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Antioxidant Capacity and Bioactive Compounds Accumulation in Peach Breeding Germplasm

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Abstract

Antioxidant capacity and accumulation of bioactive compounds (total phenolics, flavonoids, and anthocyanins) were evaluated in 132 peach/nectarine cultivars, representing modern peach [*Prunus persica* (L.) Batsch] breeding germplasm available and/or produced in the U.S. market. Accumulation of bioactive compounds and antioxidant capacity was highly variable and influenced by flesh color, ripening season, release date, genotype, and environment. Peach germplasm accumulated on average 50.9 mg GAE/100 g FW of total phenolics (ranging from 22.9 – 110.7), 15.2 CE/ 100 g of FW flavonoids (ranging from 2.1 to 56), and 6 mg C3GE/ kg FW anthocyanins (ranging from 1.6 – 19) with an observed average antioxidant capacity of 652.6 µg TE/ g FW (ranging from 93.3 – 2,115.3). Ripening season influenced accumulation of bioactive compounds, with antioxidant capacity and phytochemical content increasing as the season progressed. The highest antioxidant capacity (> 1000) and accumulation of bioactive compounds were measured in heirloom cultivars, such as yellow-fleshed 'Elberta' and 'Jerseyqueen', and white-fleshed 'Belle of Georgia'. Different climatic conditions during the years of study resulted in higher antioxidant capacity in 2013 than in 2012 and 2014. However, anthocyanin accumulation was significantly higher in 2012 than in the other two years, while accumulation of both total phenolics and flavonoids were higher in 2014. Antioxidant capacity and accumulation of flavonoids and total phenolics ($r = 0.838$ and $r = 0.501$, $P < 0.01$, respectively) were positively correlated. These results clearly show the high nutritional value of peaches available on the U.S. market and suggest that the peach breeding germplasm offers a valuable resource for increasing nutritional value in breeding programs. Additional knowledge of nutritional quality existing in modern peach germplasm will facilitate development of new peach cultivars with increased phytochemical composition, therefore providing tastier and healthier food choices for consumers.

Many fruits and vegetables are considered functional foods and have always been important in the human diet. Numerous studies showed that increased consumption of fruits and vegetables has a protective effect against chronic-degenerative diseases and improves antioxidant defenses of the human body (Cantín et al., 2009b; Prior and Cao, 2000; Vizzotto et al., 2007). These protective properties in fruits and vegetables are the results of antioxidative properties of phenolic compounds against the damaging effects of reactive oxygen species (ROS) and /or reactive nitrogen species (RNS) (Blokhina et al., 2003; Gill and Tuteja, 2010; Kaur and Kapoor, 2001).

The health benefits of the fruits and their role in the healthy daily diet have been the major focus in the mainstream media and governmental policies driven by the increase in unhealthy eating habits (Slavin and Lloyd, 2012; www.choosemyplate.gov). Breeding for improvement of any trait requires understanding of the existing variation and quite often available information is restricted to a limited number of genotypes. Reports on accumulation of phenolic compounds and their antioxidant capacity in peach (*P. persica*) have often focused on breeding germplasm or cultivars important for a specific market (Cantín et al., 2009b; Ceccarelli et al., 2016;

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Reig et al., 2013; Vizzotto et al., 2007; Zhao et al., 2015). On occasion when the broader germplasm was investigated the cultivars included were either inferior and no longer grown or relevant for only a narrow geographical region, e.g. Europe or China (Font i Forcada et al., 2014; Zhao et al., 2015). Beside analyzing accumulation levels in the peach germplasm, studies have also been directed towards the effect of peach nutritional compounds on human body (Cevaloss-Casals et al., 2006; Dalla Valle et al., 2007; Noratto et al., 2009). Peach phenolic compounds exhibited high antioxidant kinetics, indicating their potential for faster removal of free radicals (Cevaloss-Casals et al., 2006), improved total radical-trapping potential of plasma in humans (Dalla Valle et al., 2007), and selectively killed breast cancer cells (Noratto et al., 2009).

Bioactive compounds are excellent scavengers of free radicals and could be affected/elicited by numerous abiotic environmental stresses, such as chilling, salinity, heavy metals, water deficiency, or UV-B irradiation (Agati et al., 2012). For example, UV light induced anthocyanin biosynthesis and accumulation in different peach and nectarine tissues (Ravaglia et al., 2013). In addition to environment, the genotype, horticultural practices, maturity date, pre-harvest applications, and post-harvest conditions, season, and plant organ also influence accumulation of phytochemicals and fruit quality traits (Brown et al., 2014; Eduardo et al., 2011; Gil et al., 2002; Lee and Kader, 2000; Martínez-Espla et al., 2014; Sivaci and Duman, 2014; Zhang et al., 2012). Phenolic compounds are widely distributed within the peach fruit, with higher concentration in the exocarp (Cevallos- Casals et al., 2006; Zhang et al., 2015). Even though the fruit skin (exocarp) has the higher concentration of phytochemical compounds, it only represents ~8% of the total fresh flesh weight, accumulating ~30% of total phenolic compounds per fruit (Cevallos-Casals et al., 2006; Remorini et al., 2008). A recent study on the accumulation of total phenolics and antioxidant capacity between peach fruit flesh and skin (peel and flesh, separately and together) revealed no significant differences in the total phenolic content among different tissues (based on concentration per fresh weight), suggesting that regardless of how peach fruit is consumed, with or without peel, approximately the same amount of total phenolics is consumed (Abdelghafar et al., 2015).

Breeding for fruit size and appearance has for a long time been one of the major foci of peach breeding programs (Byrne, 2005). Increased awareness of health benefits of bioactive compounds in fruits and vegetables has intensified breeder's efforts to enhance these compounds in newly developed cultivars. Consequently, there is growing interest in breeding programs to obtain information on phenolic compounds and antioxidant capacity of existing germplasm and its potential to provide enhanced health benefits to consumers (Brown et al., 2014; Cantín et al., 2009b; Font i Forcada et al., 2014; Vizzotto et al., 2007). Therefore, the goal of this study was to evaluate different factors that affect antioxidant capacity and bioactive compound accumulation in peach breeding germplasm for its potential to provide sources for improvement of these compounds in a peach breeding program. Increased knowledge of nutritional quality of modern peach germplasm will facilitate the development of new peach cultivars with increased bioactive compounds.

Materials and Methods

Plant material: One hundred and thirty-two peach and nectarine cultivars and one advanced selection, maintained within the *Prunus* collection at Clemson University, were included in this study (Table 1). Fruit quality and accumulation of phytochemical compounds were evaluated over two years (2013 and 2014). A subset of 21 peach and nectarine cultivars and one red-fleshed advanced selection were analyzed over three seasons (2012 - 2014) to facilitate evaluation of genotype and yearly effects on phy-

Table 1. Characteristics of peach breeding germplasm. Accessions are ordered by the ripening time.

Name	Fruit type	Flesh color	Ripening time[z]	Ripening season	Release date	Origin
Rich May	Peach	Yellow	142	Early	1991	USA, CA
Crimson Lady[y]	Peach	Yellow	149	Early	1992	USA, CA
Spring Snow	Peach	White	149	Early	1997	USA, CA
Carored	Peach	Yellow	153	Early	2005	USA, SC
'NJ350' Desiree®	Peach	Yellow	153	Early	2008	USA, NJ
Springold	Peach	Yellow	153	Early	1966	USA, GA
Westbrook	Nectarine	Yellow	153	Early	2002	USA, AR
Arctic Star	Nectarine	White	156	Early	1995	USA, CA
FA 101	Peach	Yellow	156	Early	1972	USA, CA
Manon	Peach	White	156	Early	1988	France
PF 1	Peach	Yellow	156	Early	1995	USA, MI
Arctic Glo	Nectarine	White	160	Early	1992	USA, CA
Candor	Peach	Yellow	160	Early	1965	USA, NC
Honey Blaze	Nectarine	Yellow	160	Early	1998	USA, CA
PF 5D Big	Peach	Yellow	160	Early	2007	USA, MI
Sugar May	Peach	White	160	Early	1991	USA, CA
Sweet Scarlet	Peach	Yellow	160	Early	1996	USA, CA
Country Sweet	Peach	Yellow	163	Early	1999	USA, CA
Early Red Free	Peach	Yellow	163	Early	1938	USA, CA
Garnet Beauty	Peach	Yellow	163	Early	1958	Canada
Glenglo	Peach	Yellow	163	Early	1996	USA, WV
Sentry	Peach	Yellow	163	Early	1980	USA, WV
7 Ball	Peach	Yellow	167	Early		USA, MI
Arctic Sweet	Nectarine	White	167	Early	1996	USA, CA
'NJ354' July Rose™	Peach	White	167	Early	2013	USA, NJ
Easternglo	Nectarine	Yellow	167	Early	1992	USA, CA
Gala	Peach	Yellow	167	Early	1992	USA, LA
Jade™ Momee cv.	Nectarine	White	167	Early	1993	France
PF-7A	Peach	Yellow	167	Early	2006	USA, MI
FA 47	Peach	Yellow	167	Early	1997	USA, MI
Snowbrite	Peach	White	167	Early	1993	USA, CA
Vulcan	Peach	Yellow	167	Early	1994	Canada
Galaxy	Peach	White	174	Early	1995	USA, CA
Karla Rose	Nectarine	White	174	Early	1975	USA, CA
PF 8 Ball	Peach	Yellow	174	Early	2006	USA, MI
FA 12	Peach	Yellow	177	Early	1998	USA, MI
Caroking	Peach	Yellow	177	Early	1987	USA, SC
PF 11 Peach	Peach	Yellow	177	Early	2006	USA, MI
Redhaven	Peach	Yellow	177	Early	1940	USA, MI
FA 11	Peach	Yellow	177	Early	1998	USA, MI
Coronet	Peach	Yellow	181	Mid	1953	USA, GA
Klondike White	Peach	White	181	Mid	1999	USA, CA
PF 15A	Peach	Yellow	181	Mid	1994	USA, MI
FA 52	Peach	Yellow	181	Mid	1997	USA, MI
Reliance	Peach	Yellow	181	Mid	1964	USA, NH

Summer Beaut	Nectarine	Yellow	181	Mid	1979	USA, CA
White Lady	Peach	White	181	Mid	1986	USA, CA
Raritan Rose	Peach	White	184	Mid	1936	USA, NJ
Zin Dai	Peach	White	184	Mid		China
10 Ball	Peach	Yellow	188	Mid		USA, MI
11 Ball	Peach	Yellow	188	Mid		USA, MI
Burpeachfive	Peach	Yellow	188	Mid	2002	USA, CA
Carolina Belle	Peach	White	188	Mid	1987	USA, NC
FA 59	Peach	Yellow	188	Mid	1998	USA, MI
Crimson Rocket	Peach	Yellow	188	Mid	2004	USA, WV
Ernie's Choice	Peach	Yellow	188	Mid	1991	USA, NJ
Glohaven	Peach	Yellow	188	Mid	1963	USA, MI
Harrow Beauty	Peach	Yellow	188	Mid	1983	Canada
John Boy II	Peach	Yellow	188	Mid	2000	USA, PA
PF 17	Peach	Yellow	188	Mid	1993	USA, MI
Sunhigh	Peach	Yellow	188	Mid	1938	USA, NJ
White County	Peach	Yellow	188	Mid	2007	USA, AR
Winblo	Peach	Yellow	188	Mid	1972	USA, NC
9 Ball	Peach	Yellow	191	Mid		USA, MI
FA 80	Peach	Yellow	191	Mid	1997	USA, MI
Arctic Belle	Nectarine	White	191	Mid	1998	USA, CA
Arctic Jay	Nectarine	White	191	Mid	1991	USA, CA
Bounty	Peach	Yellow	191	Mid	1988	USA, WV
Flavortop	Nectarine	Yellow	191	Mid	1969	USA, CA
FlavrBurst	Peach	Yellow	191	Mid	2010	USA, WV
Intrepid	Peach	Yellow	191	Mid	2002	USA, NC
Sweet Dream	Peach	Yellow	191	Mid	1998	USA, CA
Beaumont	Peach	Yellow	195	Mid	2007	USA, MI
Beekman	Peach	Yellow	195	Mid		USA, NJ
Canadian Harmony	Peach	Yellow	195	Mid	1968	Canada
Contender	Peach	Yellow	195	Mid	1987	USA, NC
Early Loring	Peach	Yellow	195	Mid	1977	USA, PA
Julyprince	Peach	Yellow	195	Mid	2004	USA, GA
Late Large 23	Peach	Yellow	195	Mid		USA, MI
Loring	Peach	Yellow	195	Mid	1946	USA, MO
Majestic	Peach	Yellow	195	Mid	1979	USA, LA
PF-24-007	Peach	Yellow	195	Mid	1997	USA, MI
Redglobe	Peach	Yellow	195	Mid	1954	USA, MD
Summer Gold	Peach	Yellow	195	Mid	1970	USA, GA
Suncrest	Peach	Yellow	195	Mid	1959	USA, OR
99p4388	Peach	Red	198	Mid		USA, GA
Diamond Princess	Peach	Yellow	198	Mid	1989	USA, CA
Late 24-007	Peach	Yellow	198	Mid	1997	USA, MI
PF Lucky 24B	Peach	Yellow	198	Mid	2003	USA, MI
Redrose	Peach	White	198	Mid	1940	USA, NJ
Sweet Breeze	Peach	Yellow	198	Mid	2004	USA, PA
Arctic Gold	Nectarine	White	202	Late	1995	USA, CA
'NJ351' Gloria®	Peach	Yellow	202	Late	2007	USA, NJ

