first SPME extraction and randomly repeated during each series. Quantitative comparisons of relative peaks areas were carried out between the same chemicals in the different samples.

GC-EIMS analyses were executed with a Varian (Palo Alto, CA) CP3800 gas chromatograph equipped with a DB-5 capillary column (30 m x 0.25 mm x 0.25  $\mu$ m; Agilent) and a Varian Saturn 2000 ion trap mass detector. The analytical settings were as follows: injector and transfer line temperatures were 250 and 240°C, respectively; oven temperature was programmed from 60 to 240 °C at 3 °C min<sup>-1</sup>; carrier gas was helium at 1 mL min<sup>-1</sup>; splitless injection. Constituents identification was based mainly on a comparison of the retention times with those of authentic samples, comparing their linear retention indices relative to the series of n-hydrocarbons (C7-C28) and on 176 computer matching against the commercial (NIST 2014 and Adams 2007) and homemade libraries of mass spectra,

177 and MS literature data (Adams, 1995; Davies, 1990; Jennings and Shibamoto, 1980; Stenhagen et al., 1974; Swigar

178 and Silverstein, 1981). Results were expressed as % of total volatile compounds.

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### 180 2.3. Statistical analysis

181 All values are expressed as the means ± SD. Data were analysed using SPSS program, release 17.0 for Windows 182 (SPSS, Chicago, IL, USA). Analysis of Variance (ANOVA) was used to determine differences among means. 183 Duncan multiple comparison test was used to discriminate among mean values of the five P. persica cultivars. 184 Statistically significant differences between groups were defined at p < 0.05.

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186 3. Results

#### 187 3.1. Macro and micronutrients

188 The mineral composition of agricultural by-products (young, mature leaves and stems) from the five peach cultivars 189 (Sweet Cap, Early Maycrest, O'Henry, Flordastar and Rubirich) is reported in Table 1. Comparing the different 190 minerals, N showed higher contents in peach leaves among macronutrients, while Mn showed higher contents 191 among micronutrients. Stems also recorded high levels of Ca, comparable to those found in young leaves, an 192 exception observed for Rubirich. Traces were recorded for micronutrient levels in the stems. In average, young 193 leaves contained 1.5 to 3-times higher amounts from Ca and Mn than mature leaves, from 14 to 85-times higher 194 amounts from Na, while levels of Zn, Cu, K, and N were approximately identical thought ripening process of peach 195 leaves. Magnesium contents differed between young and mature leaves according to cultivars (Table 1). Mg values 196 for the different cultivars of peach leaves ranged from 5.88 to 24.20 mg g<sup>-1</sup> DW in young leaves for Rubirich and 197 Early Maycrest cultivars, respectively and from 7.88 to 12.01 mg g<sup>-1</sup> DW in mature leaves for Flordastar and 198 Rubirich cultivars, respectively. Comparing cultivars, accumulation of leaf and stems nutrients was cultivar-199 dependent, but inconsistent. In fact, leaf N concentration was not significantly affected by the cultivar.

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3.2. Spectrophotometric analysis

202 Variations in total flavonoids, flavonols, o-diphenols and total phenols in the by-products of five peach cultivars 203 (young and mature leaves and stems) are presented in Table 2. Total phenols showed values ranging from 2534.06 to 4593.01 mg 100 g<sup>-1</sup> MS. Total flavonoids varied from 649.25 to 1011.26 mg 100 µg-1 MS and the lowest values 204 205 were found in leaves. Early Maycrest, Flordstar and Rubbirich cultivars did not show statistically significant differences among phenolic compounds (flavonoids, flavonols, *o*-diphenols and total phenols contents). In addition,
 the stems showed the highest levels of total phenols, total flavonoids and *o*-diphenols among the peach by-products
 studied but the lowest flavonol contents. These results are cultivar-dependent with the richest Sweet Cap cultivar.

Table 2 showed that among cultivars, no statistical difference (p>0.05) between mature leaves were observed.

210 However, young leaves of Sweet Cap and Rubbirich cultivars showed the highest contents of *o*-diphenols (1534.85

and 1546.97 mg 100 g<sup>-1</sup> DW, respectively), flavonoids (1011.26 and 869.13 mg 100 g<sup>-1</sup> DW, respectively) and total

212 phenols (4593.01 and 4326.83 mg 100g<sup>-1</sup> DW, respectively). O'Henry and Flordstar leaves had the highest contents

- 213 of flavonols (1309.39 and 1413.05 mg 100  $g^{-1}$  DW, respectively).
- 214 The antioxidant and antiradical activity of the agricultural by-products of the five peach cultivars were investigated 215 using two methods, ABTS+•, and reducing power assays (Table 2). All young leaf extracts were more effective at 216 reducing power capacity (82.26 - 177.51 mg 100 g<sup>-1</sup> MS, respectively for Rubirich and Sweet Cap) than ripe leaf 217 extracts. ABTS values showed no variation between the two types of leaves (p > 0.05), with the exception of Sweet 218 Cap. In addition, the comparison with the stem extracts, potent activities were observed for both tests evaluated 219 among the peach by-products studied. Comparing the cultivars, O'Henry showed the highest ABTS value, while no 220 significant difference was shown between the other cultivars. For stem extracts, the highest activity was observed in 221 Early Maycrest; This result can be explained by its high content of phenolic compounds.
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### 3.3. Individual phenolic compounds

Table 3 illustrates the quantity of individual phenolic compounds detected in the methanol extract of peach leaves and stems. Thirteen compounds were identified and quantified including hydroxycinnamic acid derivatives (neochlorogenic acid, chlorogenic acid, caffeic acid, coumaric acid and ferulic acid), flavan-3-ols (catechin and epicatechin), hydroxybenzoic acids (gallic acid), flavonols (quercetin-3-rutinoside and quercetin-3-galactoside) and anthocyanins (cyanidin-3-glucoside and cyanidin-3-galactoside).

Quantitatively, among the individual compounds identified, in addition to chlorogenic acid (127.34-178.25 mg 100g<sup>-1</sup> DW), flavonols were the most concentrated compounds in the leaves, with quercetin-3-galactoside contents ranging between 73.43 mg 100g<sup>-1</sup> DW and 106.09 mg 100g<sup>-1</sup> DW, followed by quercetin-3-rutinoside (73.85-115.84 mg 100 g<sup>-1</sup> DW) whereas flavan-3-ol were the most abundant compounds in the stems, with catechin (170.09-199.98 mg 100g<sup>-1</sup> DW) being the most concentrated, but no statistical differences were observed between cultivar (Table 3). Quercetin-3-galactoside, also known as hyperoside, was statistically (P < 0.01) higher concentrated in young leaves of O'Henry and Early Maycrest cultivars than in the other cultivars (Table 3). 236 Anthocyanins showed the least contents among the peach agricultural by-products. As shown in Table 3, the cultivar 237 (P < 0.05) had a significantly remarkable impact on individual phenolic compounds of peach by-products. Cyanidin-238 3-rutinoside was not detected in the two types of leaves of the Flordastar and Rubirich cultivars as well as in Early 239 Maycrest young leaves, and cyanidin-3-glucoside was undetectable in Flordastar mature leaves.

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#### 241 3.4. Volatile compounds

242 The study allowed the characterization of 59 volatile compounds at different percentages and occurrences in the 243 peach by-products (Table 4), of which 43 were present in the leaf. GC-MS analysis revealed that benzaldehyde was 244 detected as the major volatile leaf component (70-95%), whereas myrcene (18-21%) and terpinolene (18-26%) were 245 found to be the most important compounds in stems. The other major compounds of leaves were 1-pentanol (2.4-246 4.2%), (Z)-2-hexenal (3.3-5.3%), (E)-2-hexenal (0.6-4.1%), 2,6-dimethylnonane (1.6-2.7%) and 2,9-dimethylnonane 247 (2.3-4.5%). The levels of these compounds varied according to the cultivar and the maturity of the leaves (Table 4). 248 The compounds  $\alpha$ -pinene, 1-pentanol, (Z)-2-hexenal, 2,6-dimethylnonane, 2,9-dimethylnonane, 1-octanol, 1-nonen-249 3-ol, 6-methyldodecane, (E)-2-tridecane, (Z)-2-tridecane and 3-methyldecanol were found only in mature peach 250 leaves. On the contrary, myrcene, limonene, (E)- $\beta$ -ocimene,  $\gamma$ -terpinene,  $\beta$ -bourbonene,  $\beta$ -caryophyllene, (Z)-3-251 hexenyl acetate, benzyl alcohol, 1-nonanol, n-tetradecane, 1-cyclohexyloctant and n-hexadecane were exclusive of 252 young peach leaves. Apocarotenoids (theaspirane I and theaspirane II) were detected only at the stems (Table 4).

