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EFFECT OF SOME HORMONAL TREATMENTS ON GROWTH AND CHEMICAL COMPOSITION OF *RHAPHIOLEPIS UMBELLATA* SHRUB

F.M. Saadawy; A.S. El-Fouly and A.W. Sayed

Ornamental Plants and Landscape Gardening Res. Dept., Hort. Res. Inst., ARC, Giza, Egypt

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ABSTRACT: A pot experiment was consummated on *Rhaphiolepis umbellata* seedlings grown under seran greenhouse at Al-Zohriya Garden, Hort. Res. Inst., ARC, Giza, Egypt during the two consecutive seasons 2017 and 2018, to study effects of five foliar spray treatments of a mixture containing three growth regulators (PGR), at the same concentration for each, on plant growth and chemical composition. The three-tested PGR were NAA, GA_3 and BA, while the five concentrations for each were 0 (control treatment), 100, 200, 300 and 400 ppm. Obtained results indicated that, generally, all recorded vegetative and root growth traits*;* i.e., plant height, stem diameter, numbers of branches and number of leaves/plant, leaf area, root length and number of roots/plant increased significantly under the effect of all tested PGR treatments, compared to the control plants. The highest increases in this regard were recorded in plants sprayed with PGR at 200 ppm, while the least values were found in the untreated control plants, followed by plants sprayed with the highest PGR concentration of 400 ppm for the two seasons. The same trend was noticed in respect of leaf chemical constituents, as plants that recorded the highest vegetative and root growth (treated 200 ppm PGR) also contained the highest values of total chlorophyll and carotenoids (mg/g f.w.) as well as percentages of total carbohydrate, N, P and K in leaf tissues. Accordingly, it could be recommended to apply the mixture of these three growth regulators 200 ppm on seedlings of *Rhaphiolepis umbellata*, two times monthly during the growing season to get the best plant growth and quality.

Key words: *Rhaphiolepis umbellata*, foliar spraying, plant growth regulators, NAA, GA_3, BA.

INTRODUCTION

Ornamental plants hold an important status in the horticultural industry of the world (Sajjad *et al.*, 2017). The flowering shrub *Rhaphiolepis umbellata* is one of the rarest unknown plants despite the beauty of its flowers and vegetative growth. Its growth is very slow. It belongs to Fam. *Rosaceae*, native to Korea and Japan. The common name hawthorn is often called "Yeddo hawthorn". This species vary in size, some reaching 1.0-1.5 m.

The exogenous application of plant growth regulators plays an important role in the modern production of ornamentals as they help to improve the different economically important and market desirable characteristics of them. The use of plant growth regulators is being widely practiced by the commercial growers of ornamental plants as a part of cultural practice. Bergstrand (2017) reported that chemical plant growth regulators (PGRs) are used in the production of ornamental potted and bedding plants, as growth control is needed for maximizing production per unit area, reducing transportation costs and to obtain a desired visual quality. Plant growth regulators consist of a large group of naturally occurring or synthetically produced organic chemicals and considered as helping tool in ornamentals production system. There are various factors contributing to the efficacy of plant growth regulators.

Benzyladenine (BA) is a synthetic cytokinin comprises the N6-substituted aminopurine. It stimulates protein synthesis. It is perhaps for this reason can promote the maturation of chloroplasts (Taiz and Zeiger, 2002). Taiz and Zeiger (2010) stated that although apical dominance may be determined primarily by auxin; physiological studies indicated that cytokinins play important role in initiating the growth of lateral buds. Direct application of cytokinins to the axillary buds of many species stimulates cell division activity.

Gibberellic acid (GA_3) regulates stem elongation, stimulates plant growth and development, also, it plays important roles in transition from meristem to shoot growth, juvenile to adult leaf stage, vegetative to flowering and determines sex expression along with an interaction of different environmental factors, viz. light, temperature and water (Gupta and Chakrabarty, 2013).

Naphthaleneacetic acid (NAA) is an organic compound with the formula $C_{10}H_7CH_2CO_2H$. NAA is a plant growth regulator belongs to the auxin group and it is an ingredient in many commercial postharvest horticultural products. It is also a rooting agent, used for enhancing vegetative propagation of plants by stem and leaf cuttings (Dimitrios *et al.*, 2008 and Mohammed *et al.*, 2009).

This research aimed to overcome the problem of slow growth in *Rhaphiolepis umbellata*, which limits its spread in gardens and nurseries despite the beauty of flowers and vegetative growth.

