



Proceeding Paper

Foliar Application of Gibberellic Acid and Methyl Jasmonate Improves Leaf Greenness in *Hesperantha coccinea* (syn. *Schizostylis coccinea*), a Rare Ornamental Plant [†]

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- † Presented at the 1st International Electronic Conference on Plant Science, 1–15 December 2020; Available online: https://iecps2020.sciforum.net/.

Abstract: *Hesperantha coccinea* (Iridaceaea) is a little-known ornamental plant recommended for cultivation in pots, gardens and for cut flowers. The species produces narrow, decorative green leaves that sometimes turn yellow as the plants begin to anthesis. A solution to the problem may be the application of plant growth regulators (PGRs), and so far none have been studied in *Hesperantha* cultivation. The study assessed the effect of foliar application of gibberellic acid solutions (GA₃; 50,100 and 200 mg dm⁻³) and methyl jasmonate (MeJA; 100,500 and 1000 µmol dm⁻³) on the leaf greenness index (SPAD index), leaf number, leaf length and leaf width of *H. coccinea*. Control plants were not treated with PGRs. It was found that both GA₃ and MeJA applied at all tested concentrations significantly increased the greenness index of *H. coccinea* leaves. In addition, GA₃ increased the number of leaves without affecting the length and width of the leaves. MeJA had a growth inhibitory effect. When compared to the control, plants sprayed with MeJA solutions produced fewer leaves that also had reduced length and width. These findings indicate that both regulators (GA₃ and MeJA) positively affected plant quality by improving leaf greenness.

Keywords: kaffir lily; river lily; geophytes; phytohormones; greenness index



Citation: Salachna, P.; Łopusiewicz, Ł.; Dymek, R.; Matzen, A.; Trochanowicz, K. Foliar Application of Gibberellic Acid and Methyl Jasmonate Improves Leaf Greenness in Hesperantha coccinea (syn. Schizostylis coccinea), a Rare Ornamental Plant. Biol. Life Sci. Forum 2021, 4, 97. https://doi.org/10.3390/IECPS2020-08622

Academic Editor: Yoselin Benitez-Alfonso

Published: 30 November 2020

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1. Introduction

Production of ornamental plants is an important branch of horticulture that is growing dynamically as a result of increasing wealth of developed societies [1]. Regular expansion of the offer with new species is the factor influencing the development of the ornamental plant market [2,3]. The assortment of ornamental geophytes blooming in autumn is still very modest [4]. A particularly attractive plant belonging to this group is the species *Hesperantha coccinea* (Backh. & Harv. Goldblatt & J.C. Manning) known until recently under the Latin name *Schizostylis coccinea* [5]. *H. coccinea* belongs to the family Iridaceaea and occurs naturally in South Africa [6]. The species is marketed under the popular names of kaffir lily, river lily or crimson flag lily. Plants produce numerous linear or lanceolate leaves, within which 50–70 cm long spikes composed of several flowers develop in late summer and autumn. Single flowers are large (up to 6 cm), red, pink or white in color (Figure 1a), and are pollinated by butterflies. The fruit is an egg-shaped capsule. The storage organs of plants are short rhizomes [7]. *H. coccinea* is recommended for cultivation in pots, gardens and for cut flowers. The available scientific literature lacks research on the cultivation of plants of the genus *Hesperantha*.

During flowering, some geophytes show yellowing, chlorosis and drying of the leaf tips [8]. Similarly, *H. coccinea* leaves often have dry tips already before anthesis, which

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significantly reduces the quality of plants (Figure 1b). This is particularly a problem when plants are sold in pots. The purchase is determined by the quality and appearance of the plant in the pot. Garden owners and landscape architects most often choose plants characterized by fresh green leaves [9,10]. In order to increase the quality of ornamental plants, various types of plant hormones, growth regulators [11,12] and biostimulators [13,14] are used in production. Thus far, no regulators have been studied in the cultivation of *H. coccinea*.



