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ORIGINAL ARTICLE **OPEN ACCESS**

Okra growth, Chlorophyll Content and Fluorescence Yield as Affected by Naphthalene Acetic Acid as Growth Regulator

ABM Sharif Hossain
Department of Biology, College of Science, Imam Mohammad Ibn Saud Islamic University, Riyadh, Saudi Arabia
Correspondence: ashossain@imamu.edu.sa

ABSTRACT
The study was conducted to evaluate the influence of different concentration of NAA (Naphthalene acetic acid) on okra growth and development. The stem injection innovative method of application was used in this experiment rather than spray as common and traditional method. Stem injection method was applied on the stem of okra plant using NAA at different concentrations. The higher concentrations (25 mg/l) of NAA greatly increased the plant height compared to the control. NAA application at 25 mg/l induced the highest value of stem girth over the control. The chlorophyll content in leaves was affected significantly by different concentrations of NAA. It was found that lower concentrations of NAA (25, 50 mg/l) increased chlorophyll content and fluorescence yield *as compared to the higher concentration of 100, 200 mg/l and control. The pod per plant, pod length, pod diameter, pod size, per pod weight and healthy seeds percentage were significantly higher in the lower concentration of 25 and 50 mg/l than the higher concentrations of 100 and 200 mg/l of NAA. The 100 and 200 mg/l concentrations of NAA had increased the production of aborted seeds as compared with the lower concentrations of 25, 50 mg/l and control. Finally it seemed that 25 and 50 concentrations of NAA were the more effective for the okra growth, Chlorophyll content and fluorescence yield.*
Keywords: NAA, okra, growth regulator, Chlorophyll, stem injection

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INTRODUCTION

Okra is one of the important and popular vegetable crops in tropical and subtropical area in Asia [1] and Africa [2,3]. It belongs to the family Malvaceae, genus, *Abelmoschus and species, esculentus.* Cultivated okra is suitable for agriculture as garden crop as well as on commercial farms. It is one of the vegetables which grown commercially in many countries such as India, Westren Africa, Iraq, United States and other countries around the world. It was reported that total production of okra was 5.9 m tons in the world [4]. It was reported [5,6]that okra pods are considered nutritious, providing some human supplementary vitamins such as vitamin C, A, B- complex, calcium, potassium, iron and other minerals. Okra pod contains many nutritional contents which important for human health. One hundred gram of fresh pod has around ; moisture (89.6 percent). K (103 mg), Ca (90 mg), Mg (43 mg), P (56 mg), vitamin C (18 mg) and some important metals such as iron and aluminum [7].

The application of plant growth regulartors are known as one of the most important treatments used nowadays in agriculture. Some horticulture crops production were increased by application of different growth regulartors [8]. Growth regulators mainly regulate the plant physiological and biochemical processes. For example, play a major role in dormancy, organ size, crop improvement, flowering and fruit set, regulation of chemical composition of plants [9]. The phytohormone auxin affects approximately all developmental processes in plants, including fruit improvement. However, auxin is produced in meristems and young leaves and moved to other parts of the plant in a polar fashion [10].

There are more than 100 distinct gibberellins produced primarily in roots & young leaves but GA3 or gibberellic acid is the most popular available form. GA3 has many effects on plant growth such as enhance stem and internodes elongation, produce seed germination, enzyme production during germination and fruit setting and growth [11,12] and breaking of dormancy. It was reported [13] indicated that plant growth regulartors may be used to regulate the vegetative growth of plants. Application of GA3 increased the plant height, number of internodes, leaf area, dry weight of shoot and dry weight of Gram plant respectively [14]. However, work has been done on the use of NAA to improve vegetative growth, pod size, and delay pod maturity in vegetables using spray method. But no studies

have been conducted to evaluate the vegetative growth, pod quality and seeds yield affected by NAA application in okra using stem injection. Objectives of the experiment were undertaken:
To investigate the effect of different concentration of applied NAA at different concentrations on plant height, stem girth, leaves, chlorophyll content, maximum quantum yield (Fv/Fm), pod size and yield. Also to evaluate the efficacy of this injection method (treated stem) of application on seeds for inducing parthenocarpy or Stenospermocarpy (aborted seeds) in okra pod. In addition to that find cost effective methods reducing pollution of excessive use of plant growth regulator.

