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BIOACTIVE COMPOUNDS AND PHYSICAL-CHEMICAL CHARACTERISTICS OF TWO GENOTYPES OF PEACH TREES SUBMITTED TO NITROGEN FERTILIZATION

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Abstract

Nitrogen, which is the primary nutrient peach trees need, may affect their fruit quality. Thus, this study aimed at evaluating the effect of nitrogen fertilization on two genotypes of peach trees, regarding their fruit quality, in three consecutive crops. The experiment was carried out in the experimental area that belongs to the Embrapa Clima Temperado, located in Pelotas, Rio Grande do Sul (RS), Brazil, from 2016, 2016, 2017 and 2018. Four doses of nitrogen (0, 60, 120 and 180 Kg ha⁻¹) and two peach tree genotypes ('Cascata 1513' and 'Cascata 1067') were used. For the fruit, we evaluated epidermis color, pulp firmness, epidermis firmness, soluble solids concentration, titratable acidity, concentration of total phenolic compounds, anthocyanins and antioxidant activity. Fruit underwent physical, chemical and bioactive compound analyses. Results showed that the highest dose of nitrogen (180 Kg ha⁻¹) applied to the soil retards fruit ripening, while no application of nitrogen fertilization brings fruit maturation forward. Nitrogen fertilization via soil does not favor anthocyanins in fruit. Doses of 60 and 120 Kg ha⁻¹ of nitrogen are recommended because they lead to improvement in peach color, epidermis firmness and acidity. Peach tree genotypes influence soluble solids, juice pH, phenolic compounds and antioxidant activity of their fruit.

Keywords: Fruit quality. Mineral nutrition. Prunus persica. Ripening.

1. Introduction

Peach quality is fundamental to commercialization, since some attributes, such as color, taste, flavor and texture, are required by consumers (Trevisan et al. 2010; Cuquel et al. 2011). These quality parameters are directly related to the use of techniques of crop management (Gonçalves et al. 2014), cultivar genetics (Cremasco et al. 2016; Ferreira et al. 2016) and orchard fertilization (Dolinski et al. 2018; Ferreira et al. 2018).

Adequate fertilization management in peach tree orchards impacts fruit productivity and quality. Concerning peach trees, nitrogen (N) is their main nutrient (Dolinski et al. 2018), since it performs specific functions in plants, such as amino acid, protein and chloroplast constitution and regulation of metabolic reactions (Kerbauy 2012; Taiz et al. 2017).

N affects vegetative growth, productivity and fruit quality directly (Falguera et al. 2012; Olivos et al. 2012; Pascual et al. 2013; Ferreira et al. 2018; Dolinski et al. 2018). However, the right dose must be defined

so as to ensure nutritional balance of plants and keep their ratio of vegetative and productive growth. Excess of N may stimulate vegetative growth of peach trees (Della Bruna and Back 2014; Ferreira et al. 2018) and delay fruit ripening (Crisosto and Costa 2008; Rufat et al. 2011), while N deficiency affects plant photosynthesis (Leal et al. 2007; Taiz et al. 2017).

N fertilization may influence fruit quality parameters, such as acidity (Ferreira et al. 2016), soluble solids (Dolinski et al. 2018), epidermis color (Crisosto and Costa 2008; Dolinski et al. 2018), phenolic compounds, antioxidant activity (Ferreira et al. 2016; Vashisth et al. 2017) and carotenoids (Ferreira et al. 2016). Fruit quality parameters may undergo changes, not only because of the mount of N provided to plants, but also because of peach tree genotypes (Segantini et al. 2012; Santos et al. 2013; Vashisth et al. 2017).

Therefore, to improve peach quality, doses of N applied to the soil must be adjusted to the genotype. This study aimed at evaluating physical-chemical attributes and bioactive compounds of fruit borne by two peach tree genotypes submitted to different doses of N fertilization in three consecutive crops.

