# GIBBERELLIC ACID (GA3 AND NPK NANO-FERTILIZERS CAN ENHANCE THE GROWTH AND DEVELOPMENT OF *BUXUS* SEMPERVIRENS 'SUFFRUTICOSA' PLANT

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#### ABSTRACT

This study was conducted in the greenhouse of the College of Agricultural Engineering Sciences, University of Duhok for the period between 15<sup>th</sup> March 2022 to 15<sup>th</sup> March 2023. The study aimed to test the effect of Gibberellic acid (GA<sub>3</sub>) with three concentrations (0, 200, 400) mg  $L^{-1}$ , and (1, 2, 4) g  $L^{-1}$  of NPK Nano fertilizers sprayed two times on the growth and development of Buxus sempervirens 'Suffruticosa' plant and the experimental units were organized by using (RCBD) Randomized Complete Block Design, with two factors  $3 \times 3 = 9$  treatments and three replications then taking three plants for each replication. The results showed that the 400 mg  $L^{-1}$  of GA<sub>3</sub> significantly increased the number of branches, plant weight, number of leaves, growth index and leaf area compared with 200 mg L<sup>-1</sup> which resulted in increasing plant height, vegetative dry weight, root dry weight, total chlorophyll content compared with control. Increasing of Nano fertilizer concentration up to 4 g L<sup>-1</sup> significantly increased the plant height, number of branches, plant weight, number of leaves, vegetative dry weight, root dry weight also leaf area compared with 2 g L<sup>-1</sup> which affected on increasing total chlorophyll content. The highest branches number (17.33) branch/plant, highest plant weight (189.20) g, number of leaves (143.33) leaf/plant, vegetative dry weight (42.30) g leaf area (2755.00) cm<sup>2</sup> was obtained from the interaction of (4 g L<sup>-1</sup> + 400 mg L<sup>-1</sup>) of NPK Nano fertilizer and GA<sub>3</sub> while plant height gave the same result (58.67) at both concentration (4 g L<sup>-1</sup> + 200 mg L<sup>-1</sup>) and (4 g L<sup>-1</sup>  $^{1}$  + 400 mg L<sup>-1</sup>), but in other side maximum total chlorophyll and growth index were from interaction of (2 g L<sup>-1</sup> + 200 mg L<sup>-1</sup>) and gave (87.97), (74279) respectively. Overall, the interaction between (gibberellin at 400 mg L<sup>-1</sup> + Nano-NPK fertilizer 4 g L<sup>-1</sup>) provided best results of most studied characteristics. Also, both factors individually provided best results when 400 mg L<sup>-1</sup> gibberellin and 4 g L<sup>-1</sup> Nano- NPK fertilizer were used.

*KEY WORDS:* Buxus SPP., Gibberellic acid Nano- fertilizers, Seedling growth, Development, NPK Dormancy

### INTRODUCTION

uxus sempervirens L. 'Suffruticosa' D plants belong to Buxaceae family which identified as a member of the genus Buxus that contains around 70 species. It is considered as an evergreen sclerophyllous that developed into smaller trees or shrubs, they have ability to grow in different environmental condition from deep shade of the forest beech to high elevation in the Mediterranean mountains (Koldobika et al.,2005). Buxus spp. usually at the beginning of spring can produce only single flush of growth after that other signs of growth and development occur like bud break and stem elongation while at other times of the year the plant will stay at dormant condition (Musselwhite et al., 2004). The dormancy occurs inside plant structure they

organized by physiological factors which are due to outside condition like what happens with apical para-dormancy, the dormancy happens due to the endogenous condition of plants itself because hormones levels control and coordinate the metabolism process, growth and development of plant parts (**Taiz** *et al.*, **2002**).