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### 254 4. Discussion

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# 4.1. Macro and micronutrients

256 The mineral composition of agricultural by-products (young, mature leaves and stems) from the five peach cultivars 257 (Sweet Cap, Early Maycrest, O'Henry, Flordastar and Rubirich) showed significant variations in the levels of Mg, 258 Ca, Mn, K and Na with respect to harvests, which confirms that the uptake of mineral elements by peach trees varies 259 with the plant physiological stages over the production cycle (Leonel et al., 2011). Abadia et al. (1985) found 260 different results of the mineral composition in peach leaves which are the following : Ca (1.37 mg 100g<sup>-1</sup> DW), Mg (0.48 mg 100g<sup>-1</sup> DW), K (2.09 mg 100g<sup>-1</sup> DW), Na (0.06 mg 100g<sup>-1</sup> DW), Mn (28 mg 100g<sup>-1</sup> DW), Cu (10 mg 100g<sup>-1</sup> 261 262 <sup>1</sup> DW), and Zn (34 mg 100g<sup>-1</sup> DW). In addition, our results are higher than those found in *Prunus cerasifera* leaves 263 (C 448.6; N 14.7; P 1.5; K 24.8; Ca 15.7; Mg 2.4; Na 1.2; Mn 44.4; Cu 7.7; Zn 10.2 mg g<sup>-1</sup> DW) (Kuppusamy et 264 al., 2016) but lower than those found in plums (Mg 0.35%; Ca 2.24%; K 2.59%; Zn 23.6 mg kg<sup>-1</sup> DW; Mn 31.6 mg 265 kg<sup>-1</sup> DW; Cu 5.27 mg kg<sup>-1</sup> DW) (Reig et al., 2018). Comparing cultivars, accumulation of leaf and stems nutrients

was cultivar- dependent, but inconsistent, and resulted in nutrient deviations from normal values which confirm previous findings on *Prunus avium* (Milošević et al., 2015). In fact, leaf N concentration was not significantly affected by the cultivar. These results are consistent with previous works (Balal et al., 2011; El-Jendoubi et al., 2014).

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## 4.2. Spectrophotometric analysis

Total phenols and total flavonoids values of the *P. persica* leaves found in our study are higher than those of the leaves of *Prunus cerasifera* and *Prunus laurocerasus* reported by other works (Karabegovic et al., 2014; Kuppusamy et al., 2016). Early Maycrest, Flordstar and Rubbirich cultivars did not show statistically significant differences among phenolic compounds (flavonoids, flavonols, *o*-diphenols and total phenols contents). In addition, the stems showed the highest levels of total phenols, total flavonoids and *o*-diphenols among the peach by-products studied but the lowest flavonol contents. These results are cultivar-dependent where Sweet Cap cultivar was found to be the richest. These findings confirmed the work of Eftekhari et al. (2017) on leaves and stems of *Vitis vinifera*.

Our findings of the by-product in the five peach cultivars indicate that peach leaves had important contents of polyphenol compounds, which could have potential biological importance. Similar results were observed in apricot leaves (Vieira et al., 2016; Zeb et al., 2017), where the total phenolic content increased with maturation. The increase was highly correlated with the variation in the polyphenols profile of the leaves. According to these authors (Vieira et al., 2016; Zeb et al., 2017), variations in the polyphenol compounds of the leaves during maturation may be correlated with fruit development. Furthermore, these variations may be affected by the variety, climate, soil and environmental conditions.

Quantitatively proved, stems showed higher levels of phenols than young and mature leaves. However, differences
between cultivars in phenol levels of mature leaves were almost inexistent.

According to our results, Sweet cap showed an important antioxidant and antiradical activity mainly in young leaves comparing to the other peach cultivars. These results can be explained by the higher phenol contents of Sweet Cap; this confirms previous work (Dabbou et al., 2015). For stems extracts, the highest activity was observed in Early Maycrest, consistent with the high content of phenolic compounds.

## 292 4.3. Individual phenolic compounds

According to the HPLC analysis, chlorogenic acid was the major phenolic compound found in peach leaves, that contained also other hydroxycinnamic acid derivatives, flavan-3-ols acids, hydroxybenzoic acids, flavonols and anthocyanins. This phenolic profile was proved by previous works on leaves extracts from *Prunus* genus and especially *Prunus dulcis* Mill. (Bottone et al., 2018) and *Prunus avium* (Dziadek et al., 2019). However, in other

297 leaves from the same genus *Prunus* the major compound found was the neochlorogenic acid (Jesus et al., 2019).

298 Karabegović et al. (2014) also identified luteolin-7-glucoside, apigenin-7-glucoside, kaempferol-3-glucoside and

299 naringin in leaf extracts of *Prunus laurocerasus* L. Furthermore, Dziadek et al. (2019) found myricetin compound

300 in addition to caffeic, cholorogenic, *p*-coumaric and ferulic acids in *Prunus avium* L. leaves, while 36 compounds

301 were identified in leaves of *Prunus cerasus* L. and 26 compounds in stem extracts of *Prunus avium* L. (Bastos et al.,

302 2015; Oszmianski and Wojdylo, 2014).

303 Quantitatively, the O'Henry cultivar seemed to be the richest one for young leaves and stems compared to the other304 cultivars, indicating a genotype-depending variability in the phenolic content of the different plant organs.

Anthocyanins, the most important group of water-soluble vacuolar pigments, appear as red, blue, or purple and occur in all plant tissues, including flowers, stems, leaves, roots, and fruits (Xu et al., 2017). These substances showed the least contents among the peach agricultural by-products. Conversely, flavonols were the most concentrated compounds in the leaves of the five cultivars studied.

309 According to our results, phenolic compounds vary depending on cultivar (P < 0.05). Early Maycrest had the highest 310 catechin content among the analysed peach cultivars in mature leaves, though no statistical differences were 311 observed for young leaves (Table 3).

The analysis of stems methanolic extract showed the abundance of catechin. These results confirm those found previously in the stems of *Prunus avium* L. (Aires et al., 2017; Bastos et al., 2015; Jesus et al., 2019). However, values were higher than those of *P. persica* stems studied. It was shown that catechin is one of the flavan-3-ol acids compounds with the highest antioxidant activity (Frankel et al., 1997).

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## 317 *4.4. Volatile compounds*

GC-MS analysis of the volatile compounds from leaves and stems of *P. persica* revealed that benzaldehyde was detected as the major volatile leaf component, whereas myrcene and terpinolene were found to be the most important compounds in stems. Benzaldehyde levels were higher than those reported by other studies (Horvat and Chapman, 1990; Verma et al., 2017). Benzaldehyde probably arises from the cyanogenic glycoside amygdalin, a typical constituent of many *Prunus* species (Takeoka et al., 1990). Based on market demand, it represent the second most important molecule in the industry as ingredient for flavouring foods and beverages; it is also requested by fragrance industries (Verma et al., 2017). Furthermore, Farré-armengol et al. (2016) showed a physiological importance of benzaldehyde from green leaf in plant protection against microbe infection or predators, constituting arelevant role in ecological signalling.

327 Comparing young to mature leaves,  $\alpha$ -pinene, 1-pentanol, (Z)-2-hexenal, 2,6-dimethylnonane, 2,9-dimethylnonane, 328 1-octanol, 1-nonen-3-ol, 6-methyldodecane, (E)-2-tridecane, (Z)-2-tridecane and 3-methyldecanol were found in 329 mature peach leaves only and myrcene, limonene, (E)- $\beta$ -ocimene,  $\gamma$ -terpinene,  $\beta$ -bourbonene,  $\beta$ -caryophyllene, (Z)-330 3-hexenyl acetate, benzyl alcohol, 1-nonanol, n-tetradecane, 1-cyclohexyloctant and n-hexadecane were detected 331 exclusively in young peach leaves. These results confirm previous works indicating that developmental stages and 332 phenological processes affects the emission of VOCs (Bracho-Nunez et al., 2011). The profiles of the emitted VOCs 333 is strongly specie-specific, so their accumulation in plants may results constitutively different (Vieira et al., 2016). 334 Concerning terpinolene, 1,8-cineole, (E)-2-hexenal and benzaldehyde, their levels remained approximately the same 335 during leaf maturation, which confirm previous studies (Horvat and Chapman, 1990). Biosynthesis of mono- and 336 sesquiterpenes in plants is often related to the presence of specialized secretory structures (e.g. glandular trichomes, 337 oil and resin ducts and glandular epidermis); consequently, each part of the plant may have typical aroma 338 compounds composition (Lewinsohn et al., 1998).

Takahashi et al. (2006), when analysing volatiles from leaves of the subgenera (*Cerasus, Padus, Laurocerasus*, and
 *Prunus*) in the genus *Prunus* found linalool, phenethyl alcohol, and coumarin compounds. These compounds were
 not found in *P. persica* leaves studied in the present work.

According to our bibliographic researches, there is no previous work published to date on the volatile compounds of*P. Persica* stems.

