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Physicochemical characteristics and phytochemical contents of peach trees (*Prunus persica* (L.) Batsch) grafted on different rootstocks during cold storage

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Abstract

Peach is highly perishable and need cold storage to have their preservation period extended. Rootstocks may affect both phenology and the development of plant, interfering with the potential of fruit preservation. Therefore, this study aimed at evaluating physicochemical characteristics and phytochemical contents of grafted Maciel var. peaches, on eight rootstocks in an orchard and in cold storage and commercialization simulation conditions. The measurements were carried out after 7, 14 and 21 days of cold storage at 1±0.5°C, followed by a 3-day commercialization simulation period at 22±1°C. The experimental design was a completely randomized design in factorial scheme 8x3 (8 rootstocks and 3 storage periods) with 4 repetitions of 20 fruits. Peaches were analyzed for pulp browning, total phenol content and antioxidant activity, mass loss, rot, firmness, ripening index, color, total soluble solids and titratable acidity. Lower total phenol content and lighter browning were observed in fruits harvested from trees grafted on both 'Seleção Viamão' and 'Umezeiro' rootstocks. Regardless of their rootstocks, peaches fruits retain their quality up to 14+3 days. After that, there was high mass loss, occurrence of rot, browning and softening. However, rootstocks interfered in the variables of peach quality throughout the cold storage period followed by commercialization simulation.

Keywords: enzymatic browning; fruit quality; postharvest.

Introduction

Due to high perishability of peach fruits, either they are commercialized or consumed right after harvest or storage, the method is being used for extending retail sale is very important (Barbosa et al., 2010; Pegoraro et al., 2015). Among all of commercial methods for fruit preservation, refrigeration has been the most common one (Brackmann et al., 2009; Pegoraro et al., 2010; Pinto et al., 2012).

In general, from the 15th to the 20th storage day, when peaches are removed from the cold rooms and kept at room temperature, there are significant losses due to rotting, woolliness, pulp browning, mass loss and softening (Santana et al., 2011; Pinto et al., 2012). Although, there are several studies of this nature, the exact causes of these problems have not been found yet (Pegoraro et al., 2015). Measures that prevent the metabolism of peaches and associated microorganisms causing deterioration need to be investigated to increase preservation periods (Martins et al., 2004).

The factors that usually affect the metabolism of fruits are mostly due to the inherent characteristics of the cultivar itself, orchard management (Martins et al., 2004), harvest time (Seibert et al., 2010), ripening stage (Andrade et al., 2015) and preservation methods (Santana et al., 2011).

Pulp browning is one of the main factors that harm peach preservation. This disorder cannot be observed on the

external surface of the fruit, but it gets clear when the fruit is cut for consumption. According to Cantillano et al. (2008), the more susceptible the fruits are to internal pulp browning, the shorter the post-harvest period. Pulp browning is normal in peaches after storage. Some bioactive compounds such as phenolic compounds may be related to this disorder (Toralles et al., 2004; Lurie and Crisosto 2005).

Besides these quantitative losses, peaches are rich in phytochemical compounds, such as phenolic compounds and carotenoids which are found in cultivars with yellow pulp (Toralles et al., 2004). Therefore, not only classic technical losses, but also variation in contents of compounds whose metabolism is specialized should be evaluated. This is true in the case of peaches with yellow pulp, which have high antioxidant content (Santos et al., 2013). All compounds with antioxidant potential help to mitigate the incidence of degenerative diseases such as chronic ones (Lako et al., 2007).

One of the factors that can affect fruit composition, and consequently preservation potential, is the rootstock (Orazem et al., 2011; Forcada et al., 2013; Barreto et al., 2017). Picolotto et al. (2009), Forcada et al. (2013) and Barreto et al. (2017) observed that the quality of peaches throughout harvest depends on the rootstock used in the orchard. Rootstocks may affect the quality of fruits since

they may interfere in water and nutrient absorption; thus affecting the vigor of the plants and the preservation of fruits (Martínez-Ballesta et al., 2010).

Studies on orange (Hifny et al., 2012) and apple (Tomala et al., 2008) storage have already shown that rootstocks can modify physicochemical characteristics of the fruits. However, regarding peaches, this fact has not been adequately investigated. Therefore, this study aimed at evaluating physicochemical characteristics throughout cold storage and phytochemicals of fruits from Maciel. peach trees grafted on different rootstocks.

