Cobalt associated with potassium phosphite improves the post-harvest quality of mango cv. Palmer

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Abstract

Mango is a highly perishable climacteric fruit with a short post-harvest period. Therefore, to extend shelf life, the mangoes were treated pre-harvest with a potassium phosphite solution (1.4 g/mL KH₂PO₄) and a control solution (0 g/mL KHIPO) associated with different doses of cobalt (0; 48; 64; 80; 112 ml.plant⁻¹) at a density of 1.30 g/mL. The work measured physical, chemical, and biochemical variables. Cobalt and potassium phosphite favored maintaining color and firmness and reduced the respiratory rate. The treatments significantly affected the chemical variables, reduced the soluble solids content, conserved ascorbic acid, and preserved the pH. Foliar application of cobalt and potassium phosphite at pre-harvest is an efficient technique for improving shelf life and maintaining mango quality during storage at room temperature.

Keywords: *Mangifera indica,* breathing, quality, post-harvest diseases

Introduction

Mango is a tropical fruit with high worldwide acceptance (Madhavan et al., 2019). However, due to its climacteric nature, it is highly perishable and exhibits high metabolic activity after harvest, occurring within 3-4 days (Rastegar et al., 2019). Its metabolic activity causes biochemical, physiological, and chemical responses, degradation, and biosynthesis that occur concomitantly, resulting in fruit ripening. (Michelle et al., 2017).

Fruit ripening is an inevitable and irreversible physiological process during which the general quality of the fruit changes rapidly. In just a few days, the quality reaches the ideal level for consumption and subsequently declines until the fruit becomes inedible due to the senescence process (Nordey et al., 2016). Senescence is associated with the change in gas composition caused by increased respiratory and ethylene production (Jahurul et al., 2015). Increased ethylene production

initiates a series of events leading to many interactive signaling and metabolic pathways, which are responsible for the progress of ripening in climacteric fruits.

Ethylene synthesis is directly regulated by cobalt and heavy metals, such as silver ions (Ag+) (Abdelsamad et al., 2019). Cobalt regulates the enzyme aminocyclopropanecarboxylate oxidase (ACC oxidase), which inhibits the conversion of 1-aminocyclopropane-1-carboxylic acid (ACC) into ethylene and reduces its levels inside the plant cell. It has been used to delay leaf senescence by preventing the degradation of chlorophyll and reducing oxidative stress (Wi et al., 2012; Yu & Yang, 1979). Another alternative approach is using potassium phosphite as a quick source of potassium. This substance fills the fruits and increases production. Potassium phosphite can also help reduce post-harvest diseases. This product can act directly on the pathogen or act indirectly, activating the host plant's defense

mechanisms, which involve the synthesis of phytoalexins, phenolic compounds, and PR proteins (pathogenesisrelated proteins) (Spolti et al., 2015).

Several studies have demonstrated that field application of cobalt has beneficial effects during postharvest processes in many types of fruits and vegetables. However, to date, no study has yet described the benefits and implications of increasing cobalt doses and the synergistic effect of the association with phosphite in reducing ethylene synthesis and reducing damage caused by post-harvest pathogens. Considering the great importance of mango in the commercial agricultural scenario and its perishability, the work evaluated the effect of different cobalt doses, in association with potassium phosphite, on the metabolism of pigments and the quality of 'Palmer' mango fruits under field conditions.

Material and methods

Plant material and treatments

The experiment was performed in a commercial 'Palmer' mango orchard in the municipality of Matias Cardoso (15°6'11.38"S 43°48'41.66"W), Minas Gerais, Brazil, in 2019. The plants, aged 6 and 7 years, and were spaced 6.6 m x 4.4 meters (417 plants ha⁻¹) from each other. Irrigation was performed per micro aspersion, under conventional handling, with a high technical level production structure. Fertilization was performed after using soil and foliar analyses, which were carried out regularly. Pest, disease, and weed control were performed by integrating various cultural, chemical, physical, and biological practices.

The experiment consisted of 48 useful plants with internal and external borders . It was conducted in a randomized block design, with four blocks in a 2x6 factorial scheme (with and without potassium phosphite x foliar doses of cobalt). Each block was composed of 12 plants. Each plant represented a repetition.