MATERIAL AND METHODS

Plant materials, cultural operation and experimental design

The seeds of local okra variety were sown in the experimental field of the University. These seeds soaked in distilled water for 24 hours after which they were spread on moist filter paper in Petri-dish. The Petri-dish was kept in dark cupboard at room temperature of 30 ºC. Okra seeds were sown directly into the soil by hand in soil fertilized with NPK 19 g/hill 14-14-14 as basal fertilization. Thirty days after emergence, sidedress with 10g/hill 46-0-0 [6] and plots were irrigated when necessary. The experiment was laid out in randomized block design having four replications. The whole area was divided into fifteen blocks and each block into 20 unit plots. The size of the unit plot was 1 x 1 m^2. The seeds were shown in rows made by hand plough. The gaps where seeds failed to germinate were filled up within two weeks after germination of seeds. After field preparation, seeds were sown in well-prepared seedbeds in line with a distance of 70 cm when germination completed thinning was done to maintain the plant to plant distance of 30 cm. The depth of planting was 1cm from the surface of the soil. Hoeing, weeding and other cultural practices were done uniformly.

Preparation of plant growth regulators

The growth regulartors employed in the experiment were Naphthalene acetic acid (NAA). The concentrations of the NAA growth regulator were 0, 25, 50, 100 and 200 mg/L. The NAA was dissolved in 2ml of 1% ethanol to make desired concentration. Each rate of chemical NAA was added with distilled water to make 100 ml of solutions. The control plants were injected with 100ml of water mixed with 2 ml of 1% ethanol.

Growth regulator application

One and an half ml (1.5ml) of the various concentrations of NAA (0, 25, 50, 100 and 200 mg/l) were applied on the stem by injected the plant stems with needle for a surgical purpose of 1 dose at the height of 3 cm above the ground level. While control was distilled water mixed with 2ml of 1% ethanol (Fig. 1).

Fig. 1. Photo shows stem injection technique

Measurement of parameters

Data were recorded considering the following parameters:

Plant height and stem girth (cm): Plant height was measured from above ground level up to the uppermost tip of the leaves at the end of harvesting. Both plant height and stem girth were measured using a meter rule with the aid of thread.

Leaves numbers: Number of leaves: number of leaves on each treated and control plants was counted.

Leaf chlorophyll content: The chlorophyll content in the leaves was measured by SPAD value meter (Minolta Japan).

Leaf chlorophyll fluorescence measurement: Fast chlorophyll fluorescence was evaluated on the upper surface of latest fully expanded leaf by using a Plant Efficiency Analyzer (PEA, Hansatech Instruments Ltd., England). A leaf clip was appended to the leaf and kept in the dark for 15 minutes for dark adaptation. After that the shutter plate was opened and light was applied on the leaf. The initial fluorescence intensity (Fo) when all reaction centers (RCs) are open, the maximal fluorescence intensity when all reaction are close (Fm), the variable fluorescence (Fv = Fm-Fo) and the time to reach the maximal fluorescence intensity (tmax), were calculated. The quantum yield was determined according to the equation Fv/Fm. Used the ratios Fv/Fm.

Pod parameters: Five pods were randomly chosen from each treatment to determine the following characters: Green pod length (cm), Green pod diameter (cm) and Pod size (cm^2). Pod size was determined by measuring the length and diameter of pod per treatments with a Varnier caliper.
Single pod weight: Green pod weight (g) was determined with help of a digital UWE-ESP Digital Electric Balance and the average weight calculated.
Seed production: For the determination of healthy seeds from treated flowers, the number of health seeds and aborted seeds was counted after dry stage. Healthy seeds/pod (%) and seedless or Aborted seeds/pod (%) were counted.
Statistical analysis
The obtained data were statistically analysed using SPSS Computer Programme, Version16. The data were analyzed following Analysis of Variance (ANOVA) technique and mean differences were adjusted by using Duncan's Multiple Test (DMRT) at 5% level of significance.