344

## 345 **5.** Conclusion

346 Most of the studies on peach are focused on the economic, nutritional and antioxidant importance of peach fruit. 347 Conversely, very few works focused on the other organs of peach tree, as leaves, branches, stems, etc. This study 348 aims to valorize the peach agricultural by-products (young and mature leaves and stems) characterizing their 349 bioactive and mineral compounds. Decreased Ca and Na levels were observed throughout maturity. However, no 350 statistical difference (p > 0.05) for the micronutrients was observed between the leaves. The stems contained only 351 traces. In addition, the mineral and phenol contents were cultivar-dependent. Volatile compounds of leaf and stems 352 from peach trees were dominated by the benzaldehyde compound. The contents of volatile and phenolic compounds 353 were significantly affected by peach by-product as well as the cultivar. Quantitatively, total phenols were the most 354 abundant in the young leaves and stems of the O'Henry cultivar. As well, stems showed the highest levels of total

phenols, total flavonoids and *o*-diphenols among the peach by-products studied but the lowest flavonol contents.
From HPLC-DAD analysis, chlorogenic acid, quercetin-3-galactoside and quercetin-3-rutinoside were the most
concentrated compounds in the leaves whereas catechin was the most abundant in the stems.

358 Furthermore, stem extracts showed higher antioxidant activities for the two tests evaluated. In conclusion, even if an

359 essential oil is not present in peach leaves and stems, these agricultural by-products may be used as flavouring

360 agents to substitute expensive natural pure compounds, i.e. for masking undesirable odors. Furthermore, they may

- also contain secondary metabolites useful for improving health.
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# 363 References

- Abadia, J., Nishio, J.N., Monge, E., Montanes, L., Heras, L., 1985. Mineral composition of peach leaves affected by
   iron chlorosis. J. Plant Nutr. 8, 697–707. https://doi.org/10.1080/01904168509363378
- Adams, R.P., 1995. Identification of Essential Oil Components by Gas Chromatography / Mass Spectrometry.
   Allured Publishing Corp.: Carol Stream, USA.
- Aires, A., Dias, C., Carvalho, R., Saavedra, M.J., 2017. Analysis of glycosylated flavonoids extracted from sweetcherry stems, as antibacterial agents against pathogenic Escherichia coli isolates. Acta Biochim. Pol. 64, 265–
  271. https://doi.org/10.18388/abp.2016\_1374

Alagić, S., Tošić, S.B., Dimitrijević, M.D., Nujkić, M.M., Papludis, A.D., Fogl, V.Z., 2018. The content of the

potentially toxic elements, iron and manganese, in the grapevine cv Tamjanika growing near the biggest
copper mining/metallurgical complex on the Balkan peninsula: phytoremediation, biomonitoring, and some
toxicological aspects. Environ. Sci. Pollut. Res. 25, 34139–34154. https://doi.org/10.1007/s11356-018-3362-7

Balal, R.M., Gimeno, V., Shahid, M.A., Lidon, V., Garcia, A.L., Abbas, T., Garcia-Sanchez, F., Ghazanfer, U.,

376 2011. Effects of phosphorus fertilization on growth, leaf mineral concentration and xylem-phloem nutrient

- 377 mobility in two rootstocks of prunus (Prunus persica × Prunus amygdalus) and (Prunus insititia) in the
- 378 Mediterranean area. Aust. J. Crop Sci. 5, 1542–1549.
- 379 Bastos, C., Barros, L., Dueñas, M., Calhelha, R.C., Queiroz, M.J.R.P., Santos-Buelga, C., Ferreira, I.C.F.R., 2015.
- Chemical characterisation and bioactive properties of Prunus avium L.: The widely studied fruits and the
   unexplored stems. Food Chem. 173, 1045–1053. https://doi.org/10.1016/j.foodchem.2014.10.145

13

- 382 Blée, E., 2002. Impact of phyto-oxylipins in plant defense. Trends Plant Sci. 7, 315–321.
- 383 https://doi.org/10.1016/S1360-1385(02)02290-2
- Bottone, A., Montoro, P., Masullo, M., Pizza, C., Piacente, S., 2018. Metabolomics and antioxidant activity of the
  leaves of Prunus dulcis Mill. (Italian cvs. Toritto and Avola). J. Pharm. Biomed. Anal. 158, 54–65.
- 386 https://doi.org/10.1016/j.jpba.2018.05.018
- 387 Bracho-Nunez, A., Welter, S., Staudt, M., Kesselmeier, J., 2011. Plant-specific volatile organic compound emission
- rates from young and mature leaves of Mediterranean vegetation. J. Geophys. Res. Atmos. 116, 1–13.
  https://doi.org/10.1029/2010JD015521
- 390 Dabbou, S., Dabbou, S., Pandino, G., Lombardo, S., Mauromicale, G., Chahdoura, H., Gasco, L., Helal, A.N., 2015.
- 391 In vitro antioxidant activities and phenolic content in crop residues of Tunisian globe artichoke. Sci. Hortic.
- 392 (Amsterdam). 190, 128–136. https://doi.org/10.1016/j.scienta.2015.04.014
- 393 Dabbou, S., Lussiana, C., Maatallah, S., Gasco, L., Hajlaoui, H., Flamini, G., 2016. Changes in biochemical
- compounds in flesh and peel from Prunus persica fruits grown in Tunisia during two maturation stages. Plant
   Physiol. Biochem. 100, 1-11. https://doi.org/10.1016/j.plaphy.2015.12.015
- 396 Dai, L., Li, P., Shang, B., Liu, S., Yang, A., Wang, Y., Feng, Z., 2017. Differential responses of peach (Prunus
- 397 persica) seedlings to elevated ozone are related with leaf mass per area, antioxidant enzymes activity rather
- than stomatal conductance. Environ. Pollut. 227, 380–388. https://doi.org/10.1016/j.envpol.2017.04.068
- 399 Davies, N.W., 1990. Gas chromatographic retention indices of monoterpenes and sesquiterpenes on methyl silicone
  400 and C arbowax 20M phases. J. Chromatogr. 503, 1–24.
- 401 Dziadek, K., Kopeć, A., Tabaszewska, M., 2019. Potential of sweet cherry (Prunus avium L.) by-products :
- 402 bioactive compounds and antioxidant activity of leaves and petioles. Eur. Food Res. Technol. 245, 763–772.
  403 https://doi.org/10.1007/s00217-018-3198-x
- 404 Eftekhari, M., Yadollahi, A., Ford, C.M., Shojaeiyan, A., 2017. Industrial Crops & Products Chemodiversity
- 405 evaluation of grape (Vitis vinifera) vegetative parts during summer and early fall. Ind. Crop. Prod. 108, 267–
- 406 277. https://doi.org/10.1016/j.indcrop.2017.05.057
- 407 El-Jendoubi, H., Vasquez, S., Calatayud, A., Vavpetic, P., Vogel-Mikus, K., Pelicon, P., Abadia, J., Abadia, A.,
- 408 Morales, F., 2014. The effects of foliar fertilization with iron sulfate in chlorotic leaves are limited to the
- 409 treated area . A study with peach trees (Prunus persica L. Batsch) grown in the field and sugar beet (beta

- 410 vulgaris L.) grown in hydroponics. Front. Plant Sci. 5, 1-16. https://doi.org/10.3389/fpls.2014.00002
- 411 FAOstat, 2018. Food and Agricultural Organisation Statistics [WWW Document]. URL
- 412 http://www.fao.org/faostat/en/#data/QC (accessed 11.30.18).
- 413 Farré-armengol, G., Filella, I., Llusia, J., Peñuelas, J., 2016. Bidirectional Interaction between Phyllospheric
- 414 Microbiotas and Plant Volatile Emissions. Trends Plant Sci. 21, 854–860.
- 415 https://doi.org/10.1016/j.tplants.2016.06.005
- Frankel, E.N., Huang, S.-W., Aeschbach, R., 1997. Antioxidant activity of green teas in different lipid systems. J.
  Am. Oil Chem. Soc. 74, 1309–1315. https://doi.org/10.1007/s11746-997-0062-8
- 418 Horvat, R.J., Chapman, G.W., 1990. Comparison of volatile compounds from peach fruit and leaves (cv. Monroe)
- 419 during maturation. J. Agric. Food Chem. 38, 1442–1444. https://doi.org/10.1021/jf00097a002
- Jennings, W., Shibamoto, T., 1980. Qualitative Analysis of Flavor and Fragrance Volatiles by Glass Capillary
  Chromatography. Academic Press Inc., New York.
- 422 Jesus, F., Gonçalves, A.C., Alves, G., Silva, L.R., 2019. Exploring the phenolic profile, antioxidant, antidiabetic and
- 423 anti- hemolytic potential of Prunus avium vegetal parts. Food Res. Int. 116, 600-610.
- 424 https://doi.org/10.1016/j.foodres.2018.08.079
- 425 Joana Gil-Chávez, G., Villa, J.A., Fernando Ayala-Zavala, J., Basilio Heredia, J., Sepulveda, D., Yahia, E.M.,
- 426 González-Aguilar, G.A., 2013. Technologies for extraction and production of bioactive compounds to be used
- 427 as nutraceuticals and food ingredients: An Overview. Compr. Rev. Food Sci. Food Saf. 12, 5–23.
- 428 https://doi.org/10.1111/1541-4337.12005
- 429 Karabegovic, I.T., Stojicevic, S.S., Velickovic, D.T., Todorovic, Z.B., Nikolic, N.C., Lazic, Miodrag, L., 2014. The
- 430 effect of different extraction techniques on the composition and antioxidant activity of cherry laurel (Prunus
- 431 laurocerasus) leaf and fruit extracts. Ind. Crop. Prod. 54, 142–148.
- 432 https://doi.org/10.1016/j.indcrop.2013.12.047
- 433 Karabegović, I.T., Stojičević, S.S., Veličković, D.T., Todorović, Z.B., Nikolić, N.Č., Lazić, M.L., 2014. The effect
- 434 of different extraction techniques on the composition and antioxidant activity of cherry laurel (Prunus
- 435 laurocerasus) leaf and fruit extracts. Ind. Crops Prod. 54, 142–148.
- 436 https://doi.org/10.1016/j.indcrop.2013.12.047

