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Physiological Effect of Benzyl Adenine on Growth, Boll Setting and Productivity of Cotton Plant

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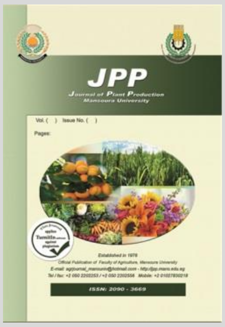

ABSTRACT

Two field experiments were conducted at El-Gemmeiza Agricultural Research Station, Agricultural Research Center, Egypt in 2018 and 2019 seasons to study the physiological effects of benzyl adenine (BA) on growth, boll setting and productivity of cotton plant cv. Giza 86. Date, number of applications and four concentrations of BA were examined used a split-plot design with three replicates.Sprayings of BA three times increased estimated elements and photosynthetic pigments in leaves in 2018 season, growth traits (except number of fruiting branches), while two sprays significantly increased number of fruiting branches, earliness measurements (except boll shedding% which decreased), seed cotton yield feddan^{-1} and open bolls number plant^{-1} in both seasons.BA at the medium level (40 ppm) gave a significant increase in the numbers of fruiting branches, total and open bolls plant^{-1}, percentages of boll setting and earliness as well as seed cotton yield feddan^{-1} in both seasons compared to the rest of treatments. The high level of BA gave a significant increase in studied elements content of leaves and photosynthetic pigments in 2018 season and growth traits with the exception of fruiting branches. Control treatment (sprayed with tap water) gave the lowest values in this respect with a significant increase in boll shedding %. Fiber traits were not significantly affected by the two factors studied and their interactions. It could be concluded that spraying with BA at 40 ppm two times under El-Gemmeiza conditions reduces the undesirable shedding and increases bolls setting which leading to an increase in cotton productivity.

Keywords: Cotton; benzyl adenine (BA); photosynthetic pigments; physiological effect.

INTRODUCTION

The cytokinins (CK) play an important role in all phases of plant development including cell division, cell enlargement, dry matter accumulation, flowers and fruit set (Cothren, 1994). Mukherjee and Kumar (2007) referred the promoting effect of kinetin on the basis that kinetin may stimulate cell division, suppresses senescence, promotes enzymes and it reduces the contents of ethylene, abscisic acid or other plant growth retardants. It similarly promotes lateral bud growth (Liu *et al*., 2009), accumulates chlorophyll, regulates cell division, differentiation and organogenesis, enhances leaf expansion, nutrient mobilization and delays senescence (Hall, 1973) and regulates apical dominance, chloroplast biogenesis, nutrient mobilization, leaf senescence, vascular differentiation, photo-morphogenic development, shoot differentiation and anthocyanin production (Mok and Mok, 2001). BA plays permissive role in the regulation of various growth processes in the plants (Ibrahim *et al*., 2010), nutrient uptake especially potassium (Guo *et al*., 1994) and photosynthetic efficiency (Oosterhuis and Zhao, 1998). In addition to their important roles in controlling and stimulating cell division, the cytokinins and their derivatives also reduced ethylene sensitivity of plants (Serek *et al*., 1994), inhibited leaf senescence (Burke, 2009 and 2011) and hormonal content which affected positively on hypocotyl diameter, breaking of apical dominance, lateral root formation, speed of lateral branch formations and enhanced boll development. Also, benzyl adenine promoted RNA and protein synthesis, whereas decreased WUE (Hopkins and Huner, 2004). It inhibited certain proteolytic enzymes (Shibaoka and Thimann, 1970). They added that the time to first flower was reduced in the treated plants, and bolls were able to more fully develop compared to control plants. These findings suggested that cytokinin treatment of young cotton seedlings may enhance overall performance and yield, especially in water-limited environments. The effect of benzyl adenine on increasing the phenolic content in cotton plants was reported (Abdel-Al *et al*., 1988). It may be attributed to the increase in carbohydrate synthesis. The increase in total phenolic content was concured with the increases in IAA contents in the plant shoots and led to the suggestion that most of phenolic compounds are diphenols and polyphenols which may inhibit IAA oxidase activity resulting in auxin accumulation, which reflected in stimulating the growth and yield of plant as reported by Sadak (2005). However, BA is used to regulate plant growth through increased meristematic activity due to enhance cell division and elongation (Sardoei, 2014).

The present study aimed to investigate the effect of exogenous application with cytokinin; 6-benzyl adenine (BA) on chemical composition, growth parameters, earliness measurements , fiber traits as well as yield and its components of cotton plant (Giza 86 cv) grown under El-Gemmeiza region.

MATERIALS AND METHODS

Two field experiments were conducted at El-Gemmeiza Agricultural Research Station, Agricultural Research Center, Egypt in two consecutive seasons (2018

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and 2019). Four levels of BA (6- benzyl adenine), three spraying number and date of applications as well as their interactions were examined.

In order to cytokinins affect branching or flowering, they must be absorbed by the meristem or on the stem below it. Spray solutions should be pH adjusted to neutral pH levels to improve absorption. Foliar sprays may be made with hand sprayers.

Soil samples were obtained from the soil surface (0-30 cm), mixtured and some properties of the soil were done according to the methods described by Jackson (1973). Data are presented in Table 1.

Table 1. The properties of the experimental soil sites in the two seasons

Particulars	Season 2018	Season 2019
Mechanical analysis		
Clay%	38.0	44.2
Silt%	38.0	33.0
Sand%	24.0	22.8
Texture	Clay loam	Clayey
Chemical analysis		
pH	8.0	8.1
EC ds/m^2	0.37	0.99
Organic matter %	1.23	1.40
Total N (mg/100g)	43.05	49.00
Available P (mg/100g)	1.19	1.28
Exchangable K (mg/100g)	21.5	31.0
Available Mg (mg/100g)	19	23
Available Fe (ppm)	6.0	12.4
Available Mn (ppm)	2.1	3.9
Available Zn (ppm)	0.70	1.12
Available Cu (ppm)	0.9	1.7

Experimental design was laid in a split plot design with three replications, where number and date of applications occupied the main plots as following**;** a_1-One spray at squaring stage, a_2-Two sprays at squaring stage and flowering initiation and a_3- Three sprays at squaring stage, flowering initiation and the top of flowering stage. While, benzyl adenine concentrations were assigned as sub plots as following; b_1-Control, b_2-20 ppm, b_3-40 ppm and b_4-80 ppm.

