

Determination the Effect of Gibberellic Acid Foliar Spray on Growth in Olive Cuttings Cv. Coratina, Chetoui, Megaron under Saline Conditions

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Received: 📅 October 25, 2018; Published: 📅 October 31, 2018

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Abstract

The study was carried out at NARC Islamabad during July 2018 to October 2018 to examine the impact of gibberellic acid on the 01-year rooted olive cuttings of three varieties i.e. Coratina, Chetoui and Megaron in tunnel under saline environment. Soil salinity was developed artificially with the mixture of different salts at 2.0dSm^{-1} . Completely randomized design was applied with three replications. Foliar spray of Gibberellic acid @ 0, 200 and 400mg l^{-1} was done. Growth parameters of plant height stem diameter internodal distance, # of leaves plant⁻¹, leaf area, fresh weight, dry weight and chlorophyll contents were recorded at the end of the experiment. Coratina olive variety attained the highest plant height, internodal distance, number of leaves plant⁻¹ and leaf area at 200 and 400mg l^{-1} GA₃ foliar spray than other two olive varieties. Stem diameter data indicated similar results in Coratina and Megaron olive varieties that was higher than control. Internodal distance was the maximum at 200 and 400mg l^{-1} GA₃ foliar spray than other two olive varieties in Coratina olive variety. There was no difference in fresh weight of leaves either in GA₃ application as well as varieties. Chetoui olive variety gained the highest dry weight of leaves. Chlorophyll contents were gained the highest position by Megaron olive variety at 200 and 400mg l^{-1} GA₃ foliar spray than other two olive varieties under artificially saline conditions.

Keywords: Gibberellic acid; Growth hormone; *Olea europaea*; Coratina; Chetoui; Megaron

Introduction

Olive is one of the fruit crops that can grow in sandy soil due to its capability to tolerate drought stress. The production of olives in these areas is generally low due to the poor soil fertility and low water holding capacity. According, it seems that trees are not only in need of macronutrients application but also the application of some microelements [1]. The botanical name of the olive plant is "*Olea europaea*". The olive specie belongs to the family Oleaceae. The history of olive plant is endless and start about 7000 years ago originated from the coastal areas of the Eastern Mediterranean Basin as well as Northern Iraq, and Northern Iran [2]. The native of olive is the Mediterranean, Asia and Africa [3]. Olives are also good source of iron which helps to transport oxygen in blood. Calcium present in olives is essential for bones and muscles [4]. Olive reduced risk of heart diseases and even help to fight cancer [5]. Olive oil is the healthy component of our diet. Oleonic acid protects liver and improves blood flow [6]. Olives release stress and boost up immune system in human body. Olives contains all essential elements that are necessary for human health. Regularly consuming olives are

good for skin health [7]. One gram of olive oil provides 8 calories energy which is a great source of energy. Olive oil is widely used in industries like food preservation, cookery items and especially in canned products of food industry. Olive oil is also used in cosmetics products and in pharmaceutical industry. 100 milliliter (ml), of one type of olive oil, contains Energy: 800 kcal with Fat: 93.3 g, or which 13.33 g is saturated, and 66.6 g is monounsaturated [8]. Olive tree is most common edible plant in the world with average life of 500 years. It can be grown in all types of soil and is moderately salt tolerant crop. Also grow on calcareous soil with pH from 5.6 to 8.5. Olive plant is very slow growing evergreen plant with extensive root system and can also tolerate drought. Mainly grow up to 50 feet .in height. Arrangement of leaves is opposite. Most of the olive plants express self-pollination due to hermaphrodite type of flowers. Some cultivars are also exhibit cross-pollination.

The trees are also persistent, easily sprouting back even when cut to the ground [9]. Olive fruit is drupe type when matured is full of edible oil. Olive plant remains productive for very long time if

manage properly. Olive propagation is done by various methods according to environment susceptibility. Olive is propagated by sexually including seed and asexually including cuttings, layers, grafting etc. Most common and modern propagation technique is cuttings for raising new plants by the help of various growth regulators and in controlled conditions Ullah et al. 2009. Cuttings types include softwood, semi hardwood and hardwood cuttings. Multiplication by hardwood cuttings is common method for raising new plants. Survival rate in hardwood cuttings is high and can tolerate environmental conditions. Desirable traits of parent plants are easily obtained in offspring by cuttings. Genetic makeup of the favorable characteristics easily obtained in new plants [10]. Olive (*Olea europaea* L.) is considered as the most extensively cultivated fruit crop due to its economic importance and nutritional values. Olive (*Olea europaea* L.) is the most cultivated and earliest fruit tree having an endless history; it covers a region of approximately 7.5 million hectares [11]. According to FAOSTAT, leading world producing countries of olive are Spain and Italy [12]. Exploration survey using morphological features of olive was conducted in Spain, which estimated 262 olive cultivars [13], and there are over 600 cultivars in Italy. Almost 25 olive cultivars are micro propagated and are under study for their performance. The most well documented olive cultivars include oueslati, cv zard, cv rowghani, moraiolo, dolce agogia, leccino, coratina, nocellara, and pendolino. Olive received maximum importance in the Mediterranean region and its cultivation in this region began 6,000 years ago.