Gibberellic Acid (GA) is a universal plant growth regulator involved in many development processes. It is worth noting their participation in xylogenesis, shoot elongation, root development, flowering and seed germination. However, in most cases, the activity of GA seems to depend on its balance with ABA. Thus, future studies should focus not only on GA modulation itself, but also on its relationship with ABA, since the ABA/GA balance is a major modulator of physiological responses (**Castro-Camba,2022**). Exogenous GA3 and GA4 significantly accelerated bud dormancy release and subsequent growth by increasing the contents of endogenous bioactive GAs, IAA, and soluble glucose such as fructose and trehalose, and accelerated cell cycle process, accompanied by decreasing ABA contents and GA3 had a more effective ability to induce cell division and starch hydrolysis (Yuxi et al., 2021). found Mohammed (2017)that Buxus sempervirens 'Suffruticosa' plant which was sprayed with a treatment combination of GA3 and BA with concentration (1000,1000) mg  $L^{-1}$  gave the highest plant height (35.39) cm, total chlorophyll content (75.21) SPAD, plant growth index (7407.87) cm<sup>3</sup>, vegetative dry weight (5.48)g, root dry weight (3.12) g with significant deference than other treatments.

Nano-NPK fertilizers are an important component in agriculture for improving crop growth, yield and quality parameters with increasing nutrient use efficiency, reducing fertilizer wastage and growing costs. Nano fertilizers increase crop growth to optimal concentrations. Further increases in concentrations may slow crop growth due to nutrient toxicity. Nano fertilizers provide more surface area for various metabolic reactions in the plant, which increases the rate of photosynthesis and produces more dry matter and yield. It also protects the plant from various biotic and abiotic stresses. (Singh et al., 2017). It is considered a new technique to improve and promote agricultural products with better quality, environmental integrity, more biological support and also monetary confirming, because Nanohyper fertilizers have very small particles therefore they have very high surface area to accelerate metabolic process, and rise of particles in unit area that facilitate their penetration (Okram, et al., 2023). Nitrogen supplies plant with suitable nutrients which are very important for its growth and development by increasing nutrient uptake and rise production. (Mirbakhsh, 2023). The plant has a seed dormancy for this reason it needs special treatments for overcoming dormancy in the seed like presoaking the seed in GA<sub>3</sub> and potassium nitrate solution (0.01 and 0.02 %) for about 24hr. 48hr or through pre-chilling (stratification)treatments for about 30 days or storing at(-20c) (Manisha, 2019). The plant has summer dormancy issues which can be overcome through using different treatments like adding fertilizers NPK, spraying plants with hormones for example (GA<sub>3</sub>), promalin and pinching process, which will make the growth and

development of the plant become better. This research aimed to cut the lone period of dormancy of this plant and increase its vegetative growth by using Nano NPK fertilizer and GA3.

# MATERIAL AND METHODS

This experiment was conducted at the University of Duhok-College of Agricultural Engineering Sciences inside a greenhouse between the period 15th March 2022 and 15th March 2023. In this study, three concentrations of NPK Nano fertilizers (1, 2, 4) g L<sup>-1</sup> were used and sprayed twice monthly, the first spray after one month of transplant and the second after two weeks from the first spray. Also, it included three concentrations of GA<sub>3</sub> (0, 200, 400) gm  $L^{-1}$  that were sprayed like Nano fertilizer twice monthly but there was one week between each factor. NPK was sprayed first then after a week GA<sub>3</sub> was sprayed. The transplants were brought from the University of Duhok Nursery then they were transported to pots with size (3 L) that were filled with a mixture of compost and loamy soil (1:1) by volume after that all the agricultural operations were done to the plants for example, irrigation and hoeing. The study was performed by using a Randomized Complete Block Design (RCBD) with two factors  $3 \times 3 = 9$  treatments and three replications and three plants for each replicate, the total number of plants was (81) plant. The studied parameters included, plant height (cm), number of branches (branches /plant), plant weight (g), number of leaves (leaves/plant), Vegetative dry weight (g), roots dry weight (g), total chlorophyll content (SPAD), plant growth index (cm<sup>3</sup>) through using equation [plant growth index  $(cm^3) = 3.14\{1 \mid 2 \times (less width)\}$ +large width) $2^2$ ×plant hight] and leaves area  $(cm^{3)}).$ 

# **RESULTS AND DISCUSSION**

The results in table (1) show that increasing GA<sub>3</sub> to 200 mg L<sup>-1</sup> increased significantly the plant height to (51.78) cm, roots dry weight of (53.05) g and total chlorophyll (83.50) SPAD compared with other treatments. And during increasing GA<sub>3</sub> to the highest concentration 400 mg L<sup>-1</sup> increased significantly the number of branches to (10.44) branch/plant, number of leaves (123.44) leaf/plant, plant growth index (52828) cm<sup>3</sup> and leaf area (2307.00) cm<sup>2</sup> compared to with the other treatments. As well as,

the two concentrations 200 and 400 mg  $L^{-1}$  of GA<sup>3</sup> increased significantly the plant weight to (174.27, and174.28) g and vegetative dry weight (37.34, 36.22) g respectively when compared with the control (0) mg  $L^{-1}$ .

