## **Notas Científicas**

## Plant growth regulators to enhance fruit color of 'Gala' apples

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Abstract – The objective of this work was to investigate the effect of plant growth regulators for enhancing fruit color of 'Gala Standard' apples (*Malus domestica*). The experiment was carried out in the 2015 and 2016 crop seasons. The treatments consisted of water, as a control; 300 mg L<sup>-1</sup> ethephon, as a positive control; 400 mg L<sup>-1</sup> prohydrojasmonate; and 400 mg L<sup>-1</sup> abscisic acid. Flesh firmness, soluble solids content, fruit weight, and red color were assessed after harvest. Plant growth regulators enhanced red color of fruit and chlorophyll degradation. Prohydrojasmonate and abscisic acid did not reduce flesh firmness, in the 2016 season.

Index terms: Malus domestica, abscisic acid, ethephon, index of absorbance difference, prohydrojasmonate.

## Fitorreguladores para o aumento da coloração de maçãs 'Gala'

Resumo – O objetivo deste trabalho foi determinar o efeito da aplicação de fitorreguladores no aumento da coloração de frutos de maçã 'Gala Standard' (*Malus domestica*). O experimento foi realizado nas safras 2015 e 2016. Os tratamentos consistiram em água, como controle; 300 mg L<sup>-1</sup> de ethephon, como controle positivo; 400 mg L<sup>-1</sup> de prohidrojasmonato; e 400 mg L<sup>-1</sup> de ácido abscísico. Após a colheita dos frutos, foram avaliadas a firmeza de polpa, o teor de sólidos solúveis, a massa e a coloração vermelha dos frutos. As aplicações dos fitorreguladores aumentaram a coloração vermelha dos frutos e a degradação da clorofila. O prohidrojasmonato e o ácido abscísico não reduziram a firmeza de polpa, na safra 2016.

Termos para indexação: *Malus domestica*, ácido abscísico, ethephon, índice de diferença da absorbância, prohidrojasmonato.

A key quality attribute of apple fruit is its skin color, which not only affects consumer preferences, but is also associated with the nutritional value of the fruits. Pigment accumulation in apple fruit can be affected by environmental conditions and cultural practices. In general, red cultivars are the most preferred, and within a cultivar, better colored fruits are in higher demand (Saure, 1990; Rabinovich, 2009).

Anthocyanin biosynthesis is regulated by a complex interaction between internal and external stimuli such as temperature, light, carbohydrates, water stress, and plant hormones (Loreti et al., 2008). A number of cultural techniques have received special attention as management tools to externally control anthocyanin formation and consequently improve skin coloration, such as summer pruning (Guerra & Casquero, 2010), hail net and reflective foil (Jakopič et al., 2010), and exogenous applications of plant growth regulators (Petri et al., 2011; Atay, 2015).

Currently in Brazil, ethylene is the main plant growth regulator used to enhance fruit red color (Steffens et al., 2006), by spraying the fruits with its commercial form Ethrel. Its application advances fruit matutrity, allows early harvest, and also increases the red color of fruit skin (Ban et al., 2007). However, ethephon application may decrease organoleptic quality and decrease the storage potential of fruit after harvest, especially in terms of flesh firmness reduction (Steffens et al., 2006; Brackmann et al., 2014, 2015).

Recently, new studies have been carried out with the objective of finding chemical products that stimulate the accumulation of anthocyanins in fruit skin without the inconveniences of maturity acceleration, ethylene production, and reduction of storage potential (Bizjak et al., 2013). Among these substances, it has been reported that plant growth regulators as abscisic acid (Lacampagne et al., 2010; Leng et al., 2014) and jasmonates (Rudell & Mattheis, 2008; Kondo et al.,

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2014), which are naturally synthesized by plants, are related to anthocyanin synthesis and consequently to fruit color development of several species.

In the main apple-producing regions of the world, studies are being carried out to determine the effect of jasmonates and abscisic acid on fruit ripening. For example, in the United States, their positive effects were detected for Fuji and Buckeye Gala cultivars (Rudell & Mattheis, 2008; Francescatto, 2013); and in Turkey and Italy, positive effects were found for Gala cultivar and its clones Mondial and Brookfield (Falchi et al., 2014; Atay, 2015). However, studies focusing on fruit maturation associated with the use of these plant growth regulators have not yet been conducted in Brazil.