MATERIALS AND METHODS

A pot experiment was consummated on *Rhaphiolepis umbellata* plant grown under seran conditions at Al-Zohriya Garden, Hort. Res. Inst., ARC, Giza, Egypt during 2017 and 2018 consecutive seasons. The research aimed to study effects of five foliar spray treatments of mixture containing three plant growth regulators (PGR) of NAA, GA_3 and BA at the same concentration for each, on plant growth and quality as well as its chemical composition. The tested PGR concentrations were 0, 100, 200, 300 or 400 ppm for each regulator.

On May 1st of the two experimental seasons, three-month-old uniform seedlings of *Rhaphiolepis umbellata* were carefully transplanted in 20-cm-diameter plastic pots filled with 3.4 kg potting mixture containing sand + clay at 1:1 (v/v), analysis of which is shown in Table (a).

Five foliar spray treatments of PGR mixture containing NAA (soluble in water), GA_3 and BA at equal concentrations for each were tested as follows:

- Tap water (PGR 0) as a control.
- PGR (NAA, GA_3 and BA) at 100 ppm for each.
- PGR (NAA, GA_3 and BA) at 200 ppm for each.
- PGR (NAA, GA_3 and BA) at 300 ppm for each.
- PGR (NAA, GA_3 and BA) at 400 ppm for each.

Table a. Physical and chemical analysis of the used potting mixture (sand+ clay 1:1 v/v) during 2017 and 2018 seasons.

Particle size distribution (%)				E.C. (ds/m)	pH	Cations (meq./l)				Anions (meq./l)			
Sand	Silt	Clay	CaO_3			Ca^{2+}	Mg^{2+}	Na^+	K^+	HCO_3^{2-}	CO_3^{2-}	SO_4^{2-}	Cl^-
69.9	20.6	9.5	25.0	6.6	7.88	17.8	14.2	33.1	0.9	2.1	-	5.5	85.4

Method of preparing PGR mixture:

The PGR solution was prepared by solving a known weight of BA (0.1, 0.2, 0.3, and 0.4 g for the four concentrations used in this study) in a few drops of HCl acid (0.5 N) to facilitate solving, then completed with tap water to the volume of 1000 ml. NAA and GA_3 (as quick solvents in water) were added thereafter (at the abovenamed weights) to the BA solution (1000 ml), shaked-up, and then the final solution containing BA, NAA, and GA_3 (at equal concentration for each) was sprayed on plant foliage till run-off after adding a little of spreading material.

Plants were treated two weeks after transplanting (on May 15th for the two tested seasons) with foliar spraying two times monthly. Spraying was achieved using hand-held sprayer to achieve coverage of the leaves and stems to enhance plant growth throughout the experimental period.

Treatments were set up in simple experiment with the above-mentioned five treatments of PGR foliar sprays in a completely randomized design. Treatments were replicated thrice, and each replicate contained three plants. Plants of all treatments were fertilized one time monthly with the irrigation process using a commercial fertilizer containing NPK (20:20:20) at 1 g/l till the end of experiment on November 15th. All plants received the usual necessary agricultural practices whenever needed.

Recorded data:

On November 15th, for the two tested seasons, the experiment was terminated and the following data were recorded:

Vegetative growth was assessed as plant height (cm), stem diameter (cm), numbers of branches and number of leaves/plant, leaf area (cm^2) root length (cm), in addition to fresh and dry weights of top growth and roots/plant). Photosynthetic pigments of total chlorophyll (a + b) and carotenoids (mg/g f.w.) were determined in fresh leaf samples according to the method described by Moran (1982). Total carbohydrates content, and percentages of N, P and K were determined in dried leaf samples at 70 $^{\circ}$C for 72 hours according to methods of Dubois *et al.* (1966), Pregl (1945), Wide *et. al.* (1985) and Jackson (1973), respectively.

Statistical analyses:

Collected data were subjected to statistical analyses using SAS Institute Program (2009) and the differences among treatments were compared according to Duncan's New Multiples Range Test. (Steel and Torrie, 1980).

RESULTS AND DISCUSSION

Effect of PGR treatments on:

1. Vegetative and root growth parameters:

Data in Tables 1 and 2 and Fig. 1 cleared that in general, values of all shoot and root growth traits (plant height, stem diameter, numbers of branches and leaves/plant, leaf area, root length, number of roots/plant, in addition to fresh and dry weights of top and root growth) increased significantly under the effect of all tested PGR's treatments compared to the control. Superiority in all characters in the two seasons was confined to using PGR's at 200 ppm, followed by PGR's at 300 ppm, in both seasons. PGR's at 100 and 400 ppm acquired the third and the fourth positions, respectively.

