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Breeding strategies for identifying superior peach genotypes resistant to brown rot

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ABSTRACT

A sustainable approach to control the incidence of brown rot in pre- and post-harvest management is to select genotypes with high contents of antioxidant compounds and tolerance to *Monilinia laxa* (Aderh. and Ruhland) Honey. In this study, 68 progenies of the 'Babygold 9' × 'Crown Princess' population from the EEAD-CSIC breeding program were screened under controlled conditions for a period of 3 years (2013–2015). Susceptibility to brown rot was evaluated after inoculating 20 healthy fruits per genotype with *M. laxa*. Brown rot incidence, lesion diameter, and colonization extent, as well as the severities of these issues, were calculated after 5 days of incubation. Physicochemical traits, such as fruit firmness and soluble solids content, were also recorded before and after storage. Titratable acidity, pH, and antioxidant composition were measured at harvest. Significant differences were found for pathogenic traits, as well as for contents of vitamin C, total phenolics, flavonoids, and anthocyanins, within genotypes in this population. Negative correlations were also found between the content of phytochemical compounds (such as anthocyanins and total phenolics), as well as disease incidence and severity. Differences in susceptibility to brown rot confirm the genetic variability available in these progeny. This allowed the selection of six genotypes highly resistant to brown rot of *M. laxa*, with high organoleptic properties and high phenol content, to be introduced in our peach breeding program.

1. Introduction

The storage life and commercial shelf life of the peach [*Prunus persica* (L.) Batsch] are negatively influenced by pre- and post-harvest diseases that are principally associated with brown rot (Sisquella et al., 2014). Brown rot of stone fruits is a disease primarily caused by *Monilinia* species, such as: *M. laxa* (Aderh. and Ruhland) Honey; *M. fructigena* Honey; *M. fructicola* (G. Winter) Honey and *M. polystroma* (G. Leeuwen) L.M. Kohn (Jansch et al., 2012). In peach, the pathogen initiates and encourages flower blights, twig and branch death, spurs, and fruit rot in the field (Gell et al., 2007). The activity of the pathogen on peach is therefore highly destructive from the flowering stage, to fruit production, and storage (Thomidis and Exadaktylou, 2010; see Obi et al., 2018b for a review).

In Spain, *M. laxa* and *M. fructicola* have been the most recurrent pathogens since the dislodgment of *M. fructigena* from Spain in 2010 (Villarino et al., 2013). These species cause over 60% fruit loss after harvest (Villarino et al., 2012; Egüen et al., 2015), mostly under favourable environmental conditions for the commencement and growth of diseases in orchards.

Host tolerance to plant pathogens is important for the development of cost effective and environmentally safe strategies for disease management (Gradziel, 1994). Similarly, according to Gell et al. (2007) the use of resistant cultivars in crop improvement is critical for crop protection, since plants and plant products are usually protected from (prophylactic) (Mooney et al., 2012), rather than cured of, diseases (chemotherapeutic) (Obi et al., 2018b). The choice of cultivar significantly influences rot incidence and severity among other potential factors in stone fruits (Tarbath et al., 2014). and, therefore, are effective at disease control (Kreidl et al., 2015). The long-term prophylactic

Abbreviations: AsA, Ascorbic acid; B9 × CP, 'Babygold 9' × 'Crown Princess'; %BRI, percentage brown rot incidence; LD, lesion diameter; C3GE, cyanidin-3-glucoside equivalents; CE, catechin equivalents; CEx, colonization extent; LS, lesion severity; CS, colonization severity; %C, percentage colonization; GAE, gallic acid equivalents; HD, harvest date; FW, fresh weight; FtW, fruit weight; FF, fruit firmness; SSC, soluble solids content; TA, titratable acidity; Vit C, Vitamin C; TPC, total phenolic content; JDs, Julian days; Vs, versus

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treatment of peach, using *M. laxa* resistant cultivars, will ensure prevention of pathogenic problems in orchards. Resistant genotypes will allow sustainable control with zero pesticide residues on fruits, improving the safety of harvesting and decreasing disease problems during storage, thereby leading to enhanced economic benefits. The total absence of pesticide residues in prophylactic resistant peach cultivars would be environmentally beneficial (Usall et al., 2016). However, disease resistant cultivars are not readily available for many fruit crops (Spiers et al., 2005), including commercial peach cultivars.

Developing peach cultivars that are resistant to *M. laxa* pathogen requires, in the first instance, the identification of existing resistant and susceptible genotypes by screening individuals from a germplasm (Rubos et al., 2008). Although most commercial peach cultivars are susceptible to *Monilinia* spp., a few resistant cultivars have been identified (Gradziel and Wang, 1993; Martínez-García et al., 2013; Oliveira-Lino et al., 2016; Obi et al., 2017). The relative tolerance or susceptibility of fruit to disease has therefore often been used to select disease resistant genotypes for the purpose of breeding peach (Gradziel, 1994). Selection within breeding descendant populations has been carried out for both peach and nectarine (Bassi et al., 1998; Pacheco et al., 2014; see Oliveira-Lino et al., 2016 and Obi et al., 2018b for details), and for other fruit germplasm such as apricot (Walter et al., 2004), plum (Pascal et al., 1994), and apple (Biggs and Miller, 2004). Previous studies have demonstrated that powerful antioxidants such as phenolic acids, flavonoids, and anthocyanins are present in the phytochemical compounds produced by peach cultivars (Giménez, 2013; Ágreda, 2016; Saidani et al., 2017). These bioactive compounds, especially chlorogenic and neochlorogenic acids, may confer important preservative functions during postharvest handling in the peach industry (Villarino et al., 2011; Pacheco et al., 2014; see Oliveira-Lino et al., 2016 and Obi et al., 2018b, in details). In addition, considering the recent drive for alternative technologies that can effectively control postharvest diseases of stone fruits (Mari et al., 2015; Usall et al., 2015, 2016), any evidence regarding compounds inhibitory to brown rot development would influence breeding schemes, and would be useful for the postharvest peach industry.

There is limited information on peach pathogenic tolerance to *M. laxa* brown rot in their breeding descendants, and their relationships with quality and phytochemical traits in fruits during postharvest handling. This study aimed to identify superior Spanish peach cultivars that exhibit high tolerance to *M. laxa* brown rot, and possess high levels of antioxidants. The specific objectives of this work, therefore, were to evaluate tolerance to *Monilinia laxa* brown rot within the breeding descendant population of 'Babygold 9' × 'Crown Princess', and to examine whether fruit quality and phytochemical composition correlate with pathogen tolerance. Finally, the identification of biochemical compounds associated with brown rot tolerance would impact breeding strategies, beneficial to the postharvest industry, and facilitate environmental sustainability.