2. Material and Methods

The experiment was carried out in the experimental area that belongs to the Embrapa Clima Temperado in Pelotas, Rio Grande do Sul (RS) state, Brazil (31º 40' 41.29" S and 52º 26' 22.05" W and altitude of 70m) from 2016 to 2018. In W. Köppen's classification, the climate in the region is humid subtropical ('cfa'), i.e., humid temperate with hot summers (Alvares et al. 2013). The soil is moderately deep and medium-textured in A Horizon and clay-textured in B Horizon, classified into Red-yellow Argissol (Santos et al. 2006).

The experiment had a randomized block design in a 2x4 factorial scheme (two selections of peach trees and four doses of N), with four replicates. Every experimental unit comprised four plants; both central ones were used for the evaluation. Two advanced selections of peach trees ('Cascata 1513' and 'Cascata 1067'), from the genetic improvement program developed by the Embrapa Clima Temperado were used in the experiment.

Fruit borne by these selections have good potential to be commercialized due to very red epidermis, which attracts consumers. Besides, Cascata 1513 has excellent crunchiness. The orchard, which was implemented in 2012, has plants conducted in a 'Y' system, whose spacing between rows is 5m and between plants is 1.5m. Density is 1,333 plants ha⁻¹.

Doses of N (0, 60, 120 and 180 kg ha⁻¹) were applied to the soil surface in the crown projection area, with no incorporation. Urea at 45%, which was the N source, was applied to trees in three periods: 50% at full bloom, 30% after thinning and 20% after harvest. All parcels got equal doses of potassium (K) and phosphorus (P), in agreement with recommendations issued by the CQFS – RS, SC (2016). Physical-chemical analyses of soil that were carried out before the beginning of the experiment exhibited the following results: water pH was 5.8; P was 17.9 mg dm⁻³; K was 155 mg dm⁻³, Ca was 1.4 cmmolc dm⁻³; Mg was 0.6 cmolc dm⁻³; clay was 260 g kg⁻¹; and organic matter was 140 g kg⁻¹.

In the three crops, 20 fruit were harvested per plot to have the following variables evaluated: epidermis color, which was evaluated on the equatorial region of fruit by a Minolta colorimeter, model CR-400, which provides parameters L* a* b*, where L* expresses luminosity (L* = 0 is black and L* = 100 is white) and matrix (Hue angle); epidermis and pulp firmness, measured by a Stable Micro Systems texture analyzer, model TA-XT Plus with a 2-mm P-2 pointer, 5 g strength and 5 mms⁻¹ speed, and results expressed as Newtons (N); soluble solids measured by an ATAGO digital hand-held refractometer, model PAL-1, and results expressed as °Brix; titratable acidity, determined in 10 mL fruit pulp added to 90 mL distilled water, and subsequent sample titration by digital burette with a solution of sodium hydroxide (NaOH) at 0.1N up to the turning point, which is pH 8.1, and results expressed as grams of citric acid.100g⁻¹ pulp; and hydrogenionic potential (pH), determined by an electrometric method and a Quimis pH meter, model Q400A.

In order to determine bioactive compounds in peach pulp, the following analyses were carried out: total phenolic compounds, determined by the method based on the reaction with the Folin-Ciocalteau reagent, in agreement with the method adapted from Swain and Hillis (1959), and results expressed as mg

chlorogenic acid in 100g sample; antioxidant activity, determined by the DPPH radical method, in agreement with the method proposed by Brand-Williams et al. (1995), and results expressed as mg Trolox equivalent in 100 g⁻¹ sample; and total anthocyanins, quantified in agreement with the methodology used by Fuleki and Francis (1968), with adaptations, and results expressed as mg cyanidin 3-glucoside/100 g sample.

Data were submitted to the analysis of variance by the F Test. When the effect of the qualitative factor (genotype) was significant, the Tukey's test at 5% probability was carried out. When the quantitative factor (doses of N fertilization) was significant, the analysis of regression was conducted. Statistical analyses were run by the SISVAR program, version 5.6 (Ferreira, 2014).

