

## Evaluation of the Antioxidant Capacity, Phenolic Compounds, and Vitamin C Content of Different Peach and Nectarine [*Prunus persica* (L.) Batsch] Breeding Progenies

CELIA M. CANTÍN, MARÍA A. MORENO, AND YOLANDA GOGORCENA\*

Departamento de Pomología, Estación Experimental de Aula Dei (CSIC), Apartado 13034, 50080 Zaragoza, Spain

Antioxidant capacity and contents of total phenolics, anthocyanins, flavonoids, and vitamin C were evaluated in 218 genotypes from 15 peach and nectarine breeding progenies. Significant differences were found among progenies on the fruit antioxidant profile, corroborated by the high contribution showed by cross to the phenotypic variance of each phytochemical trait analyzed (16–45%). Phytochemical profile varied depending on peach/nectarine and yellow/white flesh color qualitative traits. On the other hand, no significant effect of year was found on the bioactive profile of peaches and nectarines. Antioxidant capacity was linearly correlated to total phenolic content, but correlation varied depending on the progeny. No correlation was found for vitamin C versus any other phytochemical trait. The results suggest the importance of genetic background on the antioxidant profile of peaches and nectarines and stress its relevance for the ultimate objective of this work: selecting new peach and nectarine genotypes rich in bioactive compounds to benefit consumer's health.

**KEYWORDS:** *Prunus persica*; total phenolics; anthocyanins; flavonoids; vitamin C; antioxidant capacity; phytochemical profiling

### INTRODUCTION

The important role of diet in either promoting or preventing diseases has long been recognized, and in recent years, diet and human well-being have received unprecedented attention. Nowadays, there is a growing interest in bioactive compounds of fruits and vegetables due to their putative role in preventing diseases such as diabetes, cancer, stroke, arthritis, and also aging. A clear inverse relationship between the consumption of fruits and vegetables and incidence of cardio- and cerebrovascular, degenerative, and proliferative diseases and mortality has been largely proved by epidemiological studies (1). Fruits and vegetables are excellent functional foods as they are high in antioxidant compounds (2). These naturally occurring substances not only have a role in the visual appearance (pigmentation and browning) and taste (astringency) of fruits and vegetables but also have health-promoting properties, acting as antioxidants by scavenging harmful free radicals, which are implicated in most degenerative diseases (3).

The health benefits of fruits are due to their specific chemical composition, particularly to compounds of nutritional value such as phenolic acids, flavonoids, and vitamins (4). Peaches and nectarines, even though having a lower total antioxidant capacity than other fruits such as strawberry, apple, or orange (5), are nutritionally important because they are one of the most important commodities consumed worldwide. Polyphenols are secondary plant metabolites, and they are the main sources of

antioxidant capacity in peaches, although vitamin C and carotenoids also contribute to it (6). The basic feature of all polyphenols is the presence of one or more hydroxylated aromatic rings, which seemed to be responsible for their properties as radical scavengers (7). The flavonoids are a large class of phenolic compounds, present in cereals, vegetables, and fruits. Evidence is accumulating about their significant contribution to the antioxidant capacity of fruits and vegetables (8). Anthocyanins are natural colorants and, with flavanols and flavonols, are included in the flavonoid family. They are widely distributed among flowers, fruits, and vegetables and, in addition to their colorful characteristics, they have potent antioxidant properties modulated by their different hydroxylations and glycosylations (3). The main anthocyanins reported in peach are cyanidin-3-glucoside and cyanidin-3-rutinoside (9). Besides their relevance in the appearance, taste, and flavor of fruits as well as their health-promoting properties (10), phenolic compounds have been found to increase the shelf life of food and inhibit the growth of pathogenic microorganisms due to their natural antimicrobial properties (11). Vitamin C is a water-soluble antioxidant and is, as are vitamin E and  $\beta$ -carotene, referred to as an antioxidant vitamin. Humans are unable to synthesize vitamin C and are thus entirely dependent upon dietary sources to meet needs. More than 90% of the vitamin C in the human diet is supplied by fruits and vegetables (12). These benefits and the increasing consumer interest in functional foods have guided breeders of different crops to consider antioxidant compounds and other nutritional properties as interesting targets in breeding programs (13, 14).

\*Corresponding author (telephone +34 976 71 61 33; fax +34 976 71 61 45; e-mail [aoiz@eed.csic.es](mailto:aoiz@eed.csic.es)).