The olive fruit is usually oblong in shape, weighing 1 to 10 g or even more according to the cultivar. The skin of fruit is green when immature and dark blue, blue-violet or black when ripe [14]. Fresh and undressed olives are highly bitter due to a hydro-soluble glycoside called oleuropein. Some olives are naturally sweet due to low oleuropein. The glycoside is hydrolyzed with sodium hydroxide during processing and preserving. Oil can be extracted from fruit juice by hydrolyzing with water. Fresh fruits contain around 80% unsaturated fatty acids against 20% saturated ones, 20% oil and a very low quantity of cholesterol. The importance of olive oil was due to its use in cooking, salad dressing, food preparation, wool treatment, medicine, cosmetics and soap production [11]. Its wood is naturally long-lasting; in southern Italy olive is cultivated as a secondary source of profit [15]. Plant growth regulators are the chemicals that alter the plant growth and change the behavior of different crops. Produced by the plants and also produce artificially to regulate growth and development under various physiological actions. They are also called plant hormones. These hormones are applied to plants in different concentrations in order to control and regulate plant growth in different ways. All functions like normal growth, development, root and shoot growth are control by these hormones [16]. Generally, plant growth regulators are of two types, one is stimulators and other type is inhibitor. Plant growth

stimulators like auxins, gibberellins and cytokinins promote fruiting, flowering, seed development and cell enlargement. Plant growth is inhibited by abscisic acid result in dormancy and abscission. Auxins promote cell elongation and root initiation in cuttings. Natural auxins like Indole butyric acid (IBA) and Naphthalene acetic acid (NAA) hormones play an important role in root formation of stem cuttings Agusti et al., 2000. Cytokinin helps in cell division. Gibberellins are the very important plant growth regulators and promote cell elongation and also help in fruit developing. Ethylene helps in fruits ripening [17]. Gibberellic acid, also known by GA_3 is naturally occurring plant growth hormone that is harvested from fungus and can produce commercially for agriculture and other purposes. In purified form it is white to pale-yellow solid with chemical formula $C_{19}H_{22}O_6$. Gibberellic acid discovered by Kurosawa (a Japanese pathologist) from the fungus *Gibberellafujikuroi* in 1928 and most common type is GA_3 . Gibberellins generally stimulate cell division and stem elongation and standardize growth by stretching internodes. GA_3 promotes stem elongation. Gibberellic acid plays important role in germination by breaking seed dormancy. GA_3 encouraged cell elongation [18,19]. GA_3 standardizes flower initiation and development. GA_3 contributes in pollination. GA_3 improves fruit quality and promote generation of female flowers [20]. GA_3 application on olive plants diminishes the chances of alternate bearing in olive plants [21]. GA_3 performs key role in protein synthesis and in regulating nucleic acid. Higher concentration of GA_3 can suppress root initiation [22], aids plant to swing at suitable time in reproductive phase [23]. Stem elongation is response of signal transduction pathway with different environmental factors.

There is a great importance of gibberellic acid in agriculture. Foliar application of GA_3 increases yield. Also improve the quality of fruits and increase the fruit size [24]. Endogenous gibberellins influence various development processes, such as stem elongation, control various aspects of seed germination, including dormancy break and mobilization of endosperm reserves, moreover, gibberellins influence transition from juvenile stage to mature stage, induction of flowering, sex determination and fruit set establishment in the reproductive development [20]. Gibberellins (GAs) generally regulate growth and encourage stem elongation, flowering, and break seed dormancy, buds, and bulbs, influencing various developmental processes. More than 90 types of gibberellins exist, but GA_3 is the most frequently used in tissue culture. Gibberellins are natural stimulators of plant growth and development with frequent spectacular effects and help in obtaining remarkable results in agriculture production, forestry, horticulture and medicinal plants. Gibberellins promote cell division and elongation [25]. Foliar application of GA_3 and nutrients had improved the productivity and quality of flowers [26]. Spray olive trees with Gibberellic acid (GA_3) before an expected "on" year decreased the percentage of opened flower buds per shoot, number