- 437 Kazan, A., Koyu, H., Turu, I.C., Yesil-Celiktas, O., 2014. Supercritical fluid extraction of Prunus persica leaves and
- 438 utilization possibilities as a source of phenolic compounds. J. Supercrit. Fluids 92, 55–59.
- 439 https://doi.org/10.1016/j.supflu.2014.05.006
- 440 Kuppusamy, S., Thavamani, P., Megharaj, M., Nirola, R., Lee, Y.B., Naidu, R., 2016. Assessment of antioxidant
- 441 activity, minerals, phenols and flavonoid contents of common plant/tree waste extracts. Ind. Crops Prod. 83,
- 442 630–634. https://doi.org/10.1016/j.indcrop.2015.12.060
- Lanciotti, R., Gianotti, A., Patrignani, F., Belletti, N., Guerzoni, M.E., Gardini, F., 2004. Use of natural aroma
  compounds to improve shelf-life and safety of minimally processed fruits. Trends Food Sci. Technol. 15, 201–
  208. https://doi.org/10.1016/j.tifs.2003.10.004
- 446 Leonel, S., Souza, M.E. De, Tecchio, M.A., Segantini, D.M., 2011. Leaf nutritional levels in peach and nectarine

grown in subtropical climate. Rev. Bras. Frutic. 33, 752–761. https://doi.org/10.1590/S010029452011000500105

- Lewinsohn, E., Dudai, N., Tadmor, Y., Katzir, I., Ravid, U., Putievsky, E., Joel, D.M., 1998. Histochemical
  localization of citral accumulation in lemongrass leaves (Cymbopogon citratus (DC.) Stapf., Poaceae). Ann.
  Bot. 81, 35–39. https://doi.org/10.1006/anbo.1997.0525
- 452 Loreto, F., Schnitzler, J.P., 2010. Abiotic stresses and induced BVOCs. Trends Plant Sci. 15, 154–166.
  453 https://doi.org/10.1016/j.tplants.2009.12.006
- 454 Milošević, T., Milošević, N., Glišić, I., Nikolić, R., Milivojević, J., 2015. Early tree growth, productivity, fruit
- 455 quality and leaf nutrients content of sweet cherry grown in a high density planting system. Hortic. Sci. 42, 1–
- 456 12. https://doi.org/10.17221/119/2014-HORTSCI
- Montedoro, G., Servili, M., Baldioli, M., Miniati, E., 1992. Simple and hydrolyzable phenolic compounds in virgin
  olive oil. 1. Their extraction, separation, and quantitative and semiquantitative evaluation by HPLC. J. Agric.
- 459 Food Chem. 40, 1571–1576.
- 460 Niinemets, Ü., Kännaste, A., Copolovici, L., 2013. Quantitative patterns between plant volatile emissions induced
- 461 by biotic stresses and the degree of damage. Front. Plant Sci. 4, 1–15. https://doi.org/10.3389/fpls.2013.00262
- 462 Oszmianski, J., Wojdylo, A., 2014. Influence of cherry leaf-spot on changes in the content of phenolic compounds
- 463 in sour cherry (Prunus cerasus L.) leaves. Physiol. Mol. Plant Pathol. 86, 28–34.
- 464 https://doi.org/10.1016/j.pmpp.2014.03.002

- 465 Oyaizu, M., 1986. Studies on products of browning reaction -- antioxidative activities of products of browning
  466 reaction prepared from glucosamine. Jpn. J. Nutr. 3, 307–315.
- 467 Perry, L.M., Metzger, J., 1980. Medicinal plants of East and Southeast Asia: attributed properties and uses.
  468 Massachusetts Inst. Technol. Press 34, 389. https://doi.org/10.1007/BF02858311
- 469 Ranieri, A., Castagna, A., Scebba, F., Careri, M., Zagnoni, I., Predieri, G., Pagliari, M., Di Toppi, L.S., 2005.
- 470 Oxidative stress and phytochelatin characterisation in bread wheat exposed to cadmium excess. Plant Physiol.
- 471 Biochem. 43, 45–54. https://doi.org/10.1016/j.plaphy.2004.12.004
- 472 Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M., Rice-Evans, C., 1999. Antioxidant activity applying
  473 an improved ABTS radical cation decolorization assay. Free Radic. Biol. Med. 26, 1231–1237.
- 474 Reig, G., Font i Forcada, C., Mestre, L., Jiménez, S., Betrán, J.A., Moreno, M.Á., 2018. Horticultural, leaf mineral
- and fruit quality traits of two 'Greengage' plum cultivars budded on plum based rootstocks in Mediterranean
- 476 conditions. Sci. Hortic. (Amsterdam). 232, 84–91. https://doi.org/10.1016/j.scienta.2017.12.052
- 477 Rice-Evans, C.A., Miller, N.J., Paganga, G., 1996. Structure-antioxidant activity relationships of flavonoids and
  478 phenolic acids. Free Radic. Biol. Med. 20, 933–956. https://doi.org/10.1016/0891-5849(95)02227-9
- 479 Rodríguez-gonzález, S., Pérez-ramírez, I.F., Amaya-cruz, D.M., Gallegos-corona, M.A., Ramos-gomez, M., Mora,
- 480 O., Reynoso-camacho, R., 2018. Polyphenol-rich peach (Prunus persica L .) by-product exerts a greater bene
- 481 fi cial e ff ect than dietary fi ber-rich by-product on insulin resistance and hepatic steatosis in obese rats. J.
- 482 Funct. Foods 45, 58–66. https://doi.org/10.1016/j.jff.2018.03.010
- 483 Romani, A., Mancini, P., Tatti, S., Vincieri, F.F., 1996. Polyphenols and polysaccharides in Tuscan grapes and
  484 wines. Ital. J. Food Sci. 8, 13-24.
- Shashank, K., Pandey, A.K., 2013. Chemistry and biological activities of flavonoids: An overview. Sci. J. 2013, 1–
  16. https://doi.org/10.1016/j.tifs.2005.08.006
- 487 Soetan, K.O., Olaiya, C.O., Oyewole, O.E., 2010. The importance of mineral elements for humans, domestic
  488 animals and plants : A review. African J. Food Sci. 4, 200–222. https://doi.org/10.1186/s12302-017-0116-y
- 489 Stenhagen, E., Abrahamsson, S., McLafferty, F.W., 1974. Registry of mass spectral data, 1st ed. Wiley, New York.
- 490 Swigar, A.A., Silverstein, R.M., 1981. Monoterpenes: Infrared, Mass, 1H NMR, and 13C NMR Spectra, and
- 491 Koveats Indices.

- Takahashi, K., Tsutsumi, Y., Ohtani, H., Katsuki, T., 2006. Variation of fragrance constituents in the leaves of
  Prunus 34. https://doi.org/10.1016/j.bse.2005.07.022
- Takeoka, G.R., Flath, R.A., Mon, T.R., Teranishi, R., Guentert, M., 1990. Volatile constituents of apricot (Prunus
  Armeniaca). J. Agric. Food Chem. 38, 471–477. https://doi.org/10.1021/jf00092a031
- 496 Tomás-Barberán, F.A., Gil, M.I., Cremin, P., Waterhouse, A.L., Hess-Pierce, B., Kader, A.A., 2001. HPLC-DAD-
- 497 ESIMS analysis of phenolic compounds in nectarines, peaches, and plums. J. Agric. Food Chem. 49, 4748–
  498 4760.
- Tylewicz, U., Inchingolo, R., Rodriguez-estrada, M.T., 2017. Food aroma compounds, in: Nutraceutical and
  Functional Food Components. Elsevier Inc., pp. 295–332. https://doi.org/10.1016/B978-0-12-8052570.00009-0
- Verma, R.S., Padalia, R.C., Singh, V.R., Goswami, P., Chauhan, A., Bhukya, B., 2017. Natural benzaldehyde from
  Prunus persica (L.) Batsch. Int. J. Food Prop. 2912, 1-12. https://doi.org/10.1080/10942912.2017.1338728
- 504 Vieira, D.D.S.S., Emiliani, G., Michelozzi, M., Centritto, M., Luro, F., Morillon, R., Loreto, F., Gesteira, A.,
- 505 Maserti, B., 2016. Polyploidization alters constitutive content of volatile organic compounds (VOC) and
- 506 improves membrane stability under water deficit in Volkamer lemon (Citrus limonia Osb.) leaves. Environ.

