Ripening and fruit quality of 'Fortune' plums treated by pre-harvest application of

ripening stimulants

Maturação e qualidade de ameixas 'Fortune' tratadas em pré-colheita com estimulantes de amadurecimento

Maduración y calidad de la fruta de las ciruelas "Fortune" tratadas mediante la aplicación de estimulantes de la maduración antes de la cosecha

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Carine Cocco ORCID: https://orcid.org/0000-0002-1850-2266 University of Caxias do Sul, Brazil E-mail: ccocco@ucs.br Wendel Paulo Silvestre ORCID: https://orcid.org/0000-0002-9376-6405 University of Caxias do Sul, Brazil E-mail: wpsilvestre@ucs.br Gabriela Weber Schildt ORCID: https://orcid.org/0000-0001-9325-5472 University of Caxias do Sul, Brazil E-mail: gwschildt@ucs.br Felipe Afonso Tessaro ORCID: https://orcid.org/0000-0003-4109-9724 University of Caxias do Sul, Brazil E-mail: fatessaro@ucs.br

Abstract

The use of plant growth regulators that increase ethylene production is a common strategy to anticipate fruit ripening, increase peel color, and reduce the number of collections to complete the harvest of the fruits. Besides, this strategy may come true as a helping technique for the farmers to reduce costs with manpower, increasing overall profitability. This work aimed to evaluate the application of several concentrations of ethephon, associated with boric acid, on the anticipation of ripening of 'Fortune' plums, cultivated in Serra Gaúcha region, South Brazil. The study was carried out in a commercial orchard during the 2018-2019 and 2019-2020 harvests. It was used a completely randomized design with six treatments and four replicates; each replicate was composed of four plants. The following parameters were evaluated: the amount of fruits collected in each harvest, average fruit mass, diameter, length, firmness, soluble solids content, titratable acidity, SS/TA ratio, total phenolic compounds, and total anthocyanins. According to the results, the application of anthocyanin and phenolic compounds contents and fruit firmness. On the other hand, the application of boric acid increased fruit firmness. From an economic standpoint, the pre-harvest application of ethephon may be interesting to reduce manpower requirements during harvest by an earlier harvest and by reducing the number of harvest cycles.

Keywords: Prunus salicina; Peel color; Ethylene; Micronutrients.

Resumo

O uso de reguladores de crescimento vegetal que aumentam a produção de etileno é uma estratégia comum para antecipar o amadurecimento dos frutos, aumentar a cor da casca e reduzir o número de coletas para finalizar a colheita dos frutos. Além disso, essa estratégia pode se concretizar como uma técnica de auxílio aos agricultores na redução de custos com mão de obra, aumentando a lucratividade geral. Este trabalho teve como objetivo avaliar a aplicação de diversas concentrações de etefom, associado ao ácido bórico, na antecipação do amadurecimento de ameixas 'Fortune', cultivadas na região da Serra Gaúcha, Sul do Brasil. O estudo foi realizado em pomar comercial durante as safras 2018-2019 e 2019-2020. O delineamento experimental foi inteiramente casualizado com seis tratamentos e quatro repetições; cada repetição foi composta por quatro plantas. Foram avaliados os seguintes parâmetros: quantidade de frutos coletados em cada colheita, massa média do fruto, diâmetro, comprimento, firmeza, teor de sólidos solúveis, acidez titulável, relação SS/AT, compostos fenólicos totais e antocianinas totais. De acordo com os resultados, a aplicação de etefom foi capaz de acelerar e homogeneizar o amadurecimento parcial da ameixa, mas também causou redução no teor de antocianinas e compostos fenólicos e na firmeza dos frutos. Por outro lado, a

aplicação de ácido bórico aumentou a firmeza dos frutos. Do ponto de vista econômico, a aplicação pré-colheita de etefom pode ser interessante para reduzir a necessidade de mão de obra durante a colheita por uma colheita mais precoce e pela redução do número de ciclos de colheita.

Palavras-chave: Cor da casca; Etileno; Micronutrientes; Prunus salicina.

Resumen

El uso de reguladores del crecimiento de las plantas que aumentan la producción de etileno es una estrategia común para anticipar la maduración de la fruta, aumentar el color de la cáscara y reducir el número de recolecciones para completar la cosecha de las frutas. Además, esta estrategia puede hacerse realidad como una técnica de ayuda para que los agricultores reduzcan los costos con mano de obra, aumentando la rentabilidad general. Este trabajo tuvo como objetivo evaluar la aplicación de varias concentraciones de etefón, asociado con ácido bórico, sobre la anticipación de la maduración de ciruelas 'Fortune', cultivadas en la región de Serra Gaúcha, Sur de Brasil. El estudio se realizó en un huerto comercial durante las cosechas 2018-2019 y 2019-2020. Se utilizó un diseño completamente al azar con seis tratamientos y cuatro repeticiones; cada réplica estaba compuesta por cuatro plantas. Se evaluaron los siguientes parámetros: cantidad de frutos recolectados en cada cosecha, masa promedio de frutos, diámetro, longitud, firmeza, contenido de sólidos solubles, acidez titulable, relación SS / TA, compuestos fenólicos totales y antocianinas totales. Según los resultados, la aplicación de etefón fue capaz de acelerar y homogeneizar parcialmente la maduración de la ciruela, pero también provocó una reducción del contenido de antocianinas y compuestos fenólicos y de la firmeza de la fruta. Por otro lado, la aplicación de ácido bórico aumentó la firmeza de la fruta. Desde un punto de vista económico, la aplicación de etefón antes de la cosecha puede ser interesante para reducir las necesidades de mano de obra durante la cosecha mediante una cosecha más temprana y al reducir el número de ciclos de cosecha. Palabras clave: Color de la cáscara; Etileno; Micronutrientes; Prunus salicina.