Figure 1. An inflorescence of *Hesperantha coccinea* (a); yellowing of the leaf tips of *Hesperantha coccinea* at the flowering stage (b).

The greenness of the leaves is related to the content of assimilation pigments, mainly chlorophyll. The use of exogenous regulators and biostimulants often modifies the content of assimilation pigments in leaves [15,16]. For example, after treatment with gibberellic acid, an increase in leaf greenness was observed in geophytes such as \times Amarine tubergenii 'Zwanenburg' [17], Tulipa gesneriana 'Ad Rem' [18], Zantedeschia aethiopica 'Childsiana' [19] and Lilium longiflorum 'Nellie White' [20]. Methyl jasmonate is another regulator still rarely used in horticultural production [21]. The literature shows that this hormone exerts various effects on chlorophyll content in plant leaves. Under stressful conditions, the treatment of plants with methyl jasmonate inhibits the decrease in chlorophyll content [22–25].

The aim of the research was to determine the effect of various concentrations of gibberellic acid and methyl jasmonate on greenness and morphological features of *H. coccinea* leaves. The research hypothesis assumed that the tested regulators would limit the yellowing of *H. coccinea* leaves.

2. Materials and Methods

Hesperantha coccinea rhizomes were imported from the Netherlands through Ogrodnictwo Wiśniewski Jacek Junior. In order to protect against *Penicillium* and *Fusarium* fungi, the rhizomes were treated for 30 min with an aqueous suspension of the following preparations: 0.7% (w/v) Topsin M 500 SC (Nippon Soda, Tokio, Japan, active ingredient—thiophanate-methyl) and 1% (w/v) Kaptan 50 WP (Organika-Azot Chemical Company, Jaworzno, Poland, active ingredient—Captan). After drying, the rhizomes were planted

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on April 15 at a depth of 2 cm in black PVC pots, 15 cm in diameter and 1.5 L volume, filled with TS1 medium (Klasmann-Delimann, Germany). On the basis of chemical analysis carried out in an accredited laboratory, the substrate used in the experiments had a salinity of 1.86 g NaCl dm⁻³, pH 5.5 and contained 181 mg dm⁻³ N-NO₃, 131 mg dm⁻³ P and 402 mg dm⁻³ K. Pots were placed on tables in a double foil tunnel belonging to the West Pomeranian University of Technology in Szczecin (53°25′ N, 14°32′ E; 25 m asl.). Temperature and relative humidity were recorded throughout the cultivation period.

When the plants were about 10–15 cm tall, the regulator gibberellic acid (GA $_3$) was sprayed twice at concentrations of 50, 100 and 200 mg dm $^{-3}$ and methyl jasmonate (MeJA) at concentrations of 100, 500 and 1000 µmol dm $^{-3}$. Both compounds were purchased from Sigma-Aldrich Chemie GmbH (Schnelldorf, Germany). Approximately 12 cm 3 of the solution was used for one plant. Control plants were sprayed with distilled water. Ethanol was used as a solvent at a concentration of 0.04%, v/v.

The leaf greenness index (SPAD index) was measured using an SPAD 502 optical apparatus (Konika-Minolta Corporation, Osaka, Japan). The values indicated by a meter in dimensionless units called SPAD are proportional to the chlorophyll content in the leaf. They are calculated on the basis of the quantity of radiation transmitted through the leaf in two radiation ranges absorbed differently by chlorophyll. Measurements were made on 5 randomly selected, well-formed leaf blades in each replication. Moreover, the number of produced leaves was counted, and their length and width were measured.

The experiment was carried out in a random block design. Eight experimental objects were evaluated (GA $_3$ 0, 50, 100 and 200 mg dm $^{-3}$ and MeJA 0, 100, 500 and 1000 µmol dm $^{-3}$). A total of 20 rhizomes were planted in each facility, 5 in 4 replications. The results were statistically analyzed separately for each regulator using one-way analysis of variance (ANOVA) and the Statistica 13.3 software (TIBCO Software Inc. Statsoft, Cracow, Poland). Confidence half-intervals were calculated based on Tukey's HSD test.