3. Results and Discussion

The more increase in the doses of N applied to the soil, the more the °Hue values of peach epidermis color increased (Figure 1A e 1C), showing greenish fruit. In the second crop (2017), this variable exhibited interaction between genotypes and doses of N. Increase in doses of N led to linear increase in °Hue values of 'Cascata 1513' and 'Cascata 1067' peach epidermis. Increase was higher in 'Cascata 1067' (Figure 1B). This result may be related to plant vigor, since high doses of N lead to higher development of the aerial part of peach trees (Della Bruna and Back 2014; Ferreira et al. 2018), thus, interfering with luminosity and solar radiation on the crown and, consequently, yielding greenish fruit.

The lowest values of epidermis color were found in fruit borne by trees that were not submitted to N fertilization. Peaches were more reddish than the ones borne by trees that were submitted to N fertilization. Very red fruit are in their most advanced stage of ripening. It should be highlighted that fruit borne by trees that were not submitted to N fertilization ripened and were harvested about five days, on average, before the ones of the other treatments, in three crops under evaluation. N fertilization, at high doses, may delay fruit ripening (Crisoto and Costa, 2008; Rufat et al. 2011), while no N brings ripening forward, a fact that may happen due to the small aerial part of plants. Since there is small leaf area, resources are directly sent to the fruit (Duarte and Peil 2010).

Pulp firmness was not affected by the application of up to 180 kg ha⁻¹ N in the first and two crops. In the third crop under evaluation, pulp firmness of 'Cascata 1513' peaches got the highest value when 149.5 kg ha⁻¹ N was used, while 'Cascata 1067' peaches did not show any difference when distinct doses of N were applied to the soil (Figure 2A). Thus, the genotype may be related to this variable since pulp firmness of 'Cascata 1513' peaches was the highest (Table 1). Results agree with the ones found by Ferreira et al. (2016) and Cremasco et al. (2016), who observed that values of pulp firmness depended on peach tree cultivars.

Peach epidermis firmness exhibited interaction between genotypes and doses of N in the three crops under evaluation, but regression only adjusted to 'Cascata 1513' in 2017 and 2018 (Figures 2B, 2C and 2D). This genotype had quadratic behavior for epidermis firmness in 2017; the highest value was found when 178.5 Kg ha⁻¹ N was applied to the soil. In the third year (2018), the more N applied to the soil, the more linear increase in epidermis firmness. Clear differences found between 'Cascata 1513' and 'Cascata 1067' imply that genotypes respond differently to N fertilization.

The genotype 'Cascata 1067' exhibited fruit with higher values of soluble solids in 2017 and 2018 than the genotype 'Cascata 1513' (Table 1). Regarding N fertilization, doses of N do not affect contents of soluble solids in peaches. Similar results were found by Dolinski et al. (2005), who did not find any effect of N fertilization on contents of soluble solids in 'Chimarrita' peaches. Likewise, Vashisth et al. (2017) did not find such effect on the cultivar 'Tropic Beauty'. This variable has been associated with the location of fruits in the plant, light penetration into the crown, type of twigs, thinning (Picolotto et al., 2009), water absorption by plants as a response to different amounts of rainfall, which can cause sugar dilution in fruits (Brunetto et al., 2007).



Figure 1. A - Epidermis color of fruit borne by 'Cascata 1067' and 'Cascata 1513' peach trees submitted to different doses of N fertilization in 2016; B - Epidermis color of fruit borne by 'Cascata 1067' and 'Cascata 1513' peach trees submitted to different doses of N fertilization in 2017; C - Epidermis color of fruit borne by 'Cascata 1067' and 'Cascata 1513' peach trees submitted to different doses of N fertilization in 2017; C - Epidermis color of fruit borne by 'Cascata 1067' and 'Cascata 1513' peach trees submitted to different doses of N fertilization in 2017; C - Epidermis color of fruit borne by 'Cascata 1067' and 'Cascata 1513' peach trees submitted to different doses of N fertilization in 2018.