**Table 1.** Peach and Nectarine Commercial and Experimental (VAC-) Cultivars Used as Progenitors in the 15 Controlled Crosses<sup>a</sup>

cultivar	fruit type		flesh color	stone
Andross	round	peach	yellow	cling
Babygold-9	round	peach	yellow	cling
Big Top	round	nectarine	yellow	cling
Calante	round	peach	yellow	cling
Crown Princess	round	peach	yellow	cling
O'Henry	round	peach	yellow	free
Orion	round	peach	yellow	free
Red Top	round	peach	yellow	free
Rich Lady	round	peach	white	free
VAC-9510	round	peach	yellow	cling
VAC-9511	round	peach	yellow	free
VAC-9512	round	peach	yellow	free
VAC-9513	round	nectarine	yellow	free
VAC-9514	round	nectarine	white	free
VAC-9515	round	nectarine	yellow	free
VAC-9516	round	peach	white	free
VAC-9517	flat	peach	white	free
VAC-9520	round	peach	yellow	free
Venus	round	nectarine	yellow	free

<sup>a</sup> Fruit type (round or flat, peach or nectarine), flesh color (yellow or white), and stone adherence (free or cling) for each progenitor is shown.

The phytochemical content of fruit is influenced by numerous factors such as genotype, rootstock, climatic conditions, agronomic practices, harvesting time, and postharvest conditions (6, 11, 14, 15). Moreover, phenolic compounds are not uniformly distributed within the tissue of fruits, and most of them are concentrated in the epidermal and subepidermal layers of the fruit (11). Phenolic distribution is an important aspect of the overall phenolic composition and antioxidant capacity because, due to its characteristics, the peach skin is usually not eaten and therefore it does not contribute to the human diet intake.

The aim of the present work was to screen and compare 218 genotypes from 15 different peach and nectarine breeding progenies by measuring their contents of total phenolics, total flavonoids, total anthocyanins, vitamin C, and relative antioxidant capacity. We also wanted to study the influence of genotype, genetic origin, pomological traits, and year in the bioactive profile of peach and nectarine fruits. The ultimate objective of this study was to select peach genotypes with enhanced antioxidant capacity fruits that will benefit consumers with health-promoting properties.

## MATERIALS AND METHODS

**Chemicals.** All chemicals were of analytical grade. Folin–Ciocalteu's phenol reagent, 6-hydroxy-2,5,7,8-tetramethylchromane-2-carboxylic acid (Trolox), 2,2-dipyridyl-1,1-diphenyl-2-picrylhydrazyl (DPPH), 3,4,5-trihydroxybenzoic acid (gallic acid), sodium carbonate (Na<sub>2</sub>CO<sub>3</sub>), catechin, trichloroacetic acid (TCA), and ascorbic acid (vitamin C) were purchased from Sigma-Aldrich (Steinheim, Germany).

**Plant Material.** Fifteen controlled biparental crosses between 19 peach and nectarine cultivars (Table 1) were made during 2000 and 2001 to develop superior peach and nectarine cultivars for the Spanish industry. The resulting seedling trees (one tree per genotype) were grafted on the same rootstock (GF-677) and established in an experimental orchard at the Experimental Station of Aula Dei-CSIC (northern Spain, Zaragoza) in 2002. Trees were trained to the standard open vase system and planted at a spacing of 4 m × 2.5 m. Hand thinning was carried out to reduce fruit load on the heavily loaded trees. They were grown under usual conditions of irrigation, fertilization, and pest control. Vegetative and fruit quality traits were evaluated in a total of 1111 genotypes over three consecutive years (2005–2007). All traits were measured or scored for each seedling separately over the three year period, and means of three years were calculated. Phytochemical composition (total phenolics, total flavonoids, total anthocyanins, and total antioxidant capacity) was studied in 218

genotypes that were common at least for two years to estimate the seasonal effect on phytochemical profile. Vitamin C was also determined in all of the genotypes in the last year of study to corroborate the variability found in other bioactive compounds in the previous years and its contribution to the antioxidant capacity of fruits. The studied genotypes were selected among the descendants from the 15 crosses because of their higher fruit quality. For all analyses, only fruit flesh was used, as it is usually consumed. Fruits were peeled with a sharp knife, and flesh was weighed, immediately frozen separately in liquid nitrogen, and stored at –20 °C until analysis. Samples for vitamin C determination were kept at –20 °C in 5% metaphosphoric acid for preservation of ascorbic acid until analysis.

**Quality Parameters.** During the years 2005, 2006, and 2007, fruit quality parameters were measured individually in each seedling tree. Fruits were hand-picked at commercial maturity, assessed by peel fruit color and flesh firmness. Yield (kg/tree) was measured, and total number of fruits was counted for each genotype. From these variables, total average fruit weight was calculated. Ten fruits from each plant were randomly selected for the quality evaluations. Some quality traits such as fruit type (peach/nectarine), flesh color (yellow/white), and endocarp staining were scored. Fruit type was scored on a 1–2 scale as peach (1) or nectarine (2). Similarly, flesh color was scored as (1) yellow or (2) white. Endocarp staining (redness around stone) was scored on an increasing scale from no color (1) to high redness (10). The soluble solids content (SSC) of the juice was measured with a temperature-compensated refractometer (model ATC-1, Atago Co., Tokyo, Japan), and data are given as °Brix. The titratable acidity (TA) was determined by titration with 0.1 N NaOH to pH 8.1. Data are given as grams of malic acid per 100 g of fresh weight (FW), because this is the dominant organic acid in peach.