Seed sowing and date of spraying plant leaves are presented in Table 2. Dates of foliar spraying coincide with the planting physiology stages; squaring stage, flowering initiation sand peak of flowering stage.

Table 2. Date of sowing and spraying with benzyl adenine in the two growing seasons, 2018 and 2019.

Sowing dates	Dates of spraying		
	Squaring stage	Flowering initiation	Peak of flowering stage
8/4/2018	2/6	3/7	20/7
10/4/2019	1/6	2/7	18/7

The experimental plot area was 14 m^2 (3.5 m × 4 m), (5 ridges, with 70 cm width). Hills were 25 cm apart and sowing was tooke place on one side of the ridge (16 hills per ridge). Seedlings were hand thinning at 2 leaf stage to leave two vigour's seedlings hill^{-1}, providing plant density of 48,000 plants fad^{-1}.

Phosphorus in the form of ordinary superphosphate (15.5% P_20_5) was applied during land preparation at the rate of 22.5 kg P_20_5 fad^{-1}. Nitrogen fertilizer in the form of ammonium nitrate (33.5% N) was applied at the rate of 45 Kg N fad^{-1} in two equal amounts (22.5 + 22.5 Kg N fad^{-1}). Half of nitrogen fertilizer was applied before the irrigation post thinning and the rest with the next irrigation. Potassium fertilizer in the form of Potasin-P at the rate of 1 liter fad^{-1} was applied as foliar application three times (at squaring stage, flowering start and the top of flowering). Other agronomic practices were adopted as recommended by CRI, ARC, Egypt.

Studied characters:

A- Leaves chemical composition: In 2018 season, after 116 days from sowing (after 15 days from the last spraying), a leaf sample of 20 leaves (blade + petiole) was taken at random from the youngest fully matured leaves (4th leaf from the apex of the main stem) to determine the percentages of N, P, K and Mg according to Chapman and Parker (1981). Chlorophyll (a), (b) and total chlorophyll (mg/g dry weight) were determined as reported by Fadeel (1962). Carotenoids concentration (mg/g dry weight) was also determined as described in A.O.A.C. (1995).

B-Growth parameters: In both seasons, six guarded plants were taken at random from each sub-plot carefully after 116 days from sowing to determine dry weight of assimilatory parts (g plant^{-1}). They were washed and dried to a constant weight in a forced air oven at 70 °C and weighted. Leaf area index (LAI) was determined as proposed by Hunt (1978). At harvesting, ten representative plants from each sub-plot were taken at random to determine final plant height (cm) and its number of fruiting branches.

C-Earliness measurements: Ten representative plants of each-sub plot were taken at random. The following earliness measurements were calculated according to Richmond and Radwan (1962) and Kadapa (1975); 1- Number of total flowers plant^{-1}, 2- Number of total bolls set plant^{-1}, 3- Boll setting percentage, 4- Boll shedding percentage and 5-Earliness% (percentage of the first picking yield to total yield).

D- Seed cotton yield and its contributory characters: At harvesting, data were taken from the above ten representative plants to determine the following yield characters; 1- Number of open bolls plant^{-1}, 2- Boll weight (g), 3- Lint percentage as percentage of lint cotton to seed cotton after ginning and 4-Seed index (g). The seed cotton yield fad^{-1} was estimated as the weight of seed cotton in kilograms picked twice from each sub-plot and transformed to kentars fad^{-1} (one kentar = 157.5 kg)

E- Fiber measurements:

Fiber tests were made at the laboratories of the Cotton Technology Research Division, Cotton Research Institute, Agricultural Research Center, Giza, Egypt. Measurements included; (i) 2.5% fiber span length (millimeters) and uniformity ratio (%) as measured by a digital fibrograph, (ii) Micronaire reading (a combined measure of fiber fineness and fiber maturity) measured by a micronaire instrument, and (iii) Fiber strength measured by the Pressley tester and expressed as Pressley index (according to Annual Book of A.S.T.M. Standards, 2012).

Statistical Analysis: The obtained data were subjected to statistical analysis of variance for each season according to the procedure described by Snedecor and Cochran (1990). Significance of differences among variables was done according to Least Significant Differences test (LSD) at 5% level of probability.

RESULTS AND DISCUSSION

A-Leaf chemical composition:

1-Effect of number and date of applications:

Table 3 shows that application of benzyl adenine three times at squaring, flowering initiation and the top of flowering stages exhibited a significant increase in the content of N, P, K and Mg, as well as the photosynthetic pigments [chlorophyll (a and b) and carotenoids] compared with single or twice treatments.

2- Effect of benzyl adenine concentration:

Results in the same table show that the differences in leaves percentages of nitrogen, phosphorus, potassium and magnesium were statistically significant as a result of spraying cotton plants with different concentrations of BA, where the highest percentages of N, P, K and Mg were recorded in the plants sprayed with BA at the high concentration 80 ppm. The lowest percentages of these four nutrients were recorded in the leaves of control plants.

Moreover, it was found that the leaves content of three photosynthetic pigments chlorophyll (a and b) and carotenoids were gradually increased as the concentration of BA was increased. The highest values of chlorophyll (a and b) and carotenoids were obtained at the highest concentration (Table 3). Higher concentrations of BA (40 or 80 ppm) had a favourable effect on chlorophyll synthesis and accumulation in the leaves, and gave higher values than the control, or plants sprayed with the lower concentration (20 ppm BA). The highest total chlorophyll content was found in the leaves of plant sprayed with the high BA concentration (80 ppm).