507 Exp. Bot. 126, 1–9. https://doi.org/10.1016/j.envexpbot.2016.02.010

- 508 Xu, C.-C., Wang, B., Pu, Y.-Q., Tao, J.-S., Zhang, T., 2017. Advances in extraction and analysis of phenolic
- 509 compounds from plant materials. Chin. J. Nat. Med. 15, 721–731. https://doi.org/10.1016/S1875-
- 510 5364(17)30103-6
- Zeb, A., Khadim, N., Ali, W., 2017. Changes in the polyphenolic profile, carotenoids and antioxidant potential of
  apricot (Prunus armeniaca L.) leaves during maturation. Agriculture 7, 1-12.
- 513 https://doi.org/10.3390/agriculture7020009
- Zhishen, J., Mengcheng, T., Jianming, W., 1999. The determination of flavonoid contents in mulberry and their
   scavenging effects on superoxide radicals. Food Chem. 64, 555–559.
- 516 Compliance with Ethical Standards
- 517 **Conflict of interest**
- 518 The authors declare that they have no conflict of interest.

			Young lea	ves				Mature	eaves		Stems					
Minerals	Sweet Cap	Early Maycrest	O'Henry	Flordastar	Rubirich	Sweet Cap	Early Maycrest	O'Henry	Flordastar	Rubirich	Sweet Cap	Early Maycrest	O'Henry	Flordastar	Rubirich	
Macronutrients (mg g <sup>-1</sup> MS)																
Ν	148.13 <sup>b</sup>	157.79 <sup>a.b</sup>	143.43 <sup>ab</sup>	158.80 <sup>ab</sup>	167.38 <sup>a</sup>	150.73 <sup>p</sup>	141.49 <sup>pq</sup>	131.80 <sup>q</sup>	139.08 <sup>pq</sup>	133.19 <sup>q</sup>	0.06 <sup>vw</sup>	0.08 <sup>v</sup>	0.05 <sup>v</sup>	$0.06^{vw}$	$0.05^{w}$	
Na	8.17 <sup>b</sup>	15.55ª	5.22 <sup>cd</sup>	7.78 <sup>bc</sup>	3.06 <sup>d</sup>	0.25 <sup>p</sup>	0.19 <sup>p</sup>	0.26 <sup>p</sup>	0.26 <sup>p</sup>	0.23 <sup>p</sup>	8.19 <sup>bc</sup>	12.14 <sup>w</sup>	6.21 <sup>wx</sup>	5.28 <sup>x</sup>	23.83 <sup>v</sup>	
Mg	12.88 <sup>b</sup>	24.20 <sup>a</sup>	7.47 <sup>c</sup>	7.72 <sup>c</sup>	5.88 <sup>c</sup>	8.75 <sup>q</sup>	8.98 <sup>q</sup>	7.88 <sup>q</sup>	7.96 <sup>q</sup>	12.01 <sup>p</sup>	12.90 <sup>b</sup>	24.05 <sup>v</sup>	6.95 <sup>x</sup>	5.53 <sup>x</sup>	7.94 <sup>x</sup>	
Ca	66.45 <sup>a</sup>	52.21 <sup>b</sup>	30.87 <sup>c</sup>	40.05 <sup>c</sup>	19.02 <sup>d</sup>	23.69 <sup>q</sup>	18.96 <sup>q</sup>	22.37 <sup>q</sup>	24.96 <sup>q</sup>	33.45 <sup>p</sup>	66.59 <sup>a</sup>	60.22 <sup>vw</sup>	41.39 <sup>wx</sup>	30.09 <sup>x</sup>	76.89 <sup>v</sup>	
K	18.24 <sup>a</sup>	20.20 <sup>a</sup>	20.16 <sup>a</sup>	20.65 <sup>a</sup>	15.49 <sup>a</sup>	23.36 <sup>q</sup>	21.45 <sup>q</sup>	20.41 <sup>q</sup>	19.41 <sup>q</sup>	30.16 <sup>p</sup>	18.28 <sup>a</sup>	4.01 <sup>w</sup>	3.86 <sup>w</sup>	4.29 <sup>w</sup>	3.6 <sup>w</sup>	
Micronutrients (µg g <sup>-1</sup> MS)																
Zn	14.68 <sup>a</sup>	15.35 <sup>a</sup>	18.22 <sup>a</sup>	14.23 <sup>a</sup>	18.85 <sup>a</sup>	12.38 <sup>p</sup>	14.80 <sup>p</sup>	9.95 <sup>p</sup>	11.28 <sup>p</sup>	11.94 <sup>p</sup>	$0.02^{v}$	0.02 <sup>v</sup>	$0.02^{v}$	$0.02^{v}$	$0.02^{v}$	
Mn	60.83 <sup>a</sup>	46.92 <sup>abc</sup>	43.23 <sup>bc</sup>	54.79 <sup>ab</sup>	39.34 <sup>c</sup>	90.96 <sup>p</sup>	78.50 <sup>p</sup>	74.17 <sup>p</sup>	55.74 <sup>p</sup>	69.80 <sup>p</sup>	$0.01^{vw}$	0.01 <sup>v</sup>	$0.01^{v}$	0.01 <sup>w</sup>	0.01 <sup>w</sup>	
Cu	4.12 <sup>vw</sup>	2.48 <sup>c</sup>	4.80 <sup>ab</sup>	3.14 <sup>c</sup>	6.61 <sup>a</sup>	6.27 <sup>p</sup>	4.99 <sup>p</sup>	5.64 <sup>p</sup>	4.98 <sup>p</sup>	4.47 <sup>p</sup>	0.01 <sup>v</sup>	$\mathrm{tr}^{\mathrm{v}}$	tr <sup>v</sup>	tr <sup>v</sup>	$0.01^{v}$	

Table 1. Mineral nutrients evaluated in young, mature leaves and stems of five Prunus persica cultivars grown in the center of Tunisia

Values are the means of the five different *Prunus persica* by-products samples (n=3). Different superscripts for the same quality parameter mean significant differences among cultivars p < 0.05. For any parameter, different letters a – e, p-t and v-z indicate significant differences among young, mature leaves and stems, respectively of the five cultivars.

**Table 2.** *Total* phenolic compounds and antioxidant activities (EC50) expressed as mg 100g<sup>1</sup> DW and evaluated in young, mature leaves and stems of five Prunus persica cultivars

				Young leaves				Ν	Iature leaves		Stems					
		Sweet Cap	Early Maycrest	O'Henry	Flordastar	Rubirich	Sweet Cap	Early Maycrest	O'Henry	Flordastar	Rubirich	Sweet Cap	Early Maycrest	O'Henry	Flordastar	Rubirich
o-diphenols		1534.85 <sup>a</sup>	1285.01 <sup>b</sup>	1522.74 <sup>a</sup>	1177.36 <sup>b</sup>	1546.97 <sup>a</sup>	991.15 <sup>p</sup>	1211.93 <sup>p</sup>	1048.36 <sup>p</sup>	1239.19 <sup>p</sup>	1289.45 <sup>p</sup>	2792.58 <sup>v</sup>	3084.37 <sup>v</sup>	2670.10 <sup>v</sup>	2226.28 <sup>w</sup>	1957.57 <sup>w</sup>
Flavonols		1155.64 <sup>bc</sup>	1169.63 <sup>bc</sup>	1309.39 <sup>ab</sup>	1036.43°	1413.05 <sup>a</sup>	747.72 <sup>q</sup>	935.96 <sup>pq</sup>	872.33 <sup>pq</sup>	982.38 <sup>pq</sup>	1051.71 <sup>p</sup>	234.16 <sup>w</sup>	210.97 <sup>w</sup>	247.22 <sup>w</sup>	328.40 <sup>v</sup>	266.27 <sup>w</sup>
Flavonoides		1011.26 <sup>a</sup>	691.86 <sup>d</sup>	790.36 <sup>c</sup>	689.82 <sup>d</sup>	869.13 <sup>b</sup>	732.29 <sup>p</sup>	809.02 <sup>p</sup>	649.25 <sup>p</sup>	721.74 <sup>p</sup>	778.72 <sup>p</sup>	1038.22 <sup>v</sup>	1057.02 <sup>v</sup>	1071.22 <sup>v</sup>	829.42 <sup>w</sup>	782.76 <sup>w</sup>
Phenols		4593.00 <sup>a</sup>	3656.54°	4120.01 <sup>b</sup>	3226.23 <sup>d</sup>	4326.83 <sup>ab</sup>	2534.06 <sup>q</sup>	3448.68 <sup>p</sup>	3071.47 <sup>pq</sup>	3100.84 <sup>pq</sup>	3564.31 <sup>p</sup>	5145.38 <sup>v</sup>	5249.23 <sup>v</sup>	5206.12 <sup>v</sup>	3976.91 <sup>w</sup>	3952.06 <sup>w</sup>
EC50																
	ABTS	316.74 <sup>a</sup>	350.58ª	225.30 <sup>b</sup>	384.88 <sup>a</sup>	337.67 <sup>a</sup>	155.35 <sup>t</sup>	335.07 <sup>q</sup>	300.45 <sup>rs</sup>	347.78 <sup>p</sup>	293.33 <sup>rs</sup>	$140.34^{\mathrm{w}}$	85.86 <sup>x</sup>	104.93 <sup>wx</sup>	101.89 <sup>wx</sup>	215.93 <sup>v</sup>
	Reduced power	137.38 <sup>bc</sup>	147.32 <sup>ab</sup>	114.87 <sup>c</sup>	169.45 <sup>a</sup>	82.26 <sup>d</sup>	177.51 <sup>q</sup>	171.54 <sup>q</sup>	272.09 <sup>p</sup>	145.0 <sup>q</sup>	166.92 <sup>q**</sup>	62.48 <sup>vw</sup>	64.07 <sup>vw</sup>	72.71 <sup>v</sup>	60.63 <sup>w</sup>	45.54 <sup>x</sup>

Values are the means of the five different *Prunus persica* by-products samples (n=3). Different superscripts for the same quality parameter mean significant differences among cultivars p < 0.05. For any parameter, different letters a – e, p-t and v-z indicate significant differences among young, mature leaves and stems, respectively of the five cultivars.