The treatments consisted of the use of potassium phosphite (1.4 g/ mL KH $_{2}$ PO $_{4}$) and a control solution (0 g/ mL KH $_{\rm 2}$ O $_{\rm 4}$) associated with different doses of a commercial product (1.3 g/ml – Cobalt in doses 0; 48; 64; 112 ml -1), applied via foliar spray. The first application occurred when the fruit reached stage 1 of maturation, where the pulp was cream-colored, according to the classification of Assis & de LIMA (2008); the other two applications occurred with an interval of seven days between each application. The fruits were harvested at the physiological maturity stage. Fruits were selected according to size, color, shape, and damage or fungal infection absence for standardization. The harvested fruits were taken to the laboratory and washed in water and 0.2% neutral

detergent for latex coagulation and surface cleaning. Subsequently, the fruits were immersed in a fungicide Imazalil suspension at a dose of 2mL.1000mL-1 of water at room temperature and dried in the open air. Three fruits from each treatment were placed in polyvinyl chloride (PVC) trays and then stored at 20 °C. Assessments were carried out on the experiment setting day and, subsequently, on the 5th and 10th day of storage.

Determination of physical attributes

Fruit firmness was determined by maximum force, measurements were taken in two equidistant regions on the opposite sides of the fruit, and the results were expressed in Newton (N). Peel and pulp colors were evaluated using a digital colorimeter (Color Flex model 45/0-2200) to determine values of three parameters: luminosity (L*), Hue angle (°h), and chroma saturation index (C*), calculated from the a* and b* values. Fresh mass loss was calculated by relating the weight of each sample before and during each evaluation period and expressed as a percentage of fresh matter weight loss to the initial weight of the fruit.

Biochemical and chemical variables determination

The pigment content was measured using 0.1g of pulp via solvent extraction (acetone 80%), as per Macedo et al. (2013). The plant tissue was placed in tubes containing 10 ml of 80% acetone, then covered with aluminum foil and kept in the dark for 72 hours. After this time, the liquid was spectrophotometer-tested at wavelengths of 470, 645, and 663 nm for carotenoids and chlorophylls a and b, respectively. The results were expressed in milligrams of pigment per gram of fresh pulp mass (mg g⁻¹). Vitamin C levels were determined by titration, using Tillmans reagent (2.6 DFINa) and expressed in mg of ascorbic acid per 100 ml of juice (IAL, 2008). Respiration was assessed by titration, placing fruits and fixative solution (0.5N NaOH) in a hermetically closed package for 12 hours, adding two drops of phenolphthalein and 10mL of 0.2N $\mathtt{BaCl}_{2'}$, and titrating with 0.1N hydrochloric acid. The results were expressed as mg Cokg⁻¹.h⁻¹ on each evaluation day.

The soluble solids content was determined by a digital refractometer (HI96801 Hanna) calibrated with distilled water. The mango pulp was crushed in a food processor. An aliquot was removed for direct reading in the refractometer, expressing the result in °Brix. The titratable acidity (TA) was determined using the analyte (10 g of homogenized pulp and diluted to 100 ml), titrated with a standardized solution of sodium hydroxide (NaOH) at 0.1 N, using phenolphthalein as the indicator. The results were expressed in grams of citric acid per 100g of

fresh pulp. Pulp pH was measured according to LUTZ–IAL (2008). The maturation index (RATIO) was calculated by examining the relationship between soluble solid content and titratable acidity.

Pathological variables determination and species identification.

Anthracnose intensity evaluation in the fruits was assessed, considering the incidence and severity of the disease for 15 days, with an interval of three days between assessments. The incidence was obtained by the number of fruits affected per repetition, with these values expressed as a percentage per treatment.