3. Results and Discussion

Foliar chlorosis is known to reduce the aesthetic value of the plants and therefore regulators are sought that could reduce leaf yellowing. In this study, two phytohormones with a different mechanism of action were used: gibberellic acid and methyl jasmonate. They are natural compounds with a broad spectrum of activity, completely metabolized by plants and therefore safe for the environment.

Both GA₃ and MeJA increased the greenness of H. coccinea leaves, thereby increasing the visual quality of the plants (Tables 1 and 2). Compared to the control, plants treated with gibberellic acid at the concentration of 50, 100 and 200 mg dm⁻³ had a clearly increased SPAD index. The positive effect of GA on chlorophyll content in leaves may be caused by an increase in the number and size of chloroplasts. Gibberellic acid applied to extend the life of cut flowers and cut greenery inhibits leaf chlorosis. There are hypotheses that GA₃ limits aging of leaf tissues through slower degradation of chlorophyll. Perhaps gibberellins influence the activity of enzymes that mediate chlorophyll degradation, such as chlorophyllase and magnesium dechelatase, but this requires further research.

Table 1. Effect of gibberellic acid (GA₃) on the leaf greenness index (SPAD), number of leaves, leaf length and leaf width of *Hesperantha coccinea*. Means over each column not marked with the same letter are significantly different at $p \le 0.05$.

GA_3 (mg dm ⁻³)	SPAD	Leaves (No.)	Leaf Length (cm)	Leaf Width (cm)
0 (Control)	55.6 b	32.3 b	34.5 a	0.88 a
50	63.8 a	37.0 a	32.9 a	0.83 a
100	62.4 a	35.9 a	33.0 a	0.84 a
500	60.7 a	37.9 a	32.4 a	0.86 a

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Table 2. Effect of methyl jasmonate (MeJA) on the leaf greenness index (SPAD), number of leaves, leaf length and leaf width of *Hesperantha coccinea*. Means over each column not marked with the same letter are significantly different at $p \le 0.05$.

MeJA (μmol dm ⁻³)	SPAD	Leaves (No.)	Leaf Length (cm)	Leaf Width (cm)
0 (Control)	51.6 с	26.3 a	45.2 a	1.08 a
100	53.6 b	22.5 b	41.5 b	0.88 b
500	56.7 ab	21.2 b	41.0 b	0.88 b
1000	59.0 a	19.3 b	42.0 b	0.93 b

On the basis of the results, it was found that the application of MeJA at a concentration of 100, 500 and 1000 μ mol dm⁻³ increased the SPAD index and, unlike GA₃, the concentration of MeJA had a significant impact on the greenness index value (Tables 1 and 2). The highest SPAD index (59.0) was observed in plants after MeJA application at 1000 μ mol dm⁻³. The obtained results were consistent with other studies [17], where the stimulating effect of MeJA spraying with 1000 μ mol dm⁻³ on the greenness index of λ amarine tubergenii 'Zwanenburg' leaves was demonstrated. It can be assumed that MeJA inhibits chlorophyll degradation in leaves, delaying their aging. It was found in tomato grown under stress conditions caused by lead treatment that the application of jasmonic acid improved chlorophyll content by reducing the expression of chlorophyllase. Vainstein et al. [26] showed that cut rose (*Rosa hybrida*) 'Frisco' treated with methyl jasmonate retained petal color mainly by delaying carotenoid degradation. The cited authors believed that methyl jasmonate could induce photosynthetic pigment synthesis.

The applied regulators had an opposite effect on the number of *H. coccinea* leaves. Under the influence of gibberellic acid, irrespective of the concentration, plants produced significantly more leaves than the untreated plants. In the case of MeJA, the opposite reaction was observed—plants sprayed with this regulator produced fewer leaves. When analyzing the length and width of *H. coccinea* leaves, it was found that exogenous gibberellic acid had no effect on these two features. Both MeJA and gibberellic acid are involved in the regulation of plant germination, morphogenesis and aging, and they can affect growth and physiological activity to varying degrees.

