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Original article

Plant growth regulators modulate the growth, physiology, and flower quality in rose (*Rosa hybirda*)



Anam Zahid^a, Gao Yike^{a,*}, Stepan Kubik^b, Fozia^a, Muhammad Ramzan^c, Hasan Sardar^d, Muhammad Tahir Akram^e, Muhammad Ahsan Khatana^f, Sana Shabbir^g, Sulaiman Ali Alharbi^h, Saleh Alfarrajⁱ, Milan Skalicky^j

^a School of Landscape Architecture and Ornamental Horticulture, Beijing Forestry University, PR China

^b Department of Zoology and Fisheries, Faculty of Agrobiology, Food and Natural Resources, Czech University of Life Sciences Prague, Kamycka 129, 165 00 Praha 6-Suchdol, Czech Republic

^c School of Soil and Water Conservation and Desertification Combating, Beijing Forestry University, PR China

^d Department of Horticulture, Bahauddin Zakariya University, Multan, Pakistan

^e PMAS-Arid Agriculture University, Rawalpindi, Pakistan

^f Institute of Horticultural Sciences, University of Agriculture Faisalabad, Pakistan

^g Beijing Advanced Innovation Center for Food Nutrition and Human Health, College of Food Science and Nutritional Engineering China Agricultural University, Beijing, PR China

^h Dept. of Botany & Microbiology, College of Science, King Saud University, P.O. Box 2455, Riyadh 11451, Saudi Arabia

¹Zoology Department, College of Science, King Saud University, Riyadh 11451, Saudi Arabia

¹Department of Botany and Plant Physiology, Faculty of Agrobiology, Food and Natural Resources, Czech University of Life Sciences Prague, Kamycka 129, 165 00 Prague, Czechia

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This experiment was conducted to investigate the effect of different plant growth regulators as a foliar spray on greenhouse rose cv. *Rosa hybirda* to increase the growth and vase life. Main objective of this research is exogenous application of plant growth regulators that significantly increase the flower quality of rose. A total of 5 treatments $T_1 = \text{control}$, $T_2 = \text{Thiourea} @ 60 \text{ mg/L}$, $T_3 = \text{Ascorbic acid @ 60 mg/L}$, $T_4 = \text{Salicylic acid @ 60 mg/L}$ and $T_5 = \text{Humic acid @ 60 mg/L}$ were tested in complete randomized design with three replications. All the treatments improved the yield, flowering quality and vase life as compared to control treatment. Although, various treatments, salicylic acid showed the maximum plant height (123.5 cm) and days to first flower harvest (87 days). Results declared that foliar spray of plants growth regulators increased the chlorophyll *a*, chlorophyll *b* and total chlorophyll contents. Additionally, flowering parameters like flower stalk length (16.533 cm), fresh weight of flower (15.95 g), dry weight of flower (2.20 g), flower diameter (10.133 cm), number of flowers per plant (10.133), flower quality (4.6) and vase life (2.86 days) were significantly increased in plants sprayed with salicylic acid and humic acid @ 60 mg/L.

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1. Introduction

Abbreviations: HA, humic acid; TU, thiourea total; SA, salicylic acid; AA, ascorbic acid; PGRs, plant growth regulators.

* Corresponding authors.

E-mail address: gaoyk@bjfu.edu.cn (G. Yike).

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Roses are considered important floral crops in the world and used as raw material in various industries like cosmetics and perfume (Ghadimian and Danaei, 2020). The number of species of genus *Rosa* varies among 200 species and it contained more than 35,000 commercial cultivars most of which are hybrids. The perfume, medicine and floriculture industries are mainly depended on the commercial cultivation of Roses (Gil et al., 2019). Besides, its use as ornamental plant, Rose also produces essential oil and vitamin C, which categorically increased its value among cash crops. Plant growth regulators (PGRs) play key role in life cycles of plants and these can be produced naturally by plants or

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synthetically by chemists (Davies, 2013). Different growth factors such as, light and temperature effect on the good quality production. Control and management of these factors are not only difficult but also could be expensive. Plants quickly respond to change in hormonal balance due to growth substances' (PGS). Floriculture industry has been revolutionized through identification of various growth regulators which control growth and flowering for high quality end product (Sajid et al., 2015).