FA 17	Peach	Yellow	202	Late	1997	USA, MI
PF 2050	Peach	Yellow	202	Late		USA, MI
Sweet N Up	Peach	Yellow	202	Late	2004	USA, WV
Belle of Georgia	Peach	White	205	Late	1875	USA, GA
FA 18	Peach	Yellow	205	Late	1998	USA, MI
China Pearl	Peach	White	205	Late	2001	USA, NC
Cresthaven	Peach	Yellow	205	Late	1963	USA, MI
Fantasia	Nectarine	Yellow	205	Late	1969	USA, CA
'NJ352' Messina®	Peach	Yellow	205	Late	2007	USA, NJ
Blake	Peach	Yellow	209	Late	1953	USA, NJ
Carolina Gold	Peach	Yellow	209	Late	2007	USA, NC
Summer Fire	Nectarine	Yellow	209	Late	1991	USA, CA
SummerFest	Peach	Yellow	209	Late	2010	USA, WV
Zephyr™ Monphir	Nectarine	White	209	Late	1992	France
Glacier	Peach	White	211	Late	2001	USA, CA
Madison	Peach	Yellow	211	Late	1963	USA, VA
PF 25	Peach	Yellow	211	Late	1997	USA, MI
PF 28-007	Peach	Yellow	211	Late	2004	USA, MI
Ambre™ Monam cv.	Nectarine	Yellow	216	Late		France
Elberta	Peach	Yellow	216	Late	1889	USA, GA
Jerseyqueen	Peach	Yellow	216	Late	1964	USA, NJ
Lady Nancy	Peach	White	216	Late	1989	USA, NJ
PF 27A	Peach	Yellow	216	Late	1997	USA, MI
Redskin	Peach	Yellow	216	Late	1944	USA, MD
Augustprince	Peach	Yellow	219	Late	2006	USA, GA
FA 42	Peach	Yellow	219	Late	2003	USA, MI
Burpeachfour	Peach	Yellow	219	Late	2002	USA, CA
Flameprince	Peach	Yellow	219	Late	1993	USA, GA
PF 30-007	Peach	Yellow	219	Late	2003	USA, MI
Snow Giant	Peach	White	219	Late	1993	USA, CA
Encore	Peach	Yellow	223	Late	1980	USA, NJ
Snow King	Peach	White	226	Late	1993	USA, CA
Laurol	Peach	Yellow	230	Late	1994	USA, NJ
Victoria	Peach	Yellow	230	Late	2007	USA, NJ
Autumn Red	Peach	Yellow	233	Late	1996	USA, CA
Caro Tiger	Peach	Yellow	237	Late	2012	USA, SC
September Snow	Peach	White	237	Late	1992	USA, CA
Snow Gem	Peach	White	237	Late	2000	USA, CA
Tra-Zee	Peach	Yellow	244	Late	1988	USA, CA
Autumn Flame	Peach	Yellow	251	Late	1996	USA, CA

[z] Ripening time expressed in Julian date.
[y] Bold - accessions analyzed in three years (2012- 2014).

tochemical profiles. Analyzed material was comprised of 103 yellow and 29 white flesh peach and nectarine cultivars available and/ or grown in the U.S. market with addition of the red-flesh advanced selection. Breeding germplasm consisted of 116 peach and 17 nectarine accessions. The red-fleshed advanced selection was included to provide the variability in flesh colors and to allow assessment of health benefits that red flesh might bring to the market. The peach and nectarine germplasm collection is maintained at the Musser Fruit Research Center, Seneca, SC (34.605202 latitude and -82.877995 longitude) under a warm, humid temperate climate and standard commercial practices for irrigation, fertilization, and pest and disease control. The trees were at least 8-years-old, grafted on Guardian® rootstock, grown in triplicate with a 2 × 6 m spacing and a perpendicular-V training system. The maturity groups were based on average Julian ripening date recorded at the Musser Fruit Research Center over 6 years. Climatic data were collected using a WatchDog 2000 Series (Spectrun Technologies Inc., Aurora, IL) weather station.

Fruit sampling: One half bushel (9 – 11 kg) of fruits at commercial maturity stage were picked randomly from 3 trees for each accession. Index of Absorbance Difference (I_{AD}), calculated as the difference between fruit absorbance at the wavelengths of 670 and 720 nm (chlorophyll-a absorbance peak and background of the spectrum) (Ziosi et al., 2008), was used to ensure uniform fruit maturity for all accessions included in the study. Five fruits in the ripening stage equivalent to $I_{AD} = 0.6$ were selected and analyzed for fruit quality and phytochemical content.

Fruit quality measurement: Fruit quality traits were measured according to Frett et al. (2012) phenotyping protocol. A fruit texture analyzer (FTA) [GÜSS Manufacturing (Pty) Ltd., South Africa], equipped with an electronic fruit size measurement (EFM) device and electronic scale, was used to measure fruit size (diameter in mm), fruit weight (g), and fruit firmness (kg). Soluble solids concentration (SSC) was determined using a digital refractometer (Atago 3810 PAL-1). Titratable acidity (TA, Malic acid %) was measured by titration with NaOH 0.1 N to pH 8.2 (862 Compact Titrosampler, Metrohm, Riverview, FL). Additionally, ripening index (RI) was calculated as the sugar/acid ratio (SSC/TA).

Phytochemical analyses: Two longitudinal slices of mesocarp per individual fruit were excised from opposing cheeks to ensure consistent sampling for phytochemical analyses. The exocarp and red tissue around the pit was removed and tissue sliced, individually packed, frozen in liquid nitrogen (LN2), and stored at -20 ºC until needed. To ameliorate high variability within fruit and between different fruits from the same tree and/or between the different trees of the same cultivar (Brown et al., 2014), a composite sample per accession was prepared by combining equivalent amount of tissue from each of five fruits, and grinding in liquid nitrogen using Freezer/Mill (SPEX® SamplePrep, Metuchen, NJ, USA). Ground tissue was stored at -80 ºC until needed. One ml of 80% methanol was used to extract 500 mg of frozen composite powder in five replicates. The extract was incubated at 4 ºC overnight and then centrifuged for 10 min at 4 ºC and 12,000 g to collect the supernatant. The hydroalcoholic extract was stored at -80 ºC and used in subsequent analyses. The total phenolics, flavonoid content, anthocyanin, and antioxidant capacity were measured following the protocols described in Cantín et al. (2009b). The total phenolics, expressed in mg of gallic acid equivalents (GAE) per 100 g of fresh weight (FW), were determined according to the Folin−Ciocalteu method (Singleton and Rossi, 1965). The flavonoid content was measured using Zhishen et al. (1999) protocol and the results were expressed as mg of catechin equivalents (CE) per 100g of FW. Total anthocyanin content of the hydroalcoholic extract was determined using the method of Fuleki and Francis (1968) adapted to peach tissue and quantified as mg of cyanidin-3-glucoside per kg of FW. The antioxidant capacity was measured using an 1,1-Diphenyl-2-picryl-hydrazyl (DPPH) method (Brand-Williams et al., 1995) and expressed as μg of Trolox (6-hydroxy-2, 5, 7, 8-tetramethylchromane-2-carboxylic acid) Equivalent Antioxidant Capacity (TEAC) per g of FW.

Statistical analysis. Data were expressed as means and standard deviations (± SD) using yearly data as replicates. Data were analyzed for each year separately to take out variance between years for each of fruit type, flesh color, ripening season, release date, and environmental factors. The data for 22 genotypes were averaged over 3 years to investigate the genotypic effect on the accumulation of phytochemical compounds. The fresh peach market in the U.S. is almost exclusively yellow and white flesh peach and nectarines; thus, the red flesh accession was included only in the flesh color analysis. For the purpose of statistical analysis, cultivars ripening from May to the end of June (< 180 Julian days) were considered early, from July 1 – 20 (181-201) mid-season, and after July 21st (> 202) late maturing. Release date of 9 cultivars could not be found; therefore, these cultivars were not included in the release date analysis. Analysis of variance was performed by season, and means were compared using the Student-Newman-Keuls's test and t test ($P < 0.05$). A *t* test ($P < 0.05$) was used for comparing two different fruit types (peach vs. nectarine) or years (2013 vs. 2014). The focus of this study are bioactive compounds; therefore, fruit quality traits were included only in Pearson's correlation coefficients and principle component analysis to investigate the relationships between bioactive compounds and fruit quality traits. Pearson's correlation coefficient at $P < 0.01$ was used to assess the correlation between analyzed traits. Principal Components Analysis (PCA) was performed to study the relationship among the analyzed traits. All statistical analyses were performed in SPSS v. 24 (IBM SPSS Statistics for Windows, Version 24.0. Armonk, NY: IBM Corp).

Results and Discussion

Variation in accumulation of phytochemical compounds was observed within the analyzed material and between experimental years. Data for each year separately, and mean of the two years, for each cultivar, are shown in tables 2 and 3. Accumulation of bioactive compounds and their antioxidant capacity was highly influenced by flesh color, ripening season, release date, genotype, and environmental conditions during the study years.

Total phenolics. Overall, accumulation of total phenolics across 2 years ranged from 22.9 to 110.7 mg GAE per 100 g of FW with the most of the accessions accumulating < 70 mg GAE per 100 g of FW (Tables 2 and 3). Total phenolics in the peach breeding germplasm were within the range reported in the literature for peach, i.e. 12.7 to 111 mg GAE per 100 g of FW (Cantín et al., 2009b; Gil et al., 2002; Tavarini et al., 2008). Cantín et al. (2009b) reported similar total phenolic content in 218 peach genotypes from several breeding progenies, ~13 to 71 mg of GAE per 100 g of FW with an average accumulation of 36.4 mg of GAE per 100 g of FW, as well as Tavarini et al. (2008), of 14 to 50 mg GAE per 100 g of FW, and Gil et al. (2002), 21 to 111 mg GAE per 100 g of FW. It was to be expected that our data for accumulation of total phenolics in peach fruit encompassed those previously reported. Given peach's narrow genetic base (Byrne, 1999; Scorza and Okie, 1990; Scorza et al., 1985), any fresh market peach breeding germplasm would include material closely related to those used in other studies. The observed differences are most likely due to the level of diversity in the evaluated material in each study.

Accumulation of the total phenolics was associated with flesh color. The red-fleshed selection had the highest accumulation in 2013 and 2014, 57.3 and 89.6 mg GAE per 100g of FW, respectively (Tables 4 and 5), whereas white flesh cultivars accumulated on average less total phenolics (48.6 mg GAE per 100g of FW) than yellow flesh cultivars (53.1 mg GAE per 100g of FW). Interestingly, white flesh cultivars included in the study had a wider range of accumulated total phenolics (29.3 to 110.7 mg GAE per 100g of FW) than yellow flesh cultivars (22.9 to 98.3 mg GAE per 100g of FW) (Tables 2 and 3). Among white flesh cultivars, 'Sugar May' had the lowest accumulation of total phenolics, whereas the 'Belle of Georgia' had the highest (Table 2). 'Loring' had the highest accumulation of total phenolics among yellow flesh cultivars, whereas 'FA 12' had the lowest (Table 3). Differences in accumulation of total phenolics between white and yellow flesh peach cultivars were previously reported; however, higher accumulation was reported in white fleshed material than in yellow (Cantín et al., 2009b; Gil et al., 2002). Reported ranges for white flesh peach cultivars were very similar to our data (28 - 111

Table 2. Total phenolics, flavonoids, anthocyanins and antioxidant concentrations in white flesh cultivars in different ripening seasons in two years (2013 and 2014). GAE, Gallic acid equivalents; CE, Catechin equivalents; C3GE, Cyanidin-3-glucoside equivalents; TE, Trolox equivalents.