**Phytochemical Analysis.** The frozen fruit material (5 g) was homogenized with a Polytron (2 min on ice) with 10 mL of extraction solution, consisting of 0.5 N HCl in methanol/Milli-Q water (80% v/v). The mixture was incubated overnight at 4 °C and then centrifuged for 20 min at 4 °C and 20000g. Supernatant was recovered and the volume measured. This hydroalcoholic extract was used for total phenolics, anthocyanins, flavonoids, and antioxidant capacity assays.

The content of phenolic compounds in methanol extracts was determined according to the Folin–Ciocalteu method (16). The method consisted of mixing 500 μL of the extract diluted in water with 500 μL of Folin–Ciocalteu's reagent. After 3 min of reaction, 1 mL of 1 N sodium carbonate (Na<sub>2</sub>CO<sub>3</sub>) was added. The tubes were mixed for 15 s and then allowed to stand for 60 min at 20 °C. Absorbance was measured at 725 nm using a spectrophotometer (Beckman Coulter DU 800). The standard calibration curves were daily prepared using gallic acid (3,4,5-trihydroxybenzoic acid). The phenolic content was expressed in milligrams of gallic acid equivalents (GAE) per 100 g of FW.

Total flavonoids content was determined using a colorimetric assay based on the method of Zhishen et al. (17). One milliliter of the methanolic extract was diluted with water (1:2), and 0.3 mL of 5% NaNO<sub>2</sub> was added. After 5 min, 0.3 mL of 10% AlCl<sub>3</sub> were added. After 1 min, 2 mL of 1 N NaOH was added, and the solution was mixed by vortex. Absorbance at 510 nm was measured against a blank with a spectrophotometer (Beckman Coulter DU 800). The results were expressed as milligrams of catechin equivalents (CE) per 100 g of FW on the basis of a standard curve using catechin as standard.

Total anthocyanin content of the hydroalcoholic extracts was determined using the method of Fuleki and Francis (18) adapted to peach tissue. Aliquots of the clear methanol extract were used for spectrophotometric readings at 535 nm by subtracting the absorbance at 700 nm (due to turbidity). The spectrophotometer was zeroed with the anthocyanins extraction solvent as the blank. Anthocyanins were quantified as milligrams of cyanidin-3-glucoside per kilogram of FW using a molar extinction coefficient of 25965 cm<sup>-1</sup> M<sup>-1</sup> and a molecular weight of 494.

Vitamin C was determined using the method for the spectrophotometric determination of ascorbic acid (vitamin C) as described by Zaharieva and Abadia (19). Samples were homogenized with 5% metaphosphoric acid at 4 °C. Then, they were centrifuged at 20000g for 15 min at 4 °C, and the supernatant was immediately used for vitamin C analysis. Absorbance was measured at 525 nm using a spectrophotometer (Beckman Coulter DU 800). The standard calibration curve was daily prepared using ascorbic acid as standard. Vitamin C was expressed as milligrams of ascorbic acid (AsA) per 100 g of FW.

**Table 2.** Basic Statistics Based on Single Plant Observations for the Seedlings from 15 F1 Peach and Nectarine Progenies Studied over 3 Years, for Total Phenolics, Flavonoids, Anthocyanins, Vitamin C, and Antioxidant Capacity (RAC)<sup>a</sup>

trait	N	min	max	mean	MSE	SD
total phenolics (mg of GAE/100 g of FW)	218	12.7	71.3	36.4	1.0	15.2
flavonoids (mg of CE/100 g of FW)	218	1.8	30.9	8.8	0.4	6.0
anthocyanins (mg of C3GE/kg of FW)	218	0.1	26.7	3.0	0.3	4.0
vitamin C (mg of AsA/100 g of FW)	218	1.2	9.1	3.7	0.1	1.5
RAC ( $\mu$ g of Trolox/g of FW)	218	227.3	629.9	405.0	4.9	73.0

<sup>a</sup> For each trait, number of observed seedlings (N), minimum, maximum, mean value, mean standard error (MSE), and standard deviation (SD) are presented. Abbreviations: GAE, gallic acid equivalents; CE, catechin equivalents; C3GE, cyanidin-3-glucoside equivalents; AsA, ascorbic acid; RAC, relative antioxidant capacity.

