Universidade Federal Rural do Semi-Árido
Pró-Reitoria de Pesquisa e Pós-Graduação
https://periodicos.ufersa.edu.br/index.php/caatinga

ISSN 0100-316X (impresso)
ISSN 1983-2125 (online)
http://dx.doi.org/10.1590/1983-21252021v34n104rc

RESISTANCE ELICITORS ON PRODUCTION AND POST-PRODUCTION PERFORMANCE OF POTTED LISIANTHUS[1]

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ABSTRACT - Lisianthus is an important ornamental species, whose cultivation in pot still requires studies, mainly regarding the use of alternative management. Thus, the aim of this work was to evaluate the influence of resistance elicitors on the production and post-production quality of potted lisianthus. Plants grown in pots were treated with two elicitors: (E1) phosphorylated mannano-oligosaccharide and (E2) citric bioflavonoids and phytoalexins, in the doses (D) of 1 and 2μL.L^{-1} (E1D1 1μL.L^{-1}, E1D2 2μL.L^{-1}, E2D1 1μL.L^{-1}, E2D2 2μL.L^{-1} and control), with 5 replications, in a completely randomized design. After opening the first flower buds, the plants remained in the greenhouse for 15 days, until they reached at least 3 open flowers, when were transferred to the room conditions (24 ± 2 ^{o}C and 72 ± 2%) for another 15 days. The application of elicitors did not influence the culture cycle or the size of the stem. There was an increase, however, in the number of viable buds and open flowers, in addition to an increase in flower's useful life paralleled to the reduction of senescent flowers, in relation to the control. The dose of 2μL.L^{-1} of the E2 kept the percentage of viable open flowers in relation to the control for at least another 5 days, reducing the percentage of senescent flowers. Together, with the application of the E2 elicitor in potted lisianthus, the viability of the flowers can be prolonged, maintaining quality and delaying senescence, and, in this way, increasing the market period.

Keywords: *Eustoma grandiflorum* (Raf.) shinn. Ornamental plant. Quality. Resistance induction. Potted flowers viability.

ELICITORES DE RESISTÊNCIA NO DESEMPENHO DE PRODUÇÃO E PÓS-PRODUÇÃO DE LISIANTHUS EM VASO

RESUMO - O lisianthus é uma importante espécie ornamental, cujo cultivo em vaso ainda requer estudos, principalmente quanto ao emprego de manejo alternativo. Assim, o objetivo deste trabalho foi avaliar a influência de elicitores de resistência na produção e na qualidade pós-produção de lisianthus em vaso. Plantas de lisianthus cultivadas em vaso foram tratadas com dois elicitores: (E1) manano-oligossacarideo fosforilado e (E2) bioflavonóides e fitoalexinas cítricas, nas doses (D) de 1 e 2μL.L^{-1} (E1D1 1 μL.L^{-1}, E1D2 2 μL.L^{-1}, E2D1 1 μL.L^{-1}, E2D2 2 μL.L^{-1} e controle), com 5 repetições, em delineamento inteiramente casualizado. Após a abertura dos primeiros botões florais, as plantas permaneceram na estufa durante 15 dias, até atingirem pelo menos 3 flores abertas, quando foram transferidas para a condição ambiente (24 ±2 ^{o}C e 72 ±2%) durante mais 15 dias A aplicação dos elicitores não influenciou o ciclo da cultura nem o tamanho da haste. Entretanto, observou-se aumento nos números de botões e de flores abertas viáveis, além de aumento dos dias de vida útil, paralelo à redução de flores senescentes em relação ao controle. A dose de 2μL.L^{-1} de E2 manteve em, pelo menos, mais 5 dias o percentual de flores abertas viáveis com relação ao controle, reduzindo o percentual de flores senescentes. Em conjunto, com a aplicação do elicitor E2 em lisianthus em vaso pode-se prolongar a viabilidade das flores, por manter a qualidade e retardar a senescência e, assim sendo, aumentar o período de comercialização.

Palavras-chave: *Eustoma grandiflorum* (Raf.) shinn. Planta ornamental. Qualidade. Indução de resistência. Viabilidade de flores em vaso.