2. Leaf chemical composition:

Data in Tables 3 and 4 exhibited that all tested PGR treatments markedly increased total chlorophylls and carotenoid pigments (mg/g f.w.) as well as percentages of total carbohydrate, N, P and K in leaf tissues. The highest values in this respect were recorded in plants received PGR's at 200 ppm, followed by PGR's at 300 ppm. On the other hand, the lowest contents were found in plants sprayed with PGR's at 100 ppm and PGR's at 400 ppm. These results were confirmed during the two experimental seasons.

Table 1. Effect of foliar spray of plant growth regulators on some vegetative and root growth traits of *Rhaphiolepis umbellata* plant during 2017 and 2018 seasons.

Treatments (PGR levels)	Plant length (cm)	Stem diameter (cm)	branches No./plant	leaves No./plant	leaf area (cm^2)	root length (cm)	roots No./plant
			First season: 2017				
Control	13.13 d	0.28 d	1.00 c	15.67 c	5.68 d	17.94 d	12.67 d
PGR 100 ppm	22.74 bc	0.40 c	2.00 b	26.67 b	12.03 b	29.83 b	22.33 bc
PGR 200 ppm	35.37 a	0.73 a	4.33 a	61.33 a	14.52 a	34.47 a	36.33 a
PGR 300 ppm	29.33 ab	0.57 b	2.33 b	53.67 a	11.24 b	29.30 b	27.67 b
PGR 400 ppm	20.00 cd	0.33 cd	1.00 c	23.33 bc	8.09 c	25.33 c	20.00 c
			Second season: 2018				
Control	15.74 e	0.30 d	1.00 c	19.67 d	6.26 c	20.27 d	13.33 d
PGR 100 ppm	23.80 c	0.42 c	2.00 b	33.67 c	12.06 b	48.10 a	23.33 bc
PGR 200 ppm	35.57 a	0.77 a	5.00 a	74.00 a	14.63 a	39.73 ab	39.67 a
PGR 300 ppm	32.17 b	0.63 b	2.67 b	54.67 b	13.51 a	31.00 bc	28.33 b
PGR 400 ppm	20.97 d	0.41 c	1.00 c	27.00 cd	11.86 b	25.97 cd	21.33 c

Means having the same letter/s within a column are not significantly different according to Duncan's Multiple Range Test at 5% level. PGR = NAA, GA_3 and BA.

Table 2. Effect of foliar spray of plant growth regulator treatments on top and root fresh and dry weights of *Rhaphiolepis umbellata* plant during 2017 and 2018 seasons.

Treatments (PGR levels)	Top fresh weight (g)	Top dry weight (g)	Root fresh weight (g)	Root dry weight (g)
		First season: 2017		
Control	8.00 d	1.23 d	1.14 d	0.34 c
PGR 100 ppm	15.51 c	4.09 c	1.69 cd	0.87 c
PGR 200 ppm	40.84 a	14.57 a	10.74 a	5.40 a
PGR 300 ppm	30.20 b	11.45 b	6.95 b	3.25 b
PGR 400 ppm	12.98 c	4.23 c	3.34 c	0.83 c
		Second season: 2018		
Control	8.87 d	1.51 c	1.15 d	0.61 c
PGR 100 ppm	16.51 c	4.70 b	1.82 cd	0.96 c
PGR 200 ppm	42.67 a	15.01 a	11.63 a	5.83 a
PGR 300 ppm	31.06 b	13.46 a	7.37 b	3.29 b
PGR 400 ppm	12.94 cd	7.03 b	3.69 c	0.86 c

Means having the same letter/s within a column are not significantly different according to Duncan's Multiple Range Test at 5% level. PGR = NAA, GA_3 and BA.