The antioxidant capacity was measured using the DPPH method adapted from Brand-Williams et al. (20). Briefly, 100  $\mu$ L of the methanolic extract was added to 2.9 mL of fresh DPPH radical solution (98.9  $\mu$ M in methanol) and mixed in the dark by vortex at room temperature. The absorbance of the samples was measured at 515 nm after 10 min. These readings were used for calculation of the relative antiradical capacity (RAC), which indicates the antiradical capacity of the sample compared to Trolox for a specific reaction time (10 min). For each sample, three separate determinations were carried out. The standard calibration curves were prepared daily using Trolox (6-hydroxy-2,5,7,8-tetramethylchromane-2-carboxylic acid). Results were expressed in micrograms of Trolox per gram of FW.

**Statistical Analyses.** All statistical analyses were performed using SPSS 17.0 (SPSS Inc., Chicago, IL). To obtain basic statistics for the entire plant material studied, maximum and minimum values, mean, mean standard error (MSE), and standard deviation (SD) were calculated for each trait. Data for each genotype in the three years of study were averaged, and mean values were used as estimated genotypic values. The significance of cross, year, and cross  $\times$  year interaction effects on phytochemical profile was tested on the 218 genotypes by analysis of variance (ANOVA). Duncan's multiple-range test ( $P \leq 0.05$ ) was used to estimate progeny means and to find differences in phytochemical profile among crosses. A *t* test ( $P \leq 0.05$ ) was run to compare different fruit types. Finally, correlations were calculated with raw data of the three years, according to Pearson's test at  $P \leq 0.01$ .