1. Introduction

The Japanese plum (*Prunus salicina*), native to China, is destined mainly for *in natura* consumption, being highly appreciated by its taste and nutritional properties. Plums are rich in anthocyanins, which give the fruit its characteristic color, and whose contents vary according to the cultivars (Roussos et al., 2016).

World plum production is concentrated in China, corresponding to 17% of total world production (6.78 million tonnes), followed by Romania and the USA. Among the countries of South America, Chile is the largest producer, occupying the seventh position in the ranking of the largest world producers (FAO, 2018). In Brazil, about 64,000 t are produced each year in 4,415 ha of acreage (Kleina et al., 2019). The region of South Brazil, composed of the states of Rio Grande do Sul, Santa Catarina, and Paraná, highlights itself, in which plums are cultivated between the end of October and February (IBGE, 2017).

There was an increase in the Brazilian production of plums in the last years, however, imports still occupy an important part of the traded volume, especially in off-season months. In 2017, 12,600 t of plums were imported from neighboring countries, such as Chile and Argentina (Kleina et al., 2019). In Rio Grande do Sul state, the 'Serra Gaúcha' region highlights itself, with an annual production of 15,000 t, where the plum cultivation is carried out in family properties with small acreages, about 0.5-2.0 ha. The cultivars 'Laetitia', Santa Rosa', 'Reubennel', and 'Fortune' are the most cultivated in this region (Fioravanço, 2015).

In the last years, the farmers have concentrated efforts in cultural practices to produce high-quality fruits, anticipate harvesting, and extend the shelf-life of the fruits. In this sense, the use of growth regulators may be an important tool to standardize fruit ripening and to reduce the harvest time with the doses being adjusted for the desired effects (Bisht, 2018). In the countries that have a long experience in the production of temperate fruits, growth regulators are widely used to manipulate the most important phenological stages of the plant, aiming to reduce costs with manpower and increase rentability (Schott; Walter, 2020).

The plum is a climacteric fruit, presenting an important increase in the respiratory rate at the start of the physiological maturation, caused by an increase in the endogenous synthesis of ethylene; this unleashes changes in color, taste, aroma, and texture of the fruit (Fioravanço et al., 2007). Due to the sensibility of the plum to ethylene, several substances may interfere in

the ripening process, anticipating or retarding this physiological process.

The plant hormone ethylene is associated with several processes, such as stimulation or inhibition of cell growth, induction of flowering, fostering of tissue and organ senescence, ripening of fruits, and abscission of flowers and fruits (Rademacher, 2015). Its synthesis occurs in all plant cells, not having a specific location of production, however, it depends on respiratory activity, temperature, and is also directly related to stress. Its translocation occurs by tissue diffusion, as a gas dissolved in the xylem (Petri et al., 2016).

Ethylene is capable to accelerate the phenological characteristics during the ripening phase in apples and other climacteric fruits, being the only phytohormone produced in the gaseous form, synthesized by the amino acid methionine (Schwambach and Cardoso Sobrinho, 2014). Its synthesis may be stimulated by synthetic products, such as ethephon (2-chloroethylphosphonic acid). When absorbed by leaves and flowers, this substance enters in cell cytoplasm, liberating ethylene and stimulating the plant to produce more of this phytohormone (Schott; Walter, 2020).

According to Petri et al. (2016), ethephon, by promoting ethylene biosynthesis, reduces the number of collections in each harvest, but also induces a quick fruit softening, ultimately causing an early fruit fall, as well as lower post-harvest durability and a reduced storage time under refrigeration. Given that, the storage of ethephon-treated fruits in pre-harvest is not advised.

Flores-Cantillano (2003) cited that a significant part of the production costs is related to manpower, especially at harvest, which requires training, since the degree of ripening of the fruit is a determinant factor, and conditions good post-harvest conservation. Normally, it is necessary to carry out several collections in each plant during harvest due to the disuniformity in fruit ripening. In general, about three collections are performed, avoiding collecting fruits with deficient ripening (Flores-Cantillano, 2005).

In this sense, boron is a micronutrient that acts in several phenological stages, including fruit ripening. Leaf treatments based on boron in the time before the harvest of 'Galaxy' apple increase the fruit respiratory rate and ethylene production. Then, it becomes possible to anticipate the harvest (Brackmann et al., 2016, Nachtigall and Czermainski, 2016; Nunes, 2016). However, there are many controversies relative to the efficiency of leaf application of boron, as highlighted by Coldebella et al. (2016).

As exposed, the objective of the present work was to evaluate the effect of ethephon, boron, and a commercial plant stimulant (Physiogrow Color[®]) on fruit ripening, quality, and content of bioactive compounds in 'Fortune' plums.

2. Methodology

2.1 Field experiment conditions

The experiments were conducted during the 2018/2019 and 2019/2020 harvests in a commercial orchard located in the municipality of Antônio Prado, ecoclimatic region of Northeast Mountain Range of Rio Grande do Sul, located in the geographical coordinates 28°50' W and 51°24' W, with an average altitude of 635 m. The climate of this region is classified as temperate, with cold winters and uniform rainfall, and mild summers, according to the Cfb climate by Köppen classification. The average yearly temperature is 16.6 °C, with an average yearly rainfall of 1,800 mm.

The study was carried out using six-year-old 'Fortune' Japanese plum trees, grafted on the 'Okinawa' rootstock, with a spacing of 4.5 m between lines and 1.8 m between plants, with a density of 1,235 plants ha⁻¹, conducted on 'Y' system. The pollinator cultivar was the 'Reubennel', implanted in a 15% proportion, acting only as a pollinator.