Genotype	Year	Total phenolics (mg GAE/100g) ^{z}	Flavonoids (mg CE/100g)	Anthocyanins (mg C3GE/k)	Antioxidants (µg TE/g)
			Early season		
Arctic Glo	2013	18.72 x	2.92 x	18.26 y	214.16 x
	2014	46.31 y	43.96 y	9.13 x	690.59 y
	Mean	32.52 **a**	23.44 **d-g**	13.70 **e**	452.37 **b-f**
Arctic Star	2013	14.99 x	6.07 y	2.62 y	501.96 y
	2014	60.69 y	4.99 x	1.14 x	32.52 x
	Mean	37.84 **ab**	5.53 **ab**	1.88 **a**	267.24 **a-c**
Arctic Sweet	2013	16.46 x	4.46 x	2.51 x	284.45 x
	2014	60.25 y	19.11 y	9.82 y	578.15 y
	Mean	38.35 **ab**	11.79 **a-e**	6.16 **a-d**	431.30 **b-f**
Early Red Free	2013	18.13 x	10.66 x	2.62 x	616.40 x
	2014	79.42 y	36.32 y	7.08 x	1070.98 y
	Mean	48.77 **a-c**	23.49 **d-g**	4.85 **a-c**	843.69 **h-l**
Galaxy	2013	26.99 x	5.22 x	1.71 x	449.00 x
	2014	77.66 y	26.08 y	3.77 y	689.72 y
	Mean	52.32 **a-c**	15.65 **a-f**	2.74 **a-c**	569.36 **d-h**
Jade™ Momee cv.	2013	32.49 x	22.03 x	7.88 y	811.99 y
	2014	31.49 x	40.49 y	4.91 x	738.78 x
	Mean	31.99 **a**	31.26 **g**	6.39 **a-d**	775.39 **g-k**
Karla Rose	2013	31.11 x	5.34 x	4.68 y	436.68 y
	2014	42.49 y	6.17 x	2.74 x	243.12 x
	Mean	36.80 **ab**	5.76 **ab**	3.71 **a-c**	339.90 **a-e**
Manon	2013	15.29 x	8.42 x	3.19 x	601.87 x
	2014	45.25 y	16.39 y	3.31 x	604.83 x
	Mean	30.27 **a**	12.40 **a-e**	3.25 **a-c**	603.35 **e-i**
'NJ354' July Rose™	2013	40.47 x	8.80 x	12.79 y	668.40 y
	2014	49.71 y	11.68 y	3.42 x	521.52 x
	Mean	45.09 **a-c**	10.24 **a-d**	8.11 **a-e**	594.96 **d-i**

Snowbrite	2013	28.66 x	14.71 x	4.34 x	318.70 y
	2014	31.98 x	1.96 x	3.08 x	88.73 x
	Mean	30.32 **a**	8.34 **a-d**	3.71 **a-c**	203.72 **ab**
Spring Snow	2013	21.87 x	15.49 x	3.88 x	707.76 y
	2014	39.88 y	24.81 y	10.73 y	688.89 x
	Mean	30.88 **a**	20.15 **b-g**	7.31 **a-e**	698.32 **f-j**
Sugar May	2013	26.72 x	4.03 x	5.82 y	446.56 x
	2014	31.94 y	10.09 y	2.97 x	424.24 x
	Mean	29.33 **a**	7.06 **a-c**	4.40 **a-c**	435.40 **b-f**
Mid-season					
Arctic Belle	2013	27.93 x	7.36 x	2.39 x	454.74 x
	2014	56.28 y	15.35 y	5.25 y	800.52 y
	Mean	42.11 **a-c**	11.36 **a-e**	3.82 **a-c**	627.63 **f-i**
Arctic Jay	2013	39.27 x	16.01 y	8.22 y	967.14 y
	2014	64.78 y	12.79 x	3.65 x	547.81 x
	Mean	52.02 **a-c**	14.40 **a-e**	5.94 **a-d**	757.48 **g-k**
Carolina Belle	2013	50.51 x	9.69 x	4.45 x	747.36 x
	2014	89.22 y	23.00 y	11.87 x	910.11 y
	Mean	69.86 **a-c**	16.35 **a-f**	8.16 **a-e**	828.73 **h-k**
Klondike White	2013	14.30 x	13.09 x	2.96 x	701.64 x
	2014	50.53 y	15.58 y	4.34 x	682.22 x
	Mean	32.42 **a**	14.33 **a-e**	3.65 **a-c**	691.93 **f-j**
Raritan Rose	2013	25.70 x	6.28 x	1.94 x	427.27 x
	2014	42.32 y	21.98 y	5.25 y	883.87 y
	Mean	34.01 **ab**	14.13 **a-e**	3.60 **a-c**	655.57 **f-j**
Redrose	2013	23.80 x	4.07 x	4.34 x	299.96 x
	2014	110.36 y	27.99 y	19.86 y	338.58 x
	Mean	67.08 **a-c**	16.03 **a-f**	12.10 **de**	319.27 **a-d**
White Lady	2013	41.10 x	3.03 x	3.88 x	529.26 y
	2014	42.22 x	6.74 y	3.54 x	504.22 x
	Mean	41.66 **a-c**	4.89 **ab**	3.71 **a-c**	516.74 **c-g**
Zin Dai	2013	15.79 x	0.18 x	7.76 y	20.12 x
	2014	74.52 y	21.26 y	5.60 x	617.42 y
	Mean	45.16 **a-c**	10.72 **a-e**	6.68 **a-d**	318.77 **a-d**
Late season					
Arctic Gold	2013	17.23 x	10.93 x	2.51 x	589.36 x
	2014	147.76 y	54.01 y	15.75 y	1248.90 y
	Mean	82.49 **c**	32.47 **g**	9.13 **a-e**	919.13 **j-l**
Belle of Georgia	2013	77.25 x	43.50 x	8.22 x	1646.95 x
	2014	144.08 y	56.60 y	18.49 y	1711.68 y
	Mean	110.67 **d**	50.05 **h**	13.36 **e**	1679.31 **n**

China Pearl	2013	29.12 x	14.33 x	5.02 x	929.70 x
	2014	126.27 y	30.48 y	6.62 x	1097.08 x
	Mean	77.69 **bc**	22.41 **c-g**	5.82 **a-d**	1013.39 **kl**
Glacier	2013	30.48 x	2.86 y	1.94 x	207.56 y
	2014	60.76 y	1.26 x	2.51 x	66.21 x
	Mean	45.62 **a-c**	2.06 **a**	2.22 **a**	136.89 **a**
Lady Nancy	2013	23.15 x	11.74 x	2.39 x	675.06 x
	2014	111.16 y	40.11 y	17.47 y	1502.28 y
	Mean	67.16 **a-c**	25.93 **e-g**	9.93 **b-e**	1088.67 **l**
September Snow	2013	36.72 x	11.66 x	3.88 x	1068.90 y
	2014	68.43 y	14.95 y	6.85 y	657.02 x
	Mean	52.57 **a-c**	13.30 **a-e**	5.37 **a-d**	862.96 **i-l**
Snow Gem	2013	18.85 x	3.75 x	2.51 x	465.01 x
	2014	44.17 y	11.55 y	2.51 x	756.96 y
	Mean	31.51 **a**	7.65 **a-c**	2.51 **ab**	610.98 **e-i**
Snow Giant	2013	54.70 x	19.50 x	5.02 x	1261.11 x
	2014	87.23 x	38.30 y	21.92 y	1680.05 y
	Mean	70.97 **a-c**	28.90 **fg**	13.47 **e**	1470.58 **m**
Snow King	2013	20.81 x	4.70 x	1.82 x	474.11 x
	2014	79.66 y	10.55 y	18.49 y	453.79 x
	Mean	50.23 **a-c**	7.62 **a-c**	10.16 **c-e**	463.95 **b-f**
Zephyr™ Monphir	2013	37.07 x	28.66 y	6.17 x	1306.00 y
	2014	41.81 x	14.47 x	6.39 x	692.39 x
	Mean	39.44 **ab**	21.56 **c-g**	6.28 **a-d**	999.20 **kl**

[z] Means within columns and harvest season followed by common letters do not differ at the 5% level by Student-Newman-Keul's' test and *t* test. Different letters indicate significant differences at $P < 0.05$ according to Student-Newman-Keul's' test (a-n) for differences between cultivars, and *t* test (x, y) between years.

mg GAE/100g FW) even though a smaller sample was analyzed (e.g., 5 cultivars in each category of white flesh nectarines and peaches and yellow flesh nectarines and peaches), while yellow flesh peach cultivars had a smaller range than observed in our study (21 - 61 mg GAE/100g FW) (Gil et al., 2002).

Ripening season exhibited a positive influence on accumulation of total phenolics with increased accumulation in later season cultivars (Tables 4 and 5). Early ripening peach and nectarine cultivars accumulated on average 37.6 (±19.7) mg GAE per 100g of FW of total phenolics; mid-season cultivars 52.2 (± 33.6) mg GAE per 100g of FW, and late season cultivars 65.8 (± 40.5) mg GAE per 100g of FW. Increasing accumulation of total phenolics in peach and nectarine cultivars as the ripening season progressed was observed in both 2013 and 2014 (Tables 4 and 5). However, in other *Prunus* species, such as apricot, the lowest levels of gallic acid were measured in a late ripening cultivar (Gundogdu et al., 2013). This is the first observation on positive influence of ripening season on phenolic compound accumulation in peach and nectarine. Peach germplasm evaluated in this study bloomed within 2 - 3 weeks but harvest season stretched over 3 months (May to August). Therefore, the differences observed in accumulation of total phenolics in fruit might be explained by the fruit development period, e.g. days after full bloom, and the influence of the various environmental factors the tree was exposed to which could be reflected in the fruit.

In both years, older and / or heirloom cultivars (release date prior to 1960) accumulated more phenolics [37.2 (± 20.5) and 86.0 (± 38.7) in 2013 and 2014, respectively] in comparison with those recently released (Tables 4 and 5), indicating tendencies to select for lower accumulations of total phenolics in newly released material. Fresh market peach breeding germplasm, although variable in its potential to accumulate total phenolics, had lower levels in these compounds than its ancestors (Tables 4 and 5). Breeding for fruit quality, size, appearance and productivity may have inadvertently decreased the phenolic value of modern peach cultivars.

Flavonoids. Peach breeding germplasm had a wide range of flavonoid accumulation from 2.1 to 56 mg of CE per 100 g of FW with an average accumulation of 15.1 mg of CE per 100 g of FW. Interestingly, the average flavonoid accumulation (15.1 mg CE/ 100 g FW) observed in 132 peach and nectarine accessions in this study was higher than previously reported for peach. For example, Cantín et al. (2009b) reported an average of only 8.8 mg CE per 100 g of FW, ranging from 1.8 to 30.9 mg of CE per 100 g of FW, in 218 progeny derived from 15 controlled bi-parental crosses. Similar observations were reported by Abidi et al. (2011) with an average 12.5 mg CE per 100 g of FW reported in an F_1 nectarine population (‘Venus’ × ‘Big Top’). These discrepancies reveal the potential within breeding germplasm for enhancing flavonoid accumulation in peach to much higher levels than previously thought. Accumulation of flavonoids in white- and yellow-fleshed genotypes did not differ (Tables 4 and 5). However, the red flesh selection had on average twice the level of flavonoids (33.1 mg CE/100g FW) detected in white and yellow flesh material. There was a large variation in the accumulation of flavonoids in both white and yellow cultivars regardless of ripening season (Tables 2 and 3). White flesh cultivars accumulated on average from 2.1 – 50 mg of CE per 100g of FW, with ‘Glacier’ exhibiting the minimum and ‘Belle of Georgia’ the maximum values (Table 2). Similarly, yellow flesh cultivars differed significantly for accumulation of flavonoids with the highest accumulation detected in ‘Jerseyqueen’ (56 mg of CE/100g FW) and lowest in ‘Country Sweet’ (2.5 mg of CE/100g FW) (Table 3). Genotype effect on the accumulation of flavonoids observed in our study was previously reported for peach, nectarine and plum (Tomás-Barberán et al., 2001).

Similar to total phenolics, influence of ripening season on accumulation of flavonoids was evident (Tables 4 and 5), with flavonoid content increasing as the season progressed. On average, early- and mid-season peach and nectarine cultivars had the lowest accumulation of flavonoids (10.8 – 12.4 mg CE/100g FW) while late-season cultivars had the highest; with a 2-fold increase (21.0 mg CE/100g FW). Opposite effect of the ripening season on accumulation of individual flavonoid compounds was recently reported for apricot (Gundogdu et al. 2013). Interestingly, the highest levels of the flavonoids catechin and epicatechin were observed for mid-season cultivars, and the lowest for the late-season cultivars. Apricot cultivars had wide variability in levels of catechin and epicatechin accumulation, despite a small sample size. The decrease in accumulation of flavonoids and other compounds was attributed to the cooler temperatures in fall and reduced exposure to sunlight leading to the decrease in respiration rate and O_2 concentration (Gundogdu et al., 2013).