2. Materials and methods

2.1. Plant material

The plant materials are progenies from a controlled biparental cross of two commercial cultivars, 'Babygold 9' × 'Crown Princess' (B9 × CP). These genotypes were propagated during 2000 and 2001 in collaboration with Agromillora Catalana S.L. (Barcelona, Spain). Both the progenitors and the entire progeny are yellow fleshed, clingstone peach. The resulting seedlings were budded on GF677 rootstock, and established in 2002 at the Estación Experimental de Aula Dei-CSIC (Zaragoza, Spain). Trees were trained to the standard open vase system, hand thinned, and subsequently grown under standard conditions of irrigation, fertilization, and pest and disease control chemical spray programmes. For the 3 years of the study (2013–2015), any fungicide treatment was applied in the field prior to harvest with adequate consideration to the free entry period and harvesting for evaluation. A total of 68 genotypes were harvested in the 2013 and 2014 seasons (Supplementary Table 1). Seventeen genotypes with lesion severity (LS) < 40 mm were then pre-selected, either in 2013 or 2014, or when the mean value for both years was below 40 mm (Obi et al., 2017), and harvested in 2015 to validate results concerning *M. laxa* tolerance. The pathogenic traits [percentage of brown rot incidence (%BRI), lesion diameter (LD), and colonization extent (CEx)] were measured for each seedling tree separately over the 3-year period, and the means of the 17 selected genotypes were calculated. Fruits were subjectively selected and harvested based on optimum maturity [(Cantín et al., 2009) (expressed on visual colour change and manual evaluation of firmness, favouring apparently healthy fruit of uniform ripeness and size)]. Fruits were disinfected as described by Obi et al. (2017).

2.2. Pathogen culture, conidia production, and inoculation

The procedure adopted is as described by Obi et al. (2017). Briefly, the culture of *Monilinia laxa* (Aderh. & Ruhland) Honey, isolate number: CPML02, used in this study was supplied by the Collection of Postharvest Pathology Group of IRTA (Lleida, Spain). Conidia from wounded fruits were sampled into a solution of sterile distilled water and Tween® 80 (0.0005%) surfactant. Quantification of conidia in suspension was as in Obi et al. (2017), and adjusted to $25 \times 10^3\ mL^{-1}$ spore for fruit inoculation. To evaluate tolerance to brown rot, 20 disinfected fruits were inoculated with 25 µL of spore load of the virulent pathogen. Five fruits used as control were inoculated with 25 µL of sterile water. Both treatment and control were then incubated for five days in darkness at 23 °C.

2.3. Brown rot disease evaluation

Pathogenic traits were evaluated according to Obi et al. (2017). In brief, inoculated fruits were observed daily during the five days of incubation. The %BRI was assessed using the percentage fraction infected over the total number of inoculated fruits. Percentage of colonization (%C) was assessed using the percentage colonised over the total number of fruits. LD and CEx were also measured. These parameters were used in the determination of brown rot disease severity for genotype tolerance rating, as has been reported previously (Martínez-García et al., 2013; Obi et al., 2017). LS was calculated by the %BRI × LD/100 and colonization severity (CS) by the %C × CEx/100.

2.4. Fruit quality trait evaluation

During the 2014 and 2015 seasons, twenty fruits were harvested to evaluate fruit quality individually for each tree seedling. Harvesting date (Julian days, JDs) ranged from late-May to mid-September, depending on the genotype of the population. Fruit weight (FtW) and physicochemical traits were determined for each genotype. Titratable acidity (TA) and pH were determined at harvest, as detailed in previous studies (Abidi et al., 2015; Zeballos et al., 2016).

Fruits were evaluated for firmness (FF) and soluble solids content (SSC) at three different levels: at harvest and after 5 days of storage (inoculated and uninoculated) at 23 °C. At harvest, firmness was determined for 5 fruits and genotypes on opposite sides of the equator of each fruit, after a section of the peel (approximately 2 cm^2) was removed using a penetrometer fitted with an 8-mm diameter probe (Effegi, Milan, Italy). Both measures were averaged for each fruit, and data are given in Newton (N). Firmness of uninoculated and inoculated fruits were determined on 5 and 20 fruits and genotypes, respectively, in the undamaged part of the fruit after 5 days of incubation. The SSC of the juice was also measured at harvest and after incubation using a temperature compensated refractometer (model ATC-1, Atago Co., Tokyo, Japan); and data are presented as °Brix.

2.5. Antioxidant compounds analysis

For biochemical analysis on fruit pulp and peel, out of the 20 fruits used for the study, 10 were randomly selected, peeled using a mechanical peeler, and later cut into smaller pieces for relative homogeneity. Then, 3 g of peel and 5 g of fresh fruit were weighed into 50 mL transparent polypropylene jars, frozen in liquid nitrogen, and conserved at −20 °C for later use in total phenolics (TPC), flavonoids, anthocyanins assays. For vitamin C (Vit C) determination, samples were stored with metaphosphoric acid (HPO_3) and subsequently conserved at −20 °C prior to analysis. Biochemical extractions were performed as described in Cantín et al. (2009).

Vit C, TPC, flavonoid, and anthocyanin contents were determined using colorimetric methods (Cantín et al., 2009) and measured using a spectrophotometer ([BIOCHROM ASYS UVM 340 microplate reader (see details in Ágreda, 2016)]. Standard calibration curves were prepared daily for all determinations. For Vit C, absorbance was measured at 525 nm, and the amount of Vit C was expressed as milligrams (mg) of ascorbic acid (AsA) per 100 g fresh weight (FW). For TPC, the colorimetric method based on the chemical reduction of the Folin-Ciocalteu reagent was used. Absorbance was measured at 725 nm, and the phenolic content was expressed in mg of gallic acid (3,4,5-trihydroxybenzoic acid) equivalents (GAE) per 100 g of FW. Total flavonoid content was determined by measuring the absorbance at 510 nm, and the results were expressed as milligrams of catechin equivalents (CE) per 100 g of FW. The total anthocyanin content was evaluated using a hydroalcoholic extract, and the absorbance was measured at 520 and 700 nm. Anthocyanin concentration was calculated using the molar extinction absorptivity coefficient $\varepsilon = 26{,}900/\text{cm}$ and was expressed in milligrams of cyanidin-3-glucoside equivalents (C3GE) per 100 g of FW (Liu et al., 2015; Saidani et al., 2017).

2.6. Statistical analysis

Means, standard errors (SE), and Pearson's correlation were calculated using SPSS 25 (IBM Inc, Armonk, NY, USA) statistical software. The incidence and severity of brown rot, including the influence of quality parameters, were also analysed using an analysis of variance (ANOVA) with SPSS 25 statistical software. Statistical significance was set at the $p < 0.05$ level, and the Duncan's test was used for the comparison of means.