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[1]Received for publication in 08/08/2019; accepted in 09/11/2020.
Paper extracted from the Thesis of the first author.
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INTRODUCTION

Lisianthus (*Eustoma grandiflorum* (Raf.) Shinn) is an ornamental plant that has been gaining space across the world due to the high attractiveness and vivid colors of the flowers (SAEEDI et al., 2015), standing among the 10 best selling products worldwide (LÓPEZ-GUERRERO et al., 2019). This species is popular due to its beautiful morphological characteristics (YAN et al., 2019) and stands out for the production of single or double flowers in a great diversity of colors (AHMAD et al., 2017) and bicolors (ALMEIDA; CALABONI; RODRIGUES, 2016) with high productivity (SHEIKH, F. et al., 2014). It is preferred for decorative arrangements and bridal bouquets due to the durability of the flowers as well as the length and firmness of the stems (LÓPEZ-GUERRERO et al., 2019). Lisianthus is grown as a cut flower in soil, or as a potted plant (ÖZKAN; ÖZEN, 2016). The cultivars 'Echo' and 'Mariachi' are preferred by the Brazilian market, with folded flowers being the most commercialized, mainly those of white and white with blue edges colors (CAVASINI et al., 2018), which are grown mainly as cut flowers (AHMAD et al., 2017).

However, the production of lisianthus from the cultivar 'Echo' in pots has been showing increasing demand in the market, and it has been characterized as an important option for floriculture. On the other hand, information about lisianthus cultivation in pots is still scarce both in Brazil (ALMEIDA; CALABONI; RODRIGUES, 2016) and in other producing countries (AHMAD et al., 2017; CASTILLO-GONZÁLEZ; VALDEZ-AGUILAR; AVITIA-GARCÍA, 2019), as well as the post-production behavior of potted plants (ÖZKAN; ÖZEN, 2016), which consequently compromises the commercialization process. In this context, aiming at lisianthus pot cultivation and the best yield, parameters related to types of pruning (BACKES et al., 2006), management (CASTILLO-GONZÁLEZ; VALDEZ-AGUILAR; AVITIA-GARCÍA, 2019) must be defined, in addition to criteria to guarantee the best quality of the flower demanded by the markets (FERRANTE et al., 2015). However, to reduce the negative impacts of agriculture, it is important to reduce dependence on fertilizers, making it necessary to use safe practices, of lower-cost and ecologically correct management in agricultural production (AHMED et al., 2020).

The limitation of information on quality criteria in post-production has been responsible for difficulties in sales and increased losses of potted flowers (FERRANTE et al., 2015). Such losses occur mainly from to the rapid deterioration of the flowers due to the degrading physiological processes because of the rapid use of carbohydrate reserves that occur more intensely after the transfer of the pot from the greenhouse to the marketing environment, making the flowers' durability very limited (PALIYATH et al., 2009). The quality of potted plant flowers is essentially defined by the visual appearance, which depends on the shape, size, renewal, and turgidity of the flowers and leaves (FERRANTE et al., 2015), whose main quality parameters are represented by the maintenance of color, number of flowers and if these are healthy, if are semi-open or fully open without any symptoms of senescence (NOWAK; RUDNICK, 1990).

Over time, strategies have been used to extend the life of cut lisianthus through floral preservatives and ethylene action inhibitors (NOORDERGRAAF, 1994; SHEIKH et al., 2014; SAEEDI et al., 2015; CAVASINI et al., 2018). However, considering the cultivation of potted flowers, sustainable strategies that are safe for the producer and the consumer must be explored, such as those involving induced resistance in plants (ROMANAZZI et al., 2016; LÓPEZ-GUERRERO et al. 2019; AHMED et al., 2020), not yet studied in potted lisianthus.

Resistance induction uses biotic and abiotic inducers as an alternative control method, in order to activate the defense mechanisms of plants (ALENCAR et al., 2020). The use of plant resistance inducers has been reported in tropical flowers to reduce the incidence of diseases that compromise quality (GURGEL et al., 2018), reducing the useful life. Among the commercial products used as resistance elicitors, have been highlighted the compounds based on phosphorylated mannan-oligosaccharide from the *Saccharomyces cerevisae* cell wall (SARDINHA et al., 2019) and those containing bioflavonoids and citric phytoalexins, vitamins, and organic acids, which are antioxidant substances that, even when applied in very small doses in production or pre-harvest, promote changes in the metabolism of plants, helping to prevent diseases and maintaining nutritional balance (ARANEGA-BOU et al., 2014). Furthermore, the bio-based resistence inducers, such as inducers based on citric biomass, rich in flavonoids, polyphenols, phytoalexins, and organic acids, can improve the use of inputs, balancing the metabolism of phenolic compounds to induce systemic acquired resistance (SAR) in plants, providing lower disease rate in pre- and postharvest, increasing resistance to transport, in addition to acting against water stresses and phytotoxicity. Its main function is to induce plant tissues to synthesize their own phytoalexins, which are responsible for reducing the damage caused by pathogens (BURKETOVA et al., 2015).