As with total phenolics, flavonoid accumulation was lower in newly released cultivars, 8.1 (± 7.0) mg of CE per 100g of FW in 2013 and 18.6 (± 12.7) mg of CE per 100g of FW in 2014, compared to older cultivars, 13.7 (± 12.2) and 29.2 (± 16.2) in 2013 and 2014, respectively (Tables 4 and 5).

Anthocyanins. Cyanidin is the main pigment responsible for red coloration in peach and nectarine, and cyanidin 3- glucoside (C3GE) is the main anthocyanin reported in peach (Tomás-Barberán et al., 2001; Wu and Prior, 2005). As previously observed for total phenolics and flavonoids, anthocyanin accumulation, measured as cyanidin-3-glucoside, was highly influenced by genotype (Figures 2 and 3). No significant difference in the anthocyanin accumulation between white and yellow flesh cultivars was observed (Tables 4 and 5); however, wide ranges within flesh colors were evident. White flesh cultivars ranged from 1.9 mg of C3GE per kg of FW in 'Arctic Star' to > 13 mg of C3GE per kg of FW in 'Arctic Glo', 'Snow Giant' and 'Belle of Georgia' (Table 2). Overall, the lowest and highest accumulation of anthocyanins developed in yellow-fleshed cultivars with 1.6 mg C3GE/kg FW for 'Candor', 'Crimson Lady', and 'Vulcan' and 19 mg C3GE/kg FW for 'Ernie's Choice' (Table 3). Although the average anthocyanin accumulation in white and yellow flesh peach and nectarine cultivars was similar to those previously reported, 1.5 - 5 mg C3GE/100g FW in peach and 1.2 - 9.5 mg C3GE/kg FW in nectarine (Abidi et al., 2011; Vizzotto et al., 2007), the range

Table 3. Total phenolics, flavonoids, anthocyanins and antioxidant concentrations in yellow flesh cultivars in different ripening seasons in two years (2013 and 2014). GAE, Gallic acid equivalents; CE, Catechin equivalents; C3GE, Cyanidin-3-glucoside equivalents; TE, Trolox equivalents.

Genotype	Year	Total phenolics (mg GAE/100g) [z]	Flavonoids (mg CE/100g)	Anthocyanins (mg C3GE/k)	Antioxidants (µg TE/g)
			Early season		
7 Ball	2013	23.97 x	5.60 x	8.90 x	453.78 x
	2014	54.66 y	15.17 y	5.71 x	564.22 y
	Mean	39.32 **a-c**	10.39 **a-g**	7.31 **f**	509.00 **e-i**
Candor	2013	26.37 x	5.42 x	2.05 x	415.46 y
	2014	37.75 y	8.44 y	1.14 x	327.27 x
	Mean	32.06 **a-c**	6.93 **a-d**	1.60 **a**	371.37 **b-g**
Caroking	2013	28.72 x	6.63 x	2.74 y	445.60 y
	2014	68.79 y	9.41 y	1.37 x	211.06 x
	Mean	48.75 **a-d**	8.02 **a-e**	2.05 **a-c**	328.33 **b-f**
Carored	2013	29.24 x	2.74 x	1.94 x	280.39 x
	2014	55.67 y	5.24 y	3.77 y	345.20 y
	Mean	42.46 **a-c**	3.99 **a-c**	2.85 **a-c**	312.79 **b-e**
Country Sweet	2013	16.01 x	1.53 x	3.54 x	138.02 x
	2014	43.19 y	3.56 y	2.74 x	174.75 x
	Mean	29.60 **ab**	2.54 **a**	3.14 **a-c**	156.39 **ab**
Crimson Lady	2013	17.59 x	1.98 x	1.25 x	173.30 y
	2014	56.81 y	5.57 y	1.94 x	13.39 x
	Mean	37.20 **a-c**	3.78 **ab**	1.60 **a**	93.34 **a**
Easternglo	2013	8.06 x	2.52 x	7.99 y	279.49 x
	2014	75.90 y	5.79 y	5.14 x	337.53 y
	Mean	41.98 **a-c**	4.15 **a-c**	6.57 **d-f**	308.51 **b-e**
FA 11	2013	36.42 x	9.56 y	1.48 x	507.51 y
	2014	67.80 y	7.00 x	5.25 y	108.09 x
	Mean	52.11 **a-d**	8.28 **a-f**	3.37 **a-c**	307.80 **b-e**

FA 12	2013	9.66 x	1.65 x	2.05 x	181.94 x
	2014	36.23 y	5.60 y	1.71 x	308.04 y
	Mean	22.95 **a**	3.63 **ab**	1.88 **a-c**	244.99 **a-c**
FA 47	2013	33.78 x	1.60 x	8.10 x	278.39 x
	2014	48.54 y	5.81 y	1.82 x	406.61 y
	Mean	41.16 **a-c**	3.70 **ab**	4.96 **c-e**	342.50 **b-f**
FA 101	2013	35.06 x	10.97 x	1.82 x	591.09 x
	2014	52.91 y	20.18 y	2.51 x	611.64 x
	Mean	43.99 **a-c**	15.57 **e-h**	2.17 **a-c**	601.36 **h-j**
GaLa	2013	34.87 x	5.15 x	2.17 x	464.93 x
	2014	82.57 y	18.65 y	4.34 y	889.04 y
	Mean	58.72 **cd**	11.90 **b-g**	3.25 **a-c**	676.98 **i-k**
Garent Beauty	2013	22.23 x	6.31 x	1.82 x	497.37 y
	2014	40.80 y	14.02 y	2.62 x	477.58 x
	Mean	31.52 **a-c**	10.16 **a-g**	2.22 **a-c**	487.48 **d-i**
Glenglo	2013	27.86 x	11.73 x	3.19 x	886.12 y
	2014	61.81 y	23.22 y	6.39 y	701.73 x
	Mean	44.83 **a-d**	17.48 **gh**	4.79 **b-e**	793.92 **k**
Honey Blaze	2013	25.68 x	7.26 x	1.82 x	462.61 x
	2014	43.29 y	24.97 y	3.65 y	716.36 y
	Mean	34.49 **a-c**	16.12 **f-h**	2.74 **a-c**	589.48 **h-j**
‘NJ350’ Desiree®	2013	23.97 x	2.40 x	1.71 x	246.09 x
	2014	32.05 y	7.48 y	2.51 y	288.55 x
	Mean	28.01 **ab**	4.94 **a-c**	2.11 **a-c**	267.32 **a-d**
PF 1	2013	16.47 x	4.46 x	1.60 x	395.61 x
	2014	50.02 y	14.45 y	2.71 x	426.57 x
	Mean	33.25 **a-c**	9.46 **a-f**	2.15 **a-c**	411.09 **c-h**
PF 11 Peach	2013	20.38 x	10.88 y	2.62 x	726.43 y
	2014	55.66 y	7.17 x	2.74 x	406.61 x
	Mean	38.02 **a-c**	9.03 **a-f**	2.68 **a-c**	566.52 **g-j**
PF 5D Big	2013	13.49 x	2.14 x	1.48 x	307.10 y
	2014	34.94 y	5.15 y	1.94 x	226.67 x
	Mean	24.22 **a**	3.65 **ab**	1.71 **ab**	266.88 **a-d**
PF 7A	2013	22.55 x	3.95 x	6.74 y	252.57 x
	2014	33.19 y	8.55 y	1.83 x	409.82 y
	Mean	27.87 **ab**	6.25 **a-d**	4.28 **a-d**	331.20 **b-f**
PF 8 Ball	2013	12.44 x	0.83 x	9.47 y	81.26 x
	2014	42.39 y	20.03 y	4.34 x	598.77 y
	Mean	27.42 **ab**	10.43 **a-g**	6.91 **ef**	340.01 **b-f**
Redhaven	2013	35.40 x	5.68 x	3.65 x	378.35 x
	2014	100.47 y	36.12 y	2.97 x	744.93 y
	Mean	67.93 **d**	20.90 **h**	3.31 **a-c**	561.64 **g-j**
Rich May	2013	15.78 x	16.19 y	7.31 y	748.47 y
	2014	41.04 y	12.70 x	1.94 x	348.83 x
	Mean	28.41 **ab**	14.45 **d-h**	4.62 **a-e**	548.65 **f-j**

Sentry	2013	28.93 x	14.50 x	3.88 x	908.94 y
	2014	41.90 y	20.68 y	3.20 x	554.74 x
	Mean	35.42 **a-c**	17.59 **gh**	3.54 **a-c**	731.84 **j-k**
Springold	2013	39.19 x	19.07 y	2.51 x	1059.00 y
	2014	43.46 x	5.77 x	6.22 x	182.36 x
	Mean	41.33 **a-c**	12.42 **c-g**	4.37 **a-d**	620.68 **h-k**
Sweet Scarlet	2013	16.45 x	5.02 x	2.39 x	493.01 y
	2014	39.59 y	9.52 y	2.17 x	456.97 x
	Mean	28.02 **ab**	7.27 **a-d**	2.28 **a-c**	474.99 **c-i**
Vulcan	2013	26.09 x	5.42 x	1.48 x	374.96 y
	2014	82.16 y	6.36 x	1.62 x	347.83 x
	Mean	54.12 **b-d**	5.89 **a-c**	1.55 **a**	361.40 **b-g**
Westbrook	2013	11.75 x	2.14 x	2.74 x	321.56 x
	2014	35.52 y	10.87 y	1.60 x	415.15 y
	Mean	23.63 **a**	6.51 **a-d**	2.17 **a-c**	368.35 **b-g**
Mid-season					
10 Ball	2013	30.25 x	3.99 x	2.28 x	423.10 y
	2014	61.11 y	5.30 x	7.65 y	273.79 x
	Mean	47.39 **a-d**	4.72 **a-c**	5.26 **a-c**	340.15 **ab**
11 Ball	2013	14.96 x	1.59 x	2.28 x	257.92 x
	2014	31.61 y	5.80 y	1.82 x	305.26 y
	Mean	23.29 **a**	3.70 **ab**	2.05 **ab**	281.59 **ab**
9 Ball	2013	15.78 x	4.19 y	1.82 x	396.30 y
	2014	51.36 y	1.24 x	2.85 x	235.40 x
	Mean	33.57 **ab**	2.72 **a**	2.34 **ab**	315.85 **ab**
Beaumont	2013	22.28 x	5.08 x	1.71 x	400.84 x
	2014	168.88 y	14.03 y	15.75 y	281.53 x
	Mean	95.58 **d**	9.55 **a-h**	8.73 **a-c**	341.18 **ab**
Beekman	2013	39.61 x	17.40 x	3.65 x	818.84 x
	2014	114.67 y	28.05 y	17.35 y	775.69 x
	Mean	77.14 **a-d**	22.73 **hi**	10.50 **a-c**	797.27 **e-i**
Bounty	2013	36.31 x	11.24 x	2.05 x	676.63 x
	2014	58.76 y	17.83 y	1.71 x	736.71 x
	Mean	47.53 **a-d**	14.54 **a-i**	1.88 **ab**	706.67 **c-g**
Burpeachfive	2013	41.06 x	4.35 x	5.71 x	367.79 y
	2014	56.91 y	8.30 y	6.85 x	193.18 x
	Mean	48.98 **a-d**	6.32 **a-e**	6.28 **a-c**	280.48 **ab**
Canadian Harmony	2013	28.68 x	4.92 x	4.45 x	400.09 x
	2014	54.90 y	10.96 y	10.27 y	377.36 x
	Mean	41.79 **a-c**	7.94 **a-f**	7.36 **a-c**	388.73 **a-c**
Contender	2013	29.17 x	3.99 x	1.94 x	279.84 x
	2014	69.65 y	19.58 y	4.56 x	992.37 y
	Mean	49.41 **a-d**	11.78 **a-h**	3.25 **ab**	636.10 **b-g**