3. Results

We studied a total of 68 descendants from the 'Babygold 9' × 'Crown Princess' population over a period of 3 years (2013, 2014, and 2015) for tolerance to *Monilinia laxa* brown rot (Supplementary Table 1). The disease parameters used included: %BRI, LD, LS, %C, CEx, and CS. As previously mentioned, we selected 17 genotypes that exhibited a *M. laxa* LS of < 40 mm, either in 2013 or 2014, or with the mean value for both years (Supplementary Table 2), to evaluate and validate the *M. laxa* tolerance of these genotypes in 2015.

For the 17 genotypes studied, the harvest date (HD) was recorded and the physicochemical traits [FtW, FF, SSC, pH, and TA] were evaluated over a period of 3 years [2013–2015 (Table 1)], and a parametric test of Pearson correlation was conducted within pairs of fruit quality traits (Table 2, Supplementary Table 3). We also determined phytochemical trait compounds as Vit C and total phenolic, flavonoid, and anthocyanin contents in flesh (2014–2015, Table 3) and in peel (2015 only, Table 4, Supplementary Table 4).

3.1. Effect of phytopathogen activities

The evaluation of the 68 genotypes of 'Babygold 9' × 'Crown Princess' for brown rot tolerance in 2013 and 2014 is presented in Supplementary Table 1. The %BRI in both years was between 50–100%.

Table 1
Effect of storage and inoculation on FF, SSC, and physicochemical traits in the 17 descendants of the 'B9' × 'CP' population. Data are mean ± SE of the 3 years (2013–2015). Resistant genotypes are shown in bold.

Genotype	HD (JDs)	FtW (g)	FF at harvest (N)[1]	FF uninoculated (N)[1]	FF inoculated (N)[1]	SSC at harvest[a] (°Brix)[1]	SSC after storage uninoculated (°Brix)[1]	SSC after storage inoculated (°Brix)[1]	pH[a]	TA (%)[a]	RI[a]
BC1	**175 ± 5**	**175 ± 21.1**	**26.50 ± 1.9 ab**	**33.40 ± 1.9 bcde**	**28.10 ± 0.8 b**	**8.2 ± 0.4**	**8.0 ± 0.5ab**	**8.0 ± 0.3 b**	**3.62 ± 0.0**	**0.6 ± 0.0**	**13.29 ± 0.8**
BC11	227 ± 6	209 ± 27.1	51.16 ± 3.8 d	48.13 ± 4.6 f	41.55 ± 3.0 d	9.8 ± 0.7	10.0 ± 0.4 cd	9.2 ± 0.2 c	3.96 ± 0.1	0.5 ± 0.1	17.70 ± 1.3
BC19	175 ± 5	186 ± 21.0	18.58 ± 2.3 a	24.05 ± 1.2 a	22.31 ± 0.8 ab	9.7 ± 0.1	8.0 ± 0.6 ab	7.5 ± 0.3 ab	3.82 ± 0.1	0.4 ± 0.0	20.05 ± 0.5
BC24	226 ± 7	208 ± 32.6	39.31 ± 3.3 c	34.18 ± 2.1 cde	34.35 ± 1.9 c	10.4 ± 0.4	9.2 ± 0.3 bc	9.2 ± 0.3 c	3.96 ± 0.1	0.6 ± 0.1	17.31 ± 1.6
BC44	175 ± 5	171 ± 21.5	20.35 ± 2.1 ab	19.07 ± 1.5 a	17.65 ± 0.6 a	8.2 ± 0.2	7.9 ± 0.3 ab	7.5 ± 0.2 ab	3.84 ± 0.3	0.5 ± 0.1	15.88 ± 3.1
BC48	**224 ± 4**	**208 ± 15.7**	**47.51 ± 2.9 cd**	**36.23 ± 1.4 e**	**35.11 ± 0.9 c**	**11.1 ± 0.7**	**11.1 ± 0.4 d**	**9.3 ± 0.2 c**	**3.88 ± 0.1**	**0.6 ± 0.1**	**16.56 ± 0.9**
BC51	175 ± 5	181 ± 21.0	24.17 ± 4.4 ab	25.15 ± 2.8 ab	25.50 ± 1.2 b	8.9 ± 0.9	8.0 ± 0.5 ab	7.9 ± 0.2 b	3.71 ± 0.1	0.5 ± 0.1	16.17 ± 2.9
BC53	178 ± 3	143 ± 16.2	19.22 ± 0.9 a	24.50 ± 0.8 a	23.73 ± 0.7 b	8.5 ± 0.3	7.1 ± 0.4 a	7.6 ± 0.3 ab	3.68 ± 0.0	0.5 ± 0.1	14.63 ± 1.2
BC57	227 ± 6	241 ± 18.0	48.19 ± 4.9 cd	46.76 ± 3.9 f	49.82 ± 3.1 e	9.3 ± 0.5	9.6 ± 0.5 c	7.9 ± 0.3 b	3.95 ± NA	0.5 ± NA	17.44 ± NA
BC58	**180 ± 6**	**187 ± 15.5**	**23.79 ± 2.1 ab**	**27.20 ± 1.1 abcd**	**24.27 ± 0.7 b**	**7.7 ± 0.9**	**7.6 ± 0.5 a**	**7.1 ± 0.3 a**	**3.62 ± 0.0**	**0.6 ± 0.1**	**12.98 ± 1.3**
BC59	176 ± 8	163 ± 24.3	29.08 ± 1.5 b	34.06 ± 1.7 cde	28.07 ± 0.8 b	9.7 ± 1.8	8.0 ± 0.3 ab	8.0 ± 0.3 b	3.76 ± 0.1	0.5 ± 0.0	17.16 ± 3.0
BC60	224 ± 7	187 ± 13.2	51.09 ± 3.1 d	52.29 ± 3.6 f	42.00 ± 1.6 d	11.0 ± 0.6	10.3 ± 0.3 cd	9.0 ± 0.2 c	3.92 ± 0.2	0.7 ± 0.2	14.40 ± 2.7
BC61	227 ± 6	220 ± 22.4	41.27 ± 2.6 c	36.26 ± 3.2 e	33.56 ± 1.5 c	9.2 ± 0.7	10.0 ± 0.3 cd	9.6 ± 0.2 cd	4.17 ± 0.0	0.4 ± 0.1	21.20 ± 2.1
BC63	**222 ± 2**	**235 ± 18.0**	**43.28 ± 1.9 cd**	**35.91 ± 1.6 de**	**36.37 ± 1.0 c**	**9.4 ± 1.0**	**9.5 ± 0.4 c**	**9.1 ± 0.2 c**	**3.89 ± 0.1**	**0.6 ± 0.1**	**14.28 ± 0.6**
BC66	216 ± NA	151 ± NA	29.29 ± 2.1 b	33.87 ± 1.6 bcde	27.22 ± 1.3 b	8.8 ± NA	9.5 ± 0.5 c	10.1 ± 0.3 d	3.87 ± NA	0.6 ± NA	12.68 ± NA
BC67	**180 ± 6**	**169 ± 10.6**	**21.70 ± 1.1 ab**	**25.60 ± 1.2 abc**	**24.73 ± 0.5 b**	**8.5 ± 1.2**	**7.6 ± 0.5 a**	**6.9 ± 0.2 a**	**3.67 ± 0.0**	**0.5 ± 0.1**	**17.01 ± 0.3**
BC68	**180 ± 6**	**186 ± 4.2**	**17.51 ± 1.0 a**	**25.32 ± 0.7 abc**	**24.05 ± 0.5 b**	**9.4 ± NA**	**7.2 ± 0.5 a**	**7.1 ± 0.2 a**	**3.76 ± 0.0**	**0.4 ± 0.1**	**18.01 ± NA**