Concerning the effect of Nano-NPK fertilizer in the same table (table 1) it is obvious that the most of studied characteristics were significantly increased as a result of increased the concentration of fertilizer to (4) g L<sup>-1</sup> such as plant height, number of branches, plant weight, number of leaves, vegetative dry weight, and leaf area which gives 55.89 cm, 12.00 branches, 181.71 g, 134.44 leaf/plant, 39.09 g, and 2392.30 cm<sup>2</sup> respectively, comparison with the lowest values (41.11 cm, 6.22 branches, 153.58 g, 107.67 leaf/plant, 33.80 g, and 1950.48 cm<sup>2</sup>) respectively for the lowest concentration (1) g L<sup>-1</sup>. While the growth index gave a maximum value of 63254 cm<sup>3</sup> when (2) g L<sup>-1</sup> was used. On the other side the root dry weight and total chlorophyll increased significantly when treated seedlings with 2 and 4 g L<sup>-1</sup> and provided (49.99, 52.61 g) and (80.52, 79.41 SPAD) respectively for the two characteristics comparison with the lowest value (43.58 g and 73.98 SPAD) for treated seedling with 1 g L<sup>-1</sup>.

Table (1): Effect of Nano-NPK fertilizer and GA3 on th	growth and development of <i>Buxus sempervirens</i> plant.

Characteristics	GA₃ mg L⁻¹			Nano NPK fertilizer g L <sup>-1</sup>			
	0	200	400	1	2	4	
Plant height (cm)	43.56°	51.78ª	47.78 <sup>b</sup>	41.11°	46.11 <sup>b</sup>	55.89ª	
Number of branches branch/plant	7.11 <sup>b</sup>	7.67 <sup>b</sup>	10.44 <sup>a</sup>	6.22 <sup>b</sup>	7.00 <sup>b</sup>	12.00ª	
Plant weight (g)	156.86 <sup>b</sup>	174.27ª	174.28ª	153.58°	170.12 <sup>b</sup>	181.71ª	
Number of leaves (leaf/plant)	113.56°	118.11 <sup>b</sup>	123.44ª 107.67 <sup>c</sup>		113.00 <sup>b</sup>	134.44ª	
Vegetative dry weight (g)	33.38 <sup>b</sup>	37.34ª	36.22ª	33.80 <sup>b</sup>	34.06 <sup>b</sup>	06 <sup>b</sup> 39.09 <sup>a</sup>	
Root dry weight (g)	45.78 <sup>b</sup>	53.05ª	47.35 <sup>b</sup>	43.58 <sup>b</sup>	49.99ª	52.61ª	
Total chlorophyll (SPAD)	72.28°	83.50ª	78.13 <sup>b</sup>	73.98 <sup>b</sup>	80.52ª	79.41 <sup>a</sup>	
Plant growth index (cm <sup>3</sup> )	46935°	49440 <sup>b</sup>	52828ª	42539 <sup>b</sup>	63254ª	43411 <sup>b</sup>	
Leaves area (cm <sup>2</sup> )	1976.51°	2101.72 <sup>b</sup>	2307.00 <sup>a</sup>	1950.48 <sup>b</sup>	2042.46 <sup>b</sup>	2392.30ª	

Means with the same letter for each factor and for each characteristic are not significantly different at 5% level based on DMRT.

Means with same letter for the interaction between the factors for each characteristic are not significantly different at 5% level based on DMRT.