In the main apple-growing regions of Southern Brazil, it is common to find situations not favorable for achieving good skin color, such as the planting of poorcolored cultivars (Gala Standard and Fuji Standard) (Epagri, 2006); the adoption of vigorous rootstocks, which normally result in fruit shading (Pasa et al., 2016); excessive rainfall and cloudiness, resulting in lower availability of ultraviolet radiation, as well as high daytime and nighttime temperatures (above 18°C), especially in lower altitude producing regions (Cardoso et al., 2012). All of these conditions are associated with lower accumulation of anthocyanins in fruit skin (Saure, 1990).

Due to the lack of information regarding the use of abscisic acid and jasmonates to enhance apple red color in Southern Brazil, the objective of the present work was to investigate the effect of different plant growth regulators on skin color development and others aspects of fruit quality of cv. Gala Standard.

The study was performed in a commercial orchard in the region of São Joaquim, in the state of Santa Catarina (28°11'43"S, 49°59'41"W, at an altitude 1,200 m) during the 2015 and 2016 crop seasons. The climate of the region is mesothermal humid (Cfb) according to the Köppen-Geiger classificaton, i.e, constantly humid temperate climate, without dry season and with a cool summer (Benez, 2005). Average accumulation of temperatures bellow 7.2°C is 900 hours. The soil of the experimental field is a Cambissolo Húmico (Inceptisol), according to the Brazilian soil classification system (Santos et al., 2013). Plant material consisted of 16 year-old 'Gala Standard' apple trees, grafted on 'Marubakaido' rootstock. Trees

were spaced at 6 m between rows and 2 m within the row (833 trees per hectare) and were trained in a central-leader system, without supporting structure. Orchard management was performed according to recommendations for the apple production system (Epagri, 2006).

The treatments consisted of water, as a control; 300 mg L<sup>-1</sup> ethephon, as a positive control; 400 mg L<sup>-1</sup> prohydrojasmonate; and 400 mg L<sup>-1</sup> abscisic acid. The experiment was arranged in a randomized complete block design with six replicates (plots). Each plot consisted of three plants, but only the central plant was used in the evaluations, leaving one plant at each end as a border. For laboratory evaluations, samples of 30 fruits were randomly selected from the central plant of each plot. Data were subjected to the analysis of variance (Anova) at 5% probability, and when treatment effects were detected, Tukey's test was performed at 5% probability.

The application of plant growth regulators was carried out approximately 20 days before harvest (1/22/2015 and 1/20/2016). They were sprayed using the Stihl SR430 powered backpack sprayer (40 psi working pressure). At the time of application, the volume limit applied was the runoff point. Fruits were harvested at commercial maturity, according to the starch-iodine index (4–5), flesh firmness (80–90 N), and soluble solids content (11–12°Brix) (Epagri, 2006). Fruit color, red color intensity, index of absorbance difference (I<sub>AD</sub>), fruit weight, flesh firmness, and soluble solids concentration were evaluated.

The intensity (%) of red color was visually estimated, considering the percentage of reddish area relative to fruit total surface (Argenta et al., 2010). Red color intensity was evaluated using the Minolta CR-300 Chroma Meter (Minolta Camera Co., Osaka, Japan), which measured two color parameters (a\* and b\* values). Hue angle (arctan b\*/a\*), a color-intensity indicator for determining color differences in fruit that ripen from green to either yellow or shades of red, was obtained using the formulas provided by McGuire (1992) and expressed in degrees (0°, red-purple; 90°, yellow; and 180°, bluish-green). Two measurements were performed in the equatorial regions of the fruits, one in the region less exposed to the sun and less red, and the other in the reddish region.

The index of absorbance difference (I<sub>AD</sub>), which measures chlorophyll degradation, was determined

through a portable vis/NIR device, the DA-Meter (Sinteleia, Bologna, Italy), based on fruit absorbance spectra. It was calculated as  $I_{AD} = A670 - A720$ , where A670 and A720, near the chlorophylla absorbance peak, are the A values at the wavelengths of 670 and 720 nm, respectively. Measurements were taken in two opposite equatorial points at the transition area between the red and green color of the fruit (Toivonen, 2014).

The weight of the sampled fruits was measured on a digital scale and the results were expressed in grams. Flesh firmness was determined by removing fruit skin on two opposite sides of each fruit, using the Fruit Texture Analyzer with an 11-mm diameter probe (Güss Manufacturing, Strand, South Africa), and was expressed in Newton. To measure the soluble solids (SS), the sample was juiced and placed on the prism of the PR-32 digital refractometer (Atago Co., Ltd., Tokyo, Japan), the results were expressed as °Brix (Pasa et al., 2016).