The orchard was irrigated by dripping, with a line of drippers in each plant line, with a spacing of 0.6 m between drippers and a flow rate of $0.6 \text{ L} \cdot \text{h}^{-1}$. When necessary, fertigation was also applied, according to the demand of the culture and the climatic conditions during the productive cycle. The winter pruning was carried out in July, and it aimed to remove

misplaced branches and replace some productive branches. During vegetative growth two green prunings were performed, the first 72 days after complete flowering and the second soon after harvesting to eliminate vigorous branches. It was carried out haul fertilization through canopy projection area; the amount of fertilizer was calculated based on soil and plant tissue analysis.

Two experiments were performed, the stimulants were applied when the plums presented a green-whitish color, with some fruit already starting the change to a reddish color.

Experiment One was carried out in the 2018/2019 harvest. Six treatments were applied; they were constituted by combined ethephon and boric acid applications. The first applications were carried out 20 days before the predicted harvest time, on November 30, 2018. The second applications were performed seven days after the first applications, on December 06, 2018. The treatments were: T1 - control (no application); T2 - one application of ethephon (100 mg·L⁻¹) + boric acid (10 mg·L⁻¹); T3 - two applications of ethephon (100 mg·L⁻¹) + boric acid (10 mg·L⁻¹); T4 - one application of ethephon (200 mg·L⁻¹) + boric acid (10 mg·L⁻¹); T5 - one application of ethephon (200 mg·L⁻¹) + boric acid (10 mg·L⁻¹); T5 - one applications of boric acid (10 mg·L⁻¹).

Experiment Two was carried out in 2019/2020 harvest; six treatments were evaluated; these were constituted by different ripening stimulants in plums. The treatments were: T1 - control (no application); T2 - Physiogrow[®] Color (6 L·ha⁻¹); T3 - Physiogrow[®] Color (8 L·ha⁻¹); 4 - ethephon (200 mg·L⁻¹); T5 - boric acid (30 mg·L⁻¹); T6 - ethephon (200 mg·L⁻¹) + boric acid (10 mg·L⁻¹). All applications were carried out 20 days before the predicted harvest date.

The experimental design in both experiments was by randomized blocks, with four replicates. Each replicate was constituted of four plants; only the two central plants of each replicate were evaluated. The source of ethephon in both experiments was Ethrel[®] (Bayer, USA), with 24 wt.% of ethephon; it was used boric acid P.A. (99.8%, Labsynth, Brazil) as the source of boron. The applications were carried out using a backpack sprayer, using a spray volume of 1,000 L·ha⁻¹.

The plums were collected when presented with a uniform purplish color. Three collections were needed in successive weeks, due to the uneven ripening. At harvest, the collected fruits were weighed using a digital scale with a capacity of 30 kg and resolution of 1 g. The production of each plant (kg·plant⁻¹) was calculated by the sum of the weight of the fruits in each collection.

2.2 Evaluation of fruit quality parameters

Ten fruits were randomly selected for each replicate to determine the average fruit mass. The fruits were weighted using an electronic scale with a capacity of 10 kg and resolution of 1 g; the data was presented as grams per fruit (g·fruit⁻¹). The diameter (equatorial length) and length (longitudinal length) of the fruits were measured using a digital caliper, with a measuring range of 0-150 mm and a resolution of 0.1 mm; the results were expressed in millimeters.

Pulp firmness was determined using a digital penetrometer with a 08 tip. Two readings were measured, on opposite sides, in the equatorial region of the fruit after removal of fruit peel by a shallow cut of two disks with a diameter of 1 cm. The results were expressed as kilograms per square centimeter (kg·cm⁻²).

The juice from ten fruits was extracted using an electric juice extractor. Soluble solids (SS) content was determined using an analog refractometer, with a measurement range of 0-30 °Brix and a resolution of 0.5 °Brix. Titratable acidity was measured by the method IAL 310/IV, described by IAL (2008); the results were described as the mass of equivalent malic acid by volume percentage (% m/v). The SS/TA ratio was calculated by dividing the SS by the TA values.

For the determination of the contents of anthocyanins and phenolic compounds, it was carried out a hydroalcoholic extraction using ethanol 70% v/v. Approximately 30 g of homogenized pulp and peel of the samples were macerated using a porcelain mortar and pestle. The macerated samples were transferred to a 125 mL Erlenmeyer and 100 mL of ethanol 70% v/v were added. The samples were kept at rest for 24 h at room temperature and away from sunlight, according to Bucic-Kojic et

al. (2009).

Total anthocyanin content was determined by the differential pH method, according to the AOAC procedures (Lee et al., 2005). The results were expressed as milligrams of equivalents of cyanidin-3-glucoside per kilogram of pulp ($mg \cdot kg^{-1}$). Phenolic compounds content was determined according to the procedures described by Gardin et al. (2012); the results were expressed as milligrams of equivalents of gallic acid per 100 g of pulp ($mg \cdot 100 g^{-1}$).

2.3 Statistical analysis

The obtained data underwent analysis of variance (ANOVA), and the means were compared by Tukey's multiple range test at 5% probability ($\alpha = 0.05$). The principal component analysis (PCA) was carried out using the same variables as the ANOVA; the correlation matrix of the data was used in the analysis. The production and quality parameters also underwent Spearman correlation analysis at 5% probability ($\alpha = 0.05$) relative to ethephon and boric acid concentrations and the commercial plant stimulant doses. The statistical analyses were carried out using the Statistica 12 (Statsoft, USA) software.