Figure 2. A - Pulp firmness of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in 2018; B - Epidermis firmness of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in 2016; C - Epidermis firmness of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in 2017; D - Epidermis firmness of fruit borne by Cascata 1067 and Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in 2017; D - Epidermis firmness of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in 2017; D - Epidermis firmness of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in 2017; D - Epidermis firmness of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in 2017; D - Epidermis firmness of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in 2017; D - Epidermis firmness of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in 2018.

Genotypes	Firmness of pulp (N)			Soluble solids (°Brix)			pH of the juice		
	2016	2017	2018	2016	2017	2018	2016	2017	2018
Seleção 1513	1.96 a	3.77 a	2.82 a	11.05	11.33b	13.07 b	4.60 a	4.42 a	4.55 a
Seleção 1067	1.62 b	3.34 b	2.59 b	11.32	12.11 a	13.52 a	3.94 b	4.10 b	3.96 b
Significativo	*	*	*	ns	*	*	*	*	*
Doses of N (kg ha⁻¹)									
0	1.67	3.19	2.44 b	11.42	11.71	12.86	4.28	4.50	4.37
60	1.89	3.71	2.63 ab	11.23	12.17	13.47	4.31	4.21	4.39
120	1.65	3.66	2.93 a	11.32	11.57	13.37	4.10	4.21	4.34
180	1.97	3.71	2.82 ab	10.76	11.42	13.47	4.27	4.14	3.91
Significant	ns	ns	*	ns	ns	ns	ns	ns	ns
Interaction	ns	ns	*	ns	ns	ns	ns	ns	ns

Table 1. Pulp firmness, soluble solids and pH of juice extracted from fruit borne by Cascata 1067 and Cascata1513 peach trees submitted to different doses of N fertilization in Pelotas, RS, Brazil in 2016, 2017 and 2018.

Means followed by same letters do not differ by the Tukey's test, at 5% probability ($p \le 0.05$). ns = not significant at 5% probability of error. * significant at 5% probability of error.

Concerning juice pH, 'Cascata 1513' peaches had higher values than 'Cascata 1067', but there was no difference in doses of N applied to the soil in the three crops under evaluation (Table 1). This result agrees with Ferreira et al. (2016), who did not find significant differences in pH of peaches submitted to different doses of N. Results found by Alcobendas et al. (2013) imply that pH of fruit juice is more related to the

orientation of the fruit in the plant. The authors stated that titratable acidity of peaches may depend on climate factors.

In the first two crops, quadratic behavior for titratable acidity was found in relation to doses of N (Figure 3A e 3B). In the third year (2018), there was interaction among factors, but regression only adjusted to 'Cascata 1513'; the highest value of acidity was found when the dose of N was about 100 kg ha⁻¹ in the soil (Figure 3C). Ferreira et al. (2016) found interaction between cultivars ('BRS Kampai', 'BRS Rubimel' and 'Cascata 805') and doses of N (0, 100 and 200 kg ha⁻¹) in the first crop, but treatments did not have any effect in the second crop. According to Barreto et al. (2020), it is possible that a variable depends on other factors, such as climatic factors, which are variable each year of cultivation.

Fruit of genotype 'Cascata 1513' exhibited the highest concentration of total anthocyanins, by comparison with the one of 'Cascata 1067' in 2016 (Table 2), even though they are white pulp peaches. It happens because the part of the pulp that involves fruit stones of these peach tree selections range from pink to red.

Genotypes		Anthocyanins ¹		Total phenolic compounds ²			
	2016	2017	2018	2016	2017	2018	
Seleção 1513	6.05 a	7.56	5.38	180.11 b	123.65 b	112.36 b	
Seleção 1067	4.31 b	6.36	5.34	286.68 a	182.18 a	204.56 a	
Significant	*	ns	ns	*	*	*	
	A	ntioxidant activit	γ ³				
Genotypes	2016	2017	2018	-			
Seleção 1513	339.23 b	397.51 b	307.19 b	-			
Seleção 1067	555.67 a	672.37 a	646.22 a				
Significant	*	*	*				

Table 2. Total anthocyanins, total phenolic compounds and antioxidant activity of fruit borne by Cascata 1067 and Cascata 1513 peach trees in Pelotas, RS, Brazil in 2016, 2017 and 2018.