4. Conclusions

In this study, the following growth regulators were used for the first time: gibberellic acid (GA₃; 50, 100 and 200 mg dm⁻³) and methyl jasmonate (MeJA; 100, 500 and 1000 μ mol dm⁻³) in *Hesperantha coccinea* cultivation. *H. coccinea* is a little-known ornamental plant with great potential. It was demonstrated that both GA₃ and MeJA, at all concentrations applied, increased the leaf greenness index and thus improved the quality of *H. coccinea* plants. MeJA inhibited the growth of *H. coccinea*, showing an effect similar to the retardants commonly used in horticulture.

Author Contributions: P.S. conceived and designed the experiments; P.S. and Ł.Ł. analyzed the data; R.D., A.M. and K.T. performed the experiments; P.S. wrote the paper. All authors have read and agreed to the published version of the manuscript.

Conflicts of Interest: The authors declare no conflict of interest.

References

- 1. Seaton, K.; Bettin, A.; Grüneberg, H. New ornamental plants for horticulture. In *Horticulture: Plants for People and Places*; Dixonand, G.R., Aldous, D.E., Eds.; Springer: Dordrecht, The Netherlands, 2014; pp. 435–463.
- 2. Salachna, P.; Grzeszczuk, M.; Soból, M. Effects of Chitooligosaccharide Coating Combined with Selected Ionic Polymers on the Stimulation of *Ornithogalum saundersiae* Growth. *Molecules* **2017**, 22, 1903. [CrossRef]
- 3. Piechocki, R.; Salachna, P. Effect of flurprimidol on growth and flowering of Chilean ornamental geophyte *Leucocoryne coquimbensis* F. Phil. ex Phil. *World Sci. News* **2019**, *133*, 34–44.
- 4. Salachna, P.; Zawadzińska, A.; Piechocki, R.; Wośkowiak, A. The growth and flowering of *Rhodohypoxis baurii* (Baker) Nel cultivars depending on rhizome weight. *Folia Hortic.* **2015**, 27, 169–173. [CrossRef]