Cut flowers are metabolically active plant organs which face many growth-related issues from primordial initiation to full blooming at morphological, environmental and physiological levels (Aziz et al., 2020). Vase life is prime parameter of interest in cut flower industry and defined as time taken to wilt in water. It has more important role in crop value Effect of plant growth regulator and stimulators on growth and quality of ornamental plants is well documented. Various PGRs are being used commercially in cut flower industry to modify plants behavior like compactness of growth, number and aesthetic value of flowers (Pal, 2019).

Among these, ascorbic acid (AA) is an important chemical which plays significant role in growth, development and quality of cut flowers. It acts as co-factor thus modifying phyto-hormone facilitated signaling involve in shifting of plant from vegetative phase to reproductive phase involving floral initiation, growth and ultimately floral drop (Ghadimian and Danaei, 2020). Exogenous application of ascorbic acid improves the nutrient contents of sweet pepper and significantly enhanced the growth and quality of Italian cypress (Farahat et al., 2007).

(Rajpar et al., 2011) reported that application of humate in plants grown with foliar spray or in nutrients solutions enhanced chlorophyll contents, soil fertility and crop productivity. It stated that photosynthetic capacity of creeping bent grass, marginally stimulated by foliar application of humic acid solution during the summer when even photosynthetic activity declined due to harsh environment.

Exogenous application of Salicylic acid (SA) improved and showed better results such as, heat tolerance, chilling and salt stress in dicotyledons (Basit et al., 2018). Abdi and Karami (2020) expressed that SA brings a substantial increase on the germination and biomass accumulation.

Thiourea has major significant group of non-purines cytokinins and sulf-hydral compound famous for breaking dormancy and regulating the growth, are broadly distributed in plants and regulate growth and phonological development of plants (Srivastava et al., 2011).

Hence, yield and flower growth of rose plants in response to foliar application plant growth regulators (PGR) was evaluated by determining agronomic and physiological parameters to improve the vase life and enhance the productivity of *Rosa hybrida* L.

Very limited information is available regarding effect of exogenous application of growth regulators HA, SA, TU, AA on growth, development and physiology of rose CV rose cheeks. Hence present study was planned to investigate the effect of PGRs on plant growth, vigor, production and quality of flowers in rosy cheeks, when applied exogenously.

2. Materials and methods

This experiment was carried out during 2017–2019 in green house at Beijing Forestry University, Beijing, China. Cutting of Rosy Cheeks was grown in pots. Used pot dimensions were 20 cm diameter and 35 cm in height. The experimental was laid out in triplicated completely randomized design (CRD) consisting of 5 treatments.

Plant growth regulators were applied at three different stages, first 2–3 leaf stage, second 6–7 leaf stage, and third at flowering

stage. Application of plant growth regulators were 60 mg/L of H_2O as (T_1) control (no spray), (T_2) ascorbic acid, (T_3) humic acid, (T_4) thiourea and (T_5) salicylic acid.

2.1. Vegetative and floral examination

The vegetative and floral parameters for each treatment were observed in selected healthy plants such as plant height (cm), stalk length (cm), No of flowers per plant, Fresh and Dry weight of flower (g), flower size and Days to first floral harvest etc. Digital Image Analysis was done as described by O'Neal et al. (2002).

2.2. Vase life of flower (days).

Vase life of flowers was estimated by counting the days taken to observable wilting and twisting of cut stems more than 90° (He et al., 2006).

2.3. Total chlorophyll contents, chlorophyll a, chlorophyll b, carotenoids

Chlorophyll and carotenoids contents were estimated as described by Krishnan et al. (1996). 100 mg of leave sample was mixed with 20 mL of acetone (80%) in graduated tube and incubated at $4C^{\circ} \pm 2C^{\circ}$ in dark place. The mixture was shaken for efficient extraction of pigments. The extract liquid was filtered by glass wool after 48 h of incubation and transferred to another tube by removing leaf pieces. The extracted liquid then was marked volume up to 25 mL with 80% acetone. 3 mL of this solution was taken in a sealed quartz-glass cuvettes and absorption was recorded at 1 nm wavelength in UV visible spectrophotometer (Optizen Pop, Mecasys – Korea). The quantity of pigments was calculated using the equations cited by Dere et al. (1998).