Severity refers to the proportion of colonized tissue area. It was determined by a specific diagrammatic scale for anthracnose in mangoes (Corkidi *et al.*, 2006), with disease severity ranging from 0 - 1% (no disease); 1 - 5% (mild disease); 6 - 9% (moderate disease); 10 - 49% (severe disease), and 50- 100% (very severe disease) of injured area/fruit. The results allowed us to calculate the area under the incidence progress curve (AACPI) and the area under the severity progress curve (AACPS) (Sharner & Finney, 1977)

The results were used to calculate the area under the incidence progress curve (AACPI) and the area under the severity progress curve (AACPS) (Sharner and Finney, 1977), according to the formula:

$$
AACPD = \sum_{1}^{n-1} [(xi + xi + 1) / 2 * (ti + 1 - ti)] i)
$$

Where n is the number of evaluations, x is the proportion of the disease, and $(t^i+1 - ti)$ is the interval between assessments.

To identify the species of *Colletotrichum*, pieces of fruit tissue measuring 0.5 mm in diameter were removed from lesions in the transition region between injured and healthy areas. These fragments were superficially disinfected with 70% alcohol for 30 seconds and 1.5% sodium hypochlorite for 3 minutes and rinsed three times in sterilized distilled water. Then, the tissue fragments were transferred to Petri dishes containing water agar (AA) medium and incubated at $25 \degree$ C for 48 hours in a BODtype chamber. After this period, the fragments of plant tissue that showed fungal growth, the hyphal tip of the fungus, were placed in a Petri dish containing potatodextrose-agar (BDA) medium. After the fungus covered the entire surface of the medium, discs measuring 0.5 mm in diameter were removed from the edges of the colonies and transferred to tubes containing PDA medium. Subsequently, the isolates obtained from mango were

identified through analysis of partial gene sequences: APN2-MAT1-2; APN2, and GAPDH-IG, at the Mycology Laboratory of the Federal Rural University of Pernambuco.

Production and Productivity

The production was assessed by weighing all the fruits of each plant subjected to treatments with the aid of a digital scale. Productivity was subsequently estimated. ha $^{-1}$.

Statistical analyses

The variable data were tested for homogeneity of variance analysis by Burr & Foster, normality of residuals by Shapiro & Wilk, and non-additivity of the Tukey model. The variables titratable acidity, chlorophyll a, and carotenoids did not meet the homogeneity and normality requirements and were transformed into $\sqrt{(x+1)}$. The results were subjected to analysis of variance (ANOVA), considering the sources of variation, cobalt doses, use of potassium phosphite, and days of storage at 5% probability. The characteristics were studied using multiple linear regression analysis for the triple interaction (Phosphite x Cobalt dose x Storage days and double (Cobalt dose x Storage days), whose statistical model selection procedure was based on the following criteria: significant F test for regression (p < 0.05); non-significant F test for lack of adjustment (p > 0.05); significant Student's T-test for regression coefficients (p < 0.05) and coefficient of adjusted determination (R2aj). The program used to carry out the statistical analyses was Genes . The analysis used the linear and quadratic statistical models, respecting the biological response. Later, the analyses considered the Person correction using the Corrplot pack of the R-Studio Software. The *Principal Component Analysis – PCA* was to reduce the dimensionality of the data set and identify the variables explaining the most significant proportions in the total variance and was performed using the statistical program Minitab ® (Minitab Inc., Philadelphia).

Results and Discussion

Association of cobalt and potassium phosphite in the physical attributes of mango

The fruit's firmness is one of the main variables determining the mango's quality and shelf life. It indicates resistance and directly impacts exportation (Aquino et al., 2015). Cobalt significantly reduced the firmness loss during storage; the increase in cobalt concentration restricted the firmness loss rates, especially at the 112 mL dose. Regardless of whether the use of potassium phosphite (FK) or not, all treatments with cobalt (0, 48, 64, 80, 96, 112 ml) displayed a firmness reduction during storage (**Figure**

1). However, firmness loss was lower in the treatments including potassium phosphite (5%), compared to fruits without added phosphite.

Figure. 1 – Average firmness values (N) depending on the different concentrations of Co applied pre-harvest (0, 48, 64, 80, 96, and 112 ml) and use of phosphite (without phosphite (A) and with phosphite (B) under different post-harvest times, 0, 5 and 10 days after harvest, cultivated in 2019.