Thus, the present work aimed to evaluate the influence of resistance elicitors in the production and during the post-production period of lisianthus from the cultivar 'Echo Blue Picotee' cultivated in pots.

MATERIAL AND METHODS

Lisianthus young seedlings, cultivar Echo Blue Picotee (Sakata Seed Sudamerica) (white folded flowers with blue edges), were purchased from the Isabel Yamagushi (Atibaia - São Paulo/ Brazil) nursery, with approximately two pairs of leaves. Seedlings establishment were carried out in the Association for the Sustainable Development of Macacos and Furnas (Areia – Paraíba/Brazil), in a 3.5 m-high commercial production greenhouse, covered with transparent film. The transplant was done using two seedlings per pot (750 mL) and a substrate mixture of soil (70%), carbonized rice husks (25%) and bovine manure (5%). The irrigation system was dripped daily, performed in the morning, applying approximately 150 mL per pot of nutrient solution. After 15 days of transplanting, when the plants were about 15 cm high, pruning was carried out above the second and third pair of leaves (BACKES et al., 2006), aiming at stimulating the side shoots to obtain a greater production of flowers in pots.

The experimental design was completely randomized in a split-time plot with 5 treatments, two elicitors (E) in two doses (D) and one control (E1D1 - 1 $\mu L.L^{-1}$; E1D2 - 2 $\mu L.L^{-1}$; E2D1 - 1 $\mu L.L^{-1}$; E2D2 - 2 $\mu L.L^{-1}$ and control) with 5 replications over a period of 15 days at the post-production (after transferring from greenhouse to room condition), with daily evaluations. Resistance inducers were applied 58 days after transplanting, a period that preceded the production of flower buds. Two applications of the elicitors were performed, with an interval of 15 days, using a manual spray with a capacity of 1 liter, applying 100 $mL.planta^{-1}$, approximately. Two commercial elicitors were used, one based on phosphorylated mannano-oligosaccharide (E1) from the *Saccharomyces cerevisiae* cell wall, and the other (E2) containing bioflavonoids (vitamin P), citric phytoalexins, ascorbic acid, citric acid and lactic acid. The elicitors were used in the doses of 1 and 2 $\mu L.L^{-1}$, diluted in distilled water.

The potted lisianthus market condition was established after the opening of the first flower buds, by keeping the pots in the greenhouse for a 15 days period, until a mean number of 3 flowers per pot was reached, meeting the quality standards of the region's consumer (several opened flowers per vase). At this point, the potted plants were transferred to the laboratory benches of the Laboratory of Postharvest Biology and Technology, Center of Agrarian Science, Universidade Federal da Paraíba, Areia – PB/Brazil and kept under room conditions (24 ± 2 °C and 72 ± 2% RH) for 15 days, simulating the locals of sale, and watered with water on alternate days.

The production cycle was determined by the number of days spent by the seedlings from the transplanting to pots until the point of exposure to the market conditions (HALEVY; KOFRANEK, 1984); the size of the floral stem (cm) was measured from the insertion of the stem into the root to the peduncle; the units of buds, viable open flowers, senescent flowers, and flowers' useful life were measured by the number of days between the first 3 flowers opening in the greenhouse and the beginning of senescence of the last 3 flowers opened in the stems, in pots kept under room conditions. Under this experiment condition, based on the preference of the regional consumer, the minimum limit for viable open flowers was set at 20% relative to flower buds, and the maximum limit for senescent flowers in 80% relative to viable open flowers.

The data were subjected to variance analysis, and the treatment means were compared with the control by Dunnett's test up to 5% of error probability, using the SAS program. For the split of time within each treatment, polynomial regression analysis up to 2nd degree was used, considering the coefficient of determination greater than 0.7.