3-The interaction effect:

Regarding the interaction effect, the results in Table 3 showed that there was a significant effect on leaves percentages of N, P, K and Mg and concentration of the three photosynthetic pigments [chlorophyll (a and b) and carotenoids] in 2018 season, where the highest values were recorded with application of benzyl adenine three times at the high concentration. While, the lowest ones were obtained from the control (one water spraying).

The increasing effects of BA on the photosynthetic pigments noticed in this study may be due to its protecting effect on chlorophylls against photo-oxidation (Petrenko and Biryukova, 1977), retention of chlorophyll and inhibits its degradation (Van Staden and Joughin, 1988), delaying the aging process (Xu *et al*., 2011) and enhancing formation of one or more proteins to which chlorophyll binds and becomes stabilized (Salisbury and Ross, 1990) and inhibiting de novo synthesis of the enzyme (chlorophyllase enzyme) that is involved in the degradation of chlorophyll (Beyer, 1981). It maintains a high protein content by retarding the rate of breakdown rather than enhancing the rate of synthesis (Sacher, 1973) and delays the onset of rising respiration associated with leaf senescence (Thimann, 1980).

Table 3. Effect of number and date of foliar spraying applications with benzyl adenine at different concentrations as well as their interactions on certain chemical composition of cotton leaves in 2018 season.

Traits / Treatments		N	P	K	Mg	Chlorophyll a	Chlorophyll b	Total chlorophyll	Carotenoids
		%				(mg /g dry weight)			
A- *Number and date of applications:									
a_1- One spray		2.48	0.39	2.73	0.446	3.98	1.36	5.34	1.46
a_2-Two sprays		2.63	0.43	2.94	0.463	4.06	1.40	5.46	1.47
a_3-Three sprays		2.72	0.47	3.02	0.498	4.14	1.42	5.56	1.51
LSD at 0.05		0.06	0.01	0.04	0.011	0.01	0.01	0.01	0.03
B- Benzyl adenine concentrations:									
b_1 - Control		2.41	0.34	2.62	0.397	3.92	1.32	5.24	1.40
b_2 – 20 ppm		2.61	0.40	2.76	0.460	4.05	1.39	5.44	1.48
b_3 = 40 ppm		2.66	0.47	3.06	0.504	4.12	1.42	5.54	1.52
b_4 - 80 ppm		2.76	0.50	3.15	0.516	4.16	1.44	5.60	1.52
LSD at 0.05		0.04	0.01	0.03	0.018	0.03	0.01	0.04	0.03
AXB Interaction									
a_1	b_1	2.40	0.33	2.60	0.400	3.92	1.31	5.23	1.40
	b_2	2.46	0.35	2.70	0.430	3.97	1.36	5.33	1.47
	b_3	2.50	0.42	2.78	0.467	4.01	1.37	5.38	1.46
	b_4	2.57	0.46	2.85	0.487	4.03	1.39	5.42	1.50
a_2	b_1	2.41	0.34	2.62	0.400	3.90	1.32	5.22	1.42
	b_2	2.69	0.40	2.76	0.450	4.07	1.39	5.46	1.43
	b_3	2.69	0.45	3.12	0.503	4.12	1.43	5.55	1.53
	b_4	2.75	0.50	3.25	0.500	4.16	1.44	5.60	1.51
a_3	b_1	2.42	0.36	2.63	0.390	3.93	1.33	5.26	1.39
	b_2	2.68	0.44	2.82	0.500	4.11	1.41	5.52	1.54
	b_3	2.81	0.53	3.27	0.543	4.23	1.47	5.70	1.56
	b_4	2.98	0.55	3.35	0.560	4.29	1.48	5.77	1.54
LSD at 5%		0.08	0.02	0.05	0.031	0.12	0.02	0.04	0.05

***a_1-One spray at squaring stage, a_2-Two sprays at squaring stage and flowering initiation and a_3- Three sprays at squaring stage, flowering initiation and the top of flowering stage.**

Repeated BA treatments induced more higher concentration of leaves N, P, K, Mg and the photosynthetic pigments compared with single or twice treatments. It may be due to enhanced chlorophyll retention which attributed to one or more of the followings; 1-BA increases the absorption and translocation of the elements necessary for plant growth through increasing the width of conductive tissues (xylem and phloem) (Krishnamoorthy, 1981). 2- BA enhances translocation and rate of ion entry through the membrane permeability and the mechanism of ions uptake (Van-Steveninck, 1976). 3-BA promotes the opening of plant leaves stomata, thus enhancing gas (CO2) exchange, increasing photosynthesis and accumulation of carbohydrate in the treated plants leaves (Wareing and Phillips, 1981). 4-It promotes formation of grana (the stacks of thylakoids, which contain photosynthetic pigments) and an increase in the volume of grana coupled with a high chloroplast DNA content (Momotani *et al*., 1991) increases the rate of chlorophyll formation (Lew and Tsuji, 1982). Kassem *et al*. (2009) reported that cotton plants treated with various treatments of kinetin (10 and 20 ppm) showed higher leaves content of chlorophyll a, b and total chlorophylls than those of untreated ones. 5-It activates the synthesis of two proteins of the chloroplast; RUBP carboxylase and the chlorophyll a/b protein complex (Funkees-Shippy and Levine, 1985). 6- The increase in the leaf's contents of these four nutrients due to BA application is attributed to the influence of BA in augmenting the leaves dry weights to a higher degree than the leaves of control plants (Table 4).

B-Growth traits:

1- Effect of number and date of applications:

Data in Table 4 indicated significant differences in growth parameters of cotton plants as indicated by leaf area index, dry weight (g plant^{-1}) after 116 days old, plant height and number of fruiting branches at harvesting compared with the control (untreated plants) due to number and date of benzyl adenine applications. Plant height, leaf area index and dry weight (g plant^{-1}) were significantly reduced under one spraying at squaring stage (141.00 cm, 2.61 and 104.08 g;152.62 cm, 2.44 and 105.67 g) in the first and second seasons, respectively, whereas application of benzyl adenine three times at squaring stage, flowering initiation and the top of flowering exhibited the maximum values of these traits (150.33 cm, 2.81 and 120.82 g; 160.09 cm, 2.65 and 116.26 g). Tow foliar applications of benzyl adenine at squaring stage and flowering initiation being in the middle and recorded the highest number of fruiting branches (17.13 and 15.75 branch).