Fruit weight and soluble solids contents were not affected by the application of plant growth regulators in both evaluated seasons. Similar results were obtained by Francescatto (2013), who verified that prohydrojasmonate did not affect the fruit weight and soluble solids contents of Buckeye Gala cultivar in the United States.

For flesh firmness, no significant differences were found in the 2015 crop season. However, in the 2016 season, the fruits treated with ethephon showed lower flesh firmness compared with the other treatments (Table 1). These results confirm the findings of other authors, who reported that ethephon had the

disadvantage of accelerating the fruit ripening process, resulting in reduced flesh firmness (Steffens et al., 2006; Brackmann et al., 2014, 2015).

In both evaluated seasons, fruits treated with prohydrojasmonate, ethephon, and abscisic acid showed lower values of Hue angle (Ho) than the control, that is, they showed a higher intensity of red color, whereas the control had a higher intensity of yellow color (Table 1). There was greater chlorophyll degradation in the skin (IAD) of fruits subjected to the application of plant growth regulators. In both evaluated seasons, the lowest chlorophyll degradation was observed in control fruits. Research results obtained in Turkey also showed the efficiency of prohydrojasmonate to enhance red color of Mondial Gala cultivar (Atay, 2015), and the results obtained in Italy with Gala Brookfield cultivar also confirmed the effect of abscisic acid on improving the red color of fruits (Falchi et al., 2014).

Regarding the percentage of fruits with red color, control fruits presented the lowest fruit surface with red color (Table 1 and Figure 1). In the 2015 season, the applications of ethephon, prohydrojasmonate, and abscisic acid resulted in an increase of 16, 25, and 47% of fruit surface with red color, respectively, compared with the control. In the 2016 season, an increase of 82, 90, and 95% of fruit surface with red color was observed when prohydrojasmonate, ethephon, and abscisic acid were applied, respectively. In another study, with the 'Buckeye Gala', there was an increase of 15% in fruit surface with red color in response to the application of prohydrojasmonate (Francescatto, 2013).

**Table 1.** Characteristics of 'Gala Standard' apple (*Malus domestica*) fruits, subjected to the application of prohydrojasmonate (Prohy), abscisic acid, and ethephon, in the 2015 and 2016 crop seasons<sup>(1)</sup>.

Treatment	Fruit weight (g)		Flesh firmness (N)		SS (°Brix)		Hue angle (H°)		$I_{AD}$		Red color (%)	
-	2015	2016	2015	2016	2015	2016	2015	2016	2015	2016	2015	2016
400 mg L <sup>-1</sup> Prohy	130.0	122.7	70.8	80.5a	10.9	11.9	29.3a	64.4b	0.37ab	0.44ab	64.1b	64.1b
400 mg L <sup>-1</sup> Abscisic acid	121.0	127.8	69.9	80.1a	11.2	12.0	30.9a	63.1b	0.40b	0.40a	59.3b	66.9ab
300 mg L <sup>-1</sup> Ethephon	129.9	132.4	69.9	76.5b	11.0	11.7	28.7a	59.6a	0.30a	0.47b	75.3a	68.6a
Control	123.9	124.1	70.3	80.1a	10.8	11.5	37.9b	77.2c	0.65c	0.80c	51.1c	35.1c
p-value	0.095	0.756	0.590	< 0.001	0.705	0.571	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001

<sup>(1)</sup> Means followed by equal letters, in the columns, do not differ by Tukey's test, at 5% probability.

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The plant growth regulators prohydrojasmonate and abscisic acid were as efficient as ethephon to enhance red coloration of 'Gala' fruits in both evaluated seasons. Based on the 2016 crop season, the results suggest

2015 2016 300 mg L1 ethephon 300 mg L' ethephon 400 mg L1 abscisic acid 400 mg L-1 abscisic acid 400 mg L-1 prohydrojasmonate 400 mg L<sup>-1</sup> prohydrojasmonate

**Figure 1.** Color appearance of the sun-exposed side of 'Gala Standard' apple (*Malus domestica*) fruits, at commercial harvest, subjected to the application of different plant growth regulators in the 2015 and 2016 crop seasons.

that prohydrojasmonate and abscisic acid intensify the red coloration of the fruits without stimulating the ripening process and without impairing fruit quality.

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