RESULTS AND DISCUSSION

The production cycle of the Echo lisianthus cultivar was 88 days, starting from transplanting, followed by a period in the greenhouse for the potted plants of all treatments (Table 1), thus verifying that the application of elicitors did not negatively influence the culture cycle. A similar result was reported by Backes et al. (2006), evaluating the same cultivar with different nutrient solutions, which reported an 87-day cycle. However, according to Ahmad et al. (2017), the lisianthus cultivation cycle is four to six months and varies depending on the environment and the cultivar. Therefore, the use of resistance elicitors maintained the production cycle satisfactorily, compared to the use of nutrient solutions, or the cycle was inferior to those reported for this culture in other continents.

Regarding the stem size (Table 1), it was found that the treatments with elicitors did not differ among treatments, varying from 22.9 to 29.4 cm. However, this stem size range is satisfactory, meeting Brazilian Northeast consumers' quality standards, which prefer stems up to 30 cm. This also results in a lower production cost, as there is no need to apply growth regulators. According to Castillo-González, Valdez-Aguilar and Avitia-García (2019), for growing potted plants, the growth regulator must be applied two to three weeks after topping, when the sprouts are 2.5 to 5 cm, repeating the application two to three weeks later, which is costly and requires greater input of labor. Larger stem sizes were reported by Backes et al. (2006) for the cultivar 'Flamenco Blue' grown in pots with different nutrient solutions and ways of conducting, whose values were 47.71 and 43.62 cm for the absence of pruning and pruning in the second pair of leaves, respectively. This much higher size of stems may be due, however, to the supply of nutrients resulting from the nutritional solutions of that experiment.

Regarding the post-production period, the mean number of buds and the number of viable flowers, as well as the useful life of the flowers increased, parallel to the reduction of senescent flowers, compared to the control (Table 1), clearly characterizing the positive effect of the elicitors in maintaining the post-production quality of potted lisianthus flowers. The highest means of the number of buds, number of viable open flowers, and senescents were provided by E2 applications, probably because it is a multi-action organic complex based on bioflavonoids and phytoalexins, indicated to compliment and increase the efficiency of the nutritional treatment (BURKETOVA et al., 2015). According to the manufacturer's bulletin, the crops most favored by this elicitor are those whose plants have a bush, creeping, or climbing habit, with a greater amount of non-lignified tissues, as the lisianthus plants.

Table 1. Mean values for units of flower bud, viable flower, senescent flower, viable days of post-production useful life and stem size in potted lisianthus (*Eustoma grandiflorum* (Raf.) Shinn, cv. Echo Blue Picotee) plants treated with two doses of E1 and E2 elicitors, followed by exposure to room conditions (24±2 ^{o}C and 72±2% RH).

Treatments	Production Cycle (Days)	Flower Buds (Units)	Viable Flowers (Units)	Senescent Flowers (Units)	Useful Life** (Days)	Stem Size (cm)
Control	88	0.45	3.85	5.89	26	29.4
E1D1 1µL.L^{-1}	88 ^{ns}	0.40^{ns}	4.87*	3.93*	28 ^{ns}	26.9 ^{ns}
E1D2 2µL.L^{-1}	88 ^{ns}	0.16*	5.87*	3.24*	30*	26.9 ^{ns}
E2D1 1µL.L^{-1}	88 ^{ns}	1.24*	7.51*	3.65*	>30*	29.5 ^{ns}
E2D2 2µL.L^{-1}	88 ^{ns}	1.55*	7.11*	4.20*	>30*	28.2 ^{ns}
CV (%)	6.22	11.21	10.12	13.51	6.36	11.99

*Means differ from Control (p≤0.05) by Dunnett's test; ^{ns} Not significant. n=5.
**Mean sum of days from 3 flowers opened in a greenhouse (15 days) added to the days with up to 3 viable flowers in the exposure to the room conditions. E1 = phosphorylated mannano-oligosaccharide; E2 = Bioflavonoids and citrus phytoalexins.

Regarding the useful life, it was found that the plants treated with elicitors remained with viable flowers for a longer period of life than the control (considering that these had three flowers open 15 days before exposure), except for E1 at a dose of 1 µL L^{-1}. However, E2, in the two doses applied, stood out for maintaining the useful life of the potted flowers for more than 30 days, which is very significant to guarantee the successful commercialization of the plants. Backes et al. (2006) reported for the potted 'Flamenco Blue' cultivar a 22 -day longevity, much lower than those obtained in this work. Lower shelf life was also reported by Halevy and Kofranek (1984) for the potted 'Blue' cultivar, whose longevity was 18.9 days. Therefore, the results of this work indicate that the use of E2 provided an improvement in the post-production of potted lisianthus, reflected in the significant increase in the useful days of viable flowers. This response was probably due to this elicitor acting as a growth and development phytoregulator, improving the performance of plants and adjusting their defense and resistance responses against pathogens (LÓPEZ-GUERRERO et al. 2019).