2- Effect of benzyl adenine concentration:

Data presented in Table 4 show that, the foliar application of different concentrations of benzyl adenine (BA) had significantly stimulatory effect on growth parameters of cotton plants in term of leaf area index as well as dry weight (g plant^{-1}) after 116 days old compared with the control (untreated plants). Treatment with BA at 80 ppm gave the highest values of LAI (2.88 and 2.72) and plant dry weight (121.60 and 120.14 g) compared with the other treatments and control.

Data in the same table indicated significant effects of benzyl adenine concentration on plant height and number of fruiting branches at harvest in both seasons Plant height was taller (151.37 and 162.52 cm) with application of benzyl adenine at the high concentration (80 ppm), while shorter plants were recorded under control (137.91 and 149.17 cm) in the first and second seasons, respectively. Increasing the concentration resulted in significant increase in plant height. However, the highest number of fruiting branches plant^{-1} (17.07 and 16.66 branch) was obtained from using the medium concentration (40 ppm). Least number (14.41 and 14.83 branch) was recorded under control in the first and second seasons, respectively.

Table 4. Effect of number and date of foliar spraying applications with benzyl adenine at different concentrations as well as their interaction on cotton growth traits studied in 2018 and 2019 seasons

Treatments		Leaf area index		Above ground dry weight(g plant^{-1})		Plant height(cm)		No. of fruiting branches plant^{-1}	
		After 116 days old				At harvesting			
		2018 season	2019 season	2018 season	2019 season	2018 season	2019 season	2018 season	2019 season
A-*Number and date of applications:									
a_1- One spray		2.61	2.44	104.08	105.67	141.00	152.62	15.10	15.73
a_2-Two sprays		2.72	2.56	113.04	111.69	147.40	157.85	17.13	15.75
a_3-Three sprays		2.81	2.65	120.82	116.26	150.33	160.09	15.24	15.48
LSD at 0.05		0.04	0.02	0.80	1.53	0.53	0.73	0.32	0.15
B-Benzyl adenine concentrations:									
b_1 - Control		2.53	2.33	99.82	100.11	137.91	149.17	14.41	14.83
b_2 – 20 ppm		2.66	2.53	112.27	109.67	146.31	156.12	16.02	15.26
b_3 = 40 ppm		2.78	2.63	116.89	114.90	149.39	159.60	17.07	16.66
b_4 - 80 ppm		2.88	2.72	121.60	120.14	151.37	162.52	15.78	15.89
LSD at 0.05		0.03	0.03	1.32	0.96	0.28	0.60	0.32	0.36
A x B interaction:									
a_1	b_1	2.51	2.30	97.40	99.00	138.05	150.20	14.28	14.90
	b_2	2.57	2.41	102.00	104.13	139.82	151.27	15.22	17.00
	b_3	2.65	2.50	104.80	108.90	142.22	153.30	16.61	16.10
	b_4	2.69	2.56	112.10	110.63	143.89	155.70	14.28	14.97
a_2	b_1	2.54	2.33	100.20	99.90	137.89	149.03	14.56	14.97
	b_2	2.68	2.53	115.37	109.43	148.33	157.13	17.39	13.20
	b_3	2.79	2.64	117.73	115.67	150.56	163.43	18.17	17.63
	b_4	2.87	2.73	118.87	121.77	152.83	161.80	18.39	17.20
a_3	b_1	2.55	2.37	101.87	101.43	137.70	148.27	14.39	14.63
	b_2	2.73	2.63	119.43	115.43	150.78	159.97	15.44	15.57
	b_3	2.89	2.76	128.13	120.13	155.39	163.70	16.44	16.67
	b_4	3.06	2.86	133.83	128.03	157.39	168.43	14.67	15.07
LSD at 0.05		0.05	0.05	2.29	1.66	0.83	1.04	0.56	0.63

***a_1-One spray at squaring stage, a_2-Two sprays at squaring stage and flowering initiation and a_3- Three sprays at squaring stage, flowering initiation and the top of flowering stage.**

3-The interaction effect:

Table 4 showed that, there was a significant effect on growth parameters of cotton plants in term of leaf area index as well as dry weight (g plant^{-1}) after 116 days old compared with the control (untreated plants). Application of benzyl adenine three times at the high concentration gave the highest values of LAI (3.06 and 2.86) and plant dry weight (133.83 and 128.03 g) compared with the other treatments and control.

Also, the interaction gave a significant effect on plant height and its number of fruiting branches in both seasons, where the longest plants (157.39 and 168.43 cm) were recorded with application of benzyl adenine three times at the high concentration. While, the shortest ones (137.7 and 148.27 cm) were obtained from untreated (three water sprayings) in the first and the second seasons, respectively. The highest numbers of fruiting branches plant^{-1} (18.39 and 17.63 branch) were given with two foliar sprayings at the high concentration (80 ppm) and at the medium concentration (40 ppm), while the lowest ones (14.28 and 13.20 branch) were obtained from untreated (one water spraying) in the first season and from two foliar spraying with benzyl adenine at the low concentration (20 ppm) in the second season.

The enhanced dry matter accumulation due to BA application is mainly attributed to a high leaf area index in BA treated plants and a largest leaf area for CO_2 fixation, resulting in enhanced carbohydrate metabolism and created new source-sink relationships and promoted development rate, hence accumulation of dry matter increased (Gardner *et al*., 1985).

The positive effect of the high concentration compared to the untreated plants is mainly due to the effect of BA on increasing meristematic activity, cellular division, cell elongation and differentiation, resulting in increased plant height, dry matter accumulation, LAI, photosynthesis rate through increasing leaf surface. Kassem *et al*. (2009) reported that cotton plants treated with various treatments of kinetin (10 and 20 ppm) significantly increased plant height and number of fruiting branches plant^{-1}in both seasons.