Chlorophyll $a = 11.75 \ A662 - 2.350 \ A645$ Chlorophyll $b = 18.61 \ A645 - 3.960 \ A662$ Chlorophyll total= (Chl.a + Chl.b)

Carotenoids = 1000 A470 – 2.270 Chlorophylla-81.4 Chlorophyllb/227

2.4. Proline ($\mu m \ mg^{-1}$)

Proline content was measured in leaves at three growth stages as described by Bates et al. (1973).

2.5. Phenols (mg GAE/g of extract)

Folin-Ciocalteu method was used to estimate phenolic contents in rose extract (Marquele et al., 2005). 1 mL of extract was mixed with 2 mL of 1 M Folin-Ciocalteu agent and allow to rest for 5 mins. Then 2 mml of sodium carbonate (7.5%) was mixed to the solution and kept for 30 mins at room temperature followed by centrifugation @ 2000 rpm for 10 mins spectrophotometric absorbance of supernatant was noted at 760 nm wavelength. 1 M Folin-Ciocalteu reagent was used as blank. Gallic acid curve was used to estimate the phenolic contents expressed as gallic acid equivalents.

2.6. Flower quality

Flower quality (flower shape, growth of bud, diameter size and color) were evaluated by using method showed by Cooper and Spokas (1991).

2.7. Statistical analysis

The data recorded was subjected to Analysis of variance following Steel *et al.* (1997) through Statistix 8.1. Significance of differences among treatment means was checked at α = 5% using LSD test.

3. Results:

3.1. Vegetative and floral examination

In this experiment, statistically data was recorded that plant height, showed significant variances amongst PGR treatments. Application of both Ascorbic acid and HA caused significant increase in plant height. Among the four levels of PGR in first year maximum plant height was showed T2 (123.5 cm \pm 0.1a) followed by T3 (118.4 cm \pm 0.1a) as compared to T1 (88.5 cm \pm 0.3d). In second year, maximum plant height was observed in T2 followed by T4 as compared to control. The treatment T2 was expressed max flower diameter in first year than control. In second year, max flower diameter revealed by T2 (12.4 cm \pm 0.1a) and min in T1 (5.3 cm \pm 0.2c). In both years flower stalk length, no of flower per plant was recorded highest in T2 as compared to T1 (Fig. 1).

3.2. Fresh and dry weight of flower

Different PGR at same concentration (60 mg L⁻¹) showed max fresh and dry weight of flower as compared to control. The highest

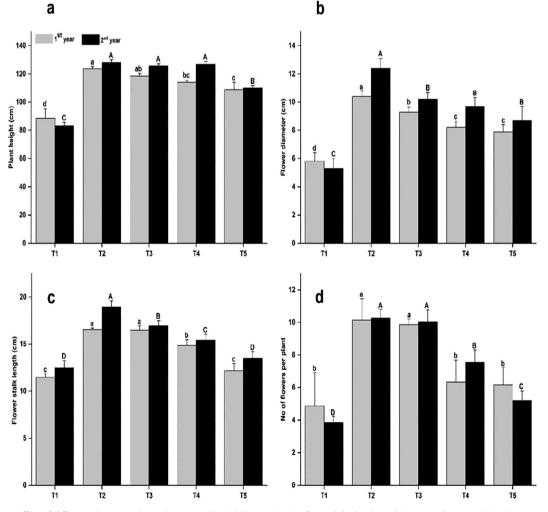
fresh weight of flower exhibited by T2 (15.95 g \pm 0.8a) in first year and T2 (17.45 g \pm 0.8a) in second year. In both years, rose flower treatment T2 showed the max dry weight (2.20 g \pm 0.2a) and (1.7 4 g \pm 0.6a) as compared to control T1.

3.3. Vase life of flower (days)

Statistical analysis of the data for vase life of the spikes revealed significant differences among the PGRs, their concentrations and interaction between these two factors. Both the PGRs significantly improved the vase life of flower as compared to control and were similar in their effect. Among the PGR concentrations, the ascorbic acid @ (60 mg L⁻¹) resulted in maximum vase life of flower in both years. Some PGR increase the vase life but some decrease the vase life of flower. The minimum vase life was recorded in the case of control (T1), followed by (T5) in both years (Table 1).