RESULT AND DISCUSSION

Plant height (cm), number of branches, stem girth and Chlorophyll content

NAA applied at different concentrations influenced the plant height significantly ($P<0.05$). The lower concentrations (25 and 50 mg/l) of NAA increased the plant height compared to the control. The higher concentrations had lesser plant height but higher than the control (Table 1).
A significant variation was evident in the number of branches per plant, stem girth, number of leaves and leave area due to the application of NAA at different concentrations (Table 1). The treated plants generated higher number of branches over control. Among the NAA application, 25mg/l of NAA induced maximum number of branches (2.5) followed by 50 mg/l (2.0) and 100 mg/l (1.75) as compared to the control (1.50). In the contrary, NAA application at 25 and 50 mg/l induced the highest value of stem girth (2.73 and 2.29cm) over the control (2.20cm). The highest number of leaves per plant (23.25) was obtained by 25 mg/l followed by 50 mg/l (22.75) and the lowest leaves number/plant (22) was observed with control treatment. The data indicated that the lower concentration of NAA increased number of leaves more efficiently than the higher concentrations. Results in Table 2, indicated that the highest leaf area was found at the concentration of 25 mg/ml NAA (Table 2). The leaves content of chlorophyll was affected significantly by different concentrations of NAA. The results showed that 25, 50, 100 concentrations of NAA increased chlorophyll content compared to the control and 200mg/l of NAA concentration. Chlorophyll fluorescence yield (Fv/Fm yield) was found higher in the lower concentrations of NAA (25 and 50 mg/l) and lower in the higher concentration (100 and 200 mg/l) and control.

Pod (fruit) production, yield contributing characters and seeds yield

Results in Table 2, indicated that pod length, pod diameter, pod size, individual pod weight and healthy seeds percentage per pod were significantly affected by different concentrations of NAA. Among the concentrations treatments, 25 mg/l had the maximum number of pod per plant (4.24 cm) followed by 50, 100, 200 mg/l (4.23 cm) in comparison with control (4.22 cm). The data revealed that 25 mg/l produced the longest pod followed by 50 and 100 mg/l. Pod diameter was found maximum with 25 mg/l and 100 mg/l (3.57cm) and followed by 200 mg/l of concentration. Significantly highest pod size was obtained in 25 and 50 mg/l ($5.27cm^2$) followed by 100 mg/l ($4.21 cm^2$). In this respect, pod weight recorded higher in 25, 50 and 100 mg/l compared to the 2000 mg/l and control. Pod harvested from 100and 200 mg/l treated plants had significant highest aborted seeds percentage (4.66%) followed by 25 mg/l (4.65%), 50 mg/l (4.65%) and control (4.64%). Concentration of 25 and 50 mg/l treatments had increasing the production of healthy seeds compared with control (Table 3). Concentration of 25 mg/l had showed the maximum healthy seeds (95.34%) per pod followed by 500 mg/l (95.34%) while minimum healthy seeds was observed in 200 mg/l treatment (95.32%).

Table 1. Effect of stem injection treatment at different concentrations of NAA on some vegetative parameters of okra

Concentrations (mg/l)	Plant height (cm)	No. of branches/plant	Stem girth (cm)	No. of leaves/plant
0	78.81±0.01c	1.50±0.57b	2.20±0.02d	22.00±1.41d
25	78.86±0.05a	2.50±0.57a	2.73±0.02a	23.25 ±1.50a
50	78.85±0.01b	2.00±0.00ab	2.29±0.03c	22.75±1.26b
100	b 02±0. 78.83	1.75±0.50ab	2.24±0.01b	22.25±2.21c
200	78.82±0.01c	1.25±0.50b	2.16±0.02e	21.25±0.96e
LSD (0.05)	0.05	0.73	0.03	2.30
	*	*	*	NS

Values are means ± standard deviation. Means followed by same letter or no letter do not differ significantly at 5% level by Duncan's Multiple Range Test (DMRT) N .S. : no significant difference.