a: No replication (data from pooled fruits of 5). Abbreviations: HD, harvest date; JDs, Julian days; FtW, fruit weight; FF, fruit firmness; SSC, soluble solids content; TA, titratable acidity; RI, ripening index (SSC/TA); SE, standard error; NA, not available, because replications were less than 3 or harvested once a year. [1] Different letters show differences among genotypes at $P \leq 0.05$ (Duncan test).

Table 2
Pearson correlations (parametric test) within pairs of fruit quality traits in the 'B9' × 'CP' population studied over a period of 3 years (2013–2015).

	FtW	FF at harvest	FF uninoculated	FF inoculated	SSC at harvest	SSC uninoculated	SSC inoculated	pH	TA	RI
HD (JDs)	0.554**	0.602**	0.385*	0.552**	0.677**	0.630**	0.687**	0.759**	0.092	0.497**
FtW		0.220*	0.200*	0.319**	0.334**	0.463**	0.445**	0.464**	0.167	0.421**
FF at harvest			0.833**	0.800**	0.418**	0.514**	0.363**	0.316**	0.261*	0.115
FF uninoculated				0.837**	0.367**	0.547**	0.386**	0.369**	0.245*	0.175
FF inoculated					0.391**	0.562**	0.407**	0.415**	0.260*	0.173
SSC at harvest						0.786**	0.829**	0.667**	0.174	0.586**
SSC uninoculated							0.810**	0.667**	0.133	0.518**
SSC inoculated								0.696**	0.199	0.514**

Abbreviations: HD, harvest date; JDs, Julian days; FtW, fruit weight; FF, fruit firmness; SSC, soluble solids content; TA, titratable acidity; RI, ripening index (SSC/TA). *, **: Correlations significant at $P \leq 0.05$ and $P \leq 0.01$, respectively; N = 138.

Differences exist between the years, although a similar average %BRI was found for 2013 (91.9%) and 2014 (91.6%). The average %C in 2013 was 84.8%, but was lower in 2014 (80.2%). The average LD in 2013 was 56.5 mm, while the average LD in 2014 was 48.9 mm. The mean LS was 52.5 mm in 2013, and 45.3 mm in 2014. A corresponding pattern was repeated in both years in the range of CS, with an average CS of 44.0 mm in 2013 and 36.6 mm in 2014. Almost all the associated pathological parameters indicate that the progeny showed fewer symptoms of *M. laxa* infection in 2014 than in 2013. However, only colonization extent and CS were positive correlated in 2013 *vs* 2014 (r = 0.388, and r = 0.338 at $P \leq 0.05$, respectively). Briefly, in 2015, the 17 genotypes, the average %BRI (92.9%) and %C (89.4%) was higher than in the previous years. In contrast, the %LD and LS were lower, with averages of 48.1 and 44.7 mm, respectively.

From the mean of these 17 genotypes evaluated in 2013, 2014, and 2015, only six genotypes (BC1, BC48, BC58, BC63, BC67, and BC68) showed a lesion severity of < 40 mm and a colonization severity below 32 mm (Supplementary Table 2). An analysis of the brown rot tolerance between the 3 years of the study shows that the 17 genotypes exhibited high variability in most of the pathogenic parameters studied (Supplementary Table 2). The lowest %BRI (73.3%) and %C (51.7%) occurred in the BC67 genotype, while the lowest LD (41.98 mm), LS (31.75 mm), CEx (39.05 mm), and CS (21.75 mm) were observed in the BC58 genotype. The highest values for %BRI (100%) occurred in four different genotypes (BCs: 11, 24, 61, and 66), while for LD (52.34 mm), LS (50.27 mm), and CS (42.51 mm) the highest values were recorded in the BC11 genotype. For %C (91.7%) and CEx (49.47 mm), the highest values were observed in the BC61 and BC60 genotypes, respectively.

The Pearson correlation between pairs of traits for pathological traits showed significant positive correlation coefficients ranging from 0.406 to 0.959 at $P \leq 0.01$. Among the strongest are %BRI with %C (r = 0.814, $P \leq 0.01$); LD with CEx (r = 0.859, $P \leq 0.01$), and LS with CS (r = 0.959, $P \leq 0.01$) (Fig. 1).

Within the phytopathogenic activities in fruits, we have found that genotypes with smaller fungus injury diameters correlated with smaller colonization diameters. In addition, these genotypes are also associated with a lower incidence of disease, that is, a lower percentage of damaged fruits (susceptibility).

3.2. Effect of storage, inoculation and physicochemical traits on fruits

Table 1 shows the effect of storage and inoculation on FF, SSC, and physicochemical traits in 17 selected genotypes evaluated over a period of 3 years (2013–2015). Genotypes from these progeny were harvested between 175 and 227 JDs, which is late June and mid-August, respectively. The six resistant genotypes (shown in bold in Table 1) matured between 175 and 224 JDs. The FtW ranged between 143 g and 241 g. Marked variability was encountered in the FF at harvest and after storage. In the 17 genotypes selected, the mean FF at harvest was 32.47 N. Specifically, the lowest FF at harvest (17.51 N) was recorded for BC68 genotype, while the highest FF (51.16 N) was recorded for the BC11 genotype. The mean FF at harvest (32.47 N) was lower than the mean FF at storage (33.06 N) for all 17 genotypes. The mean SSC at harvest was 9.3°Brix (from 7.7°Brix in BC58 to 11.1°Brix in BC48). Within the stored peach, the mean SSC of uninoculated fruit was 8.7°Brix, and 8.3°Brix for inoculated fruit. After storage, significant

Table 3
Antioxidant compound contents in the flesh of the 17 genotypes of the 'B9' × 'CP' population evaluated over a period of 2 years (2014–2015). Data are mean ± SE (N = 4–6 from 10 pooled fruits). Resistant genotypes are shown in bold.