			Young lea	ves			1	Mature lea	ves		Stems						
Phenolic compounds	Sweet Cap	Early Maycrest	O'Henry	Flordastar	Rubirich	Sweet Cap	Early Maycrest	O'Henry	Flordastar	Rubirich	Sweet Cap	Early Maycrest	O'Henry	Flordastar	Rubirich		
Hydroxycinnamic acid derivatives																	
Neochlorogenic acid	45.55 <sup>a</sup>	42.82 <sup>ab</sup>	40.06 <sup>ab</sup>	31.97 <sup>b</sup>	51.41 <sup>a</sup>	38.73 <sup>p</sup>	17.29 <sup>q</sup>	14.09 <sup>q</sup>	17.53 <sup>q</sup>	15.13 <sup>q</sup>	17.16 <sup>v</sup>	$14.82^{vw}$	15.93 <sup>vw</sup>	9.00 <sup>x</sup>	11.82 <sup>wx</sup>		
Chlorogenic acid	178.17 <sup>a</sup>	161.37 <sup>ab</sup>	166.48 <sup>a</sup>	142.69 <sup>b</sup>	178.25 <sup>a</sup>	139.10 <sup>p</sup>	129.21 <sup>p</sup>	128.80 <sup>p</sup>	127.34 <sup>p</sup>	135.13 <sup>p</sup>	8.01 <sup>x</sup>	59.27 <sup>w</sup>	66.59 <sup>v</sup>	10.42 <sup>x</sup>	9.71 <sup>x</sup>		
Caffeic acid	18.40 <sup>a</sup>	13.33 <sup>b</sup>	13.71 <sup>b</sup>	14.76 <sup>b</sup>	16.36 <sup>ab</sup>	22.28 <sup>p</sup>	17.11 <sup>q</sup>	12.85 <sup>r</sup>	15.20 <sup>qr</sup>	14.74 <sup>qr</sup>	5.49 <sup>v</sup>	3.33 <sup>wx</sup>	4.12 <sup>w</sup>	2.56 <sup>x</sup>	2.75 <sup>x</sup>		
Coumaric acid	12.59 <sup>b</sup>	13.16 <sup>b</sup>	10.14 <sup>b</sup>	9.80 <sup>b</sup>	19.10 <sup>a</sup>	11.90 <sup>q</sup>	9.02 <sup>q</sup>	34.17 <sup>p</sup>	7.34 <sup>q</sup>	10.28 <sup>q</sup>	12.79 <sup>v</sup>	8.07 <sup>w</sup>	11.30 <sup>vw</sup>	7.68 <sup>w</sup>	7.91 <sup>w</sup>		
Ferulic acid	1.70 <sup>d</sup>	2.60 <sup>bc</sup>	3.06 <sup>ab</sup>	2.28 <sup>cd</sup>	3.30 <sup>a</sup>	2.21 <sup>r</sup>	2.95 <sup>pq</sup>	3.00 <sup>pq</sup>	2.38 <sup>qr</sup>	3.58 <sup>p</sup>	3.59 <sup>v</sup>	3.04 <sup>v</sup>	4.06 <sup>v</sup>	3.39 <sup>v</sup>	3.50 <sup>v</sup>		
Flavan-3-ols acids																	
Catechin	24.61 <sup>a</sup>	31.29 <sup>a</sup>	36.33 <sup>a</sup>	31.98 <sup>a</sup>	30.66 <sup>a</sup>	46.59 <sup>pq</sup>	51.63 <sup>p</sup>	39.98 <sup>pq</sup>	44.33 <sup>pq</sup>	36.05 <sup>q</sup>	177.00 <sup>v</sup>	187.47 <sup>v</sup>	199.98 <sup>v</sup>	170.09 <sup>v</sup>	172.67 <sup>v</sup>		
Epicatechin	31.82 <sup>a</sup>	40.46 <sup>a</sup>	46.98 <sup>a</sup>	41.35 <sup>a</sup>	39.65ª	60.24 <sup>pq</sup>	66.77 <sup>p</sup>	51.70 <sup>pq</sup>	57.33 <sup>pq</sup>	46.62 <sup>q</sup>	42.26 <sup>v</sup>	33.80 <sup>w</sup>	37.43 <sup>vw</sup>	33.51 <sup>w</sup>	37.13 <sup>vw</sup>		
Hydroxybenzoic acids																	
Gallic acid	24.17 <sup>a</sup>	23.52 <sup>ab</sup>	18.20 <sup>b</sup>	19.60 <sup>ab</sup>	21.72 <sup>ab</sup>	30.46 <sup>p</sup>	24.47 <sup>q</sup>	17.06 <sup>r</sup>	20.19 <sup>r</sup>	19.57 <sup>r</sup>	7.29 <sup>vw</sup>	4.59 <sup>w</sup>	5.46 <sup>w</sup>	6.45 <sup>vw</sup>	8.83 <sup>v</sup>		
Flavonols																	
Quercetin-3-rutinoside	170.12 <sup>b</sup>	190.31 <sup>ab</sup>	206.37 <sup>a</sup>	183.49 <sup>b</sup>	179.56 <sup>b</sup>	130.46 <sup>p</sup>	163.49 <sup>p</sup>	153.66 <sup>p</sup>	169.12 <sup>p</sup>	158.16 <sup>p</sup>	63.88 <sup>w</sup>	72.62 <sup>vw</sup>	83.78 <sup>v</sup>	83.15 <sup>v</sup>	71.06 <sup>vw</sup>		
Quercetin-3-galactoside	188.55°	214.51 <sup>ab</sup>	232.16 <sup>a</sup>	203.36 <sup>ab</sup>	199.00 <sup>bc</sup>	159.29 <sup>p</sup>	198.91 <sup>p</sup>	168.63 <sup>p</sup>	200.42 <sup>p</sup>	175.20 <sup>p</sup>	70.79 <sup>w</sup>	98.16 <sup>v</sup>	90.92 <sup>v</sup>	89.86 <sup>v</sup>	87.52 <sup>vw</sup>		
Anthocyanins																	
Cyanidin-3-glucoside	0.19 <sup>a</sup>	0.06 <sup>bc</sup>	0.07 <sup>b</sup>	0.01 <sup>d</sup>	0.03 <sup>cd</sup>	0.44 <sup>q</sup>	0.67 <sup>p</sup>	0.25 <sup>r</sup>	nd	0.04 <sup>s</sup>	0.09 <sup>vw</sup>	0.02 <sup>x</sup>	0.02 <sup>wx</sup>	0.11 <sup>v</sup>	$0.08^{vwx}$		
Cyanidin-3-rutinoside	0.14 <sup>b</sup>	nd	0.22 <sup>a</sup>	nd	nd	1.05 <sup>p</sup>	1.13 <sup>p</sup>	0.13 <sup>q</sup>	nd	nd	0.11 <sup>w</sup>	0.09 <sup>w</sup>	0.08 <sup>w</sup>	0.32 <sup>v</sup>	0.28 <sup>v</sup>		

**Table 3.** Individual phenolic compounds (mg 100g<sup>-1</sup> DW) identified in young, mature leaves and stems of five Prunus persica cultivars grown in the center of Tunisia

Values are the means of the five different *Prunus persica* by-products samples (n=3). Different superscripts for the same quality parameter mean significant differences among cultivars p<0.05. For any parameter, different letters a – e, p-t and v-z indicate significant differences among young, mature leaves and stems, respectively of the five cultivars.