C-Earliness traits:

1-Effect of number and date of applications:

Two foliar sprayings (at squaring stage and flowering initiation) with benzyl adenine significantly increased number of total bolls set plant^{-1}, boll setting percentage and first picking% in both seasons and number of total flowers plant^{-1}in the second season (Tables 5 and 6), in comparison to foliar spraying with benzyl adenine once (at squaring stage) or thrice (at squaring stage, flowering initiation and the top of flowering stage), where the differences between the two latter treatments were insignificant. Boll shedding percentage follows the reverse trend.

2- Effect of benzyl adenine concentrations:

Significant distinctions were detected among the four concentrations of benzyl adenine as for number of total bolls set plant^{-1}, boll setting percentage and first picking% in both seasons and number of total flowers plant^{-1}in the second season (Tables 5 and 6), in a favour of applying benzyl adenine as foliar spraying at the medium concentration (40 ppm). However, the lowest values of these traits in consideration resulted from untreated plants (without benzyl adenine application). Boll shedding percentage follows the opposite trend. In respect of the concentrations of benzyl adenine effect on the boll shedding percentage, data in Tables 5 and 6 show significant differences among the four concentrations in both seasons regarding boll shedding percentage, where the boll shedding percentage significantly increased from 37.78 and 20.80% in the medium concentration (40 ppm) to 45.61 and 34.54 % in the control treatment in the first and second seasons, respectively.

Table 5. Effect of foliar spraying with benzyl adenine concentration, number of application and their interactions on earliness measurements in 2018 season

Traits / Treatments		No. of total flowers plant^{-1}	No. of total bolls set plant^{-1}	Boll setting %	Boll shedding %	Earliness %
A-*Number and date of applications:						
a_1- One spray		28.28	16.47	58.24	41.76	62.90
a_2-Two sprays		28.25	17.22	60.97	39.04	65.84
a_3-Three sprays		28.26	16.54	58.55	41.45	63.23
LSD at 0.05		NS	0.33	0.98	0.98	1.05
B-Benzyl adenine concentrations:						
b_1 - Control		28.31	15.40	54.39	45.61	58.74
b_2 – 20 ppm		28.39	17.02	59.95	40.05	64.75
b_3 = 40 ppm		28.14	17.51	62.22	37.78	67.20
b_4 - 80 ppm		28.21	17.05	60.44	39.56	65.27
LSD at 0.05		NS	0.24	0.92	0.92	1.00
A x B interaction:						
a_1	b_1	28.30	15.87	56.10	43.90	60.59
	b_2	28.37	16.70	58.87	41.13	63.58
	b_3	28.10	17.21	61.26	38.74	66.16
	b_4	28.37	16.09	56.71	43.29	61.25
a_2	b_1	28.37	15.14	53.39	46.61	57.66
	b_2	28.37	17.39	61.30	38.70	66.21
	b_3	28.17	17.73	62.94	37.06	67.98
	b_4	28.10	18.61	66.23	33.77	71.53
a_3	b_1	28.27	15.17	53.67	46.33	57.97
	b_2	28.43	16.97	59.69	40.31	64.46
	b_3	28.17	17.59	62.46	37.54	67.46
	b_4	28.17	16.44	58.38	41.62	63.05
LSD at 5%		NS	0.42	1.60	1.60	1.73

***a_1-One spray at squaring stage, a_2-Two sprays at squaring stage and flowering initiation and a_3- Three sprays at squaring stage, flowering initiation and the top of flowering stage**

3-The interaction effect:

The interaction treatments between benzyl adenine concentrations as well as number and date of its application, showed that, number of total bolls set plant^{-1}, boll setting percentage and first picking% were significantly differed over the two seasons of study and number of total flowers plant^{-1}in the second season, where two foliar sprayings (at squaring stage and flowering initiation) with benzyl adenine at the high concentration (80 ppm) gave the highest values of these traits (Tables 5 and 6). However, the lowest values resulted from untreated plants (without benzyl adenine application). Boll shedding percentage showed a reverse trend in this respect.

Application of benzyl adenine twice at the high concentration (80 ppm) produced highest earliness traits due to the delayed flower senescence. The possible role of BA on ethylene production (Serek *et al*., 1994), cell division (Vreugdenhil, 2004), chloroplast pigments biosynthesis (Bondok *et al*., 1995), uptake of nutrients especially potassium (Guo *et al*., 1994), inflorescence meristem activity and floral initiation (Werner and Schmulling, 2009), photosynthetic activity (Oosterhuis and Zhao, 1998), carbohydrate metabolism in the anther which leading to normal pollen development (Bhadula and Sawhnev, 1989), were detected. It was found that carbohydrate plays an essential role in pollen tube growth (Negi *et al*, 2009). Sandstedt (1971) found that

BA inhibited shedding through stimulating production of auxin. After 4-9 days from flowering, whereas cytokinins reaching their peak activity in cotton bolls and at 18 days their activity declined. Cytokinins are responsible for reduction in ethylene sensitivity of plants the possible reason of delayed flower senescence by BA foliar application may be achieved (Serek *et al*., 1994). In addition to CK important roles in controlling and stimulating cell division, the cytokinins and their derivatives also inhibit leaf senescence.