Coronet	2013	71.65 y	16.56 y	4.11 y	876.37 y
	2014	39.18 x	6.42 x	2.74 x	368.40 x
	Mean	55.42 **a-d**	11.49 **a-h**	3.42 **ab**	622.39 **b-g**
Crimson Rocket	2013	15.58 x	5.55 x	2.17 x	446.53 x
	2014	61.34 y	18.73 y	2.40 x	650.26 y
	Mean	38.46 **ab**	12.14 **a-h**	2.28 **ab**	548.39 **a-f**
Diamond Princess	2013	36.33 x	3.11 x	9.25 y	284.47 x
	2014	76.00 y	5.66 y	2.85 x	274.27 x
	Mean	56.16 **a-d**	4.39 **a-c**	6.05 **a-c**	279.37 **ab**
Early Loring	2013	32.16 x	2.15 x	3.08 x	409.13 x
	2014	94.31 y	18.35 y	13.24 x	709.13 y
	Mean	63.24 **a-d**	10.25 **a-h**	8.16 **a-c**	559.13 **a-f**
Ernie's Choice	2013	20.81 x	5.98 x	4.80 x	550.73 x
	2014	113.88 y	32.16 y	33.22 y	1121.79 y
	Mean	67.35 **a-d**	19.07 **e-i**	19.01 **e**	836.26 **f-i**
FA 52	2013	24.62 x	3.12 x	1.94 x	266.43 x
	2014	42.05 y	8.39 y	2.05 x	472.87 y
	Mean	33.34 **ab**	5.75 **a-e**	2.00 **ab**	369.65 **a-c**
FA 59	2013	17.82 x	5.07 x	3.31 x	388.82 x
	2014	62.48 y	29.50 y	5.71 y	590.01 y
	Mean	40.15 **ab**	17.28 **b-i**	4.51 **ab**	489.42 **a-e**
FA 80	2013	18.06 x	5.42 x	2.51 x	440.76 x
	2014	54.24 y	8.75 y	4.23 y	544.57 y
	Mean	36.15 **ab**	7.09 **a-e**	3.37 **ab**	492.67 **a-e**
Flavortop	2013	15.21 x	8.18 x	4.68 x	426.11 x
	2014	59.49 y	35.49 y	6.62 y	1271.01 y
	Mean	37.35 **ab**	21.83 **g-i**	5.65 **a-c**	848.56 **f-i**
FlavrBurst	2013	20.02 x	4.39 x	4.91 y	516.59 x
	2014	48.71 y	6.24 y	3.88 x	525.51 x
	Mean	34.37 **ab**	5.32 **a-d**	4.40 **ab**	521.05 **a-f**
Glohaven	2013	25.31 x	9.54 x	2.05 x	551.05 x
	2014	80.44 y	25.57 y	5.25 y	1064.69 y
	Mean	52.87 **a-d**	17.55 **c-i**	3.65 **ab**	807.87 **e-i**
Harrow Beauty	2013	36.77 x	2.32 x	1.82 x	181.99 x
	2014	150.82 y	10.32 y	17.81 y	241.09 x
	Mean	93.80 **cd**	6.32 **a-e**	9.82 **a-c**	211.54 **a**
Intrepid	2013	37.66 x	19.84 x	3.65 x	1011.32 x
	2014	99.38 y	23.67 x	6.17 y	1210.76 y
	Mean	68.52 **a-d**	21.76 **g-i**	4.91 **a-c**	1111.04 **i**
John Boy II	2013	35.12 x	4.98 x	3.42 x	381.89 y
	2014	56.32 y	7.17 y	8.45 y	225.37 x
	Mean	45.72 **a-d**	6.08 **a-e**	5.93 **a-c**	303.63 **ab**
Julyprince	2013	42.78 x	7.34 x	7.53 y	760.40 y
	2014	78.06 y	5.44 x	2.17 x	270.99 x
	Mean	60.42 **a-d**	6.39 **a-e**	4.85 **a-c**	515.70 **a-f**

Late 24-007	2013	12.93 x	0.64 x	4.91 x	124.15 x
	2014	63.70 y	19.42 y	6.62 y	558.31 y
	Mean	38.32 **ab**	10.03 **a-h**	5.77 **a-c**	341.23 **ab**
Late Large 23	2013	43.71 x	3.75 x	2.96 y	253.65 x
	2014	55.77 y	9.17 y	1.25 x	345.30 y
	Mean	49.74 **a-d**	6.46 **a-e**	2.11 **ab**	299.48 **ab**
Loring	2013	30.29 x	8.23 x	3.77 x	467.01 x
	2014	166.34 y	28.73 y	8.90 y	1322.60 y
	Mean	98.32 **d**	18.48 **d-i**	6.34 **a-c**	894.80 **g-i**
Majestic	2013	22.10 x	1.71 x	2.74 x	323.89 x
	2014	36.32 y	8.60 y	7.76 x	515.48 y
	Mean	29.21 **a**	5.15 **a-d**	5.25 **a-c**	419.68 **a-d**
PF 15A	2013	26.73 x	3.77 x	4.80 x	308.69 y
	2014	65.66 y	7.45 y	8.10 y	184.77 x
	Mean	46.20 **a-d**	5.61 **a-e**	6.45 **a-c**	246.73 **a**
PF 17	2013	19.10 x	6.22 x	2.17 x	412.18 x
	2014	105.13 y	19.67 y	15.07 y	512.14 y
	Mean	62.12 **a-d**	12.95 **a-h**	8.62 **a-c**	462.16 **a-d**
PF 24-007	2013	32.14 x	5.97 x	3.99 x	470.06 x
	2014	92.13 y	29.03 y	18.26 y	1021.65 y
	Mean	62.14 **a-d**	17.50 **c-i**	11.13 **bc**	745.86 **d-h**
PF Lucky 24B	2013	41.62 x	3.59 x	2.74 x	304.38 x
	2014	96.11 y	18.66 y	4.34 x	366.00 y
	Mean	68.86 **a-d**	11.13 **a-h**	3.54 **ab**	335.19 **ab**
Redglobe	2013	16.03 x	14.36 x	4.34 x	1029.40 x
	2014	69.29 y	26.85 y	6.62 x	1049.47 x
	Mean	42.66 **a-c**	20.61 **f-i**	5.48 **a-c**	1039.44 **hi**
Reliance	2013	40.04 x	4.49 x	2.39 y	286.49 x
	2014	68.15 y	15.50 y	1.03 x	299.43 x
	Mean	54.09 **a-d**	10.00 **a-h**	1.71 **a**	292.96 **ab**
Summerbeaut	2013	35.35 x	5.93 x	6.74 y	457.63 y
	2014	56.63 y	14.15 y	4.00 x	335.22 x
	Mean	45.99 **a-d**	10.04 **a-h**	5.37 **a-c**	396.42 **a-c**
Summergold	2013	26.83 x	26.18 y	8.56 x	1236.83 y
	2014	69.82 y	16.02 x	18.04 y	610.33 x
	Mean	48.33 **a-d**	21.10 **g-i**	13.30 **cd**	923.58 **g-i**
Suncrest	2013	40.00 x	17.33 x	5.48 x	1134.75 x
	2014	134.45 y	50.23 y	29.11 y	1614.24 y
	Mean	87.22 **b-d**	33.78 **j**	17.30 **de**	1374.49 **j**
Sunhigh	2013	20.98 x	3.51 x	1.71 x	331.20 x
	2014	61.27 y	10.96 y	3.42 x	509.23 y
	Mean	41.12 **a-c**	7.24 **a-e**	2.57 **ab**	420.21 **a-d**
Sweet Breeze	2013	32.07 x	5.72 x	2.05 x	384.43 x
	2014	78.70 y	17.89 y	11.64 y	709.13 y
	Mean	55.38 **a-d**	11.80 **a-h**	6.85 **a-c**	546.78 **a-f**

Sweet Dream	2013	30.20 x	8.83 x	3.08 x	798.65 y
	2014	42.59 y	9.40 x	2.74 x	673.45 x
	Mean	36.40 **ab**	9.11 **a-g**	2.91 **ab**	736.05 **d-h**
White County	2013	23.00 x	4.88 x	1.82 x	599.51 x
	2014	87.84 y	47.75 y	14.61 y	1533.00 y
	Mean	55.42 **a-d**	26.32 **i**	8.22 **a-c**	1066.25 **i**
Winblo	2013	44.52 x	34.66 y	6.85 y	1630.07 y
	2014	72.53 y	8.61 x	1.25 x	462.24 x
	Mean	58.53 **a-d**	21.64 **g-i**	4.05 **ab**	1046.15 **hi**
Late season					
Ambre™ Monam	2013	17.35 x	16.23 x	2.62 x	696.63 x
	2014	101.74 y	40.31 y	8.22 y	1423.70 y
	Mean	59.55	28.27 **b-d**	5.42 **ab**	1060.16 **e-g**
Augustprince	2013	33.44 x	8.96 x	3.19 x	755.50 y
	2014	134.79 y	29.55 y	24.32 y	118.60 x
	Mean	84.12	19.26 **a-c**	13.75 **cd**	437.05 **ab**
Autumn Flame	2013	23.14 x	6.19 x	5.14 x	428.96 x
	2014	78.10 y	20.18 y	4.79 x	760.00 y
	Mean	50.62	13.18 **ab**	4.97 **ab**	594.48 **a-d**
Autumn Red	2013	49.25 x	45.71 x	4.80 x	1282.59 x
	2014	63.41 x	43.50 x	4.80 x	1227.88 x
	Mean	56.33	44.60 **ef**	4.80 **ab**	1255.24 **gh**
Blake	2013	21.81 x	7.94 x	2.97 x	562.55 x
	2014	56.53y	31.26 y	6.85 y	1189.14 y
	Mean	39.17	19.60 **a-c**	4.91 **ab**	875.85 **c-f**
Burpeachfour	2013	41.12 x	7.20 x	7.99 x	711.55 x
	2014	117.84 y	27.30 y	7.53 x	1252.48 y
	Mean	79.48	17.25 **a-c**	7.76 **a-c**	982.02 **d-g**
Carolina Gold	2013	65.24 x	21.29 y	4.34 x	1264.29 y
	2014	91.51 y	13.73 x	16.44 y	624.91 x
	Mean	78.37	17.51 **a-c**	10.39 **a-c**	944.60 **d-g**
CaroTiger	2013	61.92 x	13.67 x	8.90 x	1439.36 y
	2014	73.80 x	30.43 y	9.93 x	1319.62 x
	Mean	67.86	22.05 **a-c**	9.42 **a-c**	1379.49 **h**
Cresthaven	2013	29.482 x	7.4307 x	3.078 x	465.036 x
	2014	40.79 y	34.40 y	12.79 y	1168.33 y
	Mean	35.14	20.92 **a-c**	7.93 **a-c**	816.68 **b-e**
Elberta	2013	46.19 x	38.29 x	7.88 x	1882.39 x
	2014	89.94 y	65.59 y	19.18 y	1855.51 x
	Mean	68.07	51.94 **fg**	13.53 **cd**	1868.95 **i**
Encore	2013	40.54 x	27.19 x	3.42 x	1251.80 x
	2014	139.26 y	53.48 y	14.04 y	1603.06 y
	Mean	89.9	40.34 **de**	8.73 **a-c**	1427.43 **h**

FA 17	2013	41.45 x	6.34 x	1.60 x	400.12 y
	2014	52.81 y	8.54 y	3.88 x	229.11 x
	Mean	47.13	7.44 **a**	2.74 **a**	314.62 **a**
FA 18	2013	22.33 x	5.45 x	2.05 x	446.70 x
	2014	82.48 y	20.14 y	21.69 y	725.69 y
	Mean	52.4	12.80 **ab**	11.87 **b-d**	586.20 **a-d**
FA 42	2013	33.27 x	14.71 x	3.19 x	935.99 x
	2014	130.40 y	47.16 y	9.42 y	1497.20 y
	Mean	81.83	30.94 **cd**	6.31 **a-c**	1216.59 **f-h**
Fantasia	2013	33.53 x	10.17 x	7.65 y	645.00 x
	2014	67.07 y	16.01 y	3.08 x	635.01 x
	Mean	50.3	13.09 **ab**	5.37 **ab**	640.01 **a-e**
Flameprince	2013	31.00 x	9.20 x	2.62 x	708.63 x
	2014	71.75 y	15.32 y	5.48 x	720.76 x
	Mean	51.37	12.26 **ab**	4.05 **ab**	714.69 **a-e**
Jerseyqueen	2013	51.87 x	35.58 x	5.94 x	1790.90 x
	2014	113.73 y	76.40 y	13.02 y	2439.68 y
	Mean	82.8	55.99 **g**	9.48 **a-c**	2115.29 **j**
Laurol	2013	47.13 x	18.56 x	6.62 x	1139.39 x
	2014	127.36 y	44.38 y	12.67 y	1664.24 y
	Mean	87.25	31.47 **cd**	9.65 **a-c**	1401.82 **h**
Madison	2013	42.56 x	9.64 x	2.39 x	604.89 x
	2014	80.95 y	20.87 y	3.54 y	741.13 y
	Mean	61.76	15.26 **a-c**	2.97 **a**	673.01 **a-e**
'NJ351' Gloria®	2013	21.13 x	3.57 x	1.71 x	404.01 x
	2014	124.32 y	21.39 y	3.65 x	906.57 y
	Mean	72.73	12.48 **ab**	2.68 **a**	655.29 **a-e**
'NJ352' Messina®	2013	35.14 x	6.34 x	3.42 y	416.79 x
	2014	69.24 y	12.59 y	1.82 x	515.90 y
	Mean	52.19	9.46 **a**	2.62 **a**	466.35 **a-c**
PF 2050	2013	35.19 x	6.44 x	3.77 x	484.56 x
	2014	140.20 y	17.62 y	4.34 x	864.69 y
	Mean	87.7	12.03 **ab**	4.05 **ab**	674.62 **a-e**
PF 25	2013	22.09 x	4.74 x	2.39 x	450.32 x
	2014	85.30 y	35.72 y	5.94 y	1459.45 y
	Mean	53.7	20.23 **a-c**	4.17 **ab**	954.89 **d-g**
PF 27A	2013	30.18 x	9.51 x	2.51 x	527.76 x
	2014	96.14 y	27.06 y	7.76 y	1027.97 y
	Mean	63.16	18.29 **a-c**	5.14 **ab**	777.87 **b-e**
PF 28-007	2013	35.92 x	3.98 x	2.62 x	306.23 x
	2014	95.29 y	23.55 y	6.62 y	1053.31 y
	Mean	65.6	13.76 **ab**	4.62 **ab**	679.77 **a-e**
PF 30-007	2013	12.84 x	6.38 x	2.17 x	385.99 x
	2014	138.71 y	15.80 y	13.24 y	419.30 x
	Mean	75.78	11.09 **ab**	7.70 **a-c**	402.64 **ab**