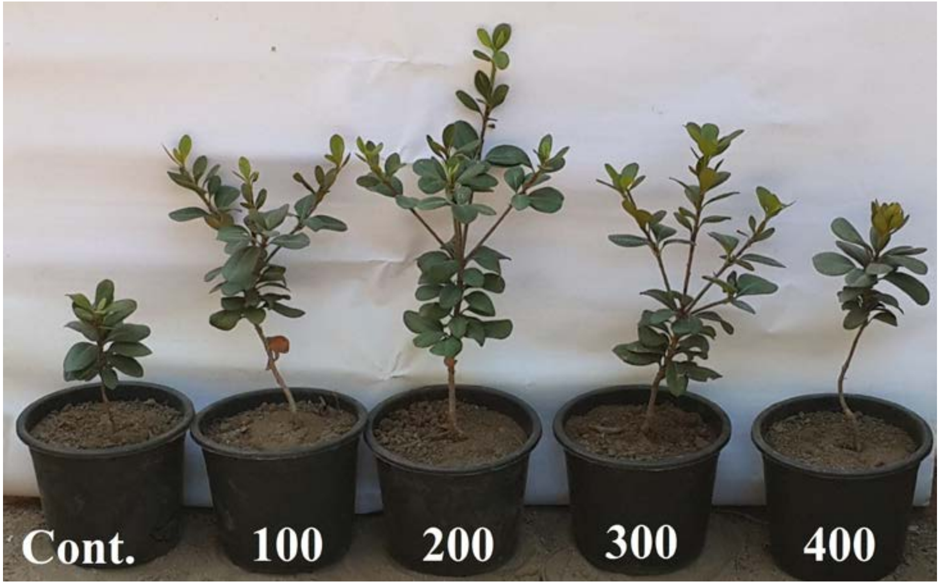

Fig. 1. Effect of foliar spraying of plant growth regulators of NAA, GA_3 and BA on *Rhaphiolepis umbellata* seedlings growth.

Treatments from left to right: control, plants sprayed with PGR at 100 ppm for each, spray with PGR at 200 ppm for each, spray with PGR at 300 ppm for each and spray with PGR at 400 ppm for each.

Table 3. Effect of foliar spray of plant growth regulator (PGR) treatments on leaf contents of total chlorophyll and carotenoids contents in leaf tissues of *Rhaphiolepis umbellata* during 2017 and 2018 seasons.

Treatments (PGR levels)	Total chlorophyll content (mg/g f. w.)	Carotenoids content (mg/g f. w.)
	The first season: 2017	
Control	2.05 e	0.27 b
PGR 100 ppm	2.37 c	0.39 ab
PGR 200 ppm	3.20 a	0.41 a
PGR 300 ppm	2.56 b	0.41 a
PGR 400 ppm	2.31 d	0.29 ab
	The second season:2018	
Control	2.48 e	0.28 c
PGR 100 ppm	2.69 c	0.44 b
PGR 200 ppm	3.28 a	0.49 a
PGR 300 ppm	2.79 b	0.49 a
PGR 400 ppm	2.64 d	0.43 b

Means having the same letter/s within a column are not significantly different according to Duncan's Multiple Range Test at 5% level. PGR = NAA, GA_3 and BA.

Table 4. Effect of foliar spray of plant growth regulator (PGR) treatments on some leaf chemical composition of *Rhaphiolepis umbellata* during 2017 and 2018 seasons.

Treatments (PGR levels)	Total carbohydrate %	Nitrogen %	Phosphorus %	Potassium %
The first season: 2017				
Control	13.95 d	1.20 d	0.14 c	1.24 c
PGR 100 ppm	21.15 b	1.61 bc	0.25 b	1.65 ab
PGR 200 ppm	24.47 a	2.17 a	0.43 a	1.82 a
PGR 300 ppm	22.26 ab	1.75 b	0.31 b	1.65 ab
PGR 400 ppm	18.28 c	1.50 c	0.16 c	1.40 bc
The second season: 2018				
Control	16.89 e	1.37 c	0.46 d	2.11 d
PGR 100 ppm	25.41 c	1.63 c	0.80 bc	2.28 bc
PGR 200 ppm	33.21 a	2.42 a	1.00 a	2.46 a
PGR 300 ppm	29.17 b	2.03 b	0.86 ab	2.41 ab
PGR 400 ppm	21.46 d	1.50 c	0.65 c	2.19 cd

Means having the same letter/s within a column are not significantly different according to Duncan's Multiple Range Test at 5% level. PGR = NAA, GA_3 and BA.