Fresh mass loss is closely linked to respiration, fruit transpiration, and water and solutes loss. It represents a main factor directly associated with the quality of mango fruit (Chitarra & Chitarra, 2005). Cobalt significantly reduced the mango fresh mass loss during the storage period, regardless of using potassium phosphite compared to the control (fruits without phosphite) (**Figure 2**). Fruits treated with cobalt alone showed a reduction of 0.78% to 1.82% for doses of 48 and 112 ml, respectively; for fruits with phosphite, the reduction was lower when compared to fruits that received exclusively the cobalt treatment.

different concentrations of Co applied pre-harvest (0, 48, 64, 80, 96, and 112 ml) and use of phosphite (without phosphite (A) and with phosphite (B)) under different post-harvest times, 0, 5 and 10 days after harvest, cultivated in 2019.

Skin color variables are crucial for post-harvest, as they are the first variables consumers evaluate at purchase (Kour, 2018). The application of potassium phosphite delayed the fruits' color change, as observed in luminosity (*L*), chromaticity (*C*), and Hue angle (*H*) (**Tables 1, 2,** and **3**). The control treatment (without

test at a 5% probability level (P<0.05) and capital letters in the row do not differ from each other by the Tukey test at a 5% probability level (P <0.05).

Table 2 - Average hue angle values for 'Palmer' mango, depending on the use of potassium phosphite and storage days (February/2019 to June/2019).

Potassium Phosphite	Storage Days			
	$\left(\right)$	\mathcal{L}	10	
With	68.93 aA	71.68 BC	68.55 BC	
Without	74.81 to A	79.85 BC	55.36 bB	
Means followed by the same lowercase letter in the column do not differ from each other by the F				

test at a 5% probability level (P<0.05) and capital letters in the row do not differ from each other by the Tukey test at a 5% probability level (P <0.05).

Table 3 - Average chromaticity values of 'Palmer' mango peel, depending on the use of potassium phosphite and storage days (February/2018 to June/2018)

Potassium Phosphite	Storage Days			
			10	
Without	16.20 BC	$16.21q$ B	29.08 aA	
With	17.78 BC	17.86 BC	23.19 _{bA}	
Means followed by the same lowercase letter in the column do not differ from each other by the F				
test at a 5% probability level (P<0.05) and capital letters in the row do not differ from each other by				
the Tukey test at a 5% probability level (P <0.05).				

phosphite) presented on the last day of storage, *L* = 39.24, *C* = 29.08, *H* =55.36. The fruits had a reddish color as a result of ripening. In turn, fruits treated with phosphite showed prolongation of these changes, as indicated by the lower reduction in luminosity (44.41), lower chromaticity (23.19), and higher Hue angle value (68.55).

Pulp color variables are the most apparent indicators of ripeness (Thongkum et al., 2018). Treatments with different doses of cobalt provided more outstanding luminosity preservation (**Figure 3**). The control treatment showed a more significant loss of luminosity, reaching 76.26 after ten days of storage. Cobalt-treated fruits displayed higher luminosity values: 77.0, 77.25, 77.5, 77.74, and 78.0 for doses of 48, 64, 80, 96, and 112 ml in treatments without potassium phosphite application. The association of potassium phosphite and cobalt increased the luminosity by 3.16% on the third day of storage. The pulp chromaticity parameter associates with the intensity of the pigments, which define the color. The application of cobalt reduced the chromaticity values (**Figure 4**). Chromaticity reduction comparing the control with the 112 ml dose was 4.80% for fruits without potassium phosphite and 8.05% for fruits with potassium phosphite on the 10th day of storage.

The days of storage and the cobalt doses influenced the pulp °Hue (**Figure 5**) (p<0.05). The treatments presented higher values on the second day of storage, with values of 86.0, 87.11, 87.48, 87.85, 88.22, and 88.59, representing tones close to yellow, decreased to 77.15, 78.25, 78.62, 78.99, 79.36, and 79.73, for cobalt doses of 0, 48, 64, 80, 96, and 112 ml, respectively, tones close to reddish-orange, which represent the pulp ripening.

Cobalt and potassium phosphite influence on the biochemical and chemical attributes.

Ripening is a complex process that promotes transformations in color, flavor, aroma, and texture, resulting in the edible state of the fruits (Tang et al., 2020).