				Young lea	ives				Mature le	aves			Stems					
Volatiles	LRI	Sweet Cap	Early Maycrest	O'Henry	Flordastar	Rubirich	Sweet Cap	Early Maycrest	O'Henry	Flordastar	Rubirich	Sweet Cap	Early Maycrest	O'Henry	Flordastar	Rubirich		
α-pinene	941	-	-	-	-	_	0.53 <sup>q</sup>	0.67 <sup>q</sup>	0.77 <sup>q</sup>	1.13 <sup>p</sup>	0.57 <sup>q</sup>	3.67 <sup>x</sup>	5.47 <sup>v</sup>	4.53 <sup>w</sup>	5.30 <sup>v</sup>	-		
Camphene	955	-	-	-	-	-	-	-	-	-	-	$0.20^{w}$	$0.30^{v}$	-	0.27 <sup>w</sup>	-		
Sabinene	977	-	-	-	-	-	-	-	-	-	-	$0.97^{\mathrm{w}}$	1.03 <sup>v</sup>	0.93 <sup>x</sup>	1.10 <sup>v</sup>	-		
β-pinene	982	-	-	-	-	-	-	-	-	-	-	2.83 <sup>w</sup>	3.27 <sup>v</sup>	2.57 <sup>y</sup>	2.67 <sup>x</sup>	-		
myrcene	933	$0.27^{ab}$	-	$0.47^{a}$	0.47 <sup>a</sup>	_	-	-	-	-	_	$20.87^{w}$	23.37 <sup>v</sup>	22.33 <sup>v</sup>	18.67 <sup>x</sup>	-		
a-terpinene	1020	-	-	-	-	-	-	-	-	-	-	0.50 <sup>x</sup>	$0.57^{v}$	0.53 <sup>w</sup>	0.50 <sup>x</sup>	-		
<i>p</i> -cymene	1028	-	-	-	-	-	-	-	-	-	-	1.77 <sup>w</sup>	$1.80^{w}$	2.20 <sup>v</sup>	1.53 <sup>x</sup>	-		
limonene	1032	0.60 <sup>a</sup>	-	0.43 <sup>ab</sup>	$0.47^{ab}$	0.27 <sup>b</sup>	-	_	-	-	_	9.37 <sup>w</sup>	10.20 <sup>v</sup>	10.27 <sup>v</sup>	8.63 <sup>x</sup>	-		
$(E)$ - $\beta$ -ocimene	1052	_	$0.17^{ab}$	_	0.13 <sup>ab</sup>	0.33 <sup>a</sup>	_	_	_	_	_	$0.77^{w}$	0.40 <sup>x</sup>	-	0.27 <sup>y</sup>	2.33 <sup>v</sup>		
γ-terpinene	1063	_	-	0.10 <sup>ab</sup>	0.17 <sup>a</sup>	-	-	_	-	-	_	1.77 <sup>y</sup>	2.43 <sup>w</sup>	2.63 <sup>v</sup>	1.93 <sup>x</sup>	-		
terpinolene	1090	0.73 <sup>a</sup>	0.30 <sup>a</sup>	1.23 <sup>a</sup>	0.97 <sup>a</sup>	0.43 <sup>a</sup>	0.73 <sup>pq</sup>	0.83 <sup>p</sup>	0.60 <sup>q</sup>	_	_	18.97 <sup>x</sup>	26.20 <sup>v</sup>	25.23 <sup>v</sup>	20.50 <sup>w</sup>	-		
Monoterpene hydrocarbones		1.60 <sup>ab</sup>	0.47 <sup>b</sup>	2.23 <sup>a</sup>	2.20 <sup>a</sup>	1.03 <sup>ab</sup>	1.27 <sup>pq</sup>	1.50 <sup>p</sup>	1.37 <sup>pq</sup>	1.13 <sup>q</sup>	0.57 <sup>r</sup>	61.67 <sup>x</sup>	75.03 <sup>v</sup>	71.23 <sup>w</sup>	61.37 <sup>x</sup>	2.33 <sup>y</sup>		
1,8-cineole	1034	_	0.43 <sup>ab</sup>	0.60 <sup>a</sup>	-	$0.40^{ab}$	0.63 <sup>q</sup>	0.70 <sup>q</sup>	1.07 <sup>pq</sup>	1.53 <sup>p</sup>	0.83 <sup>q</sup>	-	-	-	-	-		
artemisia ketone	1064	_	-	_	_	_	_	_	_	0.43	_	-	-	-	-	-		
(E)-tagetone	1141	-	-	-	-	-	-	-	-	-	-	$0.67^{w}$	0.53 <sup>x</sup>	0.57 <sup>x</sup>	0.33 <sup>y</sup>	1.23 <sup>v</sup>		
camphor	1145	_	-	-	-	0.20	-	_	0.50 <sup>x</sup>	-	_	$1.17^{w}$	-	-	-	1.50 <sup>v</sup>		
Oxygenated monoterpenes		-	0.43 <sup>ab</sup>	0.60 <sup>a</sup>	-	<b>0.60</b> <sup>a</sup>	<b>0.63</b> <sup>q</sup>	<b>0.70</b> <sup>q</sup>	1.57 <sup>p</sup>	1.97 <sup>p</sup>	0.83 <sup>q</sup>	1.83 <sup>w</sup>	0.53 <sup>x</sup>	0.57 <sup>x</sup>	0.33 <sup>y</sup>	2.73 <sup>v</sup>		
benzothiazole	1226	_	-	-	0.23	-	-	_	-	-	_	-	-	-	-	-		
cyclohexyl isothiocyanate	1236	-	-	-	0.23	-	-	-	-	-	_	0.90 <sup>x</sup>	0.60 <sup>z</sup>	0.77 <sup>y</sup>	$1.17^{w}$	1.83 <sup>v</sup>		
Nitrogen/sulfur derivatives		_	-	-	0.47	-	-	_	-	-	_	0.90 <sup>x</sup>	0.60 <sup>z</sup>	0.77 <sup>y</sup>	$1.17^{w}$	1.83 <sup>v</sup>		
Theaspirane I	1298	-	-	-	-	-	-	-	-	-	-	0.23 <sup>v</sup>	-	-	$0.20^{v}$	-		
Theaspirane II	1315	-	-	-	-	-	-	-	-	-	-	0.23 <sup>v</sup>	-	-	0.23 <sup>v</sup>	-		
Apocarotenoides		-	-	-	-	-	-	-	-	-	-	$0.47^{v}$	-	-	0.43 <sup>w</sup>	-		
$\beta$ -bourbonene	1385	_	-	$0.20^{a}$	-	$0.17^{a}$	-	_	-	-	_	-	-	-	-	-		
presilphiperfol-7-ene	1346	-	-	-	-	-	-	-	-	-	-	$0.67^{w}$	-	-	0.30 <sup>x</sup>	1.13 <sup>v</sup>		
7-epi-silphiperfol-5-ene	1345	-	-	-	-	-	-	-	-	-	-	0.90 <sup>y</sup>	0.60 <sup>z</sup>	$1.17^{w}$	1.07 <sup>x</sup>	2.03 <sup>v</sup>		
silphiperfol-6-ene	1350	-	-	-	-	-	-	-	-	-	-	0.13 <sup>w</sup>	-	-	$0.27^{v}$	-		
$\beta$ -caryophyllene	1419	1.20 <sup>a</sup>	0.10 <sup>b</sup>	0.67 <sup>ab</sup>	_	0.37 <sup>ab</sup>	_	_	-	_	_	0.87 <sup>x</sup>	0.57 <sup>z</sup>	1.53 <sup>w</sup>	0.60 <sup>y</sup>	2.17 <sup>v</sup>		
<i>trans-α</i> - bergamote	1437	_	_	_	_	-	_	_	_	_	_	0.77 <sup>y</sup>	0.53 <sup>z</sup>	1.60 <sup>v</sup>	0.90 <sup>x</sup>	$1.07^{w}$		

**Table 4**. Volatile compounds (% on total volatile compounds) evaluated in young, mature leaves and stems of five Prunus persica cultivars grown in the center of Tunisia.

Values are the means of the five different *Prunus persica* by-products samples (n=3). LRI: Linear retention indexes. Different superscripts for the same quality parameter mean significant differences among cultivars p < 0.05. For any parameter, different letters a – e, p-t and v-z indicate significant differences among young, mature leaves and stems, respectively of the five cultivars.