3. Results and Discussion

3.1 Experiment One

All treatments required three collections during harvest to carry out a complete removal of the ripe fruits (Table 1). However, the treatments that employed ethephon, regardless of its concentration, presented higher amounts of collected fruits in the first and second collections. In the first collection, about 37.92% of the fruits were harvested in ethephon-treated plants, whereas only 4.65% were collected in the control. These results indicated an eight-fold increase of fruit collection in ethephon treatments relative to the control and boric acid-only treatments. In the second collection, there was an increase in the percentage of collected fruits in both the control and the treatment of two applications of boric acid, not differing statistically from the treatment that employed ethephon 100 mg·L⁻¹ + boric acid 10 mg·L⁻¹.

In the third fruit collection, it was observed a higher percentage of fruits collected in the control treatment, with 59.4%, and in the boric acid treatment, with 58.8%. The treatments that employed ethephon have not differed among themselves, with percentages smaller than 26.5%. These results show the ripening stimulant effect of ethephon in plums, which ultimately leads to an anticipation of the harvest time.

Treatment	Harvest 1 (2018-12-11)	Harvest 2 (2018-12-19)	Harvest 3 (2018-12-27)	Overall yield
T1	1.25 b	10.93 b	17.75 a	29.88 a
T2	15.10 a	16.48 a	9.48 ab	40.94 a
Т3	11.40 a	13.60 ab	9.00 ab	33.94 a
T4	12.15 a	16.35 a	3.83 b	32,25 a
T5	17.08 a	17.73 a	4.45 b	30.13 a
T6	1.63 b	10.83 b	17.70 a	39.19 a
CV* (%)	37.52	15.41	40.81	17.21

Table 1 – Fruit yields (kg·plant⁻¹) in the three harvests and overall yield for Japanese plums in the 2018/2019 harvest.

Means in column followed by the same letter do not present statistical difference by Tukey's multiple range test at 5% probability ($\alpha = 0.05$). T1 – control; T2 – one application of ethephon (100 mg·L⁻¹) + boric acid (10 mg·L⁻¹); T3 – two applications of ethephon (100 mg·L⁻¹) + one application of boric acid (10 mg·L⁻¹); T4 – one application of ethephon (200 mg·L⁻¹) + boric acid (10 mg·L⁻¹); T5 – one application of ethephon (100 mg·L⁻¹) + two applications of boric acid (10 mg·L⁻¹); T6 – two applications of boric acid (10 mg·L⁻¹); * – coefficient of variation. Source: Authors (2021).

Petri et al. (2016) explained that ethephon generally anticipates fruit ripening when applied between one and four weeks before the estimated harvest time; its doses may vary between 100 and 200 mg·L⁻¹; concentrating the ripening in a window of 7-15 days, depending on the epoch, concentration, and temperature during and after application. The same authors

also highlighted that higher ethephon doses or temperatures higher than 25 °C intensify the action of the phytohormone, what hinders its widespread use in field conditions. This was observed in the present work since the treatments that used ethephon presented higher amounts of fruits collected in the first collections.

Fioravanço et al. (2007) applied ethephon on 'Reubennel' plums, aiming to anticipate the ripening process and to increase fruit color; the concentrations of zero, 120, 240, 360, and 480 mg·L⁻¹ were applied. All ethephon-based treatments anticipated the harvest in seven days. The authors also reported that the concentrations of 360 and 480 mg·L⁻¹ intensified the peel color of the plums, which presented a more reddish color than the plums treated with lower concentrations (zero, 120, and 240 mg·L⁻¹).

The strategy of anticipating the harvest influences directly the fruit price, since at the start of harvest time there is a lower supply and a high demand, which increases the market value of the plums, especially because harvest time coincides with Christmas and New Year celebrations, when the demand for this fruit is higher. Petri et al. (2016) also observed that marketing the fruits in the moment of highest demand may increase expressively the overall rentability; to achieve this, ethephon applications may be carried out to control/anticipate the ripening and harvesting process.

Figure 1 presents the correlation circle that shows the correlations between the application of ethephon and boric acid and their effect on the harvest.

Figure 1 – Correlation circle grouping the ethephon and boric acid concentrations with the productive parameters of the 2018/2019 harvest.



E – ethephon concentration; BA – boric acid concentration; H1 – first harvest; H2 – second harvest; H3 – third harvest; Y – overall yield. Source: Authors (2021).

According to Figure 1, it can be seen a positive relationship (angle between vectors $<90^{\circ}$) between ethephon concentration (E) and fruit yield in both the first (H1) and second (H2) collections; a negative relationship (angle between vectors $>90^{\circ}$) was also observed between ethephon concentration (E) and boric acid concentration (BA) and the yield in the third collection (H3). On the other hand, both E and BA presented a nearly orthogonal angle (90°) with the overall yield (Y),

indicating that the overall fruit yield is independent of both E and BA.

The productive parameters underwent Spearman correlation analysis to analyze possible significant relationships between the production parameters and the concentrations of growth regulators applied. The correlation coefficients are presented in Table 2.

 Table 2 – Spearman correlation coefficients of the productive parameters relative to ethephon and boric acid concentrations in the 2018/2019

 harvest.

Fruit yield	Ethephon concentration	Boric acid concentration
First harvest	0.7095	0.2174
Second harvest	0.5996	0.0934
Third harvest	-0.7639	-0.1690
Overall yield	0.2113	0.1180

The correlations that were significant at 5% probability are highlighted in bold. Source: Authors (2021).

The Spearman correlation analysis identified positive relationships between the ethephon concentration and the first and second collections, and a negative relationship between ethephon concentration and the third collection. No significant correlation was identified between ethephon concentration and the overall yield, neither between boric acid concentration nor any of the productive parameters.