of flowers per inflorescence in the subsequent year. El-Iraqy, 2001, GA₃ are responsible for cell elongation, rather than cell division [17]. Suitable in vitro protocols for olive culturing have been developed; studies indicated that the main factors for achieving elevated growth rates involved in tissue culture include medium formulation [27,28], growth hormones [29], rooting, genotype [30,31] and acclimatization conditions. Olive micro propagation was first studied and documented by [30], who proposed the olive medium (OM), which has been proved to be efficient for the micro propagation of many olive cultivars [29]. Some cultivars do not respond to in vitro conditions due to sensitivity, so their proliferation rate is prolonged, and rooting of explants is also limited, and many plantlets die at the acclimatization stage [32]. The introduction of specific olive medium (OM), [30] for axillary bud stimulation and successive shoot multiplication has paved the way for further advancement of olive micro propagation.

Auxins are the class of plant growth hormones that promotes root initiation by inducing both growth of pre-existing roots and adventitious root formation. The auxin treatment was one of the first factors [33] catch the consideration of researchers for in vitro rooting of shoots [15] as this phytohormone is involved in rooting for so many years and the positive role of various auxins on the initiation and development of rooting is well documented [33]. Exogenous auxin is used as a growth regulator in micro propagation or in vitro culture at a concentration of 0.01-10.0 mg/L. In tissue culture systems, added auxin is generally related to the promotion of growth, induction of rooting, callus induction, cell elongation, tissue swelling, cell division, inhibition of adventitious and axillary shoot formation, and induction of embryogenesis.

In Brazil, olive tree growth is still a recent rural activity in expansion [34], where this country is one of the greatest importers of olive tree products of South America, and, Argentina, one of the greatest suppliers, besides Spain and Portugal [35]. The consumption and imports increase turn the Brazilian market promising for this activity [36]. Furthermore, growth traditional areas in the world are limited to the existing crops, with no significant perspective of increase, making olive trees to gain space in South American countries [37]. In order to reach success in fruit production it is necessary, among other factors, high quality seedlings, with no phytosanitary problems, which present good development when they are combined with adequate handling. The techniques used for spreading olive trees are grafting and rooting. This last one shows lower cost and time to obtain seedling, demanding less qualified labor. But, so that the technique is feasible, according to what Fachinello [38] emphasizes, it demands that the cultivar shows high capacity to form roots, taking into consideration that its root system and later development in the area of production show good quality. Exogenous application of Phyto regulators has been one of the most studied techniques for the improvement of the hormonal balance in rooting, where the indole butyric acid

(IBA) is the most used auxin. Several researches developed towards olive trees rooting have used concentrated hydro alcohol solutions of IBA [34,39-41], therefore, the results found vary a lot according to the dosage, the cultivar, the period of rooting, the substrate and the type of cutting, among other factors. The formation of root primordium cells depends on the endogenous auxins in the cutting and on a synergic compound such as a diphenol. These substances lead to the synthesis of ribonucleic acid (RNA), which act upon root primordium initiation and gibberellins are considered substances that inhibit rooting [22].

GA₃ succeeded in inhibition of olive tree blooming in the following season. GA₃ has the potential control on growth and flowering process. In addition, increased petiole length, leaf area and delayed petal abscission and color fading (senescence) by the hydrolysis of starch and sucrose into fructose and glucose [42,43], GA₃ treatments increase the weight, length, width of olive fruits, stone weight and flesh weight than untreated ones, improvement fruit weight and flesh: seed ratio, [24]. Naphthaleneacetic acid, commonly abbreviated NAA is an organic compound, which is a plant hormone in the auxin family and is an ingredient in many commercial horticultural products; it is also a rooting agent and used for the vegetative propagation of plants from stem and leaf cutting [44]. NAA application at (100, 150) mgL⁻¹ 15 days after full bloom has been used to chemically thin olives in various countries, NAA has been reported to be useful for thinning of fruits. It has important role in fruit formation, abscission cell elongation, apical dominance, photoperiod and geotropism [45].