Table 6. Effect of foliar spraying with benzyl adenine concentration, number of application and their interactions on earliness measurements in 2019 season

Traits / Treatments		No. of total flowers plant^{-1}	No. of total bolls set plant^{-1}	Boll setting %	Boll shedding %	Earliness %
A-*Number and date of applications:						
a_1- One spray		29.63	21.88	73.79	26.21	65.63
a_2-Two sprays		30.12	22.93	76.07	23.93	68.78
a_3-Three sprays		29.80	21.61	72.41	27.59	64.83
LSD at 0.05		0.10	0.53	1.74	1.74	1.43
B-Benzyl adenine concentrations:						
b_1 - Control		29.13	19.07	65.46	34.54	57.20
b_2 – 20 ppm		30.17	22.67	75.14	24.86	68.00
b_3 = 40 ppm		30.29	23.99	79.20	20.80	71.97
b_4 - 80 ppm		29.80	22.82	76.56	23.44	68.47
LSD at 0.05		0.20	0.55	1.73	1.73	1.71
A x B interaction:						
a_1	b_1	28.87	20.00	69.28	30.72	60.00
	b_2	30.03	22.33	74.36	25.64	67.00
	b_3	30.20	23.60	78.15	21.85	70.80
	b_4	29.40	21.57	73.36	26.64	64.70
a_2	b_1	29.67	18.97	63.93	36.07	56.90
	b_2	30.27	22.97	75.88	24.12	68.90
	b_3	30.47	24.60	80.75	19.25	73.80
	b_4	30.07	25.17	83.70	16.30	75.50
a_3	b_1	28.87	18.23	63.16	36.84	54.70
	b_2	30.20	22.70	75.17	24.83	68.10
	b_3	30.20	23.77	78.70	21.30	71.30
	b_4	29.93	21.73	72.61	27.39	65.20
LSD at 5%		0.35	0.95	3.47	3.47	2.86

***a_1-One spray at squaring stage, a_2-Two sprays at squaring stage and flowering initiation and a_3- Three sprays at squaring stage, flowering initiation and the top of flowering stage**

A comparative analysis of retained and naturally abscising cotton fruits was made by Rodgers (1981) and found that cytokinins promote fruit growth and reduce shedding of flower bud and young boll. He added that abscission was negatively correlated with the cytokinins concentration. Treated with BA appear to coincide with the stimulated changes in leaf area, dry weight and photosynthetic pigments as well as the content of N, P, K and Mg. Suttle (1986) reported that after cytokinin application, production of ethylene in cotton leaves stimulated as a result of an increase in the formation and oxidation of ACC, production of ethylene 24 hours after cytokinin application reduced which due to synthesis and oxidation of inhibitors of I-amino cyclopropane-l-carboxylic acid (ACC) to ethylene; however, by 48 hours only inhibitors of ACC oxidation were effective. The increase in ethylene production was accompanied by a massive accumulation of ACC and its acid-labile conjugate. Mayeux (1985) showed that Burst (a commercial cytokinin product) applied at early bloom or applied at mid bloom significantly increased the boll set and fruiting of cotton plants. Bauer and Cothren (1986) indicated that cytokinin increased photosynthesis greatly, flowering, boll retention, and yield and they found that kinetin application as foliar spray of 10 mg L^{-1} once at the flowering stage, increased dry matter production, decreased boll shedding and increased seed cotton yield. Insignificant differences in earliness were found due to cytokinin application (Namken, 1984). Sawan *et al*. (2000) found insignificant effect on yield earliness as a result of spraying cotton plants with kinetin at 5 mg l^{-1} at 60 and 75 days after sowing. Kassem *et al*. (2009) reported that cotton plants treated with kinetin (10 and 20 ppm) significantly increased total fruiting sites plant^{-1} and decreased fruit shedding% and earliness%.

D-Yield and its components:

1- Effect of number and date of applications:

Results in Tables 7 and 8 show that, two foliar applications of benzyl adenine at squaring stage and flowering initiation significantly increased and raised the seed cotton yield fed^{-1} by more than 5.40 and 12.50%; 4.67 and 7.40% in comparison to one spraying (at squaring stage) and three sprayings (at squaring stage, flowering initiation and the top of flowering) in the first and second seasons, respectively. Also, the two foliar applications significantly increased number of open bolls plant^{-1} (17.19 and 22.93) as compared to once or three sprays which produced 16.42 and 16.46; 21.88 and 21.61 in the first and second seasons, respectively. The values of boll weight and lint% among the treatments were found to be insignificant in both seasons. However, seed index was significantly affected by number and date of applications in the second season only, in favour of three sprays followed by two sprays and at last by one spraying.

2- Effect of benzyl adenine concentration:

Concerning the effects of benzyl adenine (BA) on yield components, data recorded in Tables 7 and 8 show that foliar application of benzyl adenine at various concentrations examined (20, 40 and 80 ppm) caused a significant increase in number of open bolls plant^{-1} in both seasons, boll weight in the first season and seed index in the second season compared to the control (untreated plants) which recorded the lowest values.

With respect to benzyl adenine concentration effects, it was found that seed cotton yield fed^{-1} showed highest value (12.54 and 10.82 kentar) with the application of 40 ppm (Tables 7 and 8). It increased the seed cotton yield by 26.28 and 15.35% than the control in the first and second seasons, respectively.

Table 7. Effect of number and date of foliar spraying applications with benzyl adenine at different concentrations as well as their interactions on seed cotton yield and its attributes in 2018 season

Traits / Treatments		No. of open bolls plant^{-1}	Boll weight (g)	Seed index (g)	Lint %	Seed cotton yield (kentar fed^{-1})
A-*Number and date of applications:						
a_1- One spray		16.42	3.36	10.28	41.11	11.47
a_2-Two sprays		17.19	3.35	10.03	42.22	12.09
a_3-Three sprays		16.46	3.37	10.04	41.34	11.55
LSD at 5%		0.33	NS	NS	NS	0.25
B-Benzyl adenine concentrations:						
b_1 - Control		15.13	3.25	9.82	41.87	9.93
b_2 – 20 ppm		17.02	3.41	10.13	41.47	12.18
b_3 = 40 ppm		17.54	3.39	10.08	41.33	12.54
b_4 - 80 ppm		17.05	3.40	10.44	41.57	12.16
LSD at 5%		0.24	0.02	NS	NS	0.14
Interaction (A x B):						
a_1	b_1	15.67	3.14	9.97	41.11	9.94
	b_2	16.70	3.43	10.35	40.77	12.01
	b_3	17.21	3.42	10.16	41.84	12.41
	b_4	16.09	3.45	10.66	40.72	11.54
a_2	b_1	14.91	3.29	9.91	42.53	9.93
	b_2	17.39	3.41	10.26	42.86	12.52
	b_3	17.83	3.38	9.83	41.60	12.75
	b_4	18.61	3.32	10.11	41.91	13.15
a_3	b_1	14.81	3.31	9.57	41.97	9.90
	b_2	16.97	3.38	9.77	40.77	12.01
	b_3	17.59	3.36	10.26	40.56	12.47
	b_4	16.44	3.43	10.56	42.09	11.79
LSD at 5%		0.41	0.05	NS	NS	0.24