Regarding the number of flower buds, it was observed that E2 was also the elicitor that provided the highest number of buds, while treatments with E1 at dose 1 µL L^{-1} showed fewer flower buds when compared to the control (Table 1; Figure 1). However, the two elicitors increased the bud appearance time, since in the control the appearance of the buds ceased on the seventh day, while in the treated plants, the presence of flower buds was observed until the tenth day of room exposure. This fact can be explained, probably, by the lower production of ethylene in flower buds of plants treated with elicitors. According to Paliyath et al. (2009), in climacteric flowers such as lisianthus (HALEVY; KOFRANEK, 1984), ethylene production in the buds and young flowers is very low and stable, reaching a maximum production peak as floral maturity is reached. Thus, the control of the adverse effects of this hormone is the key factor for maintaining the quality of the flowers of potted plants (NOORDERGRAAF, 1994).

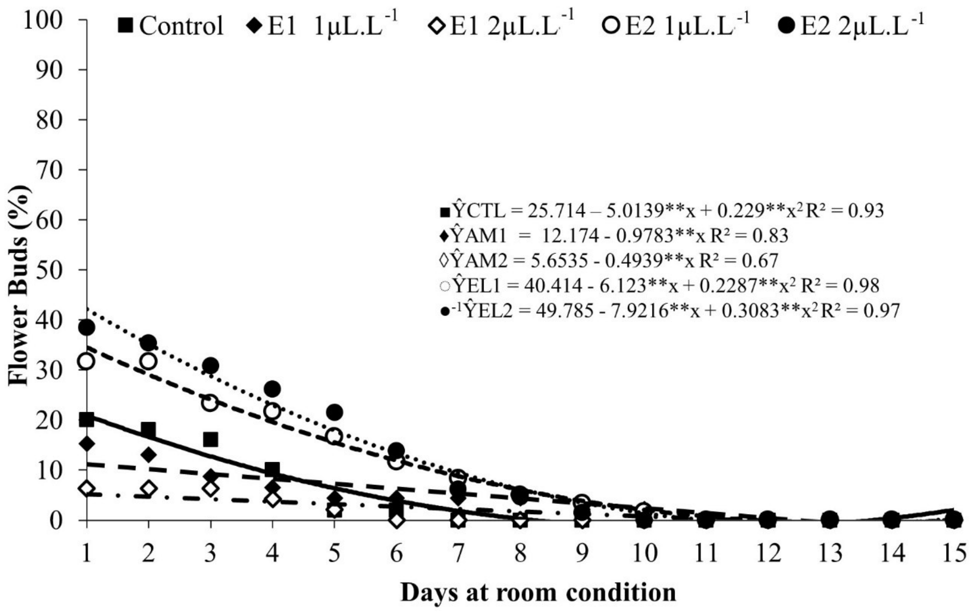

Figure 1. Percentage of flower buds relative to viable flowers in potted lisianthus (*Eustoma grandiflorum* (Raf.) Shinn, cv. Echo Blue Picotee) plants, treated with two doses (D) of E1 and E2 elicitors, after transfering from the greenhouse and maintained for another 15 days under room conditions (24±2 °C and 72±2% RH), n=5.
E1 = phosphorylated mannano-oligosaccharide; E2 = Bioflavonoids and citrus phytoalexins.

First introduced in this study, the evaluation of the percentage of open viable flowers relative to flower buds was characterized by a practical, reproducible, and efficient quality criterion to access the post-production quality of potted lisianthus. For the percentage of viable open flowers relative to flower buds (Figure 2), it was found that the elicitor E2 also promoted a more uniform distribution in the opening of the flowers until the sixth day of room storage, and linearly until the end of storage. This characteristic is desirable for the flower production chain, as it allows greater flexibility in marketing. This result indicates that elicitor E2 probably induced an increase in the natural resistance of potted lisianthus plants, improving the use of inputs (ROMANAZZI et al., 2016) and balancing the metabolism of phenolic compounds (SARDINHA et al., 2019), providing uniformity in the opening of flower buds in post-production. Still, relative to the number of open viable flowers, it was found that at 15 days under room conditions, plants treated with E2 resistance elicitors proved to be superior to the control, presenting between three and five viable flowers showing commercial conditions. These results clearly indicate the superiority of the E2 elicitor in maintaining the quality of the potted lisianthus flowers in the post-production period.