The maturity level of sprig, cutting position on the linear length of sprig, type, methods and concentrations of substances rhizogenic, the time necessary for the realization of the rooting process, has great influence on the genetic profile [46,47] Our olive varieties have the various abilities rooting. Being the genetic factor, with difficult rooting varieties continues some research such as: Minimum and maximum the number of buds and leaves, the effect of treatment with different chemical substance through foliar method, etc. [16,18]. Salinity stress negatively impacts agricultural yields throughout the world, affecting production, whether for subsistence or economic gain. At present, about 20% of the world's cultivated land and approximately half of all irrigated land and 2.1% of the dry agriculture land is affected by salinity [48]. Salinization is spreading more rapidly in irrigated lands because of inappropriate management of irrigation and drainage. Moreover, rain, cyclones and wind add NaCl to coastal agricultural lands [49]. The rapid increase in the world's population requires an expansion of crop areas to raise food production. Salinity imposes serious environmental problems that affect grassland cover and the availability of animal feed in arid and semiarid regions. Salt stress is one of the most serious limiting factors for crop growth and production in the arid regions. About 23% of the world's cultivated lands is saline and 37% is sodic [50]. Considerable research work

has been conducted on the effect of salinity on different growth characters of different crops worldwide [48,51-57]. Keeping in view this study was planned to increase the growth of olive cutting with foliar spray of Gibbeleric acid under saline conditions.

Materials and Methods

The study was carried out at NARC Islamabad during July 2018 to October 2018 to examine the impact of gibberellic acid on the 01-year rooted olive cuttings of three varieties i.e. Coratina,

Chetoui and Megaron in tunnel under saline environment. Soil salinity was developed artificially with the mixture of different salts at 2.0dSm⁻¹. Completely randomized design was applied with three replications. Foliar spray of Gibberellic acid @ 0, 200 and 400 mg l⁻¹ was done. Growth parameters of plant height stem diameter internodal distance, # of leaves plant⁻¹, leaf area, fresh weight, dry weight and chlorophyll contents were recorded at the end of the experiment. Data were statistically analyzed according to completely randomized design and compared treatment means using LSD test with statistical software, Statistix 8.1 (2005).

Results

Table1: Impact of Gibberellic acid foliar spray on olive growth under saline environment.

Treatment	Plant height (cm)			Stem diameter(cm)			Internodal distance(cm)			# of leaves plant-1			Leaf area (cm ²)			Fresh weight of leaves (g)			Dry weight of leaves (g)			Chlorophyll Contents (%)		
	V ₁	V ₂	V ₃	V ₁	V ₂	V ₃	V ₁	V ₂	V ₃	V ₁	V ₂	V ₃	V ₁	V ₂	V ₃	V ₁	V ₂	V ₃	V ₁	V ₂	V ₃	V ₁	V ₂	V ₃
T ₀	10	8.2	7	0.16	0.12	0.18	1.3	0.9	1.2	15	11	10	1.4	0.9	1.2	0.6	0.5	0.5	0.26	0.29	0.27	64	64	63
T ₁	11	9.7	7.1	0.17	0.14	0.14	1.4	1.2	1.3	14	11	8	1.5	1.4	1.2	0.6	0.7	0.7	0.28	0.35	0.35	64	57	67
T ₂	12.6	8.8	8.6	0.2	0.15	0.2	1.6	1.3	1.4	16	11	12	1.6	0.9	1.4	0.5	0.6	0.6	0.28	0.36	0.32	67	64	69
Mean	11.2	8.9	7.6	0.17	0.13	0.17	1.4	1.1	1.3	15	11	10	1.5	1.1	1.3	0.6	0.6	0.6	0.27	0.33	0.31	65	65	67

T₀ = 0 GA₃, T₁ = 200 mg l⁻¹ GA₃, T₂ = 400mg l⁻¹ GA₃ V₁= Coratina, V₂= Chetoui, V₃= Megaron

Maximum height (12.6 cm) was attained at T₂ (400 mg l⁻¹) in Coratina olive variety. Over all Coratina olive cultivar gained the highest plant height i.e., 11.2cm, and T₂ (400 mg l⁻¹) displayed second highest plant height is (10cm) followed by 9.26cm in T₁ (200 mg l⁻¹) as indicated in Table 1. Data presented in Table 1 depicted the effects of GA₃ foliar application on olive plant stem diameter. Maximum stem diameter (0.2cm) was attained at T₂ (400 mg l⁻¹) in two varieties i.e., Coratina and Megaron. Over all Coratina and Megaron olive cultivars gained the highest stem diameter i.e., 0.17cm and T₂ (400mg/l) displayed highest stem diameter (0.18cm) followed by 0.15 in T₂ (200 mg l⁻¹). Data shown in table illustrated the effects of GA₃ foliar application on olive plant internodal distance. Highest internodal distance (1.56cm) was attained at T₂ (400 mg l⁻¹) in Coratina olive variety. Overall Coratina olive variety attained the highest internodal distance (1.44cm). T₂ (400 mg/l) displayed the highest (1.43cm) internodal distance followed by 1.29cm in T₁ (200 mg l⁻¹) Data represented in Table 1 showed the effects of GA₃ foliar application on olive plant number of leaves. Maximum number of leaves were attained at T₂ (400 mg l⁻¹) in Coratina olive variety. Overall Coratina olive variety attained the maximum number of leaves (15). T₂ (400 mg l⁻¹) showed the maximum number of leaves (13.16) followed by T₁ (200 mg l⁻¹) which attained 11.10.