From the data of interaction in table (2) between the two studied factors GA3 and Nano-NPK fertilizer it is obvious that the significant maximum plant height (58.67 cm) was found from two interactions between two treatments both (GA3 at 200 mg.L<sup>-1</sup> and GA3 at 400 mg.L<sup>-1</sup> with 4 g.L<sup>-1</sup> Nano-NPK fertilizer) comparison with minimum height (37.33 cm) that were shown from interaction between (GA3 at 0 mg.L<sup>-1</sup> with 1 g.L<sup>-1</sup> Nano-NPK fertilizer). And the best number of branches (17.33 branches) was obtained from the interaction between (GA3 at 400 mg.L<sup>-1</sup> with 4 g.L<sup>-1</sup> Nano-NPK fertilizer) was

while this number was decreased to the lowest (5.33 branches) during interaction of (GA3 at 200 mg.L<sup>-1</sup> with 1 g.L<sup>-1</sup> Nano-NPK fertilizer). Likewise the significantly best plant weight (189.20 g) was found from the interaction of (GA3 at 400 mg.L<sup>-1</sup> with 4 g Nano-NPK fertilizer), whereas, during the interaction of (GA3 at 0 mg.L<sup>-1</sup> with 1 g.L<sup>-1</sup> Nano-NPK fertilizer) lest weight (144.36 g) were shown. Also both significant maximum number of leaves (143.33 leaves) and leaf area (2755 cm<sup>2</sup>) were provided from the same interaction between (GA3 at 400 mg.L<sup>-1</sup> with 4 g.L<sup>-1</sup> Nano NK

fertilizer ) comparison with minimum results (89.00 leave and 1702.13 cm<sup>2</sup>) for both mentioned characteristics that were obtained from interaction of (GA3 at 0 mg.L<sup>-1</sup> with 2 g Nano-NPK fertilizer). Also the interaction of (GA3 at 400 mg.L<sup>-1</sup> with 4 g.L<sup>-1</sup> Nano-NPK fertilizer) provided the highest vegetative dry weight (42.30 g) comparable with the lowest weight (31.01 g) that were found from interaction of (GA3 at 400 mg.L<sup>-1</sup> with 2 g.L<sup>-1</sup> Nano-NPK fertilizer). While the maximum roots dry weight (57.33 g) were observed from the interaction of (GA3 at 0 mg.L<sup>-1</sup> with 4 g.L<sup>-1</sup> Nano-NPK fertilizer). While the interaction of (GA3 at 0 mg.L<sup>-1</sup> with 4 g.L<sup>-1</sup> Nano-NPK fertilizer). While the maximum roots dry weight (57.33 g) were observed from the interaction of (GA3 at 0 mg.L<sup>-1</sup> with 4 g.L<sup>-1</sup> Nano-NPK fertilizer) compared to

the minimum result (38.66 g) that found during the interaction of (GA3 at 0 mg.L<sup>-1</sup> with 2 g.L<sup>-1</sup> Nano-NPK fertilizer). And the interaction of (GA3 at 200 mg.L<sup>-1</sup> with 2 g.L<sup>-1</sup> Nano-NPK fertilizer) caused to get best total chlorophyll (87.97 SPAD) and growth index (74279 cm3), while these results were decreased to a minimum (69.93 SPAD) for total chlorophyll during interaction of (GA3 at 0 mg.L<sup>-1</sup> with 1 g.L<sup>-1</sup> Nano-NPK fertilizer) and (31249 cm3) for growth index during interaction of (GA3 at 200 mg.L<sup>-1</sup> with 1 g.L<sup>-1</sup> Nano-NPK fertilizer).

**Table (2):** Effect of the interaction between Nano- NPK fertilizer and GA3 on the growth and development of Buxus plant.