3.4. Total chlorophyll and carotenoids contents

Statistical analysis of the data for the parameters indicated significant differences among PGRs and their interaction. The chlorophyll content was increased remarkably by the different PGRs at same level applied. Ascorbic acid application resulted in maximum leaf chlorophyll content, followed by the humic acid application in both seasons. However, the control plants (T1) had the minimum chlorophyll contents, carotenoids in their leaves. The concentration of these PGRs (i.e. 60 mg L⁻¹) resulted in significantly higher





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Treatments	Fresh weight of flower (g)		Dry weight of flower (g)		Vase life of flower (days)		Flower quality	
	1st	2nd	1st	2nd	1st	2nd	1st	2nd
T1	10.32 ± 0.7c	10.99 ± 0.4c	1.31 ± 0.1b	1.21 ± 0.04a	2.1 ± 0.3c	1.4 ± 0.4c	3.7 ± 0.2b	3.1 ± 0.5c
T2	15.95 ± 0.8a	17.45 ± 0.8a	2.20 ± 0.2a	1.74 ± 0.6a	2.8 ± 0.09a	4.1 ± 0.6a	4.6 ± 0.2a	5.0 ± 0.6a
T3	13.40 ± 0.3b	14.44 ± 0.8b	2.08 ± 0.04a	1.70 ± 0.2a	2.6 ± 0.09ab	2.8 ± 0.3b	4.1 ± 0.2ab	3.9 ± 0.2bc
T4	12.8 ± 1.0b	12.47 ± 0.5c	1.61 ± 0.3b	1.29 ± 0.1a	2.4 ± 0.2abc	2.5 ± 0.4b	4.1 ± 0.2ab	4.1 ± 0.3ab
T5	10.85 ± 0.6c	11.48 ± 0.7c	1.44 ± 0.04b	1.16 ± 0.05a	2.2 ± 0.1bc	2.4 ± 0.2bc	3.8 ± 0.2b	3.3 ± 0.3bc

Effect of foliar application of Plant growth regulators on flower quality characteristics of Rosa hybrida plants during the 2017/2018 (1st) and 2018/2019 (2nd).

T1 = Control, T2 = Thiourea, T3 = Ascorbic acid, T4 = Salicylic acid, T5 = Humic acid. Means within the same column followed by different letters are significantly different ($P \le 0.05$) and ($P \le 0.01$) based on LSD.

chlorophyll content and carotenoids in T2, T3, followed by T4 in both years' data were recorded. Among the interaction means, results revealed that ascorbic acid used at 60 mg L^{-1} resulted in maximum chlorophyll contents, carotenoids clearly indicating the dominating effect of ascorbic acid and humic acid on the rose plant as compared to control (Table 2).

3.5. Proline ($\mu m \ mg^{-1}$)

Considerable increase in proline contents of leaves was observed in response to exogenous application of plant growth regulators (Table 3). The maximum value for proline contents of leaves was recorded in plants receiving treatments (T2, T3) applied at flowering stage in both seasons (Table 3). The lowest proline content in leaves treatments (T1) followed by (T5) showed in first year values (Table 3). Second years values were recorded min proline content of rose plant in T1 followed T5 (Table 3).

3.6. Phenols (mg GAE/g of extract)

Significant variation was observed in Phenolic contents of rose Flower Extracts in response to treatment with PGRs, ranging from $86.66 \pm 1.2d$ to $101.07 \pm 3.0a$ mg GAE/g. Total phenolic content of second year treatments of the rose flower were measured, ranging from $52.6 \pm 0.1c$ to $73.1 \pm 0.2a$ mg GAE/g (Table 3).

3.7. Flower quality

Quality of cut flower also showed positive response to PGRs application at various stages of growth and development. Maximum value for flower quality index ($4.6 \pm 0.2a$) was noted in response to T2 in comparison to while lowest value ($3.7 \pm 0.2b$) in T1 (control). In second year, highest flower quality ($5.0 \pm 0.6a$) was recorded in T2 as compared to T1. Maximum scores were obtained by T2 followed by T3 and T4 while minimum scores were obtained by T1 (control) in both years (Table.1).