Genotype	Ascorbic acid (mg AsA/100 g FW)	Total phenolics (mg GAE/100 g FW)	Flavonoids (mg CE/100 g FW)	Anthocyanins (mg C3GE/100 g FW)
BC1	**9.20 ± 3.3 d**	**51.08 ± 1.9 efg**	**17.99 ± 1.8 abc**	**0.13 ± 0.0 a**
BC11	4.41 ± 0.3 abc	63.73 ± 2.6 i	33.49 ± 7.6 d	0.16 ± 0.0 ab
BC19	7.89 ± 0.5 cd	49.32 ± 0.7 def	24.46 ± 1.1 abcd	0.14 ± 0.0 a
BC24	7.74 ± 1.3 cd	58.15 ± 3.0 ghi	35.10 ± 3.5 d	0.17 ± 0.0 ab
BC44	6.12 ± 0.7 abcd	34,81 ± 1.1 ab	12.08 ± 1.2 ab	0.09 ± 0.0 a
BC48	**5.22 ± 0.5 abc**	**48.84 ± 1.4 def**	**17.69 ± 1.5 abc**	**0.09 ± 0.0 a**
BC51	3.64 ± 0.9 ab	61.26 ± 1.6 hi	35.45 ± 6.9 d	0.15 ± 0.0 ab
BC53	6.47 ± 0.9 bcd	27.90 ± 0.9 a	10.02 ± 1.9 a	0.17 ± 0.0 ab
BC57	2.76 ± 0.3 a	37.54 ± 0.5 bc	10.96 ± 0.6 ab	0.16 ± 0.0 ab
BC58	**3.17 ± 0.2 ab**	**42.98 ± 0.5 cde**	**17.48 ± 2.9 abc**	**0.22 ± 0.0 ab**
BC59	5.69 ± 1.3 abc	50.54 ± 5.1 efg	25.86 ± 9.9 bcd	0.10 ± 0.0 a
BC60	5.26 ± 1.0 abc	29.23 ± 2.3 a	09.95 ± 3.0 a	0.30 ± 0.1 bc
BC61	6.36 ± 0.4 bcd	45.33 ± 3.9 cde	13.44 ± 2.1 ab	0.20 ± 0.0 ab
BC63	**2.55 ± 0.5 a**	**53.85 ± 4.9 fgh**	**29.04 ± 8.7 cd**	**0.16 ± 0.0 ab**
BC66	3.86 ± 0.6 ab	39.10 ± 2.9 bc	19.04 ± 1.4 abc	0.12 ± 0.0 a
BC67	**4.88 ± 1.9 abc**	**50.39 ± 1.4 efg**	**19.09 ± 2.2 abc**	**0.17 ± 0.0 ab**
BC68	**4.66 ± 0.3 abc**	**41.87 ± 2.3 bcd**	**09.58 ± 0.7 a**	**0.40 ± 0.1 c**

Abbreviations: AsA, ascorbic acid; GAE, gallic acid equivalents; CE, catechin equivalents; C3GE, cyaniding-3-glucoside equivalents. For each column, different letters show significant differences among genotypes ($P \leq 0.05$, Duncan's test).

Table 4
Antioxidant compound contents in the peel of the 17 genotypes of the 'B9' × 'CP' population evaluated in 2015. Data are mean ± SE (N = 3 from 10 pooled fruits per genotype). Resistant genotypes are shown in bold.

Genotype	Ascorbic acid (mg AsA/100 g FW)	Total phenolics (mg GAE/100 g FW)	Flavonoids (mg CE/100 g FW)	Anthocyanins (mg C3GE/100 g FW)
BC1	**15.48 ± 0.7 e**	**153.54 ± 1.1 hi**	**96.42 ± 2.7 fg**	**9.66 ± 0.1 i**
BC11	9.01 ± 0.7 abcd	158.92 ± 5.0 ij	106.18 ± 3.4 g	4.17 ± 0.0 e
BC19	9.24 ± 0.9 abcd	112.17 ± 1.2 bcd	75.70 ± 4.5 de	6.00 ± 0.2 f
BC24	8.45 ± 0.5 abcd	168.24 ± 2.8 j	128.13 ± 2.4 h	0.62 ± 0.0 a
BC44	10.58 ± 0.4 bcd	116.81 ± 1.1 cde	67.74 ± 0.4 cd	2.42 ± 0.0 c
BC48	**9.87 ± 0.6 bcd**	**141.01 ± 7.5 gh**	**74.25 ± 6.7 de**	**5.99 ± 0.1 f**
BC51	8.12 ± 0.4 abcd	150.15 ± 6.7 hi	142.04 ± 5.4 hi	2.81 ± 0.0 d
BC53	11.10 ± 0.9 cd	89.98 ± 3.2 a	50.00 ± 0.8 b	4.36 ± 0.1 e
BC57	7.37 ± 0.7 abc	123.00 ± 2.4 de	61.69 ± 7.6 bcd	1.69 ± 0.0 b
BC58	**10.89 ± 0.1 cd**	**128.13 ± 5.2** ef	**86.47 ± 6.6 ef**	**8.26 ± 0.2 h**
BC59	9.48 ± 0.1 abcd	144.07 ± 3.0 gh	132.70 ± 1.3 h	2.17 ± 0.0 c
BC60	11.31 ± 0.2 d	106.19 ± 4.5 ab	56.30 ± 7.5 bc	6.94 ± 0.2 g
BC61	8.45 ± 0.3 abcd	135.70 ± 3.1 fg	105.45 ± 5.5 g	2.94 ± 0.0 d
BC63	**5.89 ± 0.3 a**	**115.61 ± 3.8 bcde**	**88.72 ± 1.4 ef**	**4.28 ± 0.1 e**
BC66	8.42 ± 0.3 abcd	103.73 ± 2.7 b	74.16 ± 1.0 de	0.68 ± 0.0 a
BC67	**16.29 ± 1.1 e**	**189.43 ± 5.7 k**	**148.61 ± 3.4 i**	**12.94 ± 0.2 j**
BC68	**6.77 ± 0.1 ab**	**148.59 ± 2.5 hi**	**36.54 ± 6.9 a**	**2.18 ± 0.0 c**

Abbreviations: AsA, ascorbic acid; GAE, gallic acid equivalents; CE, catechin equivalents; C3GE, cyaniding-3-glucoside equivalents. For each column different letters show significant differences among genotypes ($P \leq 0.05$, Duncan's test).