DISCUSSION

Khandaker *et al.* (2017) found that the application of 25 mg/l NAA significantly increased plant height, number of roots and total soluble solid content of *Mokara* Chark kuan orchid plants. However, there was no significant effect on number of leaves, chlorophyll content and number of flowers. Hakan and Kerim (2013) found that auxin group hormones (IAA, IBA, and NAA) affected morphological characteristics of the newly generated plants, especially root generation. Nisha *et al.* (2012) studied the potential effect of benzylaminopurine (BAP) on *Dendrobium hybrid* (*Dendrobium Angel* White). Plantlets were subjected to spray containing different BAP concentrations. They found that the application of BAP increased number of produced leaves per plant. Currey and Erwin (2012) assessed the efficacy of foliar spray applications of benzyladenine (75-300 ppm) on stem elongation and branching of 11 kalanchoe species. They observed that benzyladenine increased number of branches for several species. Gupta and Chakrabarty (2013) stated that gibberellic acid is a plant hormone enhances plant growth and development. Gibberellins stimulate seed germination, transitions from meristem to shoot growth, juvenile to adult leaf stage and vegetative to flowering as well as determine sex expression and grain development along with an interaction of different environmental factors, viz. light, temperature and water. The major site of bioactive GA is stamens that influence male flower production and pedicel growth. Malgorzata and Małgorzata (2013) studied the effect of gibberellic acid (GA_3) on growth and flowering of *Ajania pacifica* 'Bea'. They observed that double spraying with GA_3 accelerated buds development, thus shortened the cultivation time by about two days. Treatment with GA_3 at 500 mg/l stimulated shoot elongation more than with GA_3 at 250 mg/l. Abdel-Moniem *et al.* (2014) advised to spray the foliage of spindle tree (*Euonymus japonicus* cv. Aureus) with GA_3 at 300 ppm combined with a liquid fertilizer at 2 ml/l for the best growth performance of such foliage pot plant. Also, they noticed that all vegetative and root growth parameters improved markedly with individual application of GA_3 at 300 ppm, as this treatment gave higher growth compared to the control plants. Spraying with or GA_3 at 200 ppm improved some growth traits. Sardoei (2014) evaluated the effect of GA_3 and BAP, at 0, 100 and 200 mg/l on *Ficus benjamina*, *Schefflera arboricola* and *Dizigotheca elegantissima* plants at pot cultivation conditions. He found that the tallest plants belonged to 200 mg/l GA_3 + 200 mg/l BAP, while the shortest ones belonged to the control treatment.

Accordingly, it could be recommended to spray three-months-old seedlings of *Rhaphiolepis umbellata* with a mixture of these three growth regulators at 200 ppm for each, two times monthly to get the best plant growth and quality.

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تأثير بعض المعاملات الهرمونية على النمو والتركيب الكيماوى لشجيرة الرافيوليبيس (*Rhaphiolepis umbellata*)

فيصل محمد عبد العليم سعداوي، أمل صلاح الفولي، أحمد وهبه سيد

قسم بحوث الزينة وتنسيق الحدائق، معهد بحوث البساتين، مركز البحوث الزراعية، الجيزة، مصر

أُجريت تجربة أصص على نبات الرافيوليبيس (*Rhaphiolepis umbellata*) النامي تحت الصوبة السيران فى المزرعة التجريبية لحديقة الزهرية، معهد بحوث البساتين، مركز البحوث الزراعية، الجيزة، مصر خلال الموسمين المتتاليين ٢٠١٧ ، ٢٠١٨ لدراسة تأثيرات خمس معاملات رش ورقي بخليط محتوي على ثلاث منظمات نمو نباتية (بتركيز متساوي لكل منها) على نمو النباتات وتركيبها الكيمياوى، كانت الثلاث منظمات نمو المختبرة هي نفثالين حامض الخليك NAA، حامض الجبرليك GA_3، وبنزيل الأدنين BA، وكانت التركيزات الخمسة هي صفر(معاملة المقارنة)، ١٠٠، ٢٠٠، ٣٠٠، ٤٠٠ جزء فى المليون لكل منها. أشارت النتائج المتحصل عليها بوجه عام إلى زيادة معنوية لقيم كل القياسات المسجلة على خصائص النمو الخضري والجذري (إرتفاع النبات، قطر الساق، أعداد الأفرع والأوراق لكل نبات، متوسط مساحة الورقة، طول الجذر، وعدد الجذور الناتجة لكل نبات) تحت تأثير كل معاملات منظمات النمو المُختبرة مقارنة بنباتات المقارنة، وسُجلت أعلى زيادات بهذا الخصوص في النباتات التي تم رشها بمنظمات النمو بتركيز ٢٠٠ جزء فى المليون بينما وُجدت أقل القيم فى نباتات المقارنة الغير معاملة بمنظمات النمو وتليها النباتات المعاملة بالتركيز المرتفع من منظمات النمو (٤٠٠ جزء في المليون) خلال الموسمين، وقد لوحظ نفس الإتجاه بخصوص المكونات الكيمياوية للأوراق، حيث إحتوت النباتات التى سجلت أعلى نمو خضري وجذري أيضاً على أعلى القيم لمحتوى الكلوروفيل الكلي والكاروتينويدات وأعلى نسب مئوية للكربوهيدرات الكلية، النيتروجين، الفوسفور والبوتاسيوم بأنسجة الأوراق. وبناء عليه، يمكن التوصية برش شتلات الرافيوليبيس (*Rhaphiolepis umbellata*) بخليط من منظمات النمو الثلاثة (PGR) بتركيز ٢٠٠ جزء فى المليون لكل منها، مرتين شهرياً خلال موسم النمو للحصول على أفضل نمو وجودة للنبات.