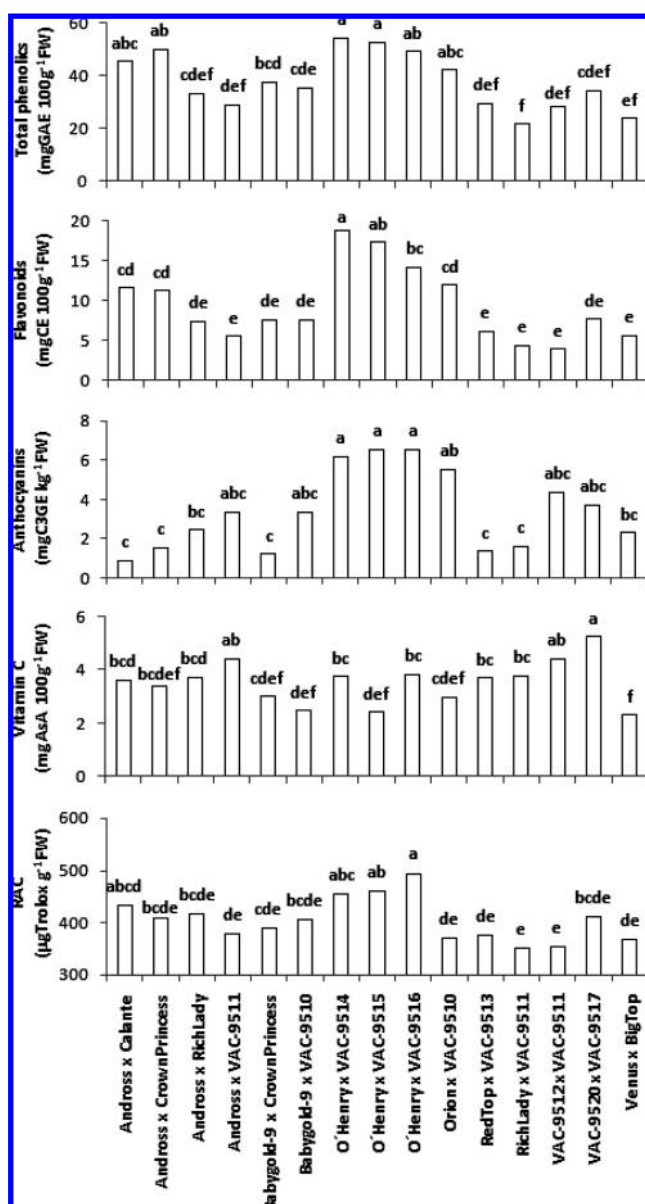
## RESULTS AND DISCUSSION

**Table 2** shows the ranges of bioactive compounds and total antioxidant activity in peaches and nectarines. Total phenolics, as determined by the Folin–Ciocalteu assay, varied among genotypes with values in the range of 12.7–71.3 mg of GAE per 100 g of FW. Values are within the range reported for peach flesh in the literature, namely, 14–77 mg of GAE per 100 g of FW (9, 14, 21). Total flavonoids content ranged from 1.8 to 30.9 mg of CE per 100 g of FW, with an average of 8.8 mg of CE per 100 g of FW. Total anthocyanins greatly varied among genotypes [0.1–26.7 mg of cyanidin-3-glucoside equivalents (C3GE) per kg of FW] depending on the red pigmentation of the flesh. Genotypes with red flesh had higher anthocyanins content. Values of flavonoids and anthocyanins in this range have been reported by other authors (6, 9). Higher values of anthocyanins content in peaches and nectarines are found in the literature when skin is included in the sample (9) due to unequal distribution of phenolic compounds in the flesh (~30%) and the skin (~70%) of the peach fruit (11, 22). On average, unpeeled fruit contained 1.5-fold higher levels of phenolics than peeled fruit (22). However, as already mentioned above, peach skin is not usually appreciated by consumers and, therefore it takes no part in the human diet. The total ascorbic acid (vitamin C) content greatly varied from approximately 1 to 9 mg of AsA/100 g of FW, with a mean value of 3.7 mg of AsA/100 g of FW. Genetic background of

the genotype is a much more important factor than climatic conditions and cultural practices in producing fruit with high vitamin C content at harvest (15). Values were in the same range as previously reported for vitamin C contents in peach flesh, namely, 1–14 mg of AsA/100 g of FW (6, 21, 23) and lower than values obtained when peach peel was included in the test (6). As for phenolic compounds, skin tissues have more vitamin C to protect the fruit from outside stress caused by light and oxidation (17). The relative antioxidant capacity (RAC) varied among genotypes, with values ranging from 227.3 to 629.9  $\mu$ g of Trolox/g of FW, with an average of 405  $\mu$ g of Trolox/g of FW. In recent years, strong attention has been given to this trait as an eligible parameter for fruit quality because many of the biological actions of phytochemicals have been attributed to it. As for anthocyanins, the antioxidant capacity observed in our study was in the range previously reported for peach flesh (100–1000  $\mu$ g of Trolox/g of FW), but lower than in other studies in which peel was included in the test sample (700–6000  $\mu$ g of Trolox/g of FW) (6, 11). Therefore, the antioxidant capacity of the fruits decreases when they are peeled.

Considerable variation was found in the content of antioxidant compounds in the fruits from different progenies (**Figure 1**). The highest total phenolic contents were shown by the three progenies descendant from O'Henry cultivar, although no significant differences were found with mean values of Andross  $\times$  Calante, Andross  $\times$  Crown Princess, Babygold-9  $\times$  Crown Princess, and Orion  $\times$  VAC-9510 progenies. The level of total phenolics in O'Henry  $\times$  VAC-9514 was > 2-fold higher than in Rich Lady  $\times$  VAC-9511, showing the wide variance of total phenolic concentrations in *Prunus persica* already reported in other studies (6, 9, 14, 21). Similarly, a high variability in the flavonoids content averaged for the 15 progenies was found in agreement with the variability among *P. persica* cultivars reported by other authors (9). A nearly 5-fold difference was measured between the lowest and highest mean values among different progenies. The highest flavonoids content was also shown by O'Henry  $\times$  VAC-9514 progeny, although no significant difference was found with O'Henry  $\times$  VAC-9515. The highest values were found in the three O'Henry progenies, resulting in the most interesting crosses from which to select peaches and nectarines with higher flavonoid content in the flesh. The O'Henry progenies also showed the highest anthocyanins content, although no significant differences were found with Andross  $\times$  VAC-9511, Babygold-9  $\times$  VAC-9510, Orion  $\times$  VAC-9510, VAC-9512  $\times$  VAC-9511, and VAC-9520  $\times$  VAC-9517 progenies. Tomás-Barberán et al. (9) reported higher anthocyanin content in the flesh of O'Henry fruits (8.1 mg/kg of FW) when compared with other commercial peach cultivars such as September Sun (3.7 mg/kg of FW), Rich Lady, and Spring Lady (no significant amounts detected in either). In agreement with all of these results, we could report an influence of O'Henry cultivar to induce higher anthocyanins content in its progeny as observed for flavonoids content. On the other hand, the highest vitamin C content was found in the VAC-9520  $\times$  VAC-9517 progeny, although differences were not significant with Andross  $\times$  VAC-9511 and VAC-9512  $\times$  VAC-9511 progenies. The lowest mean value was shown by Venus  $\times$  Big Top nectarines progeny without being significantly different from Andross  $\times$  Crown Princess, both Babygold-9 progenies, O'Henry  $\times$  VAC-9515, and Orion  $\times$  VAC-9510 progenies. A significant effect of cultivar and rootstock on the vitamin C content has been previously reported in different fruits (15). Tavarini et al. (21) found a range from 1 to 14 mg of AsA/100 g of FW in seven peach cultivars, and Nelson et al. (24) reported values from 19.3 to 71.5 mg of AsA/100 g of FW in six strawberry cultivars. Significant differences among progenies were also found for RAC (**Figure 1**), according

to previous results that have shown that antioxidant capacity changes as a function of cultivar and rootstock (6, 14, 21). As above-mentioned for other bioactive compounds, the highest