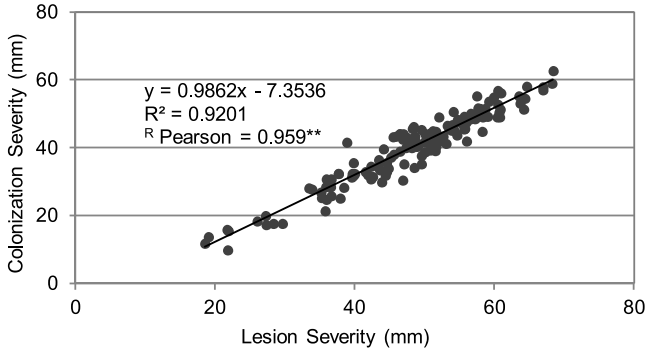

Fig. 1. Correlation between lesion and colonization severities in all the 'B9' × 'CP' genotypes evaluated over 3 years (2013–2015). N = 138.

differences were found in SSC among the 17 selected genotypes. There was also marked variability in pH (3.62–4.17), TA (0.40–0.60%), and ripening index (RI, 12.68–21.20). As shown in Table 2, there were significant positive correlations between most of the physicochemical traits. HD showed a significant positive correlation with FtW, FF, SSC, pH, and RI. The FtW showed a significant positive correlation with FF and SSC (at harvest, inoculated, and at storage), and pH and RI. The FF and SSC at harvest was highly correlated with both parameters at storage.

3.3. Effect of antioxidant compound contents

Table 3 shows the levels of all antioxidant compounds (ascorbic acid, TPC, flavonoids, and anthocyanins) in the flesh of the 17 genotypes evaluated in 2014 and 2015. In addition, we included as preliminary results the content of these compounds in the peel measured in 2015, to determine whether any compounds were associated with tolerance to *M. laxa* (Table 4). Significant differences were found between genotypes for all antioxidant contents in both flesh and peel tissues.

Among the 17 selected genotypes, the AsA content in flesh ranged from 2.55 to 9.20 mg AsA/100 g FW, TPC ranged from 27.90 to 63.73 mg GAE/100 g FW, and flavonoid contents ranged from 9.48 to 35.45 mg CE/100 g FW. The variation in anthocyanins, particularly in fruit flesh, was from 0.09 to 0.40 mg C3GE/100 g FW. A wide range of antioxidant contents were found in the peel of the 17 genotypes studied. In general, Vit C, total phenolics, and flavonoid contents were higher in the peel than in the flesh. The TPCs of the BC67 genotype, and AsA and anthocyanin contents of the BC1 and BC67 genotypes, were significantly higher than for the other genotypes. Flavonoid content was not significantly different in the resistant compared to non-resistant genotypes. As shown in Table 4, the AsA content in the peel of the 17 genotypes studied ranged from 5.89 to 16.29 mg AsA/100 g FW.

Notably. pathologic variables, %BRI, %C, LS, and CS correlated negatively with peel anthocyanin contents (r = -0.551, r = -0.552, r = -0.481, r = -0.491, $P \leq 0.05$ respectively (Fig. 2). However, only %BRI correlated negatively with fruit flesh anthocyanin contents (r = -0.219, $P \leq 0.05$).

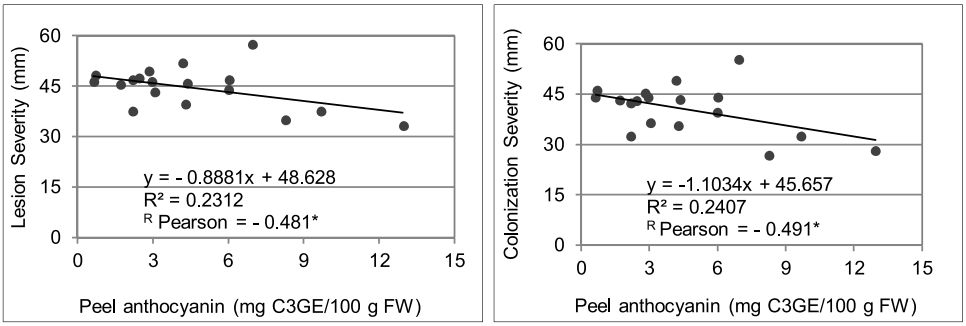

Fig. 2. Correlation between lesion severity and peel anthocyanin contents (left), and colonization severity and peel anthocyanin contents (right) in the 17 'B9' × 'CP' genotypes evaluated for 2015. N = 17.

4. Discussion

The annual disparity found in the responses of the genotypes to brown rot after inoculation may be due to different levels of cuticular cracking or fractures, as has been reported for stone fruits by other authors (Gradziel et al., 2003; Kappel and Sholberg, 2008). Cuticular cracks are considered to be the preferential portal of entry for fungi pathogens in the *Monilinia* genus (Gibert et al., 2007), and the incidence of fruit infection increases with increasing fruit cuticular crack surface area (Borve et al., 2000; Gibert et al., 2009). In the present study, fruits were not wounded prior to inoculation; therefore, the brown rot pathogen would require naturally occurring wounds or micro-cracks in the cuticle to gain entry into the fruit (Oliveira-Lino et al., 2016). The yearly variation is likely due to natural differences in surface cuticular cracks, since a uniform quantity of artificial inoculum density was used in this study; Ágreda (2016) reported similar results for a different peach population evaluated under the same conditions.

The significant positive correlation observed between pairs of pathological traits in our study is typical (Obi et al., 2017). This undoubtedly indicates that the level of infection significantly influenced the LD and CE, including the severity of the disease situation (Michailides et al., 2000). Therefore, LD and CEx are two brown rot parameters that are usually associated, and are useful in evaluating the brown rot tolerance of peach. Information for these two traits is important in the evaluation of disease tolerance from genetic or pathogenic points of view, respectively (Xu et al., 2008; Burnett et al., 2010).

Considering the physicochemical variables, the observations of HD in this study are in agreement with that of Giménez (2013), in which the studied population was harvested during 2009, 2010, and 2011, between 169 and 248 JDs. All the genotype-pathogen interactions indicated variable degrees of susceptibility, and occurred in genotypes harvested both in the early- or late-season. Nevertheless, the susceptibility of peach to brown rot depends on the interaction between the host (cultivar) and the pathogen (Obi et al., 2018a, 2018b), not on the season or ripening time. However, when fruits are harvested later in the season, they are sweeter and larger, and have higher total phenolics, flavonoids (Font i Forcada et al., 2013, Abdelghafat et al., 2018), and total sugar contents (Font i Forcada et al., 2013). Both very early-maturing and very late-maturing peach genotypes are of significant interest for the peach industry, particularly in the Mediterranean area.