Ethephon has the main effect of hastening the climacteric peak in climacteric fruits, which ends up increasing the number of ripe plums in the first harvest (Fioravanço et al., 2007a; 2007b). This is an important tool for the farmers since plums are harvested in several collections due to the staggered flowering, a common phenomenon in Brazil. Thus, it is possible to reduce manpower demand through a homogenization of fruit ripening.

Table 3 compiles the fruit quality parameters for the plums collected in the 2018/2019 harvest.

Treatment	Firmness (lbf·cm ⁻²)	Soluble solids (°Brix)	Average mass (g)	Titratable acidity (% m/v)	SS/TA ratio	Anthocyanins (mg·kg ⁻¹)	Phenolic compounds (mg· 100 g ⁻¹)
T1	8.60 a	11.55 bc	78.79 a	2.29 a	5.03 c	17.37 b	68.23 a
T2	5.77 b	11.15 cd	76.56 a	2.32 a	4.83 c	13.97 c	46.75 b
T3	5.30 b	11.13 d	79.01 a	1.87 bc	5.95 b	16.45 bc	48.88 b
T4	5.45 b	11.93 ab	77.60 a	1.90 b	6.30 b	18.50 b	53.54 b
T5	5.98 b	11.88 ab	77.85 a	1.72 bc	6.95 a	23.15 a	45.79 b
T6	8.04 a	12.08 a	66.50 a	1.69 c	7.18 a	18.39 b	65.89 a
$CV^{*}(\%)$	12.82	1.58	11.62	4.32	4.20	7.70	8.76

Table 3 – Quality parameters of Japanese plums treated with several doses of ethephon and boric acid in the 2018/2019 harvest.

Means in column followed by the same letter do not present statistical difference by Tukey's multiple range test at 5% probability ($\alpha = 0.05$). T1 – control; T2 – one application of ethephon (100 mg·L⁻¹) + boric acid (10 mg·L⁻¹); T3 – two applications of ethephon (100 mg·L⁻¹) + one application of boric acid (10 mg·L⁻¹); T4 – one application of ethephon (200 mg·L⁻¹) + boric acid (10 mg·L⁻¹); T5 – one application of ethephon (100 mg·L⁻¹); T6 – two applications of boric acid (10 mg·L⁻¹); * – coefficient of variation. Source: Authors (2021).

Relative to the fruit quality parameters, the average mass presented no statistical difference in any treatment, with an overall mean of 76.03 g. Bauchrowitz (2018), who studied the effect of ethephon in two plum genotypes, reported that the use of ethephon induced a reduction of average fruit mass. This probably occurs due to the quicker ripening, which may cause the fruit to synthesize and store water and sugars in smaller amounts than fruits that undergo a slower ripening (Macedo et al., 2016; Petri et al., 2016). However, this phenomenon was not observed in the present work.

Plums treated with two applications of boric acid (T6) have had an increase in pulp firmness and phenolic compounds content; this treatment was statistically similar to the control (T1). All other treatments (T2 to T5), in which all of them ethephon was applied, regardless of the presence of boric acid, induced lower pulp firmness and phenolic compounds content

when compared with the control, not differing statistically among themselves. Khorshidi and Davarynejad (2010), studying the effect of the application of ethephon 250 mg·L⁻¹ on cherries (*Prunus cerasifera*), reported no influence of ethephon on phenolic compounds content of the fruits.

Magrin (2016), who studied the effect of the pre-harvest application of boric acid on Pink Lady apples, reported no effect of boric acid on pulp firmness and soluble solids content. Pessoa (2016) highlighted that there is a natural reduction of pulp firmness as the fruit ripens, due to the degradation of carbohydrates, pectin, cellulose, and hemicellulose, which renders a softer pulp. The application of ethephon induces an increase in the synthesis of ethylene, accelerating the fruit decomposition rate, and reducing its storage capacity, which eventually affects pulp firmness, fruit average mass, and diameter (Bauchrowitz, 2018). Petri et al. (2016) cited that ethephon application in pre-harvest induces a decrease in pulp firmness in peaches, causing a decrease in the storage time, also inducing an early ripening of the tip of the fruit.

Soluble solids (SS) content was highest in the treatment that employed only boric acid (T6), not differing statistically from the treatments T4 and T5, which employed ethephon and boric acid in different concentrations and application times. The lowest SS content was observed in the T1 (control) and T2 (one application of ethephon 100 mg·L⁻¹ + boric acid 10 mg·L⁻¹), with 5.0 and 4.8 °Brix, respectively.

Bauchrowitz (2018) observed, for 'Reubennel' and 'Camila' plums, that the application of ethephon did not affect the soluble solids content of the fruits. Fioravanço et al. (2007) also reported similar results, citing that ethephon has not influenced the soluble solids content relative to the control. Fioravanço et al. (2007), also evaluating the effect of ethephon application on the quality of plums, observed SS values in the range of 10.1-12.1 °Brix, which were similar to the ones observed in the present work; however, the author reported the highest SS value in the control treatment, unlike the trend observed in this study.

Regarding the titratable acidity (TA), both the control (T1) and a sole application of ethephon (100 mg·L-1) and boric acid (10 mg·L⁻¹) - T2 - were statistically similar (2.29 and 2.30% m/v). The lowest TA value was observed in T6, two applications of only boric acid, with 1.69% m/v, but not differing from treatments T3 and T5 (1.87 and 1.72% m/v, respectively). These results were higher than the ones reported by Fioravanço et al. (2007), who reported a TA range of 1.31-1.42% m/v for plums treated with different doses of ethephon.