Results and Discussion

Enzymatic browning and phytochemicals in pulp

Enzymatic browning of fruits was observed 21 days after storage in cold chamber and a 3-day commercialization simulation. It was more intense in fruits obtained from peach trees grafted on Capdeboscq and 'Tsukuba' rootstocks (Fig 1).

However, it was not observed in peach fruits from trees grafted on both 'Umezeiro' and 'Seleção Viamão'. The fact that fruits from these rootstocks do not show the browning process is very important to the storage of Maciel peaches, since it is internal to the fruit and can only be noticed when the fruit is cut for consumption. This experiment does not provide the exact reason of the fact that peaches from trees grafted on Umezeiro and Seleção Viamão do not show the browning symptoms. However, it was noticed that there is low content of total phenolic compounds in the fruits harvested from the trees grafted on Umezeiro and Seleção Viamão (Fig 1 and Fig 2). Pulp browning may be the consequence of an enzymatic oxidative process which involves phenolic compounds (substrate) acted upon by oxidative enzymes (Girner et al., 2002). Lack of suppression and/or reduction in the availability of any of these components in the enzyme reaction may inhibit pulp browning.

Antioxidant activity in the pulp was higher in the case of fruits from peach trees grafted on 'Okinawa' and lower in Seleção Viamão peaches (Fig 3). The effect of the rootstock on the content of antioxidant activity was also observed by Scalzo et al. (2005), who stated that high antioxidant activity was found in peaches from rootstocks which grew vigorous plants with good root and crown development.

Mass loss and rot percentage of fruits

Mass loss in peach fruits was increased throughout the storage period. Even at cold storage, regardless of the rootstock, fruits could be stored up to 14+3 days. After this period, there is mass loss above 5%, along with pulp softening and turgidity loss in tissues (Table 1). However, the mass loss of peach fruits up to 5% is normal. When the loss exceeds 10%, fruits show symptoms related to wrinkling (Crisosto et al., 2004). At the end of the experiment, mass loss exceeded 10%, regardless of the rootstock. These results agree with the earlier report of Andrade et al. (2015), who observed 33% mass loss in Maciel fruits 30 days after cold storage, followed by a 2-day commercialization simulation.

Rotting in peaches progressed steadily throughout the storage period, regardless of the rootstock and reached 6.78% and 14.25% of rotted fruits in evaluations carried out after 14+3 days and 21+3 days, respectively (Table 1). This variable was not affected by the rootstocks either. Since rotting depends on the local biome, the fruit's susceptibility to the disease and the causal agent is expected. Prior to this, most studies of this nature had already found this kind of behavior (Seibert et al., 2010).

Ripening index and epidermis color of fruits

The ripening index obtained by portable spectrophotometer (DA meter[®]) indicates that the higher the numerical value presented, the higher the chlorophyll content and green the fruits. The ripening index of peaches decreased in all rootstocks during the storage periods, a fact that swings with the evolution of ripening, implying a decrease in the chlorophyll content of the fruit (Chitarra and Chitarra, 2005; Andrade et al., 2015). Fruits from peach trees grafted on Aldrighi had higher ripening index values than peaches harvested from the other rootstocks after 7+3, 14+3 and 21+3 days (Table 2), where the higher values indicate a greater variation in the chlorophyll content in the fruits. Fruits from peach trees grafted on Flordaguard, Tsukuba and Seleção Viamão had their lowest ripening index values after 21+3 days. These ripening index values showed that the fruits have less chlorophyll, i. e., more ripen fruits, where ripening stage is more advanced due to pectin solubilization and reduction in cohesion among cells.

Fruits from peach trees grafted on Tsukuba showed changes in the color of the epidermis after 14+3 (78.66 °Hue) and 21+3 (77.50 °Hue) days of storage, from greenish yellow to orange-ish yellow (Table 2). This parameter may be used to check the ripening stage of the fruits since ripening leads to decrease in the °Hue (Infante et al., 2011).

Pulp firmness, soluble solids and total titratable acidity of fruits

Fruits from peach trees grafted on Aldrighi had firmer pulp than fruits from the other rootstocks (Table 3). The fact that pulp firmness is affected by the rootstock is an important characteristic to choose the plant matter in the orchard (Tomala et al., 2008). In this case, the Aldrighi rootstock grafted on the cultivar Maciel produced fruits with greater pulp firmness compared to the other rootstocks tested in this study. On the other hand, fruits from peach trees grafted on 'Seleção Viamão' were the ones that had the lowest firmness, as a consequence of the solubilization of cell wall components (Billy et al., 2008; Zhang et al., 2010; Pegoraro et al., 2015). With the evolution of fruit maturation, the release of Calcium and solubilization of the pectic polymer occurs through the action of two pectinamylesterase enzymes and the polygalacturonases (Chitarra and Chitarra, 2005; Sainz et al., 2015). The pectinametersrerase responsible for the disruption of the methyl esters and polygalacturonase is active in the degradation of methylated pectins which are methylated and transforms polymers of galacturonic acid into watersoluble pectic acids (Sainz et al., 2015; Qian et al., 2016). Peaches from trees grafted on 'Tsukuba' had the highest