Redskin	2013	59.66 x	14.00 x	4.57 x	828.53 x
	2014	88.06 y	24.16 y	13.01 x	903.61 x
	Mean	73.86	19.08 **a-c**	8.79 **a-c**	866.07 **c-f**
Summer Fire	2013	11.63 x	1.94 x	2.62 x	283.50 x
	2014	109.32 y	13.76 y	14.61 y	547.73 y
	Mean	60.48	7.85 **a**	8.62 **a-c**	415.61 **ab**
SummerFest	2013	17.28 x	4.72 x	1.60 x	395.24 x
	2014	149.89 y	29.44 y	7.53 y	1167.24 y
	Mean	83.58	17.08 **a-c**	4.57 **ab**	781.24 **b-e**
Sweet N Up	2013	14.26 x	12.26 x	2.85 x	581.80 x
	2014	120.90 y	26.58 y	3.88 x	1027.00 y
	Mean	67.58	19.42 **a-c**	3.37 **ab**	804.40 **b-e**
Tra-Zee	2013	50.69 x	6.96 x	22.94 x	1078.21 y
	2014	95.64 y	26.44 y	11.30 x	74.60 x
	Mean	73.17	16.70 **a-c**	17.12 **d**	576.41 **a-d**
Victoria	2013	51.94 x	12.35 x	3.65 x	959.57 x
	2014	116.29 y	21.87 y	7.08 x	1005.01 x
	Mean	84.11	17.11 **a-c**	5.37 **ab**	982.29 **d-g**

[z] Means within columns and harvest season followed by common letters do not differ at the 5% level of significance, by Student-Newman-Keuls's and *t* test. Letters (a-k) represent significant differences between cultivars across two years according to Student-Newman-Keuls's test, while letters (x, y) represent significant differences between years according to *t* test.

was much wider in our study, which could be explained by differences in the analyzed material. As expected, the highest accumulation of anthocyanin (16.9 and 42.8 mg C3GE/kg FW in 2013 and 2014, respectively) was observed in the red flesh advanced selection, which was 5-fold higher than in white (4.9 and 8.1 mg C3GE/kg FW in 2013 and 2014, respectively) and yellow (3.9 and 7.2 mg C3GE/kg FW in 2013 and 2014, respectively) cultivars (Tables 4 and 5). Vizzotto et al. (2007) reported no significant differences in accumulation of anthocyanins between white and yellow flesh peaches and a 9 to 50-fold increase in anthocyanin accumulation in red-fleshed peaches (~ 45 – 266 mg C3GE/100g fresh tissue). Although Vizzotto et al. (2007) measured accumulation of phenolic compounds in samples comprised from both flesh and skin, accumulation in white and yellow flesh material was similar to those reported in this study regardless of the level of skin blush. Our results suggest that red-fleshed germplasm should be used to significantly increase the anthocyanin levels within the breeding program. Currently, there is no substantial red-fleshed peach market in the United States, and it is questionable if the increased health benefits of red-fleshed cultivars would be sufficient to overcome the lack of acceptance in today's market.

No significant differences in anthocyanin accumulation were observed between different ripening seasons in 2013 (Table 4). However, ripening season gradually affected the accumulation of anthocyanins as indicated by the statistically significant differences between different ripening seasons in 2014 (Table 5). The lowest accumulation [3.6 (± 2.3) mg C3GE per kg of FW] was observed in early ripening and the highest [10.1 (± 6.1) mg C3GE per kg of FW] in late ripening peach and nectarine cultivars with mid-season peaches and nectarines accumulating on average 8.2 (± 7.1) mg C3GE per kg of FW (Table 3). Increases in the level of anthocyanins as the ripening season progresses could be explained in the context of increased sugar concentration in fruit of late ripening cultivars. The link between anthocyanin levels

Table 4. Comparison of phytochemical compounds and antioxidant capacity among peaches and nectarines, flesh colors, ripening groups and cultivar realease date in 2013. No., number of observations; GAE, Gallic acid equivalents; CE, Catechin equivalents; C3GE, Cyanidin-3-glucoside equivalents; TE, Trolox equivalents.

	No.[z]	Total phenolics (mg GAE/100g)	Flavonoids (mg CE/100g)	Anthocyanins (mg C3GE/kg)	Antioxidant capacity (µg TE/g)
Fruit type					
Peach	115	30.6 ± 13.0 b[y]	9.2 ± 8.5 a	3.9 ± 2.8 a	596.7 ± 366.4 a
Nectarine	17	23.2 ± 10.1 a	9.3 ± 7.5 a	5.4 ± 4.1 a	537.6 ± 282.7 a
Flesh color					
White	29	29.6 ± 13.9 a	10.6 ± 9.1 a	4.9 ± 3.6 a	628.0 ± 361.1 a
Yellow	103	29.6 ± 12.7 a	8.8 ± 8.1 a	3.9 ± 2.8 a	578.1 ± 356.0 a
Red[x]	1	57.3 b	31.1 b	16.9 b	1689.5 b
Ripening season					
Early	40	23.7 ± 8.5 a	7.0 ± 5.2 a	4.2 ± 3.5 a	460.2 ± 224.3 a
Mid	50	29.7 ± 11.5 b	7.6 ± 6.6 a	3.8 ± 1.9 a	524.3 ± 306.3 a
Late	42	35.1 ± 15.5 c	13.2 ± 10.9 b	4.3 ± 3.6 a	789.0 ± 428.4 b
Release date					
<1960	14	37.2 ± 20.5 b	13.7 ± 12.2 a	4.1 ± 2.1 a	771.7 ± 483.7 a
1960 - 1990	28	33.9 ± 9.5 ab	10.9 ± 9.3 a	4.7 ± 4.1 a	677.7 ± 406.7 a
>1990	81	27.2 ± 11.7 a	8.1 ± 7.0 a	4.1 ± 2.8 a	545.4 ± 307.9 a

[z] Data indicates mean ± standard deviation.
[y] Means within column and fruit characteristic followed by common letters do not differ at the 5% level of significance, by Student-Newman-Keuls's and *t* test.
[x] Data obtained for red flesh accession used only in flesh color comparisons.

Table 5. Comparison of phytochemical compounds and antioxidant capacity among peaches and nectarines, flesh colors, ripening groups and cultivar realease date in 2014. No., number of observations; GAE, Gallic acid equivalents; CE, Catechin equivalents; C3GE, Cyanidin-3-glucoside equivalents; TE, Trolox equivalents.

	No.[z]	Total phenolics (mg GAE/100g)	Flavonoids (mg CE/100g)	Anthocyanins (mg C3GE/kg)	Antioxidant capacity (µg TE/g)
Fruit type					
Peach	115	76.1 ± 33.2 a[y]	19.7 ± 13.9 a	7.6 ± 6.4 a	698.9 ± 455.9 a
Nectarine	17	64.8 ± 29.9 a	21.9 ± 15.1 a	6.2 ± 4.2 a	662.0 ± 371.1 a
Flesh color					
White	29	67.6 ± 32.8 a	21.5 ± 15.8 a	8.1 ± 6.2 a	705.2 ± 422.7 a
Yellow	103	76.6 ± 32.8 a	19.6 ± 13.8 a	7.2 ± 6.2 a	691.0 ± 452.8 a
Red[x]	1	89.6 b	35.1 b	42.8 b	819.9 b
Ripening season					
Early	40	51.4 ± 16.9 a	14.5 ± 10.7 a	3.6 ± 2.3 a	449.3 ± 237.3a
Mid	50	74.7 ± 31.6 b	17.1 ± 10.6 a	8.2 ± 7.1 b	632.4 ± 358.6 b
Late	42	96.5 ± 31.3 c	28.7 ± 16.2 b	10.1 ± 6.1 b	1000.8 ± 515.3 c
Release date					
<1960	14	86.0 ± 38.7 a	29.2 ± 16.2 b	8.9 ± 6.2 a	959.3 ± 464.1 b
1960 - 1990	28	72.2 ± 29.3 a	20.8 ± 15.7 a	7.3 ± 5.5 a	709.8 ± 522.4 a
>1990	81	73.3 ± 33.0 a	18.6 ± 12.7 a	7.3 ± 6.6 a	653.3 ± 408.9 a

[z] Data indicates mean ± standard deviation.
[y] Means within column and fruit characteristic followed by common letters do not differ at the 5% level of significance, by Student-Newman-Keuls's and *t* test.
[x] Data obtained for red flesh accession used only in flesh color comparisons.

and levels of monosaccharides (glucose, galactose, rhamnose, and arabinose), di, or trisaccharides in the tissue has been proposed (Bureau et al., 2009).

Antioxidant capacity. No significant differences were observed for antioxidant capacity between peach and nectarine, nor white and yellow flesh (Tables 4 and 5). The red flesh selection had a 2-fold higher antioxidant capacity on average (1,254.7 μg Trolox/g FW) than white and yellow flesh peach or nectarine cultivars (~600 μg Trolox/g FW). White flesh cultivars ranged from 136 to 1,679.3 μg Trolox per g of FW for 'Glacier' and 'Belle of Georgia', respectively (Table 2). Yellow flesh cultivars had a wider range in antioxidant capacity than white, with the overall lowest and highest values observed in 'Crimson Lady' and 'Jerseyqueen' (93.3 and 2,115.3 μg Trolox per g of FW, respectively) (Table 3). Higher antioxidant capacity in red flesh peaches (Vizzotto et al., 2007) with no significant differences in antioxidant capacity of white and yellow, peach and nectarine cultivars is in agreement with previous reports (Tomás-Barberán et al., 2001; Vizzotto et al., 2007). The average antioxidant capacity measured in yellow and white flesh peach germplasm in this study was comparable to the values reported in peach breeding germplasm by Vizzotto et al. (2007) and higher than those reported for Spanish peach cultivars (Reig, et al., 2013) and peach (Cantín et al., 2009b) and nectarine (Abidi et al., 2011) progeny from Spanish breeding programs. This is to be expected, since variability represented in previously analyzed material was lower than that in peach breeding germplasm analyzed here.

Ripening season significantly influenced antioxidant capacity, with the late ripening peach and nectarine cultivars exhibiting the highest antioxidant capacity (789 and 1000.8 μg Trolox/ g FW) in 2013 and 2014, respectively (Tables 4 and 5). Early ripening peaches and nectarines averaged 454.7 (± 230.4) μg Trolox per g of FW, while mid-season cultivars averaged 578.2 (± 337.7) μg Trolox per g of FW (Tables 4 and 5). This trend was true regardless of fruit type, peach or nectarine, and flesh color, white or yellow; antioxidant capacity in late ripening cultivars was significantly higher than in early and mid-season ones (Tables 4 and 5). However, the opposite was observed in apricot where significantly higher levels of antioxidants, such as epigallocatechin and vitamins (A, E, b-carotene, and lycopene), were found in early-ripening cultivars (Gundogdu et al., 2013). The high antioxidant capacity in late-maturing peach cultivars could be due to a high SSC. A positive correlation between maturity date and SSC in peach was previously reported (Eduardo et al., 2011). In addition, significant and positive correlations between SSC and antioxidant capacity, phenolics, and flavonoids were reported in several studies with peach and nectarine (Abidi et al., 2011; Cantín et al., 2009b; Font i Forcada et al., 2014) supporting the essential role of sugars in the regulation of synthesis of phenolic compounds (DeJong, 1999; Font i Forcada et al., 2014).

Interestingly, the highest antioxidant capacity, on average, was observed in heirloom cultivars (with release date < 1960), such as 'Jerseyqueen', 'Elberta' and 'Belle of Georgia', (2,115.3, 1,868.9 and 1,679.3 μg Trolox/ g FW, respectively) (Tables 2 and 3), while newer cultivars had the lowest capacity (Table 5) to sequester/ scavenge reactive oxygen species (ROS). The high antioxidant capacity observed in heirloom cultivars suggests that breeding for improved fruit quality and increased bioactive compounds is possible and that breeders could consider using heirloom cultivars to achieve that goal. The significant accumulation of non-enzymatic antioxidants (total phenolics, flavonoids and their subclass anthocyanins) in peach and nectarine cultivars supports the importance of these fruits for human health.