Figure 3 – Average luminosity values depending on the different concentrations of Co applied in pre-harvest (0, 48, 64, 80, 96, and 112 ml) and use of phosphite (without phosphite (A) and with phosphite (B)) under different post-harvest times, 0, 5 and 10 days after harvest, cultivated in 2019.

Figure 4 – Average chromaticity values depending on the different concentrations of Co applied pre-harvest (0, 48, 64, 80, 96, and 112 ml) and use of phosphite (without phosphite (A) and with phosphite (B)) under different post-harvest times: 0, 5, and 10 days after harvest, cultivated in 2019.

Figure 5 – Average chromaticity values depending on the different concentrations of Co applied pre-harvest (0, 48, 64, 80, 96, and 112 ml) under different post-harvest times, 0, 5, and 10 days after harvest, cultivated in 2019.

In the experiment , potassium phosphite and cobalt significantly influenced the content of soluble solids (SS). The reduction in soluble solids content (**Figure 6**) was directly proportional to the increase in cobalt doses, reaching, at the end of storage, a decrease of 8.90%, 11.87%, 14.84%, 17.81%, and 20.78% for doses of 48, 64, 80, 96, and 112 ml of cobalt compared to control treatments with potassium phosphite. Treatments without potassium phosphite presented higher soluble solids concentrations than fruits treated with potassium phosphite.

The ascorbic acid is essential for fruit quality, affecting nutritional value (Sogi et al., 2012). The control treatment showed a reduction (15.06%) in the ascorbic acid content, while, in the highest dose of cobalt, this reduction was limited to 13.97% (**Figure 7**).

The fruit ripening process is often characterized by chlorophyll degradation and carotenoid biosynthesis (Wisutiamonkul et al., 2015). Applying potassium phosphite delayed the degradation of chlorophyll a and b (**Figure 8**) throughout the storage period at all cobalt doses. Cobalt delayed chlorophyll degradation by 0.00198 mg.g-1 for each ml of cobalt in the treatments that did

Figure 6 – Average values of soluble solids depending on the different concentrations of Co applied in pre-harvest (0, 48, 64, 80, 96, and 112 ml) and use of phosphite (without phosphite (A) and with phosphite (B)) under different post-harvest times at 0, 5, and 10 days after harvest, cultivated in 2019.

Figure 7 – Average ascorbic acid values (vitamin C) depending on the different concentrations of Co applied pre-harvest (0, 48, 64,80, 96, and 112 ml) under different post-harvest times at 0, 5, and 10 days after harvest, grown in 2019.

not use potassium sulfite. The treatments with potassium phosphite displayed a reduction of 0.00289 mg.g-1 (**Figure 8A** and 8B).

 The control treatment without potassium phosphite showed greater chlorophyll B degradation, reaching 53.05% after ten days of storage. Fruits treated with cobalt showed lower degradation: 49.33%, 48.21%, 47.12%, 46.09%, and 45.10% for 48, 64,80, 96, and 112 ml of cobalt. Fruits without potassium phosphite showed an increase until the fifth day of storage with a subsequent reduction (Figures 8C and 8D).

The carotenoid content acted in an opposite form compared to chlorophylls (Figures 8E and 8F).

Figure 8 – Average values of chlorophyll A - without phosphite (A) and with phosphite (B) chlorophyll B - without phosphite (C) and with phosphite (D) carotenoid without phosphite (E) and with phosphite (F) depending on the different concentrations of Co applied pre-harvest (0, 48, 64,80, 96, and 112 ml) and use of phosphite (without phosphite (A) and with phosphite (B)) under different post-harvest times, 0.5 and 10 days after harvest, grown in 2019.

Cobalt delayed carotenoid biosynthesis, and this effect was enhanced using potassium phosphite (IR). Regardless of the treatment, the fruits considerably increased the respiratory rate, as the mango is a climacteric fruit (Michelle et al., 2017) . Potassium phosphite significantly reduced (P≤0.05) the climacteric peak by 33% for all treatments, except for the 112 ml dose, where there was no difference between fruits with and without potassium phosphite treatment (**Table 4**).