amount of total soluble solids with 14.30 and 15.16 °Brix,

Table 1. Mass loss and rot percentage of fruits from Maciel peach trees grafted on different rootstocks in cold storage periods, followed by commercialization simulation.

Rootstocks	7+3 days		14+3 days		21+3 days	
	Mass loss (%)					
Aldrighi	4.53	aB	5.40	bB	14.17	abA
Capdeboscq	4.31	aB	7.60	abB	14.86	aA
Flordaguard	5.31	aB	7.39	abB	12.19	bcA
Nemaguard	5.00	aB	5.49	bB	13.98	abA
Okinawa	5.03	aB	5.29	bB	11.17	cA
Tsukuba	5.61	aB	9.13	aAB	15.11	aA
Umezeiro	5.48	aB	7.20	abB	14.10	abA
Seleção Viamão	6.70	aB	6.75	abB	14.24	abA
	Rotting (%)					
Aldrighi	3.40	abC	6.83	bB	12.60	cA
Capdeboscq	3.70	abC	7.43	abB	13.80	bcA
Flordaguard	2.66	bC	7.30	abB	14.53	abA
Nemaguard	3.25	abC	6.92	bB	13.10	cA
Okinawa	4.70	aC	7.89	aB	15.13	aA
Tsukuba	3.73	abC	7.80	aB	15.16	aA
Umezeiro	3.00	bC	7.80	aB	14.83	abA
Seleção Viamão	2.80	bC	7.92	aB	14.90	abA

Means followed by same letters, lowercase in the columns and uppercase in the lines, do not differ by Tukey's test, at 5% probability ($p \le 0.05$).



Fig 1. Enzymatic browning of the pulp of fruits from Maciel peach trees grafted on different rootstocks, 21-day after cold storage period, followed by a 3-day commercialization simulation.

Table 2. Ripening index and epidermis color of fruits from Maciel peach trees grafted on different rootstocks, in cold storage periods, followed by commercialization simulation.

Rootstocks	7+3 days		14+3 day	14+3 days		21+3 days	
		Ripening index					
Aldrighi	0.48	aA	0.43	aB	0.24	aC	
Capdeboscq	0.40	abA	0.39	abA	0.20	bcB	
Flordaguard	0.39	abA	0.36	abA	0.10	сВ	
Nemaguard	0.38	abA	0.32	abA	0.14	bcB	
Okinawa	0.36	abA	0.32	abA	0.17	bcB	
Tsukuba	0.37	abA	0.27	bB	0.10	cC	
Umezeiro	0.37	abA	0.31	abB	0.18	bcC	
Seleção Viamão	0.34	bA	0.24	bB	0.10	cC	
		Epide	rmis color (°۲	lue)			
Aldrighi	83.96	abA	83.08	abAB	82.81	aB	
Capdeboscq	84.94	aA	82.39	abB	82.49	aB	
Flordaguard	82.19	abAB	84.79	abA	78.85	abB	
Nemaguard	80.78	cB	86.19	aA	79.41	abB	
Okinawa	82.32	bB	81.93	abB	83.86	aA	
Tsukuba	84.91	aA	78.66	сВ	77.50	сВ	
Umezeiro	82.52	abA	80.67	abAB	78.61	abB	
Seleção Viamão	80.38	сВ	83.27	abA	82.60	aAB	



Fig 2. Total phenols of the pulp of fruits from Maciel peach trees grafted on different rootstocks, 21-day after cold storage period, followed by a 3-day commercialization simulation.

Table 3. Pulp firmness, soluble solids and total titratable acidity of fruits from Maciel peach trees grafted on different roots	tocks, in
cold storage and commercialization simulation.	