Genotype effects. The effect of genotype was studied on 21 cultivars and 1 advanced selection across three years (2012 – 2014).

No significant differences were observed within the genotypes for their total phenolics accumulation and antioxidant capacity, with ‘Elberta’, ‘99P4388’, and ‘Belle of Georgia’ exhibiting the highest accumulation of total phenolics (≥ 50 mg GAE/100g FW) and the highest antioxidant capacity (> 1000 µg Trolox/g FW) (Figure 1). Lack of statistical differences might suggest stable accumulation of total phenolics and thus antioxidant capacity in these genotypes across different seasons, or might be an artifact from the small number of replicates (three/ genotype). When comparing mean values, a wide range of genetic variability among genotypes was observed showing the potential of this material for breeders to improve levels of phytochemical compounds.

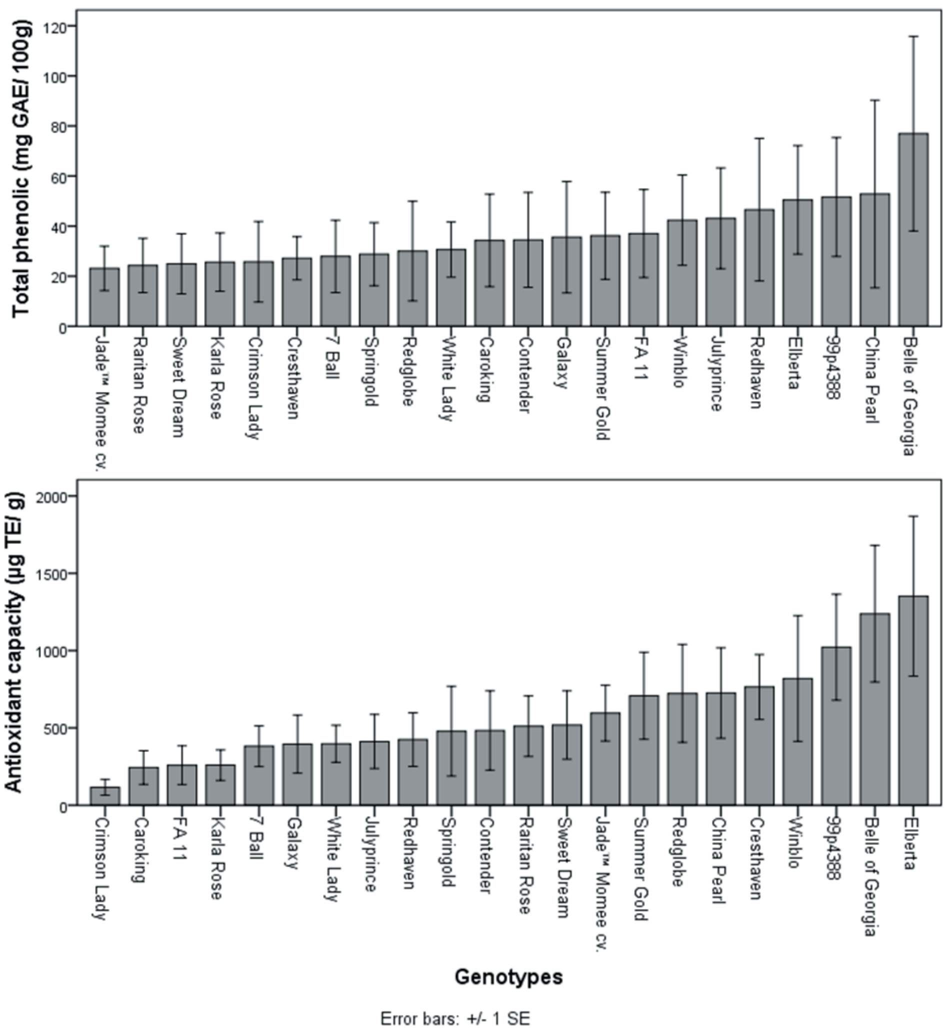

Fig. 1. Total phenolics and antioxidant capacity evaluated among 22 peach and nectarine accessions over 3 years.

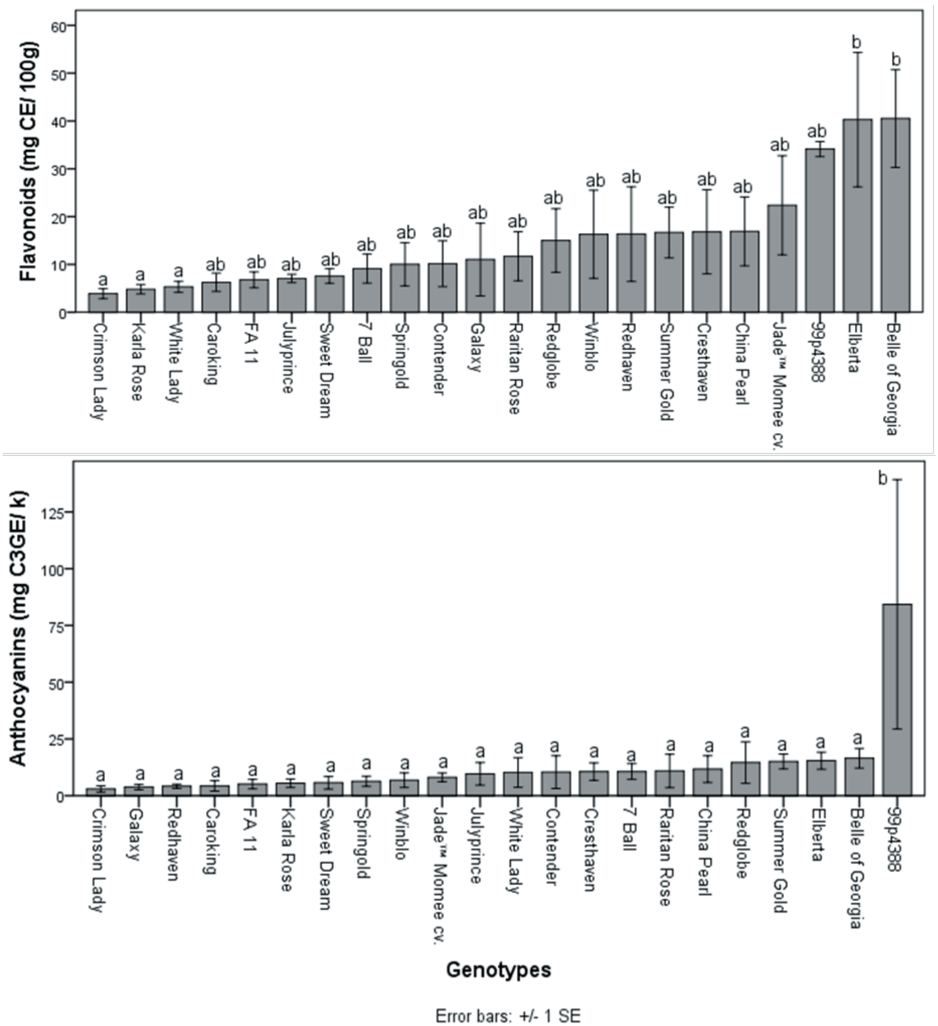

Fig. 2. Flavonoids and anthocyanins evaluated among 22 peach and nectarine accessions over 3 years. Different letters indicate significant differences at $P < 0.05$ according to Student-Newman-Keuls's test.

Wide variability was also observed among 22 genotypes for their flavonoids and anthocyanins accumulation (Figure 2). The lowest accumulation of flavonoids was observed in ‘Crimson Lady’ (3.9 mg CE/100g FW) across the three years, while the highest accumulation (> 30) was observed in advanced selection 99P4388, ‘Elberta’, and ‘Belle of Georgia’ (34.1, 40.3, and 40.5 mg CE/100g FW, respectively). Anthocyanins averaged significantly higher in red- fleshed advanced selection (84.3 mg C3GE/kg FW) than in yellow and white genotypes (3 – 16.5 mg C3GE/kg FW) across three years, which is expected given high level of anthocyanin pigments in red-fleshed peaches.

Environment effects. Different environmental conditions over the three experimental years affected the accumulation of bioactive compounds and their relative antioxidant

Table 6. Accumulation of total phenolics, flavonoids, anthocyanin, and antioxidants observed in 22 accessions across three seasons. GAE, Gallic acid equivalents; CE, Catechin equivalents; C3GE, Cyanidin-3-glucoside equivalents; TE, Trolox equivalents.

Year	Total phenolics (mg GAE/100g)[z]	Flavonoids (mg CE/100g)	Anthocyanin (mg C3GE/kg)	Antioxidants (μg TE/g)
2012	6.4 ± 3.5 a[y]	8.1 ± 7.7 a	24.6 ± 19.9 b	220.8 ± 153.5 a
2013	34.9 ± 13.5 b	14.6 ± 12.6 b	5.2 ± 3.7 a	819.1 ± 509 c
2014	69.1 ± 28.4 c	22.2 ± 17.1 c	8.1 ± 6.7 a	708.9 ± 479.9 b

[z] Data indicates means ± standard deviations.
[y] Means within columns followed by common letters do not differ at the 5% level of significance, by Student-Newman-Keul's test.

capacity (Table 6) resulting in a large deviation from the mean. During the entire ripening season (May through August), there were 25 days exceeding a maximum daily temperature of 35 ºC in 2012 compared to one day in 2013 and zero days in 2014 (Figure 3). Total rainfall from May through August was 51.3, 102.3 and 35.7 cm in 2012, 2013, and 2014, respectively. Furthermore, daily rainfall at the experimental location exceeded 3 cm 14 times during 2013 season, while only 5 times during each of the 2012 and 2014 seasons. Average antioxidant capacity (in μg Trolox / g FW) was significantly higher in

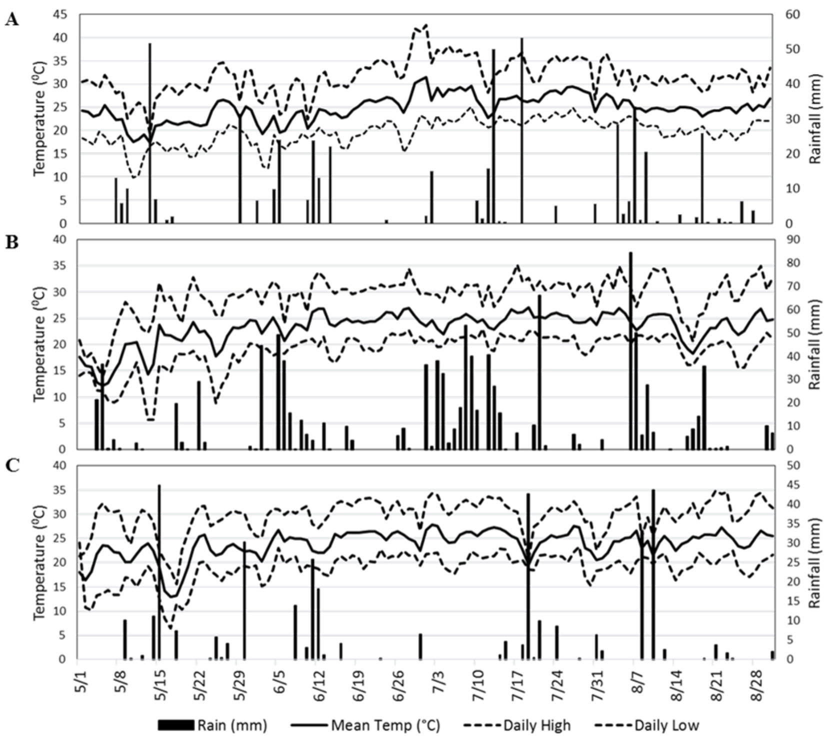

Figure 3. Weather conditions during the ripening season. Daily air temperature (°C) and rainfall (mm) at the Musser Fruit Research Center, Seneca, SC over three years [2012 (A), 2013 (B), and 2014 (C)].