**Figure 1.** Phytochemical profiles of the 15 peach and nectarine progenies. Data are the means of all of the genotypes in each progeny. For each trait, means with the same letter are not significantly different according to Duncan's test ( $P \leq 0.05$ ). Abbreviations: GAE, gallic acid equivalents; CE, catechin equivalents; C3GE, cyanidin-3-glucoside equivalents; AsA, ascorbic acid; RAC, relative antioxidant capacity.

RAC values were shown by O'Henry descendants, without being significantly different from the Andross  $\times$  Calante progeny. These results indicate the importance of cultivar and genotype for determining the antioxidant potential and phenolic content of the fruit. The most appropriate combination of phytochemical traits must be considered for the selection of new genotypes with higher nutritional value.

Bioactive compounds and antioxidant content of fruit varied depending peach/nectarine and yellow/white flesh color qualitative traits (Table 3). In this work, peaches showed higher phenolic content than nectarines, and phenolic content of white-fleshed fruit was higher than that of yellow-fleshed fruit. This shows a tendency of white-fleshed peaches to have significantly higher antioxidants content than the other genotypes tested (yellow-fleshed peaches and nectarines). No significant differences in flavonoids and anthocyanins content were found between peaches and nectarines; however, they were significantly higher in white-fleshed fruits than in yellow-fleshed fruits as previously found (9). This could be explained by the red pigmentation due to anthocyanins in the white-fleshed fruit, especially in the flesh area surrounding the stone, usually found in our studied progenies. This result is different from what occurs in the peel, where yellow-fleshed fruits are reported to produce more anthocyanin pigments than white-fleshed fruits (9). No significant differences were found for vitamin C between peach and nectarine fruits, whereas it was higher in white-fleshed fruit than in yellow-fleshed fruit. Consequently, with all of these results, white-fleshed fruits showed higher antioxidant capacity than the yellow-fleshed ones, as reported in previous works (6). In agreement with these results, significant slight positive correlations ( $P \leq 0.01$ ) were found for color flesh fruit versus phenolic compounds, flavonoids, anthocyanins, vitamin C, and RAC ( $r = 0.265$ ,  $r = 0.283$ ,  $r = 0.189$ ,  $r = 0.339$ , and  $r = 0.243$ , respectively), indicating different contents of these bioactive compounds in white- and yellow-fleshed fruits, as commented above.

In agreement with these results, the percentage of phenotypic variance explained by cross (Table 4) was high for each bioactive compound analyzed (between 15.7 and 44.6%). Contribution of cross to anthocyanins and antioxidant capacity was lower than to total phenolics, flavonoids, and vitamin C phenotypic variance. These results indicate that cultivar and genotype are decisive in determining the peach fruit antioxidant capacity. On the other hand, no significant differences were found among the three years of study for total phenolics, flavonoids, anthocyanins and antioxidant capacity (Table 4). Despite this result, slight higher flavonoids and anthocyanins contents were observed in the first year of study when compared with the two following years (data not shown), which may be due to differences of climate including temperature, sun irradiation, and/or water stress as mentioned by Tomas-Barberán and Espín (10). Sun irradiation has been demonstrated to increase anthocyanin content of different fruits such as apples and pears, whereas in cherry, grape, and plum,

**Table 3.** Total Phenolics, Flavonoids, Anthocyanins, Vitamin C, and Relative Antioxidant Capacity (RAC) in Different *P. persica* Fruit Types<sup>a</sup>

fruit type	<i>N</i>	total phenolics (mg of GAE/100 g of FW)	flavonoids (mg of CE/100 g of FW)	anthocyanins (mg of C3GE/kg of FW)	vitamin C (mg of AsA/ 100 g of FW)	RAC ( $\mu$ g of Trolox/g of FW)
peach	192	37.2 a	9.1 a	3.1 a	3.7 a	406.2 a
nectarine	26	30.5 b	6.9 a	2.2 a	3.9 a	395.7 a
yellow fleshed	176	34.5 b	8.0 b	2.6 b	3.5 b	396.5 b
white fleshed	42	44.8 a	12.3 a	4.5 a	4.8 a	442.2 a

<sup>a</sup> For each fruit type, number of observed seedlings (*N*) is presented. Data are means over the three years of study. In each trait column (peach, nectarine, yellow fleshed, white fleshed), means with the same letter are not significantly different according to *t* test ( $P \leq 0.05$ ). Abbreviations: GAE, gallic acid equivalents; CE, catechin equivalents; C3GE, cyanidin-3-glucoside equivalents; AsA, ascorbic acid.

light seems not to be essential for red color formation (10). Temperature, and in particular the difference between day and night temperatures, has been reported to have a marked effect on anthocyanin accumulation in apples, plums, grapes, and pomegranates (10).