The dose of 50.5 provided a lower respiration rate in fruits without potassium phosphite $(8.95 \text{ mg Co Kg.h}^{-1})$. Applying potassium phosphite, the dose that provided the lowest respiratory rate was 38.5 (5.96 mg Co Kg.h⁻¹).

The pH variable interacted significantly with the factors evaluated. During storage, there was an increase in pH, but the higher the cobalt dose , the lower this increase. Fruits treated with cobalt alone (**Figure 9**A) showed slight variation among treatments, and all treatments increased during the storage period. Fruits treated with cobalt and potassium phosphite (Figure 9B) displayed initial pH values between 3.24 and 3.46 at 0 and 112 ml, respectively.

 During storage, the pH values reached 4.35 and 4.12 for doses 0 and 112 ml. Acidity decreased during the mango ripening. Acidity decrease occurs due to the reduction of citric acid content, which is this fruit's most abundant organic acid (Nogueira et al., 2015). The highest titratable acidity value was observed on the fifth day of storage (Figure 9D); from this day onwards, there was an 8% reduction in all doses with potassium phosphite until the tenth day of storage. Fruits treated only with cobalt displayed this peak on the second day of storage (Figure 9C), with a subsequent reduction.

The SST/AT ratio (Ratio) is the variable used to evaluate flavor, being more representative than the isolated measurement of soluble solids or acidity (Chitarra & Chitarra, 2005). Cobalt and potassium phosphite significantly affected the ratio (Figure 9E and Figure 9F). The variable increased over the days of storage, reaching values of 66.98 to 63.94 on the 10th day, referring to doses 0 and 112, respectively. However, using potassium phosphite reduced the ratio by 10.63% and 26.93% for the control and the highest dose of cobalt used in this study.

Influence of cobalt and potassium phosphite treatments on pathological variables and species identification.

Anthracnose is a disease that causes significant damage to mangoes (Mangifera indica L.), and its control is crucial to preserve the fruit's quality. Lima et al. (2007) believed that the species that caused the disease in mangoes was *Colletotrichum gloeosporioides.* However,

Sem Fosfito

Com Fosfito

phosphite (B), titratable acidity - without phosphite (C) and with phosphite (D) and ratio - without phosphite (E) and with phosphite (F) as a function of different concentrations of Co applied pre-harvest (0, 48, 64,80, 96 and 112 ml) and use of phosphite (without phosphite (A) and with phosphite (B)) under different post-harvest times, 0, 5, and 10 days after harvest, grown in 2019.

when identifying, they found *C. siamense and C.tropicale*. The area below the incidence progress curve (AACPI) and the area below the severity progress curve (AACPS) showed a significant difference only when potassium phosphite was used. Fruits not treated with potassium phosphite (Control)displayed higher AACPS values of anthracnose caused by *C. siamense* and *C. tropicale*, having an average value of 58.67. On the other hand,

fruits treated with potassium phosphite reduced this value to 26.52 (45%). The incidence shows the same behavior, varying from 7.98 to 4.25, representing a reduction of 53.28% for fruits treated with potassium phosphite.

Influence of cobalt and potassium phosphite on production and productivity

Potassium phosphite increased production and productivity by 38.87% and 27.98%, respectively, in all doses of cobalt used. For fruits with potassium phosphite, the addition of cobalt was beneficial up to a dose of 64 ml (p<0.05), highering both production and productivity; fruits without potassium phosphite did not show a significant equation (p>0.05).

Correlation analysis of all measured characteristics

Pearson correlation analysis determined the association between the characteristics measured in mangoes treated with increasing cobalt concentration with and without the addition of potassium phosphite (**Figure 10**A and 10B). Figure 10 displays the Person's correlation matrix. Among the significant correlations, our results showed strong negative correlations between the firmness and chemical variables, such as soluble solids and ratio. Furthermore, soluble solids correlated positively and significantly with fresh mass loss and respiration. Conversely, the variable correlated negatively with the luminosity and Hue angle of the pulp. In contrast, we found that chlorophyll A correlated negatively with both C and chlorophyll B levels.