Data reported in Table 1 depicted the effects of GA₃ foliar application on olive plant leaf area. Maximum leaf area (1.58cm²) was attained at T₂ (400 mg l⁻¹) by the Coratina olive cultivar. Overall Coratina olive variety displayed the maximum leaf area (1.47cm²). T₁ (200 mg l⁻¹) showed the maximum leaf area (1.36cm²) followed by T₀ (control) attaining 1.14 cm². Data presented in Table 1

showed the influences of GA₃ foliar application on olive plant leaves fresh weight. Maximum leaves fresh weight (0.70g) was attained at T₁ (200 mg l⁻¹) by the Megaron olive variety. Overall Chetoui olive variety attained maximum leaves fresh weight (0.64g). T₁ (200 mg l⁻¹) showed the maximum olive plant leaves fresh weight (0.66g). Data presented in Table 1 showed the effects of GA₃ foliar application on olive plant leaves dry weight. Maximum olive plant leaves dry weight (0.36g) was attained at T₂ (200 mg l⁻¹) by the olive variety Chetoui. Over all Chetoui olive variety attained maximum leaves dry weight (0.33g). T₁ (200mg/l) and T₂ (400mg/l) showed the maximum olive plant leaves dry weight (0.32g). Data in Table 1 showed the effects of GA₃ foliar application on olive plant leaf chlorophyll contents. Highest chlorophyll contents (69.14%) were attained at T₂ (400 mg l⁻¹) in Megaron olive variety. Overall Megaron olive variety attained the highest chlorophyll contents (66.62%). T₂ (400 mg l⁻¹) displayed the highest (66.67%) chlorophyll contents followed by 63.04% in T₁ (200 mg l⁻¹).

Discussion

The results of this study demonstrated that GA₃ can play an important role in vegetative growth of olive cuttings. GA₃ foliar application enhances bud sprouting and shoots elongation. GA₃ was successfully used by Grigoriadou [27] for shoot proliferation of certain olive tree cultivars. From the above-mentioned results, it was noticed that GA₃ foliar sprays at 400 mg l⁻¹ concentration was more effective in plant height than spraying GA₃ at other concentrations and control. Highest stem diameter was obtained from cuttings sprayed with T₂ (400 mg l⁻¹) GA₃ while control treatment showed

the lowest diameter of olive cuttings. El-Sharkawy and Mehaisen [58] indicated that application of GA₃ on olive plants caused elongation in the primary cells in the young tissues and growth centers. The present results may be attributed to simulative impact of GA₃ on cell extension and on cell division. GA₃ is an interesting hormone for in vitro shoot elongation of many other species such as Macadamia [59], Acacia [60]. With the GA₃ application increasing cell elongation and also increases internodal distance [61]. GA₃ foliar application at the concentration of 200 mg l⁻¹ has significant effects on olive leaves fresh weight. GA₃ foliar sprays at 400 mg l⁻¹ and 200 mg l⁻¹ concentrations were more effective for olive leaves dry weight as compared to control treatment. This might be due to the fact that, GA₃ improve the rate of photosynthesis and cause greater accumulation of photosynthetic [22] which leads to increase in dry matter of plant and significant improvement in absolute growth rate. Highest chlorophyll contents were noted at the treatment of 400 mg l⁻¹ in all olive varieties because GA₃ may stimulate transport of nutrients through the phloem, modify the strength of the sink by stimulating its growth and increase the ability for sugar unloading from the phloem. Or, they may act on metabolism and compartmentalization of sugar and its metabolites. Being the genetic factor, with difficult rooting varieties continues some research such as: Minimum and maximum the number of buds and leaves, the effect of treatment with different chemical substance through foliar method, etc. [18,19].

Conclusion

It can be concluded that foliar application of gibberellic acid on olive plants stimulates the growth and development. GA₃ application regulates the growth of stems, leaves, buds, height and boost up the plant growth and development characteristics.

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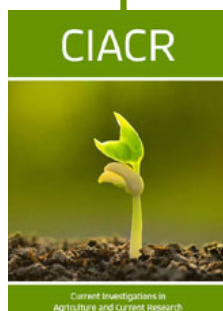
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DOI: [10.32474/CIACR.2018.05.000208](https://doi.org/10.32474/CIACR.2018.05.000208)



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