***a_1-One spray at squaring stage, a_2-Two sprays at squaring stage and flowering initiation and a_3-Three sprays at squaring stage, flowering initiation and the top of flowering stage.**

Table 8. Effect of number and date of foliar spraying applications with benzyl adenine at different concentrations as well as their Interactions on seed cotton yield and its attributes in 2019 season

Traits / Treatments		No. of open bolls plant^{-1}	Boll weight (g)	Seed index (g)	Lint %	Seed cotton yield (kentar fed^{-1})
A-*Number and date of applications:						
a_1- One spray		21.88	2.94	9.87	40.31	9.68
a_2-Two sprays		22.93	3.10	10.26	41.26	10.89
a_3-Three sprays		21.61	3.09	10.84	39.97	10.14
LSD at 5%		0.53	NS	0.40	NS	0.11
B-Benzyl adenine concentrations:						
b_1 - Control		19.07	3.01	10.07	40.40	9.38
b_2 – 20 ppm		22.67	3.12	10.72	40.62	10.41
b_3 = 40 ppm		23.99	3.02	10.45	40.10	10.82
b_4 - 80 ppm		22.82	3.02	10.05	40.94	10.32
LSD at 5%		0.55	NS	0.25	NS	0.16
Interaction (A x B):						
a_1	b_1	20.00	2.98	9.65	39.74	8.88
	b_2	22.33	3.14	10.52	41.10	10.08
	b_3	23.60	2.77	9.80	39.62	10.50
	b_4	21.57	2.86	9.50	40.78	9.23
a_2	b_1	18.97	2.97	10.04	41.04	9.74
	b_2	22.97	3.00	10.50	41.91	10.94
	b_3	25.17	3.15	10.34	40.99	11.66
	b_4	24.60	3.28	10.16	41.09	11.22
a_3	b_1	18.23	3.08	10.51	40.42	9.52
	b_2	22.70	3.21	11.13	38.86	10.22
	b_3	23.77	3.13	11.20	39.68	10.75
	b_4	21.73	2.93	10.50	40.94	10.06
LSD at 5%		0.95	NS	NS	NS	0.27

***a_1-One spray at squaring stage, a_2-Two sprays at squaring stage and flowering initiation and a_3- Three sprays at squaring stage, floweringinitiation and the top of flowering stage.**

3-The interaction effect:

All interaction treatments increased number of open bolls and seed cotton yield fed^{-1}in both seasons and boll weight in the first season only (Tables 7 and 8). In the first season, the highest number of open bolls (18.61) and seed cotton yield fed^{-1} (13.15 kentar) were obtained from two foliar sprayings with benzyl adenine at the high concentration (80 ppm), meanwhile the lowest number of open bolls (14.81) and seed cotton yield fed^{-1} (9.90 kentar) were recorded from the control. The heaviest bolls (3.45 g) resulted from one foliar spraying with benzyl adenine at the high concentration (80 ppm), while the lowest value of boll weight (3.14 g) was obtained from the control. In the second season, the highest number of open bolls (25.17) and seed cotton yield fed^{-1} (11.66 kentar) were obtained from two foliar sprayings with benzyl adenine at the medium concentration (40 ppm), meanwhile the lowest number of open bolls (18.23) and seed cotton yield fed^{-1} (8.88 kentar) were recorded from the control.

These results might be attributed to the stimulating effect of CK on cell divisions and regulation of biochemical processes, vegetative growth characteristics and plant constituents, which can influence the fruit and yield. It was found that CK had promotive effect on nutrient transport to cotton leaves (Table 3). In addition, it was found that foliar application with benzyl adenine caused a marked increase in the protein percentage and total carbohydrates, where BA activates enzymes which regulate carbohydrate metabolism reflected on increase in carbohydrate accumulation (Cao and Shannon, 1997). The tallest plants, higher number of branches plant^{-1} and heavier dry weight of plant were obtained from plants sprayed with BA may stimulate earlier cytokinin formation thus preventing premature leaf senescence and resulting in more vegetative growth with higher photosynthetic pigments.

In this respect, Abdel-Al *et al*. (1989) found little effect on lint percentage and positive response on seed index due to cytokinin application. Hedin and McCarty (1994) reported that seed cotton yield increased by 26% due to kinetin application. Abed (2001) found that BA application increased the bolls number, average boll weight and seed cotton yield. Burke (2013) found that application of cytokinin (6-benzyladenine) to cotton increased yields and reduced water stress in cotton. Similar results were reported by Namken (1984); Dhopte and Lall (1987); Sawan *et al.* (2000) and Kassem *et al*. (2009).

E-Fiber measurements:

1- Effect of number and date of applications:

Data in Tables 9 and 10 represent the effects of number and date of benzyl adenine applications regarding fibre traits under study *viz.*, fiber length, uniformity index (%), micronaire reading and fiber strength. The effect was insignificant for fiber traits during the two seasons of study.

2- Effect of benzyl adenine concentration:

The same tables represent the effect of benzyl adenine concentration concerning fibre traits under study *viz.*, fiber length, uniformity index (%), micronaire reading and fiber strength. No significant variations for these traits in 2018 and 2019 seasons were detected.