¹ mg cyanidin-3-glycoside equivalent 100 g⁻¹; ²mg Gallic acid equivalent 100 g⁻¹ fresh weight. ³mg equivalent trolox 100 g⁻¹ fresh weight. Means followed by the same lowercase letter do not differ significantly from each other ($p \le 0.05$), calculated by the Tukey's test. Means followed by the same lowercase letter do not differ significantly from each other ($p \le 0.05$), calculated by the Tukey's test.

In the first crop under evaluation, quadratic behavior for total anthocyanins was found in relation to doses of N and the lowest value was found when 108 kg ha⁻¹ N was applied to the soil. Regarding the third crop under evaluation, linear decrease in anthocyanins was found as N application was increased in the soil (Figure 4C). In general, high concentration of anthocyanins were observed in plants with no N fertilization or with low doses of N. These results corroborate the ones found by Vashisth et al. (2017), who found increase in the contents of anthocyanins in fruit with descending rates of N. Possibly; this result must be the largest vegetal mass in the plants with the use of nitrogen fertilization.

Phenolic compounds of peaches did not respond to N fertilization applied to peach trees in the three crops (Figure 4A). However, Vashisth et al. (2017) and Strissel et al. (2005) observed decrease in total phenolic compounds after N application. In the study reported by this paper, since there was no response to N fertilization, this variable may be related to genetic factors, as shown in Table 2.

Concentrations of total phenols ranged from 182.18 to 286.68mg chlorogenic acid 100g⁻¹ in the pulp of 'Cascata 1067' peaches, while lower values were found in 'Cascata 1513' ones (Table 1). Variation in phenolic compounds of peaches, depending on the cultivar, was also found by Segantini et al. (2012), Santos et al. (2013) and Ferreira et al. (2016), who attributed differences in their concentrations to genetic characteristics of genotypes.

The highest antioxidant activity was found in the pulp of 'Cascata 1067' peaches, whose values ranged from 555.67 to 672.37mg 100g⁻¹ in the three crops under evaluation (Table 2), although no effect of N treatment was found on both genotypes (Figure 4B). As was implied in the case of phenolic compounds, this study suggests that genotypes and environmental factors influence the determination of antioxidant capacity of peaches directly (Segantini et al. 2012; Santos et al. 2013).



Figure 3. A - Titratable acidity of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in Pelotas, RS, Brazil in 2016; B - Titratable acidity of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in Pelotas, RS, Brazil in 2017; C - Titratable acidity of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in Pelotas, RS, Brazil in 2017; C - Titratable acidity of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in Pelotas, RS, Brazil in 2017; C - Titratable acidity of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in Pelotas, RS, Brazil in 2018.



Figure 4. A - Total phenolic compounds of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in 2016, 2017 and 2018; B - Antioxidant activity of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in 2016, 2017 and 2018; C - Total anthocyanins of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in 2016, 2017 and 2018; C - Total anthocyanins of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in 2016, 2017 and 2018; C - Total anthocyanins of fruit borne by Cascata 1067 and Cascata 1513 peach trees submitted to different doses of N fertilization in 2016, 2017 and 2018.

4. Conclusions

The dose of 180 Kg ha⁻¹ N delays fruit ripening of the genotypes Cascata 1067 and Cascata 1513. Anthocyanins are not favored by N fertilization. Doses of 60 and 120 Kg ha⁻¹ N favor peach color, epidermis firmness and acidity. Peach tree genotypes influence soluble solids, juice pH, phenolic compounds and antioxidant activity of their fruit.

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