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- 5. Goldblatt, P.; Manning, J.C. Reduction of Schizostylis (Iridaceae: Ixioideae) in Hesperantha. Novon 1996, 6, 262–264. [CrossRef]
- 6. Wolff, K.; Knees, S.; Cubey, S. Genetic relatedness and cultivar identification in a valuable garden species, Hesperantha coccinea (*Schizostylis coccinea*). *Plant Genet. Resour.* **2009**, *7*, 281–290. [CrossRef]
- 7. Rudall, P. Taxonomic and evolutionary implications of rhizome structure and secondary thickening in Iridaceae. *Bot. Gaz.* **1984**, 145, 524–534. [CrossRef]
- 8. Ranwala, A.P.; Miller, W.B. Effects of gibberellin treatments on flower and leaf quality of cut hybrid lilies. *Acta Hortic.* **2002**, 570, 205–210. [CrossRef]
- 9. Salachna, P.; Zawadzińska, A. Horticultural therapy in the landscape architecture: People-plant interactions. *World Sci. News* **2019**, *133*, 1–11.
- 10. Zawadzińska, A.; Salachna, P. Horticultural therapy in the landscape architecture: Therapeutic garden. *World Sci. News* **2019**, *132*, 300–307.
- 11. Salachna, P. Use of Chitosan Derivatives to Improve the Growth of Ornamentals. Inzynieria Ekol. 2017, 18, 63–68. [CrossRef]
- 12. Zawadzińska, A.; Salachna, P.; Piechocki, R. Ornamental swiss chard (Beta vulgaris var. cicla) response to daminozide and flurprimidol. *World Sci. News* **2017**, *61*, 86–97.
- 13. Salachna, P.; Grzeszczuk, M.; Meller, E.; Mizielińska, M. Effects of Gellan Oligosaccharide and NaCl Stress on Growth, Photosynthetic Pigments, Mineral Composition, Antioxidant Capacity and Antimicrobial Activity in Red Perilla. *Molecules* **2019**, 24, 3925. [CrossRef]
- 14. Salachna, P. Wykorzystanie pochodnych chitozanu w celu stymulacji wzrostu roślin ozdobnych. *Inżynieria Ekol.* **2017**, *18*, 63–68. [CrossRef]
- 15. Salachna, P.; Zawadzińska, A. Effect of daminozide and flurprimidol on growth, flowering and bulb yield of *Eucomis autumnalis* (Mill.) Chitt. *Folia Hortic.* **2017**, 29, 33–38. [CrossRef]
- Grzeszczuk, M.; Salachna, P.; Meller, E. Changes in Photosynthetic Pigments, Total Phenolic Content, and Antioxidant Activity
 of Salvia coccinea Buc'hoz Ex Etl. Induced by Exogenous Salicylic Acid and Soil Salinity. Molecules 2018, 23, 1296. [CrossRef]
 [PubMed]
- 17. Salachna, P.; Mikiciuk, M.; Zawadzińska, A.; Piechocki, R.; Ptak, P.; Mikiciuk, G.; Pietrak, A.; Łopusiewicz, Ł. Changes in Growth and Physiological Parameters of × *Amarine* Following an Exogenous Application of Gibberellic Acid and Methyl Jasmonate. *Agronomy* **2020**, *10*, 980. [CrossRef]
- 18. Ramzan, F.; Younis, A.; Riaz, A.; Ali, S.; Siddique, M.I.; Lim, K.B. Pre-planting exogenous application of gibberellic acid influences sprouting, vegetative growth, flowering, and subsequent bulb characteristics of 'Ad-Rem'tulip. *Hortic. Environ. Biotechnol.* **2014**, 55, 479–488. [CrossRef]
- Mortazavi, N.; Naderi, R.A.; Majidian, N.; Naderi, B.; Sharafi, Y. The effect of GA3 and BA on the quantitative and qualitative characteristics of calla lily (Zantedeschia aethiopica cv. Childsiana). Afr. J. Microbiol. Res. 2011, 5, 4190–4196.
- 20. Han, S.S. Growth regulators delay foliar chlorosis of Easter lily leaves. J. Am. Soc. Hortic. Sci. 1995, 120, 254–258. [CrossRef]
- 21. Rohwer, C.L.; Erwin, J.E. Horticultural applications of jasmonates. J. Hortic. Sci. Biotechnol. 2008, 83, 283–304. [CrossRef]
- 22. Bali, S.; Jamwal, V.L.; Kohli, S.K.; Kaur, P.; Tejpal, R.; Bhalla, V.; Siddiqui, M.H. Jasmonic acid application triggers detoxification of lead (Pb) toxicity in tomato through the modifications of secondary metabolites and gene expression. *Chemosphere* **2019**, 235, 734–748. [CrossRef] [PubMed]
- 23. Bidabadi, S.S.; Mehri, H.; Ghobadi, C.; Baninasab, B.; Afazel, M. Morphological, physiological and antioxidant responses of some Iranian grapevine cultivars to methyl jasmonate application. *J. Crop Sci. Biotechnol.* **2013**, *16*, 277–283. [CrossRef]
- 24. Taheri, Z.; Vatankhah, E.; Jafarian, V. Methyl jasmonate improves physiological and biochemical responses of Anchusa italica under salinity stress. *S. Afr. J. Bot.* **2020**, *130*, 375–382. [CrossRef]
- 25. Yan, Z.; Chen, J.; Li, X. Methyl jasmonate as modulator of Cd toxicity in Capsicum frutescens var. fasciculatum seedlings. *Ecotoxicol. Environ. Saf.* **2013**, *98*, 203–209. [CrossRef] [PubMed]
- 26. Vainstein, A.; Meir, A.; Tadmor, Y.; Meir, S.; Glick, A.; Philosoph-Hadas, S. Methyl jasmonate enhances color and carotenoid content of yellow-pigmented cut rose flowers. *Acta Hortic.* **2007**, *755*, 243–250.