In a study with 'Galaxy' apples, Nunes (2016) evaluated the effect of crescent pre-harvest leaf applications of boric acid; the author reported that fruit TA was influenced by the number of sprayings. Relative to fruit ripening, it was reported that ripening was anticipated when four applications of boric acid were carried out in pre-harvest, which may be considered as an alternative to stagger the harvest.

The content of total anthocyanins was the highest in treatment T5 (one application of ethephon 100 mg·L⁻¹ + two applications of boric acid (10 mg·L⁻¹), with 23.15 mg·kg⁻¹. On the other hand, the treatment T2 (one application of ethephon 100 mg·L⁻¹ + boric acid 10 mg·L⁻¹) presented the lowest anthocyanin content (13.97 mg·kg⁻¹); the control (T1) presented an intermediate content, with 17.37 mg·kg⁻¹.

Anthocyanins are pigments found in plants, are generally hydrosoluble compounds that are responsible for fruit color, and may have different colorations, such as red, blue, pink, and purple, among others. These pigments are also called flavonoids, in which anthocyanins are in glycoside form, which may be hydrolyzed to sugars and aglycones. In plums, the main anthocyanins are cyanidin-3-glucoside, which has a reddish hue, and cyanidin-3-rutinoside (Redondo et al., 2021; Reque, 2012; Tomás-Barberán et al., 2001). According to Lee and Lee (1980), ethylene applications increase and accelerate anthocyanins and phenolic compounds accumulation, also stimulating sugar storage in Santa Rosa plums. Zhang et al. (2009) observed that ethephon application on Black Ambar plums fostered an increase in anthocyanins synthesis, whereas anthocyanin contents of Oishi Wase plums were similar to the control. The same authors explained that the increase in

anthocyanin content was due to a higher activity of the enzymes phenylalanine ammonia-lyase (PAL) and chalcone isomerase (CHI), both being steps of the flavonoid biosynthesis pathway.

Figure 2 presents the correlation circle that shows the relationships between the ethephon (E) and boric acid (BA) concentrations and the fruit quality parameters of the 2018/2019 harvest.

Figure 2 – Correlation circle grouping the ethephon and boric acid concentrations with the fruit quality parameters in the 2018/2019 harvest.



E – ethephon concentration; BA – boric acid concentration; F – firmness; SS – soluble solids; TA – titratable acidity; R – SS/TA ratio; AM – average mass; A – anthocyanins; P – phenolic compounds. Source: Authors (2021).

According to Figure 2, it was possible to observe that ethephon concentration (E) was positively related to the average fruit mass (AM); however, this relationship was weak, according to the length of the vector AM. On the other hand, firmness (F), phenolic compounds (P), titratable acidity (TA), and soluble solids contents (SS) were negatively related to E. Anthocyanin content (A), SS/TA ratio (R), and E presented no relation at all. The application of boric acid (BA) was positively related to R, A, and SS; it presented no relationship with AM and was negatively related to F, P, and TA.

Lee and Lee (1980b) highlighted that the phenolic compounds content in plums increased quickly in the stage of fruit coloring. However, in the present study, it was observed a reduction of phenolic compounds content in the plants treated with ethephon. On the other hand, the application of boric acid also had an inhibitory effect on phenolic compounds synthesis. Experiments with apples has shown that leaf application of boric acid some weeks before harvest increases the intensity of the red color in fruit peel, also hastening fruit ripening. However, it is unclear the exact role of boric acid in the ripening of climacteric fruits as apple and plum (Macedo et al., 2016; Ernani et al., 2010).

Table 4 presents the results of the Spearman correlation test between ethephon (E) and boric acid (BA) concentrations and the fruit quality parameters of the 2018/2019 harvest.

Parameter	Ethephon concentration	Boric acid concentration
Firmness	-0.7424	-0.2322
Soluble solids	-0.1404	0.2125
Titratable acidity	-0.2305	-0.8257
SS/TA ratio	0.1704	0.7149
Average mass	0.2075	-0.3007
Anthocyanins	0.2475	0.3502
Phenolic compounds	-0.6795	-0.3369

Table 4 – Spearman correlation coefficients of the quality parameters of Japanese plums at harvest relative to ethephon and boric acid concentrations in the 2018/2019 harvest.

The correlations that were significant at 5% probability are highlighted in bold. Source: Authors (2021).

By evaluating the Spearman correlation analysis, it was possible to observe that only fruit firmness and the content of phenolics compounds were influenced by ethephon concentration. These two parameters were negatively correlated with ethephon concentration, i.e., as the ethephon concentration increased, the content of phenolic compounds and fruit firmness decreased. Relative to the concentration of boric acid, the SS/TA ratio was positively correlated, whereas the titratable acidity was negatively correlated. In this sense, leaf application of boric acid may help reduce fruit acidity, which ends up increasing the SS/TA ratio value of the plums.

El-Kady et al. (2014), testing the application of 250, 500, 750, and 1,000 mg·L⁻¹ ethephon on Kelsey plums, reported that the use of ethephon reduced pulp firmness, the reduction was more pronounced at higher ethephon doses, presenting the same trend (negative correlation) as observed in Table 4 for the pair ethephon concentration/pulp firmness. The same authors also reported an increase in anthocyanin content and a reduction in A and B chlorophyll and carotenoid contents. The effect of ethephon on fruit ripening may explain the negative correlation between ethephon concentration and phenolic compounds content due to biochemical and metabolic changes precipitated by an earlier fruit ripening (Khorshidi and Davarynejad, 2010).