3.8. Days of first flower harvest (days)

Ascorbic acid and humic acid significantly influenced the days of first flower harvest. Among different PGRS treatment revealed maximum days in T2 followed by T3. Data were recorded during the both years of rose plant maximum days $(87.1 \pm 1.9a)$ and $(77.2 \pm 1.9a)$. Whereas, minimum days $(76.6 \pm 1.9c)$ and $(66.6 \pm 1.9c)$ were observed in T1 for *Rose hybrida* (Table 3).

4. Discussion

4.1. Vegetative and floral examination

Positive effect of salicylic acid on plant height, plant vigor, plant biomass accumulation and dry weight has been well documented

Table 2

Effect of foliar application of Plant Growth Regulators on leaf pigments of Rosa hybrida plants during the 2017/2018 (1st) and 2018/2019 (2nd) season.

Treatments	Total Chlorophyll Contents		Chlorophyll a (mg/g fw)		Chlorophyll $b (mg/g fw)$		Carotenoids (mg/g fw)	
	1st	2nd	1st	2nd	1st	2nd	1st	2nd
T1	36.01 ± 1.3d	35 ± 1.6d	14.34 ± 0.4b	13.11 ± 0.1b	21.58 ± 1.0d	22.04 ± 0.8c	0.58 ± 0.05e	0.68 ± 0.06d
T2	61.84 ± 1.2a	60.43 ± 2.1a	23.95 ± 1.3a	22.78 ± 1.3a	38.0 ± 0.62a	38.6 ± 1.4a	1.58 ± 0.04a	2.58 ± 0.02a
T3	53.23 ± 1.5b	54.66 ± 2.0b	23.82 ± 1.1a	22.63 ± 0.9a	32.0 ± 0.6b	33.07 ± 1.3b	1.2 ± 0.05b	1.15 ± 0.03b
T4	49.07 ± 1.3c	48 ± 0.8c	23.11 ± 1.3a	22.36 ± 0.4a	30.72 ± 0.8b	31.26 ± 0.7b	0.91 ± 0.04c	1.05 ± 0.03c
T5	45.78 ± 1.8c	47 ± 1.6c	$14.43 \pm 0.6b$	13.8 ± 0.6b	24.5 ± 1.3c	22.54 ± 0.9c	0.72 ± 0.02d	1.06 ± 0.02bc

T1 = Control, T2 = Thiourea, T3 = Ascorbic acid, T4 = Salicylic acid, T5 = Humic acid. Means within the same column followed by different letters are significantly different ($P \le 0.05$) and ($P \le 0.01$) based on LSD.

Table 3

Effect of foliar application of Plant Growth Regulators on Total Phenols, Proline contents of Rosa hybrida plants during the 2017/2018 (1st) and 2018/2019(2nd) season.

Treatments	Proline ($\mu m m g^{-1}$)		Phenols (mg GAE/g)		Days of first flower harvest (days)	
	1st	2nd	1st	2nd	1st	2nd
T1	216.6 ± 4d	210.6 ± 2.0e	86.66 ± 1.2d	52.6 ± 0.1c	76.6 ± 1.9c	66.6 ± 1.9c
T2	313.3 ± 3.6a	323.3 ± 2.4a	101.07 ± 3.0a	73 ± 0.2a	87 ± 1.9a	77.2 ± 1.9a
T3	283 ± 4.1b	276.3 ± 2.4b	94.10 ± 2.4bc	63.03 ± 0.2b	81.7 ± 1.5b	72.2 ± 0.9b
T4	229.6 ± 4.0c	220 ± 4.0d	97.33 ± 1.6ab	62.43 ± 0.1b	80.9 ± 2.9bc	70.5 ± 1.3b
T5	217.6 ± 3.3d	228 ± 3.2c	92 ± 3.0c	62.43 ± 0.1b	78.1 ± 2.1bc	69.6 ± 1.3bc

T1 = Control, T2 = Thiourea, T3 = Ascorbic acid, T4 = Salicylic acid, T5 = Humic acid. Means within the same column followed by different letters are significantly different ($P \le 0.05$) and ($P \le 0.01$) based on LSD.

in marigold, basil, eggplant, sweet pepper, marjoram and many other ornamental plants (Basit et al., 2018).