# Table 4. (continued)

				Young leav	/es				Mature le	aves			Stems				
Volatiles	LRI	Sweet Cap	Early Maycrest	O'Henry	Flordastar	Rubirich	Sweet Cap	Early Maycrest	O'Henry	Flordastar	Rubirich	Sweet Cap	Early Maycrest	O'Henry	Flordastar	Rubirich	
α-humulene	1455	-	-	-	-	0.30 <sup>a</sup>	-	-	-	-	-	0.47 <sup>x</sup>	-	0.77 <sup>w</sup>	0.23 <sup>y</sup>	0.83 <sup>v</sup>	
$(E)$ - $\beta$ -farnesene	1458	-	-	-	-	-	-	-	-	-	-	0.60 <sup>x</sup>	0.43 <sup>z</sup>	0.90 <sup>v</sup>	0.53 <sup>y</sup>	$0.77^{w}$	
Sesquiterpenes hydrocarbones		1.20 <sup>a</sup>	0.10 <sup>b</sup>	$0.87^{\mathrm{a}}$	-	0.83 <sup>a</sup>	-	-	-	-	-	<b>4.30</b> <sup>x</sup>	2.13 <sup>z</sup>	<b>5.97</b> <sup>w</sup>	<b>3.90</b> <sup>y</sup>	<b>8.00</b> <sup>v</sup>	
(E)-neroidol	1564	-	-	-	-	-	-	-	-	-	-	0.43 <sup>w</sup>	$0.57^{v}$	-	-	-	
Oxygenated sesquiterpenes		-	-	-	-	-	-	-	-	-	-	<b>0.43</b> <sup>w</sup>	0.57 <sup>v</sup>	-	-	-	
	<b>7</b> .00						a	0 ( <b>7</b> 1)	<b>2</b> 40 <sup>D</sup>	2.00 <sup>p</sup>	1.00 <sup>n</sup>						
1-pentanol	768	-	_ 	_ 	_ 	_ 	3.07 <sup>p</sup>	3.6 <sup>/p</sup>	$2.40^{p}$	3.23 <sup>p</sup>	4.20 <sup>p</sup>	-	-	- 	-	-	
hexanal	802	1.60 <sup>a</sup>	0.23	0.23	$0.40^{6}$	$0.40^{\circ}$	0.60 <sup>p</sup>	-	-	-	-	0.47*	0.50 <sup>x</sup>	0.57 <sup>w</sup>	0.30 <sup>y</sup>	1.67 <sup>v</sup>	
(Z)-2-hexenal	842	-	-	-	-	-	3.30 <sup>p</sup>	4.70 <sup>p</sup>	4.10 <sup>p</sup>	4.73 <sup>p</sup>	5.30 <sup>p</sup>	-	-	-	-	-	
(E)-2-hexenal	856	4.10 <sup>a</sup>	$0.60^{\circ}$	1.53 <sup>bc</sup>	1.70 <sup>b</sup>	1.00 <sup>bc</sup>	1.60 <sup>p</sup>	-	-	1.67 <sup>p</sup>	2.17 <sup>p</sup>	$0.70^{w}$	0.37 <sup>z</sup>	0.63 <sup>x</sup>	$0.47^{\rm y}$	1.97 <sup>v</sup>	
1-hexanol	869	-	-	-	-	-	0.67 <sup>p</sup>	0.73 <sup>p</sup>	-	-	-	-	-	-	-	-	
<i>n</i> -nonane	900	-	-	-	-	-	-	0.60 <sup>p</sup>	-	-	-	-	-	-	-	-	
heptanal	901	-	-	-	0.27	-	-	-	-	-	-	0.63 <sup>w</sup>	-	-	$0.60^{w}$	$0.80^{v}$	
benzaldehyde	962	85.40 <sup>b</sup>	95.13 <sup>a</sup>	86.33 <sup>b</sup>	88.73 <sup>b</sup>	91.30 <sup>ab</sup>	77.57 <sup>pq</sup>	76.23 <sup>pq</sup>	81.33 <sup>p</sup>	70.07 <sup>q</sup>	73.53 <sup>pq</sup>	$12.70^{x}$	3.70 <sup>y</sup>	2.33 <sup>z</sup>	$17.00^{w}$	48.70 <sup>v</sup>	
(Z)-3-hexenyl acetate	1008	0.37 <sup>b</sup>	0.10 <sup>c</sup>	0.63 <sup>a</sup>	_	0.20 <sup>bc</sup>	-	_	_	_	_	-	-	-	-	-	
2.6-dimethylnonane	1012	_	_	_	_	_	1.63 <sup>q</sup>	1.67 <sup>q</sup>	1.60 <sup>q</sup>	2.67 <sup>p</sup>	$2.27^{pq}$	-	-	-	-	-	
2.9-dimethylnonane	1024	_	_	_	_	_	2.53 <sup>q</sup>	2.93 <sup>q</sup>	2.23 <sup>q</sup>	4.47 <sup>p</sup>	3.47 <sup>pq</sup>	-	-	-	-	-	
benzyl alcool	1034	$0.57^{a}$	$0.47^{a}$	_	0.57 <sup>a</sup>	$0.37^{a}$	_	_	_	-	_	-	-	-	-	-	
4-methyldecane	1059	_	_	_	_	_	_	_	0.50 <sup>p</sup>	_	_	-	-	-	-	-	
1-octanol	1073	_	_	_	_	_	1 30 <sup>q</sup>	1 30 <sup>q</sup>	1 03 <sup>q</sup>	$0.20^{p}$	1 50 <sup>pq</sup>	-	-	-	-	-	
1-nonen-3-ol	1088	_	_	_	_	_	1.57 <sup>qr</sup>	1.63 <sup>qr</sup>	1.33 <sup>r</sup>	2 73 <sup>p</sup>	2 10 <sup>pq</sup>	-	_	_	-	-	
nonanal	1104	_	_	$0.40^{a}$	0.17 <sup>ab</sup>	_	-	-	-		_	$0.40^{w}$	0.53 <sup>v</sup>	$0.40^{w}$	0 33 <sup>x</sup>	_	
1-nonanol	1172	$2  10^{a}$	0.77 <sup>b</sup>	1.80 <sup>ab</sup>	$2.30^{a}$	0.87 <sup>b</sup>		_	_	_	_	10.40	9 90 <sup>y</sup>	11 13 <sup>w</sup>	0.55 7 77 <sup>z</sup>	24 90 <sup>v</sup>	
(7) 2 hexanyl butyrate	1192	2.10	0.77	0.72a	2.50	0.07	-	_	_	_	_	10.07	).)0	11.15	1.11	24.90	
decanal	1204	-	_	0.75	0 22ª	_	-	_	_	_	_	- 0.20 <sup>w</sup>	-	-	0.33 <sup>v</sup>	-	
1 avalahavulhavana	1204	-	-	-	0.23	-	-	-	-	-	-	0.50	- 0.52 <sup>x</sup>	- 0.72 <sup>w</sup>	0.33	- 1 20 <sup>v</sup>	
C mothyldodoono	1237	-	-	-	-	-	- 0.47pq	-	-	- 0.62 <sup>p</sup>	-	0.00	0.55	0.75	0.47	1.50	
(F) 2 trides are	1243	-	-	-	-	_	0.47 <sup>n</sup>	0.45	0.33	0.03 <sup>2</sup>	_	-	-	-	-	-	
(E)-2-tridecene	1305	-	-	-	-	-	0.4/11	0.301	-	0.60 <sup>4</sup>	- 0.77 <sup>00</sup>	-	-	-	-	-	
(Z)-2-tridecene	1315	-	-	-	-	-	0.83	0.604	0.531	0.97 <sup>p</sup>	0.//**	-	-	-	-	-	
3-methylundecanol	1326	-	-	-	-	-	0.53 <sup>q</sup>	0.33	0.37	$0.83^{p}$	-	-	-	-	-	-	
1-tetradecene	1393	-		-	0.17	-	-	-	-	-	-	-	-	-	-	-	
<i>n</i> -tetradecane	1400	0.53ª	0.20	0.47ª	0.40 <sup>a</sup>	-	-	-	-	-	-	0.70 <sup>y</sup>	0.93*	0.43 <sup>2</sup>	0.77 <sup>x</sup>	1.13 <sup>v</sup>	
1-cyclohexyloctane	1442	$0.20^{a}$	0.10 <sup>a</sup>	-	-	-	-	-	0.23 <sup>a</sup>	-	-	$0.40^{w}$	0.43 <sup>v</sup>	0.40 <sup>w</sup>	0.43 <sup>v</sup>	-	
<i>n</i> -pentadecane	1500	-	-	$0.50^{a}$	-	-	-	-	-	-	_	0.37 <sup>w</sup>	$0.50^{v}$	-	0.37 <sup>w</sup>	-	
(Z)-3-hexenyl benzoate	1570		-	-		0.27 <sup>a</sup>	-	-	-	-	-	-	-	-	-	-	
n-hexadecane	1600	0.57 <sup>b</sup>	$0.17^{c}$	1.03 <sup>a</sup>	0.30 <sup>bc</sup>	$0.30^{bc}$	-	-	-	-	-	$0.57^{x}$	0.73 <sup>w</sup>	0.53 <sup>y</sup>	0.83 <sup>v</sup>	-	
Others		95.43 <sup>a</sup>	<b>97.77</b> <sup>a</sup>	93.67 <sup>a</sup>	95.47 <sup>a</sup>	94.70 <sup>a</sup>	96.13 <sup>p</sup>	95.13 <sup>pq</sup>	95.77 <sup>pq</sup>	94.60 <sup>q</sup>	95.30 <sup>pq</sup>	<b>27.90</b> <sup>x</sup>	<b>18.13</b> <sup>y</sup>	<b>17.17</b> <sup>y</sup>	<b>29.67</b> <sup>w</sup>	80.47 <sup>v</sup>	
Total volatiles		98.23 <sup>a</sup>	98.77 <sup>a</sup>	97.37 <sup>a</sup>	98.13 <sup>a</sup>	97.17 <sup>ª</sup>	98.03 <sup>pq</sup>	97.33 <sup>q</sup>	98.70 <sup>p</sup>	97.70 <sup>pq</sup>	96.70 <sup>q</sup>	97.50 <sup>v</sup>	97.00 <sup>v</sup>	95.70 <sup>w</sup>	96.87 <sup>vw</sup>	95.37 <sup>w</sup>	

Values are the means of the five different *Prunus persica* by-products samples (n=3). LRI: Linear retention indexes. Different superscripts for the same quality parameter mean significant differences among cultivars p < 0.05. For any parameter, different letters a – e, p-t and v-z indicate significant differences among young, mature leaves and stems, respectively of the five cultivars.