Principal component analysis

This study used all evaluated variables to perform principal components analysis (PCA) (**Figure 11**). The PCA results show the evident influence of chemical variables (Soluble Solids and Ratio) and respiratory rate (Respiration) on mango ripening (Fig. 11A and 11B). The first component (PC1) explained 56.09% of the variation, while the second component (PC2) explained only 20.05% for fruits not treated with potassium phosphite. Fruits with potassium phosphite (IR) showed 48.7% PC1 and only 19% variation for PC2. PC1 variation explained the results best (Fig. 11A and 11B).

Component analysis separated three groups,

Figure 10 – Correlation matrix according to Person's metrics derived from the physical, chemical, and biochemical variables of fruits treated without phosphite **(A)** and with phosphite **(B)** cultivated in 2019, under different concentrations of Co applied pre-harvest (0, 48, 64,80, 96 and 112 ml), and evaluated under three post-harvest times (0, 5, and 10 days after harvest). Blue and red circles represent positive and negative correlations under significance levels *P 0.05, **P 0.01, and ***P 0.001. Abbreviations: Chll *'* a *':* Chlorophyll 'a', Chll 'b': Chlorophyll 'b,' Chll T: Total chlorophyll, Carot: Total carotenoids, Resp: Respiration, C_p: Pulp Chorma, PMF: Fresh mass loss, C_c: Chroma of the peel, ATT: Titratable acidity, VIT: Vitamin C, L_c: Luminosity of the peel, L_p: Luminosity of the pulp, H_c: Hue angle of the peel, H_p: Angle of the pulp, FF: Fruit Firmness.

Fig.11 Principal component analysis of scorplot without (A) and with phosphite (B) derived from data under different Co concentrations applied at pre-harvest (0, 48, 64,80, 96, and 112 ml), (grayscale), under different post-harvest times, 0 (frame), 5 (circle) and 10 (diamond) days after harvest, cultivated in 2019. The larger circles represent the three clusters generated by the Euclidean distance at a similarity degree of 70%. Abbreviations: PC1: Main component1, PC2: Main component, Chll *'* a *':* Chlorophyll 'a', Chll 'b': Chlorophyll 'b,' Chll T: Total chlorophyll, Carot: Total carotenoids, Resp: Respiration, C_p: Pulp chroma, PMF: Fresh mass loss, C_c: Chroma of the peel, ATT: Titratable acidity, VIT: Vitamin C, L_c: Luminosity of the peel, L_p: Luminosity of the pulp, H_c: Hue angle of the peel, H_p: Angle of the pulp, FF: Fruit Firmness

also confirmed by the Euclidean distance. Group I included all treatments on day 0, group II represented the treatments on the fifth day of storage, and group III represented all treatments on the tenth day. The analysis of variables that contributed to the groups' separation highlighted that day 0 of storage differed in the greater firmness of the fruits, luminosity, and Hue angle of the pulp, presenting higher values when the fruits were green. On the last day of storage, the fruits had a high soluble solids content, ratio, high respiratory rate, loss of fresh mass, and an increase in the skin and pulp chromaticity, displaying the fruits' ripening. Storage days were the variable that mostly influenced fruit ripening.

Cobalt has been used post-harvest to extend the shelf life of different fruits, such as sweet oranges (Hussain & Singh, 2015), apples (Mostafavi et al., 2012), pineapple (Jenjob et al., 2017), mango (Wahdan, 2011), and Tahiti lemons. The results showed that cobalt can effectively delay ripening, forming a stable compound with the sulfhydryl group of the enzyme ACC oxidase, blocking the conversion of 1-aminocyclopropane-1-carboxylic acid (ACC) into ethylene. This process accumulates ACC, thus inhibiting ethylene biosynthesis (Lau & Yang, 1976; Yang & Hoffman, 1984; Yu & Yang, 1979) and fruit ripening, hence maintaining post-harvest quality of mango.