GA <sub>3 (</sub> mg L <sup>-1)</sup> Nano NPK fertilizer (g L <sup>-1)</sup>	0			200			400		
	1	2	4	1	2	4	1	2	4
Plant height (cm)	37.33 <sup>d</sup>	43.00 <sup>c</sup>	50.33 <sup>b</sup>	42.67 <sup>c</sup>	54.00 <sup>b</sup>	58.67 <sup>a</sup>	43.33°	41.33°	58.67ª
Number of branches branch/plant	6.00 <sup>de</sup>	6.67 <sup>с-е</sup>	8.67 <sup>bc</sup>	5.33°	7.67 <sup>cd</sup>	10.00 <sup>b</sup>	7.33 <sup>⊶</sup>	6.67 <sup>c-e</sup>	17.33ª
Plant weight (g)	144.36 <sup>d</sup>	146.47 <sup>d</sup>	179.74 <sup>b</sup>	159.32°	187.29ª	176.20 <sup>b</sup>	157.04 <sup>c</sup>	176.60 <sup>b</sup>	189.20ª
Number of leaves (leaf/plant)	109.33 <sup>d</sup>	89.00 <sup>e</sup>	142.33ª	105.33 <sup>d</sup>	131.33 <sup>⊳</sup>	117.67°	108.33 <sup>d</sup>	118.67°	143.33ª
Vegetative dry weight (g)	32.43 <sup>cd</sup>	31.91 <sup>cd</sup>	35.80 <sup>bc</sup>	33.60 <sup>cd</sup>	39.26 <sup>ab</sup>	39.16 <sup>ab</sup>	35.37 <sup>bc</sup>	31.01 <sup>d</sup>	42.30 <sup>a</sup>
Root dry weight (g)	41.34 <sup>d</sup>	38.66 <sup>d</sup>	57.33ª	48.72 <sup>bc</sup>	57.28ª	53.13 <sup>ab</sup>	40.68 <sup>d</sup>	54.02ª	47.35°
Total chlorophyll (SPAD)	69.93°	75.40 <sup>cd</sup>	71.50 <sup>de</sup>	79.47 <sup>bc</sup>	87.97 <sup>a</sup>	83.07 <sup>b</sup>	72.53 <sup>de</sup>	78.20 <sup>c</sup>	83.67 <sup>b</sup>
Plant growth index (cm <sup>3</sup> )	36403°	60107 <sup>b</sup>	44296 <sup>d</sup>	31249 <sup>f</sup>	74279ª	42793 <sup>d</sup>	59966 <sup>b</sup>	55375°	43143 <sup>d</sup>
Leaves area (cm <sup>2</sup> )	1839 <sup>ef</sup>	1702 <sup>f</sup>	2388 <sup>b</sup>	2068 <sup>cd</sup>	2204 <sup>bc</sup>	2034 <sup>с-е</sup>	1944 <sup>de</sup>	2222 <sup>bc</sup>	2755ª

Means with same letter for the interaction between the factors for each characteristic are not significantly different at 5% level based on DMRT.

The significant increases of almost all characteristics from the effect of GA<sub>3</sub> factor at both concentrations (200 and 400 mg.L<sup>-1</sup>) are in accordance with the findings of (Soad et. al., on *Codiaeum variegatum* 2010) and (AbdelKader, et al., 2016) on Magnolia grandiflora L, it may be related to the effect of direct potential of GA3 on both cell division and cell enlargement under apical meristematic region which caused an increase in internode length and this increase plant height (Lokesh et al., 2018), or due to its contribution in the converting manufactured food materials in a high degree

toward growth area (AL-Khafaji, 2014), and increase in the number of branches beside is one physiological property of gibberellin, may be through their influence on stimulating enzymes and protein biosynthesis, cambial activity division of cell and cell elongation (Ashour, 2018), and increase in the number of leaves may return to the role of GA3 in increasing the formation of nucleic acids DNA and RNA that are necessary for cell division (Alwan, *et.al.* 2009) also it increase osmotic potential inside plant tissue which cause absorption of more water and nutrient elements, and then securing necessary energy to form leaves and increase its area (Sardoei, et. al., 2014) or the increase in the number of leaves is due to an increase in the branches number as mentioned by (Malik, et. al. 2019) on Antirrhinum majus L. Also the reason for high plant weight and vegetative dry weight, besides it relates to the presence of more branches number, leaves, growth index and leaf area is also related to the fact that GA3 activates cells division, stimulates their growth and enlargement through increasing activity of enzyme (aamylase) which dissolve starch and sucrose to fructose and glucose and accumulation of these sugars has direct influence on increasing osmotic potential inside cell and plant tissues, which reflect positively on absorption of water and nutrient elements (Taheri-Shiva, et.al. 2014 and Al-Abbas, et.al. 2015), or due to the role of GA3 in stimulating the formation of DNA and RNA and then increase in formation of protein and physiological process inside plant cells (Alwan, et.al. 2009). And GA<sub>3</sub> has a role in increasing the permeability of cell wall from which cell wall of roots, therefore it improving root activity and nutrient absorption from the soil (Kader, 2023), and it facilitates the formation of the IAA hormone and increases its concentration because it decreases the activity of enzymes (peroxidase and oxidase) that are inhibitory to IAA (Taha, 2001) these processes positively influence roots properties. Also, the increase in total chlorophyll may be due to the role of GA3 in preventing the degradation of chlorophyll pigment through stopping the activity of chlorophylase enzyme and collecting nutrient elements in the leaves (Abozaid, 2000). Lastly, leaf area was increased because gibberellin acid is advantageous in promoting vegetative growth and it regulates key processes such as respiration, cell division and expansion, enzyme effectiveness, and chloroplast preservation from photosynthesis oxidation (Ghosh, et al. 2009).