Relative to boric acid application, it had a significant and negative correlation only with titratable acidity (and SS/TA ratio). Wójcik (1998) commented that boron application may influence plum quality but reported that leaf applications of boron increased the soluble solids content of the fruits, not informing its influence on titratable acidity. Mahmoud et al. (2020) reported that the joint application of boron, moringa extract, and chelated calcium increased the SS/TA ratio of Hollywood plums. However, the authors did not comment if the increase was due to a reduction of titratable acidity or an increase in soluble solids content.

3.2. Experiment two

The second experiment was carried out in the 2019/2020 harvest; in this experiment, along with the application of ethephon and boric acid, a commercial growth stimulant was also tested, in two doses (6 and 8 L·ha⁻¹). The productive parameters for the 2019/2020 harvest are presented in Table 5.

Treatment	Harvest 1 (2019-12-20)	Harvest 2 (2019-12-25)	Harvest 3 (2020-01-05)	Overall yield
T1	0.00 b	9.75 ab	27.38 a	37.13 a
T2	0.00 b	7.31 b	30.25 a	37.56 a
Т3	0.00 b	10.88 ab	32.44 a	43.31 a
T4	28.13 a	15.63 ab	0.00 b	43.75 a
T5	0.00 b	10.50 ab	28.56 a	39.06 a
T6	25.69 a	17.38 a	0.00 b	42.96 a
CV* (%)	21.00	34.00	16.17	10.61

Table 5 – Fruit yields (kg·plant⁻¹) in the three harvests and overall yield for Japanese plums in the 2019/2020 harvest.

Means in column followed by the same letter do not present statistical difference by Tukey's multiple range test at 5% probability ($\alpha = 0.05$). T1 – control; T2 – Physiogrow[®] Color (6 L·ha⁻¹); T3 – Physiogrow[®] Color (8 L·ha⁻¹); T4 – ethephon (200 mg·L⁻¹); T5 – boric acid (30 mg·L⁻¹); T6 – ethephon (200 mg·L⁻¹) + boric acid (10 mg·L⁻¹); * – coefficient of variation. Source: Authors (2021).

Ethephon-treated plants had a five-day hastening of harvest, a behavior also observed in similar studies (Fioravanço et al, 2007a, 2007b). This result was observed since ethephon, when applied, is transformed into ethylene, which hastens the climacteric peak and stimulates ripening and senescence processes in climacteric fruits, as plums (Bisht, 2018; Choudhary et al., 2021; Rademacher, 2015). Moreover, ethephon application concentrated fruit harvest and reduced the need for carrying out a third collection, rendering its use economically feasible by the reduction of the manpower needed to carry out the harvest.

Figure 3 presents the correlation circle between the application of ethephon, boric acid, the commercial product Physiogrow® Color and their effect on the harvest parameters for the 2019/2020 harvest.

Figure 3 – Correlation circle grouping the ethephon and boric acid concentrations with the fruit quality parameters at harvest in the 2019/2020 harvest.



E – ethephon concentration; BA – boric acid concentration; PHY – Physiogrow[®] Color dose; H1 – first harvest; H2 – second harvest; H3 – third harvest; Y – overall yield. Source: Authors (2021).

According to the PCA (Figure 3), the ethephon concentration (E) was positively related to the first (H1) and second

(H2) collections of the harvest and also with the overall yield (Y) and was negatively related to the third collection (H3) of the harvest. Boric acid concentration (BA) was negatively related to Y and H3, unrelated to H2, and weakly related to H1. The dose of the commercial growth stimulant (PHY) was positively related to Y and H3, unrelated to H2, and negatively related to H1.

The Spearman correlation coefficients between the ethephon concentration, boric acid concentration, and Physiogrow[®] Color dose and the harvest parameters are presented in Table 6.

Table 6 – Spearman correlation coefficients of the productive parameters relative to ethephon and boric acid concentrations and doses of a commercial growth stimulant in the 2019/2020 harvest.

Fruit yield	Ethephon concentration	Boric acid concentration	Physiogrow [®] Color dose
First harvest	0.9737	0.1978	-0.4770
Second harvest	0.6129	0.3575	-0.3670
Third harvest	-0.8326	-0.2472	0.6288
Overall yield	0.3960	0.0766	0.0522

The correlations that were significant at 5% probability are highlighted in bold. Source: Authors (2021).

By verifying the Spearman correlation analysis (Table 6), it was possible to observe that no significant correlation existed between boric acid concentration and the productive parameters. On the other hand, the ethephon concentration was significantly correlated with all productive parameters, with exception of the overall yield. Ethephon concentration and the fruit yield in both the first and second collections were positively correlated, whereas the correlation with the yield in the third collection was negative. Relative to the dose of the commercial growth stimulant, it presented a negative correlation with the fruit yield in the first collection and a positive correlation with the yield in the third collection; no significant correlation was observed for both the overall yield and the yield in the second collection.

Table 7 compiles the fruit quality parameters of the plums collected in the 2019/2020 harvest.

Table 7 – Quality parameters of Fortune Japanese plums treated with several doses of ethephon and boric acid and doses of a commercial growth stimulant in the 2019/2020 harvest.