The main features of climacteric fruit ripening are cell wall softening, increased soluble solids content, color changes, and increased respiratory rate (Wahdan, 2011). The results highlighted that cobalt delays the ripening of the 'Palmer' mango, as evidenced by the maintenance of firmness, decreased fresh mass loss, reduced chlorophyll content degradation, and respiratory rate control. The joint use of cobalt and potassium phosphite (IR) strongly inhibited chemical variables, such as soluble solids and titratable acidity. Potassium phosphite may have a positive effect on ripening control. These effects may be due to the collapse delay and the extinction of pre-formed antifungal compounds present in immature fruits, besides the induction of the biosynthesis of the compounds such as phytoalexins or proteins related to pathogenesis, especially if the treated fruit has injuries resulting from harvesting or of post-harvest handling (Gómez-Merino & Trejo-Téllez, 2015).

Wounds or microbial attacks suffered by fruits increase their respiratory rate. This rise may be due to the healing meristem activity or the production of plant defense substances since the tissue produces secondary metabolism substances related to defense and macromolecules associated with constructing new tissues during healing (Ramos et al., 2013).

Potassium and cobalt phosphite significantly reduced the respiratory process. Besides, these substances increased resistance to post-harvest diseases, with the respiratory rate directly associated with ripening

and subsequent senescence, which is postponed by potassium phosphite (Dutra et al., 2018). Cobalt reduces the respiratory rate, probably due to the attenuation of steps in the respiratory process, such as oxidative phosphorylation, ATP production, and glycolysis, which occur concomitantly with ethylene biosynthesis (Dongen et al., 2011). Furthermore, in our experiment, cobalt reduced the content of soluble solids, preserved the ascorbic acid, and reduced the rise in pH and the degradation of organic acids. The respiratory rate reduction justifies these findings, as these compounds are the substrates used in the respiratory process (Saltveit, 2019).

The reduction in fruit weight loss correlates with the transpiration process and respiratory rate, which, in turn, is mainly affected by ethylene biosynthesis. Cobalt ions inhibit this ethylene synthesis, reducing the transpiration process. The maintenance of firmness of fruits treated with cobalt agrees with previous results observed by Oliveira et al. (2018) in guavas and Teodosio et al. (2018) in papaya. Cobalt doses increase reduced fruit softening. The softening of the fruit derives from pectin degradation and starch conversion into sugars. Several enzymes influence the process. Among these enzymes, the action of Pectinmethylesterase (PME) stands out, catalyzing the deesterification of pectin (Oliveira et al., 2018). The softening reduction is related to the inhibition of ethylene biosynthesis by cobalt, delaying the effect of ethylene on the activity of cell wall hydrolytic enzymes (Oliveira et al., 2018).

Chlorophyll degradation and carotenoid biosynthesis affect the fruit color changes, reducing the Hue angle (close to red) and increasing chromaticity in 'Palmer' mangoes (Liu et al., 2013). Cobalt and potassium phosphite (IR) favored the maintenance of color in treated fruits, delaying chlorophylls degradation by reducing oxidative stress triggered by ethylene synthesis, and activating resistance induced, possibly , by the increase in jasmonic acid and salicylic acid. Potassium phosphite, inducing the production of antimicrobial compounds (phytoalexins, phenylalanine - ammonialyase, and compounds such as lignin), reduces stress due to pressure from phytopathogens and the degradation of chlorophyll through the action of ethylene (Dutra et al., 2018).

Potassium phosphite provided a smaller area under the incidence and severity progress curve (AACPI and AACPS) . Phosphite-based products effectively control plant diseases (Nojosa et al., 2005) . This positive effect may be due to the action of phosphite on induced systemic resistance: the phosphite molecule triggers the synthesis of jasmonic acid and salicylic acid, stimulating natural plant self-defense substances production, such as phytoalexins, protecting it from fungal attack. The molecule also has a fungicidal effect, acting directly on fungi (Spolti et al., 2015).

Potassium phosphite increased production and productivity. This effect may be due to the plant's rapid absorption of potassium, compared to other sources, and the reduction in anthracose post-harvest loss. (Sardinha, et al., 2019) .

Conclusion

Foliar application of cobalt at pre-harvest is an efficient technique, improves shelf life, and maintains the quality of 'Palmer' mango during storage at room temperature.

Using potassium phosphite reduces the intensity of anthracnose in fruits and increases shelf life.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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