**Correlations among Phytochemical Constituents and Other Fruit Quality Traits.** A high positive correlation was found between total phenolics and flavonoids content ( $r = 0.742$ ,  $P \leq 0.01$ ), implying that flavonoids are an important group of phenolic compounds in peaches and nectarines (Table 5). Moreover, a linear positive relationship (Figure 2) was observed between antioxidant capacity and total phenolics for the flesh of the peach and nectarine genotypes, as has been observed for peaches, apricots, and plums (5, 6). However, higher correlation coefficients ( $r > 0.9$ ) were obtained by Gil et al. (6) for other peach and nectarine cultivars. This variation could be due to differences in the phytochemical profile of different peach and nectarine cultivars. In addition, the large phenotypic variability within the

**Table 4.** Factors Affecting Phytochemical Profile in 15 Peach and Nectarine Progenies Studied over 3 Years<sup>a</sup>

variable	F value	P	phenotypic variance (%)
total phenolics			
cross	16.79	0.000	33.6
year	1.22	0.295	0.5
cross × year	0.27	1.000	1.5
flavonoids			
cross	26.69	0.000	44.6
year	0.15	0.865	0.1
cross × year	0.42	0.996	2.4
anthocyanins			
cross	6.21	0.000	15.7
year	1.09	0.336	0.5
cross × year	0.36	0.999	2.0
vitamin C			
cross	8.02	0.000	35.8
year	—	—	—
cross × year	—	—	—
RAC			
cross	8.08	0.000	19.6
year	0.98	0.375	0.4
cross × year	0.33	1.000	1.9

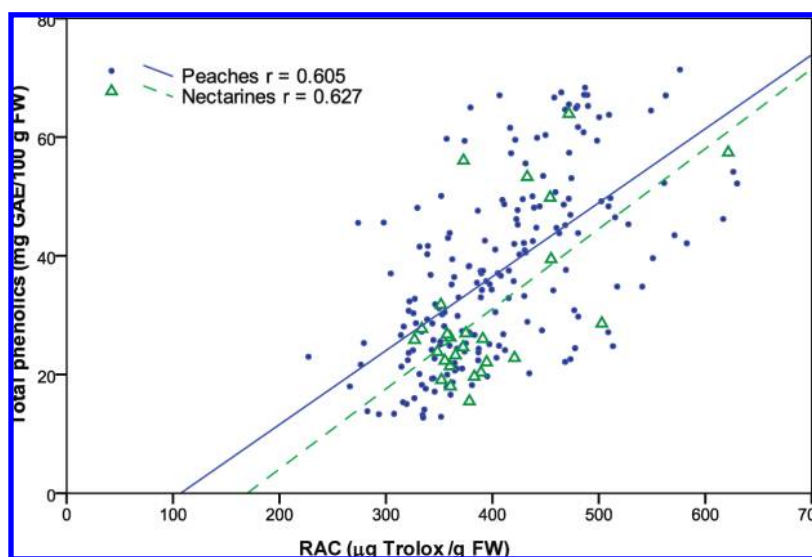
<sup>a</sup> F values and proportion (%) of phenotypic variance are indicated as determined by ANOVA. (—) no data available. Abbreviations: RAC, relative antioxidant capacity.

breeding progenies in our study could induce lower correlation coefficients between those parameters. Total phenolics and flavonoids were the only constituents that correlated significantly ( $P \leq 0.01$ ) with antioxidant capacity ( $r = 0.606$  and  $r = 0.553$ , respectively), indicating that they are important bioactive compounds contributing to the antioxidant capacity of peaches and nectarines, in accordance with previous studies on different peach, nectarine, and plum cultivars (6, 11). Indeed, correlation coefficients varied depending on the progenies. Higher correlation coefficients ( $P \leq 0.01$ ) were found between total phenolics and RAC in some progenies, such as Rich Lady × VAC-9511 ( $r = 0.835$ ) and Orion × VAC-9510 ( $r = 0.925$ ) progenies, whereas no significant correlation was found in others (Venus × Big Top, Babygold-9 × VAC-9510, Andross × VAC-9511, O'Henry × VAC-9516, and O'Henry × VAC-9514). Previous works (21, 25) have also shown these differences among peach progenies. Indeed, it is well-known that it is not only the total content of phenols but also their specific structural features, such as the number of available hydroxyl groups, that determine their antioxidant capacity (3). Proteggente et al. (23) reported that highest antioxidant capacity is found in fruits such as strawberry, raspberry, and plum due to their high content of anthocyanins. However, no significant correlation was obtained between anthocyanins and RAC in our study (Table 5). This fact is probably due to the lower content of anthocyanins in peaches and nectarines compared with contents in strawberries, raspberries, and plums. Vitamin C did not show significant correlation with RAC. All of these results suggest that phenolic compounds are mainly responsible for the antioxidant activity of peaches and nectarines, as previously described for stone fruits (6, 11), whereas vitamin C is reported as the main antioxidant compound in oranges, strawberries, raspberries, and blueberries (26). The