Also, nutrition plays a key role in plant growth and development. and it is obvious from the data in the same table (table 1) that Nano-NPK fertilizer especially 4 g L<sup>-1</sup> provided significant results of most studied characteristics, these results are in accordance with data of (**Assaf and Muhamed, 2023**) on **the** growth of *Platycladus orientalis*, (**Elshamy** *et al.*, **2019**) on potato plant and (**El-Naggar, and El-Nasharty, 2009**) on *Hippeastrum vittatum*. likely because of the particles of this fertilizer have smaller dimensions than the pores of the plant cell wall, therefore easily permeating through the plant cell wall and

reaching the plasma membrane through the gas exchange (Abdel-Aziz, et al. 2018; Mohammed, et. al. 2020), then it plays a critical role in plant physiological and biochemical processes by increasing nutrient availability, more precisely, plant height was increased due to increasing the meristematic activity and stimulation of cell elongation in plants (Mahil and Kumar, 2019), as well as, due to modification of plant gene expression and associated biological pathways which ultimately affect plant height as mentioned by (Ghormade et al., 2011). An increase in the number of branches, total chlorophyll and vegetative dry weight is related to the fact that nano-fertilizers increase the efficiency of nutrient use, have a large surface area, and slowly release, which assist in absorbing nutrients quickly and speed of penetration, representation, and movement, leading to an increase the speed of growth and quality (e.g., protein and starch) by promoting photosynthesis. (Valiki et al., 2015 and Sajyan et al., 2020). And the increase in the number of leaves is probably due to the effect of this fertilizer on increasing the plant height and metabolites activities needed for increasing leaf formation, as reported by (Singh, 2017). as well as, the best leaf area from the influence of nano-NPK spraying is attributed to the significant role of the components of this fertilizer from the major elements, particularly with the two elements of nano nitrogen and phosphorus, due to what nanomaterials are recognized by their high ability to penetrate plant tissues and promote the production of cytokinins and thus increase the effectiveness of photosynthesis and raise the efficiency of vital processes, which is reflected in plant growth and development (Mahajan et.al. 2011). Finally, the maximum achievement of plant weight and growth index, and root dry weight were related to the function of such fertilizer from the major elements (N,P,K) in formation of chlorophyll and increasing the vegetative growth and their entrance in vital process, enzymatic reactions, cell division, respiration, photosynthesis, root development and increase their density and delay their aging, that promote the need for nutrients, and increasing their accumulation in parts of plant (Valizadeh and Milic, 2016).

#### CONCLUSION

It can be concluded from this study that most of the vegetative and roots characteristics of Buxus plant can be improved through using

gibberellin hormone especially at concentration of (400 mg L<sup>-1</sup>) and using Nano-NPK fertilizer at a concentration (4 g  $L^{-1}$ ). As well as, by using both factors together as interaction (400 mg  $L^{-1}$  + Nano-NPK fertilizer 4 g L<sup>-1</sup>) best plants in terms of growth and development can be obtained. Otherwise, it can be concluded that the lowest concentration of both factors proved the least improvement in plant growth. For further research researcher recommended using other treatments with Buxus Sp. plant for overcoming summer dormancy like spraying plant with Promalin in interaction with gibberellic acid at different concentrations to provide better results and also using the pinching process to promote producing new growths.

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