The main post-production disorders in potted flowering plants are the abscission of buds and flowers (FERRANTE et al., 2015). However, in this experiment, there was no abscission of flowers or flower buds during 15 days of room exposure. For the number of senescent flowers, it was found that the application of resistance elicitors delayed the senescence of the flowers, differing from the control, probably by delaying the production of ethylene, since the production of climacteric ethylene by the flowers can be inhibited by the use of substances that inhibit both the synthesis and the action of ethylene, resulting in increased postharvest longevity (MUBAROK; SUMINAR; KAMALUDDIN, 2019). In this context, according to Ferrante et al. (2015), ethylene has marked effects on the induction of senescence, abscission, and withering of potted flowers and, especially on those that have some sensitivity to the action of this hormone (NOWAK; RUDNICK, 1990), such as lisianthus. Therefore, a probable reduction in ethylene production mediated by the action of resistance inducers led to a reduction of about 30% in the senescent flowers relative to viable flowers, in the highest dose of E1 and for E2.

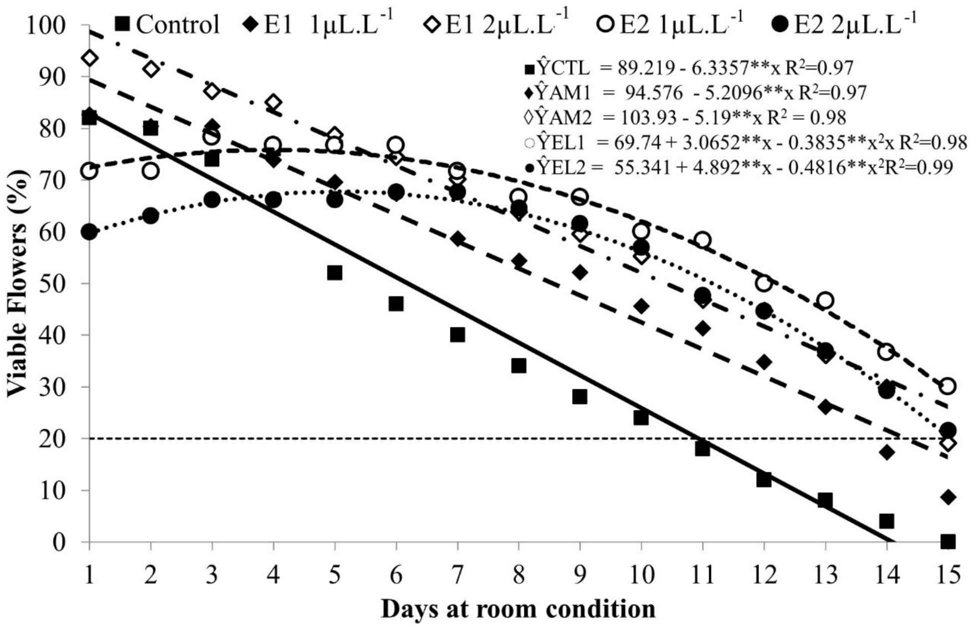

Figure 2. Percentage of viable open flowers relative to flower buds in potted lisianthus (*Eustoma grandiflorum* (Raf.) Shinn, cv. Echo Blue Picotee) plants, treated with two doses of E1 and E2 elicitors, after transfering from the greenhouse and maintained for another 15 days under room conditions (24±2 ºC and 72±2% RH), n=5.
The dotted line represents the maximum percentage of acceptance limit (80%) of senescent flowers, after thinning. E1 = phosphorylated mannano-oligosaccharide; E2 = Bioflavonoids and citrus phytoalexins.

Losses in potted flower plant quality are mainly due to flower senescence (FERRANTE et al., 2015). As with the percentage of open viable flowers, the evaluation of the percentage of senescent flowers relative to viable flowers was used for the first time in this study and was characterized as a reproducible, practical, and efficient tool to access the post-production quality of potted lisianthus and can be used to characterize the post-production quality of potted flowering plants. Considering the 80% limit on the acceptance of senescent flowers for the potted plants, which indicates that at least 20% of the flowers were viable, allowing the senescent flowers to be thinned, the control potted plants crossed the limit line of acceptance in about 10 days at room condition (Figure 3), while those treated with the highest dose of E1 and the two doses of E2 at 15 days of room exposure had not yet crossed the acceptance line after 15 days under room condition.