Contrary to our expectation, mean FF at harvest (32.47 N) was lower than mean FF at storage (33.06 N) for all 17 genotypes studied. However, there were no significant differences, indicating that the incubation conditions did not particularly affect fruit firmness. A correlation analysis indicated a significant decrease ($P \leq 0.05$) in FF during storage for uninoculated (33.06 N) vs inoculated (30.49 N). This suggests that the decrease in FF in inoculated fruit could be due to the activity of *M. laxa*, and that this may have affected the surrounding tissues. Our results may explain the observation of Yaghmour et al. (2011), who that found that rates of infection increased as the FF decreased. Our analysis revealed a broad range of FF, from 17.51 to 47.51 N, within the genotypes with a LS below 40 mm, indicating that brown rot may be dependent on fruit firmness.

The present study also revealed that the SSC decreased from the levels observed at harvest during storage, for both inoculated and uninoculated fruits. In the 17 peach genotypes studied, the mean SSC at harvest was 9.3°Brix, which ranged from 7.7°Brix in the BC58 genotype to 11.1°Brix in the BC48 genotype. In stored peach, the mean SSC in uninoculated fruits was 8.7°Brix, and 8.3°Brix in inoculated fruit. After storage, significant differences were found in SSC among the 17 selected genotypes. This trend of the decrease in SSC during storage (for uninoculated fruit) is, however, contrary to our hypothesis and contradict the results of previous studies (Amodio et al., 2007 and Liu et al., 2012), although in distinct crop populations. Conversely, the decrease in SSC observed in peach during storage (for inoculated fruit) could be attributed to the pathogenic activities of the fungus on the inoculated host; inferring that as the pathogen preys on the host, the interaction leads to the depletion of SSC as sugars can be used for mycelia biosynthesis, growth, and development.

The SSC of inoculated peaches showed a negative significant correlation with CEx, LD, and LS ($r = -0.273$, $P \leq 0.01$; $r = -0.236$, $P \leq 0.01$; and $r = -0.178$, $P \leq 0.05$; respectively). These findings are in agreement with those of Biggs and Miller, (2004), that showed negative correlations between disease severity and sugar content; they are also in agreement with Gradziel, (1994), who found that lesion development progressed as SSC content increased, becoming highest at the fully ripe stage, depending on the peach cultivar.

The relationship between disease parameters and FF within the 17 genotypes is also of interest. The BC58 genotype recorded one of the lowest FF at harvest (23.79 N), which was associated with the lowest disease parameters for LD (41.98 mm), LS (31.75 mm), CEx (39.05 mm), and CS (21.75 mm), while the BC11 genotype recorded the highest FF at harvest (51.16 N), and the highest disease parameters for LS (50.27 mm) and CS (42.51 mm). However, the BC44 genotype, which demonstrated an FF of 20.35 N, did not correspond to either a resistant or susceptible genotype (LS = 43.64 mm and CS = 34.96 mm). Hence the state of FF, especially at harvest, does not seem to significantly influence brown rot development. Consequently, the level of susceptibility to brown rot depends largely on peach genotype (Gradziel, 1994).

In the same manner the BC58 genotype, which was recorded as having the lowest SSC at harvest (7.7°Brix), was associated with the lowest disease parameters, as was the BC67 genotype (6.9°Brix). However, the BC48 genotype, which was recorded as having the highest SSC (11.1°Brix), also exhibited only low levels of damage from the pathogen. Genotypes that had intermediate SSC contents at harvest, such as BC44 (9.8°Brix), showed the highest brown rot severities.

The significant positive correlation of HD with FtW, FF, SSC, and pH observed in this study is in agreement with what has been reported by other authors (Giménez, 2013; Font et al., 2013; Ágreda, 2016). The correlation observed between FF and SSC at harvest with same parameters after storage ($r = 0.418$, $P \leq 0.01$) is similar to that reported by Giménez (2013) ($r = 0.226$, $P \leq 0.01$), who studied 100 progenies of the same population. This positive correlation between FF and SSC in resistant genotypes is important, because the genotypes with high SSC are selected aiming firstly for higher firmness, and secondly for lower pathogen susceptibility, to prevent mechanical damage during handling and transport (Crisosto et al., 2001).

The variation of pH from pH 3.62 to pH 3.89 in our six resistant genotypes are typical values for fruit acidity, since a pH lower than 4.0 at maturity is considered acidic (Abidi et al., 2015). The negative and significant correlations found between pH vs TA ($r = -0.327$, $P \leq 0.01$) and TA vs ripening index ($r = -0.665$, $P \leq 0.01$), are similar to that reported by other authors (Giménez, 2013; Abidi et al., 2015). In previous experiments, we have observed that the pH of the fruit increased as fruit maturity increased, while the TA decreased (Obi et al., 2018a). These parameters can be important, since it has been reported that acidity preserve fruits from pathogen damage (Hajilou and Fakhimrezaei, 2011; Cropotova et al., 2013; Tarabih and El-Metwally, 2014).

Regarding the bioactive compounds, the AsA content in the flesh ranged from 2.55 to 9.20 mg AsA/100 g FW, as reported by Giménez, (2013) for the same population. However, the TPC (27.90 to 63.73 mg GAE/100 g FW) among the selected 17 genotypes was in excess of the range found by Giménez, (2013) (11.22 to 37.42 mg GAE/100 g FW) for the same progeny studied over a period of 3 years (2009–2011). The differences found here may be due to the screening of genotypes for an LS that is lower than 40 mm. Flavonoid contents varied from 9.58 to 35.45 mg CE/100 g FW, and were also higher than those obtained in previous studies of different peach progenies by Giménez, (2013) (1.6 to 13.7 mg CE/100 g FW); Abidi et al., 2015 (2.3 to 18.0 mg CE/100 g FW) and Ágreda (2016) (3.8 to 27.6 mg CE/100 g FW). The average

total phenolic and flavonoid accumulation (46.23 mg CE/100 g FW and 20.04 mg CE/100 g FW, respectively) were higher than the range reported by Abdelghafar et al. (2018) in early-, mid-, and late-season peach germplasm evaluated in 2013, but below the values found in 2014, for TPC in peach harvested in any season (over 51.4 mg CE/100 g FW), and for flavonoids in late-season peach genotypes (28.7 mg CE/100 g FW). Abdelghafar et al. (2018) also found that the annual variation in the antioxidant composition was independent of season and peach germplasm. Environmental variables such as temperature, solar radiation, photoperiod, precipitation, and soil profile affect the growing environment and result in wide variations in peach fruit harvest quality (Lopresti et al., 2014). The effects of environment and orchard practices on peach fruit quality attributes are extensively reviewed by Minas et al. (2018). The anthocyanins, particularly in fruit flesh, varied from 0.09 to 0.40 mg C3GE/100 g FW. These values were below those reported by other authors [(0.7 to 12 mg C3GE/100 g FW) from a broad germplasm collection (Font i Forcada et al., 2013); (0.23–11.83 mg C3GE/100 g FW), for the same progeny (Giménez, 2013)]. These differences may be due to the flesh, with the 17 'B9' × 'CP' genotypes selected having yellow flesh, and/or due to the different methods used for quantification.