Rootstocks	7+3 days		14+3 days	s	21+3 day	S
	Pulp firmness (N)					
Aldrighi	19.39	aA	17.60	aA	15.90	aA
Capdeboscq	14.06	abA	13.56	bcA	13.11	bcA
Flordaguard	14.92	abA	12.66	cB	14.66	abcA
Nemaguard	17.81	abA	16.00	abB	15.03	abcC
Okinawa	14.31	abA	13.69	bcA	14.43	abcA
Tsukuba	14.60	abA	14.60	bcA	13.51	bcA
Umezeiro	14.48	abA	14.42	bcA	14.35	abcA
Seleção Viamão	13.53	bB	15.20	bcA	9.84	cC
	Soluble solids (°Brix)					
Aldrighi	11.90	bB	12.10	dAB	12.60	cA
Capdeboscq	12.10	abB	13.43	abAB	13.80	abcA
Flordaguard	12.65	abB	12.60	cdB	14.53	abA
Nemaguard	12.25	abB	12.26	dB	13.50	bcA
Okinawa	13.12	abB	14.13	abAB	14.55	abA
Tsukuba	13.60	aB	14.30	aAB	15.16	aA
Umezeiro	13.07	abA	13.10	bcA	13.33	bcA
Seleção Viamão	11.70	bB	12.96	bcAB	13.50	bcA
	Titratable acidity (% citric acid)					
Aldrighi	0.60	aA	0.49	cB	0.47	сВ
Capdeboscq	0.55	abA	0.52	cB	0.51	abB
Flordaguard	0.61	aA	0.55	abB	0.49	cC
Nemaguard	0.58	abA	0.58	aA	0.49	сВ
Okinawa	0.56	abA	0.56	abA	0.55	aA
Tsukuba	0.59	aA	0.58	aA	0.49	сВ
Umezeiro	0.54	bA	0.52	сВ	0.51	abB
Seleção Viamão	0.58	abA	0.56	abB	0.56	aB

Means followed by same letters, lowercase in the columns and uppercase in the lines, do not differ by Tukey's test, at 5% probability (p < 0.05).



Fig 3. Antioxidant activity of the pulp of fruits from Maciel peach trees grafted on different rootstocks, after a 21-day cold storage period, followed by a 3-day commercialization simulation.

Table 4. Characteristics of orchard soil at the Palma Agricultural Center belonging to the Universidade Federal de Pelotas in the city of Capão do Leão, in the state of Rio Grande do Sul, Brazil.

Atribute	
рН	5.80%
Organic matter (%)	1.93%
Aluminum saturation	1.50%
Base saturation	71%
CTC at pH 7	6.5
Р	11.5 mg/dm³
К	126 mg/dm ³
Са	4.4 cmol _c /dm ³
Mg	1.6 cmol _c /dm³

after 14+3 and 21+3 days of cold storage, respectively (Table 3). This confirmed the most advanced stage of fruit ripening, since ripening leads to an increase of sugar content in the fruits (Payasi et al., 2009; Jie et al., 2013). The soluble solids may increase with maturation and respiratory rate of fruit due to degradation reserve substances (Payasi et al., 2009). Low acidity was found in peaches from trees grafted on Umezeiro after 7+3 days, on Aldrighi and Capdeboscq after 14+3 days and on Aldrighi, Flordaguard, Nemaguard and Tsukuba after 21+3 days (Table 3). Chitarra and Chitarra (2005) stated that the contents of organic acids decrease as ripening takes place due to the process of respiration or the sugar conversion. Comitto et al. (2013) evaluated Aldrighi, Capdeboscq, Flordaguard, Nemaguard, Okinawa and Umezeiro rootstocks grafted on the cultivar Maciel in two harvests and reported no differences in the acidity of the fruits of the harvest. However, Barreto et al. (2016) observed that different peach rootstocks altered the fruit titratable acidity after refrigerated storage. These authors found that the fruit pesssegueiro Maciel grafted on Capdeboscq rootstock showed lower acidity 21 days after cold storage.

Materials and methods

Plant materials

The peach orchard used consisted of the *P. persica* (L.) Batsch Maciel cultivar grafted on Aldrighi, Capdeboscq,

Flordaguard, Nemaguard, Okinawa, Umezeiro, Tsukuba, and Seleção Viamão rootstocks. The orchard was planted in 2006 with a "V" type plant-organization system with 5 m of space between rows and 1.5 m space between plants, creating a density of 1.333 plants ha⁻¹.

The main characteristics of the Maciel cultivar are the following: medium plant vigor, mean productivity with 50 kg.plant⁻¹, fruits with yellow pulp, mainly aimed at industrialization, even though they are also widely accepted for consumption worldwide (Raseira et al., 2014).