Table 2. Effect of NAA injection treatment at various concentrations stem on leave area, chlorophyll contents, Fv/Fm yield and number of fruits of *okra*

Conc. mg/l)	Leave area/plant (cm^2)	Chlorophyll content (SPAD value)	Fv/Fm yield	No. of fruits/plant
0	325.73±0.01b	40.42±0.50b	0.691±0.002b	9.50±1.29c
25	327.75±0.02a	40.45±0.03a	0.693 ±0.02a	10.75 ±1.50a
50	325.74±0.02b	40.43±0.02b	0.692±0.02a	10.50±1.29b
100	325.72±0.02b	40.43±0.02b	0.691±0.02b	10.00±0.82b
200	325.72±0.01cb	40.42±0.04b	0.689±0.03c	9.00±0.82c
LSD (0.05)	0.026	0.05	0.004	0.77
	*	NS	NS	NS

Values are means ± standard deviation. Means followed by same letter or no letter do not differ significantly at 5% level by Duncan's Multiple Range Test (DMRT). N .S. : no significant difference.

Table 3. Yield and yield contributing characters of okra as influenced by with NAA at different concentrations applied by stem injection treatment..

Concentrations (mg/l)	Fruit length (cm)	Fruit diameter (cm)	Fruit size (cm^2)	Per fruit weight (g)	Healthy seeds/fruit (%)	Aborted seeds /fruit(%)
0	4.22±0.02b	1.22±0.01b	5.20±0.08b	2.72±0.02b	95.33±0.02b	4.64±0.01c
25	4.24±0.02a	1.24 ±0.02a	5.27±0.09a	2.73±0.02a	95.34±0.02a	4.65 ±0.02b
50	4.23±0.02b	1.23±0.02b	5.27±0.05a	2.73±0.02a	95.34±0.05a	4.65±0.02b
100	4.23±0.02b	1.23±0.02b	10b±0. 5.21	2.73±0.03a	95.33±0.02b	4.66±0.03a
200	4.22±0.02b	1.22±0.03b	5.18±0.10c	2.72±0.02b	95.32±0.02b	4.66±0.02a
LSD (0.05)	0.027	0.029	0.14	0.01	0.03	0.026
	NS	NS	NS	NS	NS	NS

Values are means ± standard deviation. Means followed by same letter or no letter do not differ significantly at 5% level by Duncan's Multiple Range Test (DMRT).
N .S. : no significant difference.

Several reports which indicated that application of the plant growth regulators could provide higher germination, growth, fruit set, fresh vegetables weight and seed yields quality [15]. It had been informed [16] that natural plant growth regulators or synthetic were controlled the plant activities and their productions by controlling one or more of one or more specific physiological processes within a plant. However, Gibberellic acid are safe for human health which can be used for different aims [17]. Plant growth regulators play a central role in morphology and physiology of the plants. The effect of growth regulator depends on plant species, variety, their growth stage, concentration of chemicals that used , application technique and frequency of application [18]. Application of NAA at 25, 50 and 100 mg/l increased the plant height over control in applied by stem injection method. Growth regulator is concerned to enhance cell division and elongation (Harrington et al., 1996). It was stated [19, 20] that increased stem elongation might be due to stimulating action of GA3, which alleviate the cell wall by increasing its plasticity. It found that GA3, NAA and IAA applications increased the plant height of soybean and Red sorrel, respectively [21,22]. Also, earlier studies reported that GA3 increased plant height in various crops; soybean [23]. It was stated [24] that GA3 and IAA treatment at 100 ppm increased leaves number and leaves area and chlorophyll content in Hibiscus sabdariffa L. It was mentioned that a significant increase in the leaf length in onion by application of GA3 [25]. It was informed [26] that GA3 application increases branches number by breaking apical dominance. GA3 and IAA developed yield and physiochemical characters of leafy vegetable [27]. Growth regulators allow water to enter the cells of fruits and dissolved materials which lead naturally to increase fruit size by increasing the permeability of fruit cell wall [28]. IAA and GA3 application at 100 ppm increased the yield of rice and soybean [29,30] respectively. A significant decreased of seed abortion percentage was observed after NAA treatment at 25 mg/l compared with control. The increase in seed yield due to NAA application and other treatments maybe related to improved vegetative growth (leaf area and leaf number plant. The present observations

was in confirmation with [31] who observed that growth regulator application at 100 ppm to soybean produced the highest yield of seeds per plant.

CONCLUSION

From the above discussion it can be concluded that 25 and 50 mg/l of NAA concentrations were the best for okra okra growth, Chlorophyll content and fluorescence yield. Therefore, it can be recommended that stem injection technique can be used commercially in the vegetable industry. The internal application stem injection can reduce the chemical and production cost without hazardous any environmental pollution.

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