A wide range of antioxidant contents was found in the peel of the 17 studied genotypes. In general, Vit C, total phenolics, and flavonoid contents were higher in peel than in the flesh in, which is in agreement with previous reports (Ágreda, 2016; Saidani et al., 2017). We found that around 65% of Vit C, 75% of TPC, 81% of flavonoids and 96% of anthocyanin contents are concentrated in the peel of our progeny. The TPC in the BC67 genotype, and AsA and anthocyanins in the BC1 and BC67 genotypes, were significantly higher than that of other genotypes. However, flavonoid contents were not significantly different for the resistant compared to the non-resistant genotypes. As shown in Table 4, the AsA content in the peel of the 17 genotypes ranged from 5.89 to 16.29 mg AsA/100 g FW, similar to what other investigators have recently reported (Ágreda, 2016; Saidani et al., 2017). The content of anthocyanins varied from 0.62 to 12.94 mg C3GE/100 g FW in the peel tissue of the 17 selected genotypes, and this reveals that most resistant genotypes had 27–81 times higher contents of anthocyanins in their peel than in their flesh. These values agree with previous reports (Prior et al., 1998; Gil et al., 2002; Saidani et al., 2017), supporting the inference that anthocyanins are more concentrated in the fruit peel than in the flesh.

An unequal distribution of Vit C and TPC in the flesh (≈25–30%) and peel (≈65–70%) of peach has also been documented (Ágreda, 2016; Saidani et al., 2017). It is of great significance, therefore, that the high levels of these bioactive compounds in the peel provide protection from abiotic stresses (Cantín et al., 2009), which often predispose peach fruits to pathogen invasion. Fruit peel has frequently been suggested to be important in broad range resistance against opportunistic pathogens such as *Monilinia* spp. (Pacheco et al., 2014).

Pathologic variables (%BRI and %C) and severities (LS and CS) correlated negatively with peel anthocyanin contents (Fig. 2). However, only %BRI was negatively correlated with flesh anthocyanin contents ($r = -0.219$, $P \leq 0.05$). Anthocyanins are the most common pigment in nature (Khoddami et al., 2013), a class of phytochemicals that give plants their colour and protect tissues from oxidative abiotic stress, which invariably extends the life span of the plant organ. They are therefore more concentrated in the skin portion of fruit, particularly as maturity approaches (Prior et al., 1998), to provide a protective barrier against potential phytopathogenic invaders. This could be advantageous in providing tolerance to our genotypes.

Nevertheless, only TPC from flesh showed a significant negative correlation in this progeny with LD, LS, and CEx ($r = -0.282$, $r = -0.279$, and $r = -0.225$, all at $P \leq 0.05$, respectively), as has been shown by Ágreda (2016). Other authors also have reported significant negative correlations between phenolic acids and %BRI in immature peach and nectarine cultivars (Villarino et al., 2011). High contents of antioxidants influence brown rot negatively by reducing pathogenic activities (see Supplementary Table 3); however, in the present study, the genotypes with LS < 40 mm were not those with the highest TPC, and vice-versa. Major phenolic acids such as chlorogenic and neochlorogenic acids (Villarino et al., 2011), which have highly potent antioxidant properties (Dai and Mumper, 2010; Khoddami et al., 2013), may protect the plant and plant materials against fungi and other phytopathogenic organisms (Prasad et al., 2014; Spadoni et al., 2014). However, fruit phenolic contents decrease at harvest, and their effectiveness in controlling brown rot infection can vary with peach cultivar (Cindi et al., 2016; Obi et al., 2018b).

Pearson's correlation coefficients for bioactive compounds were between 0.790 and 0.506. TPC in the flesh showed significant positive correlations with flesh and peel flavonoids ($r = 0.790$, $r = 0.718$, respectively), all at $P \leq 0.01$. Moreover, TPC and flavonoid levels in the peel were also strongly correlated ($r = 0.722$, $P \leq 0.01$). The results found for this progeny were in agreement with previous studies in this and other progenies or peach germplasm (Giménez, 2013; Font et al., 2014; Abidi et al., 2015). The strong association found in this study between the biochemical compounds implies that they are important antioxidant phytochemicals that act in coordination to induce tolerance to brown rot in peach. However, further studies are required to determine this.

Infection or incidence, sporulation, and dissemination are the three major stages of the fungal pathogen life cycle in a disease situation (Agrios, 2005). From a genetic point of view, lesion severity is a good parameter to consider during selection for breeding; although there is damage from the fungi, the dispersion of pathogens is limited by the lack of sporulation. However, from a pathogenic point of view, colonization severity is a better factor for consideration because there is the possibility of sporulation due to colonization, which leads to spore dispersal within the environment that can cause further damage.

5. Conclusions

The selection of genotypes for peach breeding that are rich in bioactive compounds, and which are possess brown rot tolerance, may avoid negative outcomes in the industry, and provide safe alternative to the use of pesticides. Based on our 3-year screening protocol, we found phenotypic differences in the susceptibility to brown rot caused by *Monilinia laxa* in the 'Babygold 9' × 'Crown Princess' population. It was also found that FF decreased due to 5 days of storage and to the activity of *M. laxa* in the surrounding tissues. It was possible to identify and select six genotypes (BC1, BC48, BC58, BC63, BC67, and BC68) for low brown rot susceptibility and high fruit quality from the germplasm of the Estación Experimental de Aula Dei-CSIC. Although genotypes that possess bioactive compounds such as AsA, phenolics, flavonoids, and anthocyanins were associated with potential brown rot tolerance, not all genotypes with a lesion of less than 40 mm contained the highest levels of bioactive compounds. The BC1 and BC67 genotypes had significantly higher levels of AsA, phenolics, and anthocyanins. However, flavonoid levels were not significantly different in the resistant compared to the non-resistant genotypes. The negative correlations observed between anthocyanin and brown rot severity highlight their potential influence on susceptibility to *M. laxa*. This interaction is of paramount importance, and consideration should be taken in breeding programs to select cultivars with high levels of bioactive compounds, health-enhancing properties, and good postharvest performance.

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Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:https://doi.org/10.1016/j.scienta.2018.10.027.

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