Treatment	Firmness (lbf·cm ⁻²)	Soluble solids (°Brix)	Average mass (g)	Titratable acidity (% m/v)	SS/TA ratio	Anthocyanins (mg·kg ⁻¹)	Phenolic compounds (mg·100 g ⁻¹)
T1	3.18 a	10.85 ab	99.69 a	2.14 bc	5.09 a	34.68 b	60.03 ab
T2	3.14 a	10.75 ab	99.07 a	2.51 a	4.29 b	9.99 d	51.74 b
T3	2.31 b	10.95 ab	84.03 b	2.15 bc	5.12 a	47.34 a	68.35 a
T4	3.29 a	10.55 b	78.16 b	2.08 c	5.09 a	26.15 c	51.57 b
T5	3.23 a	11.40 a	101.71 a	2.22 abc	5.15 a	22.10 c	53.05 b
T6	2.83 ab	10.85 ab	75.08 b	2.39 ab	4.55 ab	18.86 c	57.65 b
CV* (%)	7.82	2.92	4.52	6.15	6.21	12.79	8.16

Means in column followed by the same letter do not present statistical difference by Tukey's multiple range test at 5% probability ($\alpha = 0.05$). T1 – control; T2 – Physiogrow color[®] (6 L·ha⁻¹); T3 – Physiogrow color[®] (8 L·ha⁻¹); T4 – ethephon (200 mg·L⁻¹); T5 – boric acid (30 mg·L⁻¹); T6 – ethephon (200 mg·L⁻¹) + boric acid (10 mg·L⁻¹); * – coefficient of variation. Source: Authors (2021).

The application of Physiogrow[®] Color (8 $L \cdot ha^{-1}$) – T3 – and ethephon 200 mg·L⁻¹ + boric acid 10 mg·L⁻¹ – T6 – increased soluble solids content by 5% relative to the control (T1), whereas the average fruit mass was reduced by the application of Physiogrow[®] Color (8 $L \cdot ha^{-1}$), ethephon (200 mg·L⁻¹), and ethephon (200 mg·L⁻¹) + boric acid (10 mg·L⁻¹). The increase in soluble solids concentration in the plums may be attributed to a higher ethylene production, which in contrast increases starch hydrolysis, respiration rate, and soluble solids, as well as the increase in fruit yellowing, hastening ripening, which may or may not reduce the pulp firmness of the fruits (Choudhary et al., 2021).

The reduction of average fruit mass in the treatments containing ethephon may be explained by harvest hastening, the same effect may also be observed in other fruit, such as apples (Petri et al., 2016).

Titratable acidity was increased with the application of Physiogrow[®] Color (6 L·ha⁻¹) and ethephon (200 mg·L⁻¹) + boric acid (10 mg·L⁻¹), when compared with the control.The application of Physiogrow[®] Color (8 L·ha⁻¹) increased anthocyanins content by 36.5% and phenolic compounds content by 13.8%. The other treatments reduced total anthocyanins content in the peel of Fortune plums.

Figure 4 presents the correlation circle that shows the relationships between the ethephon (E), boric acid (BA), and Physiogrow[®] Color (PHY) concentrations and the fruit quality parameters of the 2019/2020 harvest.

Figure 4 – Correlation circle grouping the ethephon and boric acid concentrations and the doses of a commercial growth stimulant with the fruit quality parameters at harvest of the 2019/2020 harvest.



E – ethephon concentration; BA – boric acid concentration; PHY – Physiogrow[®] dose; F – firmness; SS – soluble solids; TA – titratable acidity; R – SS/TA ratio; AM – average mass; A – anthocyanins; P – phenolic compounds. Source: Authors (2021).

By analyzing Figure 4, it was possible to observe that E was negatively related to fruit quality parameters, and presented no relationship at all with F, R, and A. Regarding BA, there was a positive relationship with F, negative relationships with the other parameters, and no relation with SS and AM. The treatments PHY were positively related with SS, P, R, A, and AM, negatively related with F, and presented no relation at all with TA.

The Spearmen correlation coefficients between the treatments and the quality parameters of the fruits are presented in Table 8.

Parameter	Ethephon concentration	Boric acid concentration	Physiogrow [®] Color dose
Firmness	-0.0256	-0.0383	-0.4858
Soluble solids	-0.2734	0.4948	-0.1021
Titratable acidity	-0.7661	-0.0383	0.0229
SS/TA ratio	-0.0769	0.2450	0.1717
Average mass	-0.1149	-0.0128	-0.1595
Anthocyanins	-0.2171	-0.3320	0.1668
Phenolic compounds	-0.2043	-0.1532	0.3670

Table 8 – Spearman correlation coefficients of the quality parameters of Japanese plums at harvest relative to ethephon and boric acidconcentrations and the dose of a commercial growth stimulant in the 2019/2020 harvest.

The correlations that were significant at 5% probability are highlighted in bold. Source: Authors (2021).

According to Table 8, ethephon concentration was negatively correlated with titratable acidity, very probably due to the hastening effect on ripening, as observed by El-Kady et al. (2014). The application of boric acid was positively correlated with soluble solids content. The same behavior was observed by Wójcik (1998) for *P. domestica*. It was also possible to observe a negative correlation between the dose of the commercial product Physiogrow[®] Color and fruit firmness, probably due to an enhancement of fruit ripening caused by this product. The product Physiogrow[®] Color is a liquid leaf organic fertilizer based on free L-aminoacids and organic acids with the potential to be used to enhance fruit peel color in apples and other fruit crops since it is composed of aminoacids which are precursors in the biochemical pathways of ethylene and anthocyanins, chlorophyllase regulators, and monosaccharides.

4. Conclusion

The application of ethephon hastened and homogenized partially the ripening of Fortune Japanese plums, which implies the reduction of operational costs relative to manpower. Relative to fruit quality parameters, pre-harvest ethephon application reduced fruit firmness and phenolic compounds and total anthocyanin contents, not influencing the other parameters. On the other hand, the application of boric acid increased fruit firmness but had no effect on the other quality parameters. Thus, from an economic standpoint, the pre-harvest application of ethephon may be useful in reducing the harvest costs by reducing harvest cycles and, with it, manpower requirements.

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