**Table 5.** Pearson's Correlation Coefficients between Phytochemical Traits Observed over 3 Years in 15 Peach and Nectarine Progenies<sup>a</sup>

trait	flavonoids	anthocyanins	vitamin C	RAC
total phenolics	0.742**	0.144*	ns	0.606**
flavonoids		ns	ns	0.553**
anthocyanins			ns	ns
vitamin C				ns

<sup>a</sup> \*,  $P \leq 0.05$ ; \*\*,  $P \leq 0.01$ ; ns, not significant. Abbreviations: RAC, relative antioxidant capacity.



**Figure 2.** Linear regression ( $P \leq 0.01$ ) between relative antioxidant capacity (RAC) and total phenolics (GAE, gallic acid equivalents) in the peach and nectarine genotypes. Each value is the mean over the three years of study for each genotype.

**Table 6.** Pearson's Correlation Coefficients for Phytochemical and Quality Traits Observed over 3 Years in 15 Peach and Nectarine Progenies<sup>a</sup>

trait	fruit weight	endocarp staining	SSC	TA
total phenolics	0.298**	0.249**	0.237**	ns
flavonoids	0.345**	0.376**	0.371**	ns
anthocyanins	0.166*	0.218**	ns	ns
vitamin C	-0.450**	ns	0.154*	0.308**
RAC	ns	0.175**	0.268**	ns

<sup>a</sup>\*,  $P \leq 0.05$ ; \*\*,  $P \leq 0.01$ ; ns, not significant. Abbreviations: SSC, soluble solid content; TA, titratable acidity; RAC, relative antioxidant capacity.

demonstrated beneficial effects of antioxidant compounds on health make the antioxidant capacity of fruits an important trait to be considered in breeding programs. However, due to the lack of correlation between RAC and other important bioactive compounds such as anthocyanins and vitamin C, we suggest considering and including these traits in a breeding program for the selection of higher fruit quality genotypes.

Both total phenolics and flavonoids contents showed a slight significant positive correlation with fruit weight and sugar content (Table 6), showing a tendency of bigger and sweeter fruits to have higher levels of these bioactive compounds. This is consistent with the findings reported for most species such as plums (27), apricots (28), sweet cherries (29), and apples (30). The relationship of fruit weight with bioactive compounds could be explained by the well-known influence of the sink size (i.e., fruit weight) on the ability to attract photosynthates from the plant sources, because a sufficient accumulation of sugars in or near the fruit is essential for phenolic compounds synthesis during fruit growth (31). On the other hand, a significant positive correlation was found for total phenolics, flavonoids, and anthocyanins versus endocarp staining (redness around stone), supporting the recognized role of anthocyanin pigments in this quality trait. Higher correlation coefficients between total phenolics and endocarp staining were found in some progenies such as Andross  $\times$  Calante ( $r = 0.522$ ), Andross  $\times$  Rich Lady ( $r = 0.730$ ), and O'Henry  $\times$  VAC-9515 ( $r = 0.691$ ), whereas no correlation was found for others, suggesting that relationships between traits depend on the progeny or cultivar evaluated. A positive correlation between endocarp staining and RAC was also found as a consequence of flavonoid pigment contribution to the antioxidant capacity of fruit (3). This result indicates that higher endocarp-stained fruits have higher antioxidant capacity and, consequently, higher health benefits according to previous papers (9). Finally, the positive correlation between vitamin C and TA is due to the contribution of ascorbic acid to the fruit acidity.

These results confirm the importance of genotype on the availability of bioactive compounds and antioxidant capacity of peach and nectarine fruits and, consequently, on their benefits to health. Therefore, the peach cultivars used as progenitors in the crosses of a breeding program have a vital importance to release new cultivars with high bioactive compounds content. On the other hand, the high number of evaluated genotypes, from different genetic origins and with a large phenotypic variability, constitutes a considerable contribution on peach species and especially on breeding purposes.

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