Likewise exogenous application of ascorbic acid is reported to increase vegetative growth, flowers per plant, flower weight (fresh and dry) and bio-accumulation of nutrients in Italian cypress, cut gerbera and rose mellow (Farahat et al., 2007; Samaneh et al., 2013). Humic acid is also reported to increase plant height in canola and flower number & quality, dry weight of flower in marigold (Behzad and Zarchini, 2014; Ehsan et al., 2012). Moreover, application of thiourea is documented to increase flower number and quality in oriental lily. Foliar application of humic acid increased the number of flowers and dry weight of marigold (Ehsan et al., 2012). Application of humic acid increased the spike diameter and longer stem in gladiolus (Ahmad et al., 2013). Application of thiourea and salicylic acid increased the diameter of spike and flower stalk length in gladiolus (Pawar et al., 2018).

4.2. Vase life of flower (days)

Results showed that vase life of flower was increased by the application of different plant growth regulators at same concentration. Ascorbic acid enhanced the vase life of cut flower lisianthus (Sheikh et al., 2015). By the application of thiourea (1%) and salicylic acid (150 ppm) longer the vase life of flower in gladiolus (Pawar et al., 2018). HA and NPK application as foliar spray increase the shelf life in Essential. Humic acid act as auxin and enhance uptake & bioaccumulation of nutrients resulting in increased vase life of cut stem of gerbera (Baldotto and Baldotto, 2013).

4.3. Chlorophyll and carotenoids contents

Chlorophyll (a & b) and carotenoids contents increase significantly in response to foliar spray of humic acid. This increase may be attributed to accelerated nitrogen and phosphorus uptake from soil; increased biosynthesis of protein (Haghighi et al., 2012).

Foliar spray of ascorbic acid significantly influenced the total chlorophyll contents, chlorophyll *a*, chlorophyll *b* in *Cupressus sempervirens L* (Farahat et al., 2007). Foliar spray of thiourea helpful to increase the chlorophyll contents of leaves in coriander (Shanu et al., 2013). In *Brassica napus* foliar application of salicylic acid significantly enhanced the cholorophyll contents of leaf (Ghai et al., 2002).

4.4. Proline ($\mu m \ mg^{-1}$)

The foliar application of ascorbic acid increased the proline contents in sugarcane (Batool et al., 2012). By the application of 5% humic acid helpful to increase the proline contents in faba bean (Dawood et al., 2020). Foliar application of salicylic acid helpful to increase the proline contents in linseed (Bakry et al., 2012). Taramani et al. (2020) revealed that thiourea gives beneficial response to proline contents in maize and wheat.

4.5. Phenols (mg GAE/g of extract)

Application of 5% humic acid was helpful to increase the phenolic contents in faba bean (Dawood et al., 2020). Perveen et al. (2016) reported that thiourea increased the total phenolic contents in mung bean. Ascorbic acid at 300 ppm concentration increased the phenols in zinnia (Naglaa et al., 2018). Foliar application of Salicylic acid improved the phenols contents in maize plant (Amin et al., 2013).

4.6. Flower quality

Similarly, thiourea treatment causes internal physiological changes like assimilate distribution within the plant system which resulted in to an improvement in the quality of gladiolus flowers due to which turgidity of spikes was maintained and vase life of cut flower might have been increased (Hatamzadeh et al., 2012). Humic acid application with NPK to enhance the flower quality of gladiolus flower (Ahmad et al., 2013). Salicyclic acid and ascorbic acid significantly improved the flower quality of gladiolus (Ravanbakhsh et al., 2017).

4.7. Days to first flower harvest

Regardless, with the application of different PGR reduce the days to flower harvest. Humic acid implication proved earlier emergence of spike and more florets per spike in gladiolus (Ahmad et al., 2013). Thiourea applied at 1000 ppm concentration reduces the days to flower harvest in tuberose (Patil and Jadhav, 2010). Foliar spray of salicylic acid significantly affect the days of flowering in aster as compared to control (Kumar et al., 2017).

5. Conclusion

Vegetative growth, floral quality, vase life of flower, proline, phenols, and carotenoids contents are the important parameters that determine the quality of economical produce. The results of present have showed that quantity and quality of rose production could be increased significantly through exogenous application of different growth regulators. However, further investigation is needed for standardization of concentration and time of application to get maximum economic benefits. Moreover, efficacy of these plant growth regulators in different environments also demands for further studies. Plant growth regulators helpful to better the quality and shelf life of cut flower cultivation.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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