The Okinawa, Nemaguard and Tsukuba rootstocks have the advantage of inducing a vigorous canopy, the Flordaguard rootstock presents a low cold requirement, the Okinawa rootstock presents rapid growth initial and anticipates the entry into production (Mayer et al., 2014) and Umezeiro rootstock induces low plant vigor and may be an alternative for the management of orchard densification (Galarça et al., 2013).

Site, soil and climate conditions

The experiment was conducted in a teaching orchard at the Palma Agricultural Center (Centro Agropecuário da Palma; CAP) belonging to the Federal University of Pelotas (Universidade Federal de Pelotas) in the municipality of Capão do Leão, in the state of Rio Grande do Sul (RS), latitude 31º52'00" S and longitude 52º21'24" W, Brazil.

The soil of the orchard is moderately deep with medium texture at A horizon and clay texture at B horizon; thus, it is

classified as red-yellow clay soil. The soil characteristics can be observed in Table 4.

According to the classification of Köppen, the climate of the region is type Cfa, namely humid and temperate with hot summers (Kottek et al., 2006). The region has an average annual rainfall and temperature of 1500 mm and 17.9 °C, respectively. The experiment was conducted during the year 2014.

Treatments and experimental design

Fruits were harvested randomly from the plants in December 2014. Each rootstock had three blocks of five field plants, and the fruits were harvested only from the three central plants of each block. Afterwards, they were placed in plastic boxes which had been previously washed and disinfected. They were then taken to the laboratory. Fruits with rot and mechanical damage were discarded and the others were standardized regarding their ripening stages.

In order to observe the quality of the peaches, four repetitions per treatment were used. Each repetition comprised of 20 fruits. Evaluations were carried out after 7, 14 and 21 days in a cold chamber at 0 ± 1 °C, followed by a 3-day commercialization simulation at room temperature (22±1°C).

The experimental design was a completely randomized design in factorial scheme 8x3 (8 rootstocks and 3 storage periods) with 4 repetitions of 20 fruits.

Traits measured

Peach fruits were evaluated in relation to the occurrence of internal browning, the appearance of the fruit pulp and the numbers of fruits, with and without the browning symptom and were visually evaluated by repetition. Results were expressed as percentage and total phenolic compounds (mg Gallic acid per 100 g of sample) were determined according to the adapted method of Singleton and Rossi (1999) using the reaction with the Folin-Ciocalteau reagent. Antioxidant activity (mg trolox per 100 g fresh weight) was determined using the radical DPPH method adapted from Brand-Williams et al. (1995). The fresh mass loss was determined by the difference between the fruit mass, first when it was harvested and the second when it was evaluated in storage. The results were expressed as percentage of the occurrence of rot and was recorded when the peach fruits showed lesions with diameter over 0.5 cm. Results were expressed as percentage and ripening index. Its calculation was based on the difference of absorbance between two wavelengths near the chlorophyll-a absorption peak. Readings were standardized, two spots on both sides of the fruit were verified by a portable spectrophotometer DA meter® (Turony, Italy). The color of the epidermis was examined using a colorimeter type Minolta CR-300, with a D65 light source that measured "L" (luminosity), "a*", "b*". The hue or chromatic tonality represented by the "hue angle" and firmness of the pulp (Newtons) was measured at two opposite points in the equatorial region of peeled fruit using a manual penetrometer (model 53205, TR TURONI-Italy) with 8 mm tip. The color of the epidermis was examined using a colorimeter type Minolta CR-300, with a D65 light source that measured "L" (luminosity), "a*", "b*", and the hue or chromatic tonality was represented by the "hue

angle". The total soluble solids (PBrix) was measured using an Atago digital refractometer^{*}; and the titratable acidity (% citric acid) was quantified in 10 mL of juice diluted in 90 mL of distilled water and titrated with 0.1 mol L NaOH solution to pH 8.1 with the aid of the Quimus pHmeter^{*}.

Statistical analysis

The data collected were subjected to analysis of variance by the F-test. The means were compared using the Tukey's Test, where $p \le 0.05$ of probability using the Assitat software (Silva and Azevedo, 2016).

Conclusion

Rootstock genotypes interfered with variables of peach quality throughout the cold storage period, followed by commercialization simulation. Enzymatic browning was not found in peaches from trees grafted on Seleção Viamão and Umezeiro rootstocks. They also had the lowest content of phenolic compounds in the pulp. Maciel peaches, regardless of the rootstock, may be stored up to 14+3 days. After that, fruits lose quality, mainly due to mass loss, rotting and reduction in pulp firmness.

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