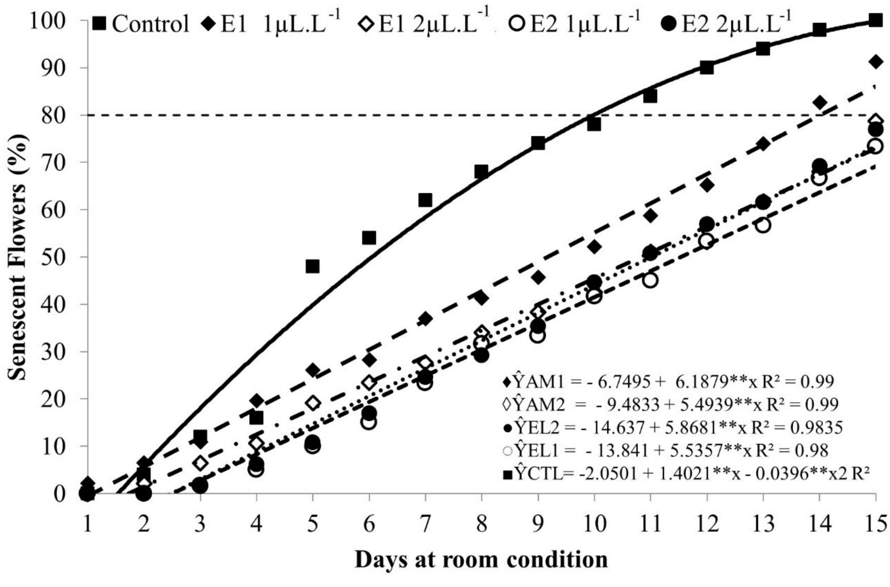

Figure 3. Percentage of senescent flowers relative to viable flowers in potted lisianthus (*Eustoma grandiflorum* (Raf.) Shinn, cv. Echo Blue Picotee) plants, treated with E1 and E2 elicitors in two doses, after transfering from the greenhouse and maintained for another 15 days under room conditions (24±2 ºC and 72±2% RH), n=5.
The dotted line represents the maximum percentage of acceptance limit (80%) of senescent flowers, after thinning. E1 = phosphorylated mannano-oligosaccharide; E2 = Bioflavonoids and citrus phytoalexins.

The influence of the application of resistance elicitors on the post-production quality of potted lisianthus after 10 days of exposure to room condition is shown in Figure 4, in which it is observed that the use of elicitors promoted the maintenance of the quality of this potted flowers and that, unmistakably, the application of 2 µL L^{-1} of E2 kept the flowers viable.

Figure 4. Potted lisiantus (*Eustoma grandiflorum* (Raf.) Shinn, cv. Echo Blue Picotee) plants, treated with elicitors (E1) and (E2) in two doses, after 10 days of maintenance at room condition (24 ± 2 °C and 72 ± 2% RH).
E1 = phosphorylated mannano-oligosaccharide; E2 = Bioflavonoids and citrus phytoalexins.

Together, these results confirm that the application of resistance elicitors, especially the dose of 2 µL L^{-1} of E2, based on bioflavonoids and citrus phytoalexins, increased the viability of potted lisianthus flowers in more than five days relative to the control, which represents a promising period for the maintenance and commercialization of this product in the markets. Given the above, the use of resistance elicitors is characterized as an ecological, safe, efficient, and low-cost strategy for the production of potted lisianthus, with superior quality to meet the emerging market of this ornamental species.

CONCLUSIONS

In potted lisianthus plants of the 'Echo Blue Picotee' cultivar, the use of resistance elicitors did not interfere with the stem size and production cycle.

The percentages of viable and senescent flowers are appropriate criteria for assessing the post -production quality of potted lisianthus.

The use of resistance elicitors promoted an extension of the useful life in the post-production of potted flowers, keeping them viable for a longer time, delaying floral senescence during exposure to room conditions.

The elicitor E2, at a dose of 2µl.L^{-1}, was more efficient in maintaining viability and delaying senescence in post-production of potted lisianthius flowers for at least another 5 days relative to the control, reducing the percentage of flowers senescent.

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