Table 9. Effect of number and date of foliar spraying applications with benzyl adenine at different concentrations as well as their interactions on cotton fiber traits in 2018 season

Treatments	Fiber traits			
	Micronaire reading	Pressley index	2.5% span length (mm)	Uniformity index(%)
	A-*Number and date of applications:			
a_1- One spray	4.62	10.42	34.02	86.17
a_2-Two sprays	4.65	10.77	34.05	86.10
a_3-Three sprays	4.62	10.52	34.00	85.37
LSD at 0.05	NS	NS	NS	NS
	B-Benzyl adenine concentrations:			
b_1 - Control	4.83	10.73	34.17	85.40
b_2 – 20 ppm	4.60	10.63	33.87	86.03
b_3 = 40 ppm	4.60	10.20	34.03	86.33
b_4 - 80 ppm	4.50	10.73	34.03	85.77
LSD at 0.05	NS	NS	NS	NS
	A x B Interaction:			
LSD at 0.05	NS	NS	NS	NS

***a_1-One spray at squaring stage, a_2-Two sprays at squaring stage and flowering initiation and a_3- Three sprays at squaring stage, flowering initiation and at the top of flowering stage.**

Table 10. Effect of number and date of foliar spraying applications with benzyl adenine at different concentrations as well as their interactions on cotton fiber traits in 2019 season

Treatments	Fiber traits			
	Micronaire reading	Pressley index	2.5% span length (mm)	Uniformity index (%)
	A-*Number and date of applications:			
a_1- One spray	4.85	10.60	34.07	86.42
a_2-Two sprays	5.02	10.77	33.85	85.95
a_3-Three sprays	4.97	10.65	34.17	86.20
LSD at 0.05	NS	NS	NS	NS
	B-Benzyl adenine concentrations:			
b_1 – Control	5.00	10.77	34.03	86.17
b_2 – 20 ppm	5.00	10.70	34.17	86.50
b_3 = 40 ppm	4.87	10.60	34.00	86.13
b_4 - 80 ppm	4.93	10.63	33.93	85.97
LSD at 0.05	NS	NS	NS	NS
	A x B Interaction:			
LSD at 0.05	NS	NS	NS	NS

*** a_1-One spray at squaring stage, a_2-Two sprays at squaring stage and flowering initiation and a_3- Three sprays at squaring stage, flowering initiation and at the top of flowering stage.**

3-The interaction effect:

The interactions between number as well as date of benzyl adenine applications and its concentration showed insignificant effects on fiber traits during both years of study, which indicates independent effect for these two factors on these traits (Tables 9 and 10).

In this respect, Namken (1984) found that cytokinin treatments gave insignificant differences in fiber properties (micronaire, length, uniformity, and strength). Abdel-Al *et al*. (1989) found that micronaire reading and flat bundle strength were not affected by application of cytokinin. Hofmann and Else (1989) found that fiber quality did not affect by kinetin application. Sawan *et al*. (2000) found insignificant effect on fiber properties as a result of spraying cotton plants with kinetin at 5 mg l^{-1} at 60 and 75 days after sowing.

CONCLUSION

It could be concluded that two foliar sprayings (at squaring stage and flowering initiation) with benzyl adenine at the high concentration (80 ppm) or at the medium concentration (40 ppm) were the best treatments for cotton production. They produced the highest yields of seed cotton fed^{-1} (13.15 and 11.66 kentar) in the two studied seasons, respectively.

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التاثير الفسيولوجى للبنزيل أدينين على النمو وعقد اللوز والإنتاجية لنبات القطن

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أجريت هذة الدراسة بمحطة البحوث الزراعية بالجميزة - محافظة الغربية - مصر خلال الموسمين المتتاليين ٢٠١٨ ، ٢٠١٩م بهدف دراسة التأثير الفسيولوجى للرش بالبنزيل أدينين على النمو والعقد والإنتاجية لصنف القطن جيزة ٨٦وتم استخدام تصميم القطع المنشقة مرة واحدة فى ثلاث مكررات حيث اشتملت القطع الرئيسية على عددالرشات وتاريخها (مرة واحدة فى مرحلة الوسواس ، مرتين فى مرحلة الوسواس وبداية التزهير ، ثلاث مرات فى مرحلة الوسواس وبداية التزهير وقمة التزهير) بينما القطع المنشقة اشتملت على اربعة تركيزات من البنزيل أدينين [بدون ، ٢٠ ، ٤٠، ٨٠ جزء فى المليون] . اعطى رش البنزيل أدينين ثلاث مرات زيادة معنوية فى محتوى الأوراق من العناصر وصبغات التمثيل الضوئى موسم ٢٠١٨م. مع زيادة معنوية في صفات النموبأستثناء الافرع الثمرية بينما أعطى الرش مرتان زيادة معنوية في عدد الافرع الثمرية و قياسات التبكير(بإستثناء النسبة المئوية للتساقط والتى إنخفضت) ، ومحصول القطن الزهرللفدان وعدد اللوز المتفتح/النبات فى الموسمين.وقد أعطي التركيز المتوسط (٤٠ جزء فى المليون) زيادة معنوية فى عدد الأفرع الثمرية واللوز الكلى والمتفتح على النبات والنسبة المئوية للعقد والتبكير ومحصول القطن الزهر للفدان فى الموسمين مقارنة بباقى المعاملات بينما أعطي التركيز المرتفع زيادة معنوية فى محتوى الأوراق من العناصر وصبغات التمثيل الضوئى وصفات النمو الخضرى بإستثناء الافرع الثمرية وقد أعطت معاملة المقارنة (الرش بالماء) اقل القيم مع زيادة معنوية فى التساقط ولم تتأثر صفات التيلة معنويا بعاملى الدراسة والتفاعل.وتوصى الدراسة بأستخدام التفاعل بين البنزيل ادينين رشا مرتين فى مرحلة الوسواس وبداية التزهير بالتركيز المتوسط (٤٠جزء فى المليون) تحت ظروف منطقة الجميزة حيث أدى لخفض نسبة التساقط وزيادة العقد وأعطى أقصى انتاجية.