2013 (819.1) than in 2012 (222.8) and 2014 (708.9). At the same time, accumulation of total phenolics (mg GAE/100 g FW) and flavonoids (mg CE/100g FW) were significantly higher in 2014 (69.1 and 22.2, respectively) when compared to 2012 (6 and 8.2, respectively) and 2013 (34.9 and 14.6, respectively). There was an opposite trend for the anthocyanin accumulation, with the highest average value in 2012 (24.6 mg C3GE/kg FW) and the lowest in 2013 (5.2 mg C3GE/kg FW) (Table 7). Wet conditions in 2013 might have contributed to the overproduction of ROS under anaerobic soil conditions causing the highest antioxidant capacity (Agati et al., 2012; Gill and Tuteja, 2010; Kassim et al., 2009). The toxic effects of ROS are counteracted by enzymatic (superoxide dismutase, catalase, and glutathione peroxidase) as well as non-enzymatic (tocopherol, ascorbic acid, glutathione, carotenoids, and phenolic compounds) antioxidative systems (Ahmad et al., 2010; Blokhina et al., 2003; Gill and Tuteja, 2010). Phenolics are diverse secondary metabolites that include simple phenols (total phenolics) and polyphenols (flavonoids and their subgroup anthocyanins) (Goleniowski et al., 2013). Phenolic compounds are ideally structured for free radical scavenging activity (Goleniowski et al., 2013), therefore their depletion is an effect of increased antioxidant productivity in protecting plants against oxidative stress damages. High anthocyanin accumulation in 2012 could be explained by the daily average temperatures of 25 ºC, which was 10 ºC warmer than in 2013. Furthermore, solar radiation (837.6 W/m²) recorded in 2012 was very high compared to 2013 and 2014 (data not shown). Influence of solar radiation, UV-B radiation in particular, on expression of genes such as bHLH3, WD40, and MYBPA1 that control anthocyanin biosynthesis in peach peel and flesh was previously reported (Ravaglia et al., 2013). However, Yang et al. (2013) documented inhibition of accumulation of anthocyanins under both high radiation and high precipitation experimental years in red currant (*Ribes* sp.). The reduced accumulation of anthocyanins during a wet cloudy season (2013) in our study was similar to the negative effect of high precipitation on anthocyanin accumulation in red currant (Yang et al., 2013).

Correlation analysis. Significant positive linear correlations were observed between different phenolic compounds across years of the study (Table 7). Antioxidant capacity was positively correlated with all other bioactive compounds. There was a positive correlation between flavonoids and total phenolics with the highest r-values in 2013 ($r = 0.544$, $P < 0.01$, data not shown). Antioxidant capacity was positively correlated with total phenolics in both years (2013 and 2014) ($r = 0.615$ and 0.484, $P < 0.01$, respectively). A similar correlation between antioxidant capacity and total phenolics was previously reported in peach and nectarine (Abidi et al., 2011; Cantín et al., 2009b) and in sweet cherry (Serrano et al., 2005). Overall, the highest positive correlation coefficient (> 0.80) was observed in 2013 between antioxidant capacity and flavonoids ($r = 0.898$). However, the lowest correlation was between antioxidant capacity and anthocyanins in 2013 ($r = 0.292$, $P < 0.01$). This could be attributed to low accumulation of anthocyanin observed in peaches and nectarines in this study. Fur-

Table 7. Pearson's correlation coefficients between antioxidant capacity and different phytochemical compounds in 132 peach and nectarine cultivars. GAE, Gallic acid equivalents; CE, Catechin equivalents; C3GE, Cyanidin-3-glucoside equivalents; TE, Trolox equivalents.

Year	Total phenolics (mg GAE/100g)	Flavonoids (mg CE/100g)	Anthocyanin (mg C3GE/kg)
2013	0.615**	0.898**	292**
2014	0.484**	0.875**	431**

** Significant correlation at $P < 0.01$.

thermore, antioxidant capacity was linearly and positively correlated with both flavonoids and total phenolics. The highest correlation coefficients (r = 0.90 and 0.86) were between antioxidant capacity and flavonoids in 2013 and 2014, respectively (3), indicating an important role flavonoids play as non-enzymatic antioxidants in protecting against free radicals and enhancing the importance of phenolic compounds in the antioxidant property of plant extracts. Correlation analysis between phytochemicals and fruit quality traits revealed total phenolics were significantly positively correlated with fruit firmness (FF; r = 0.425) and SSC (r = 0.404), but negatively correlated with fruit weight (FW; r = - 0.314) (Table 8). Flavonoids and anthocyanins were positively correlated with SSC (r = 0.425 and 0.374, respectively). Antioxidant capacity also exhibited a positive correlation with SSC (r = 0.416). SSC had the highest positive correlation ($P < 0.01$) with all phytochemical compounds reflecting the role that sugars play in complex metabolic processes, such as phenolics and flavonoids biosynthesis (DeJong, 1999; Font i Forcada et al., 2014). The positive correlation between SSC and phytochemical compounds reported in our study is in agreement with previous reports for peach (Font i Forcada et al., 2014; Abidi et al., 2011; Cantín et al., 2009 a, b).

Principal components analysis (PCA). The PCA of 132 individuals showed that 53 % of the observed variance could be explained by the first two components (PC1 and 2). PC1 includes phytochemical compounds and some fruit quality parameters, such as SSC, FF and index of absorbance difference (IAD; IAD), while PC2 represents FS, FW and RI (Figure 5). The PC1 and PC2 axes explained 31.7 % and 21.3 % of total variability, respectively. The PCA results confirmed a close relationship between bioactive compounds and SSC as observed with the Pearson correlation, supporting the hypothesis that sweeter fruit tend to have higher levels of bioactive compounds and the role of SSC in the upregulation of plant secondary metabolites (Font i Forcada et al., 2014; DeJong, 1999). In addition, FF and IAD grouped together as they significantly and positively associated with each other (Figure 5). High correlations between fruit maturity, IAD, and flesh firmness were previously reported in stone fruits particularly in peach and nectarine (Gasic et al., 2016; Infante, 2012) indicating more mature fruit being less firm and that firmness is a good indicator for determining the optimal maturity. However, IAD is highly genotype dependent (Gasic et al., 2016; Ziosi et al., 2008). In standard soft-

Table 8. Pearson's correlation coefficients between phytochemicals and fruit quality traits observed in 132 peach and nectarine cultivars. TE, Trolox equivalents; GAE, Gallic acid equivalents; CE, Catechin equivalents; C3GE, Cyanidin-3-glucoside equivalents; FS, Fruit size; FW, Fruit weight, FF, Fruit firmness; SSC, Soluble solids concentration; TA, Titratable acidity; RI, Ripening index.

Trait	FS[z]	FW	FF	SSC	TA	RI
Antioxidant capacity (µg TE/g)	ns[y]	ns	ns	0.416**	ns	ns
Total phenolics (mg GAE/100g)	ns	-0.314**	0.425**	0.404**	ns	ns
Flavonoids (mg CE/100g)	ns	ns	ns	0.425**	ns	ns
Anthocyanin (mg C3GE/kg)	ns	-ns	ns	0.374**	ns	ns

[z] Data represents average of 2 years (2013 and 2014)
[y]** Significant correlation at $P < 0.01$; ns = non significant

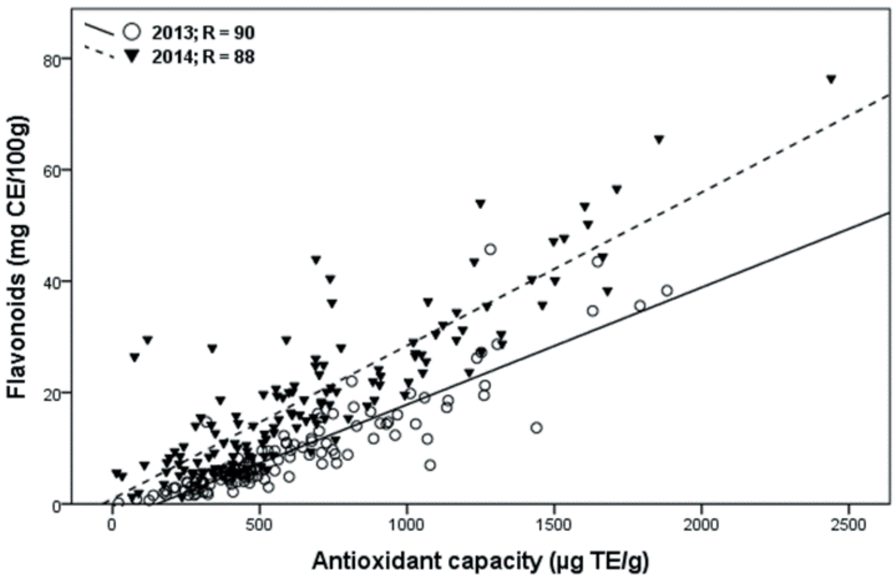

Fig 4. Relationship between antioxidant capacity and flavonoid accumulation in the 132 peach and nectarine cultivars. Slopes are signficant at the 1% level.

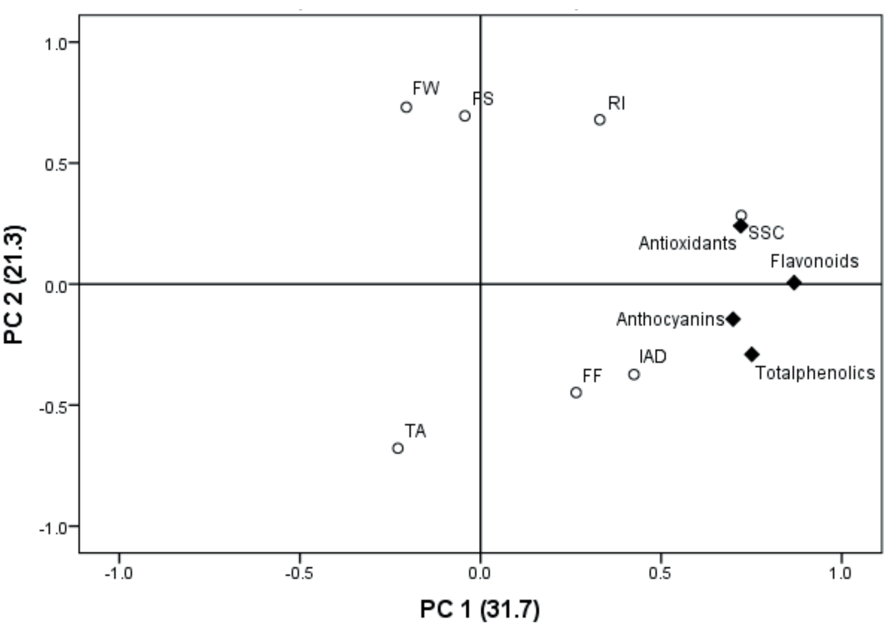

Fig. 5. Principal component analysis of the phytochemical compounds (black diamonds) and fruit quality (white circles) evaluated on 132 peach and nectarine cultivars across 2 years (2013 and 2014). Fruit quality abbreviations: FS, Fruit size; FW, Fruit weight; FF, Fruit firmness; SSC, Soluble solids concentration; TA, Titratable acidity; SSC/TA, Ripening index (RI); IAD, Index of Absorbance Difference (I_{AD}).

ening, melting, freestone peach and nectarine cultivars, FF and I_{AD} are highly correlated, while in slow-softening type cultivars, such as 'Sweet Dream', FF at the time of harvest is high but the IAD value is low (less or no chlorophyll). The distribution of traits in PC2 is likely due to a strong correlation between the traits. Indeed, the high positive correlation observed between FW and FS in this study is in agreement with previous studies in peach (Yamaguchi et al., 2002) and other fruits (Bohner and Bangerth, 1988; Ho, 1996) supporting the hypothesis that final fruit size and weight are determined by cell division and expansion beside carbohydrate dilution within cells. In addition, grouping TA and RI separately in PC2 (negative side and positive side, respectively) could be interpreted by the negative correlation between these two traits (data not shown), where the RI was determined as the SSC/TA ratio given that more mature fruit have less acidity and better taste for some consumers.

Conclusion

Variability in the antioxidant capacity and accumulation of bioactive compounds within peach breeding germplasm could provide genetic opportunities for breeding programs to continue enhancement of these healthy traits in newly developed cultivars while maintaining other fruit qualities. It also portrays peach as a valuable source of health promoting compounds for human consumption and provides valuable marketing tools to growers and retailers for delivering healthier food choices to consumers. Significant variation in the antioxidant capacity and bioactive compounds was observed in peach germplasm with different flesh color, ripening season, release date, and across different years. In general, results indicated that late ripening peach cultivars such as white flesh 'Belle of Georgia' and yellow flesh 'Jerseyqueen' and 'Elberta' have the highest accumulation of phenolic compounds and antioxidant capacity. Both 'Belle of Georgia' and 'Elberta' are heirloom cultivars and ancestors of the modern U.S. peach germplasm that played an essential role in the development of the U.S. peach industry. In addition, inclusion of the red flesh advanced selection 99p4388 in the study confirmed the potential of red flesh material for introducing variability in the fresh peach market and offering different healthy choices to consumers. Improvement of phytochemical compounds in newly developed cultivars is one of the objectives in many breeding programs besides improving fruit quality traits. Further study regarding analyzing individual phenolic compounds is needed to account for different health effects that individual phenolic compounds have on the human body.

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