Growth, Yield and Chemical Composition of Okra as Affected by Three Types and Levels of Synthetic Cytokinins under High Temperature Conditions

Sary Hassan Mostafa Ahmed Brengi
Horticulture Department, Faculty of Agriculture, Damanhour University, Egypt.

ABSTRACT

This study was carried out to evaluate the effect of three synthetic cytokinin i.e. Benzyl amino purine (BAP) at (25, and 50 ppm), 2-chloro-4-pyridyl-N-phenylurea (CPPU) at (10, and 20 ppm) and 6-furfuryl-aminopurin (Kinetin) at (25, and 50ppm) on Okra (*Abelmoschus esculentus* L.) cv .Lady's fingers. Two field experiments were executed during the two successive seasons of 2017 and 2018 in a private farm located at Abuo Homoss, El-Baharia Governorate, Egypt. The obtained results of the two seasons, generally, revealed that spraying of the three types and levels of synthetic cytokinins, significantly, increased plant height, number of leaves, number of branches and leaf area plant^{-1} compared to the untreated control. Also, synthetic cytokinins significantly enhanced the yield attributes viz., fruit length, fruit diameter, fruit weight and total yield plant^{-1}. Moreover, spraying of synthetic cytokinins were superior and significantly increased N%, P%, K%, protein % and total chlorophyll in the leaves. Likewise, it increased mean values of N%, P%, K%, protein % and mucilage in fruits. Meanwhile, spraying of BAP at 50 ppm gave the highest mean values of vegetative growth characters and chemical composition on leaves compared to other treatments. However, spraying of kinetine at 25ppm gave the highest mean values of fruit length, fruit diameter, average fruit weight and total yield plant^{-1} in addition to mean values of N%, P%, K%, protein % and mucilage in fruits compared to other treatments. On the other hand, all synthetic cytokinins increased average number of days from planting to the first flower compared to control treatment under high temperature conditions.

Keywords: Synthetic Cytokinin, high temperature, growth and yield.

INTRODUCTION

Okra (*Abelmoschus esculentus* L.) belongs to family Malvaceae. It is one of the most important summer vegetables in Egypt. Okra is a good source of amino acids, protein, vitamin A, B, C, minerals and iodine (Varmudy 2011). Although okra plants grow under a wide range of conditions, it thrives in temperatures between 20°C and 30°C (Benchasri, 2012). Okra is very sensitive crop to the temperature (Asghar, 2016). Extreme temperatures are the most likely damaging environmental factors affecting pollination and fertilization. High temperatures between 40-42°C caused decreases in the growth and yield of okra (Tripathi *et al*., 2011, Ahmad *et al*., 2016 and Hayamanesh *et al*., 2016). High temperature straight for a short duration can cause reduction in respiration, inhibition of photosynthesis, interrupts plant water status due to high rate of leaf transpiration and abortion of buds quality (Abdul-Baki, 1991). Global warming due to climate change caused a heat stress, which reduce crop production and no country is safe with it (Hall, 2001). Plant growth regulators at small amounts can adjust plant physiological process and plays a vital role in plant growth and development, such as elongation and flower development (Yamaguchi and Kamiya, 2000; Meera and Poonam, 2010). The most important types of plant growth regulators classes are grouped into plant growth promoters such as auxins, gibberellins, and cytokinins. In addition, plant growth inhibitors such as abscisic acid and ethylene.

One of important growth promoters is cytokinins, which playing a vital role from seed germination to senescence (Chatsudthipong and Muanprasat, 2009). Synthetic cytokinin such as benzyl amino purine (BAP), 2-chloro-4-pyridyl-N-phenylurea (CPPU) and 6-furfuryl-aminopurin (Kinetin) can improve plant growth by cell division, break bud dormancy and promotes the growth of the lateral bud(Hossain *et al*., 2006). Application of synthetic cytokinin on shoot increased chlorophyll formation and protein synthesis within tissues (George and Shemington, 2008). Exogenous application of synthetic CKs, such as N-(2-chloro-pyridin-4-yl)-N'-phenylurea (CPPU) and 6-benzylaminopurine (BAP), can increase plant growth, fruit set and development in vegetable crops such as snap bean, melon and watermelon (Hayata *et al*., 1995, 2000; Zaki *et al*., 2014). Kinetin at 10 ppm as a foliar spray at vegetative and flowering stages increased all growth attributes and yield of okra (Singh 2006). Cytokinin reduced heat stress effects and increased production of artichoke growing under hot fall conditions (Schrader, 2005). The endogenous cytokinin levels decreased under high temperature conditions (Hare et al., 1997; Banowetz *et al*., 1999). This research study was undertaken to migrate heat stress through studying the effect of different synthetic cytokinin types and

levels: Benzyl amino purine (BAP), 2-chloro-4-pyridyl-N-phenylurea (CPPU) and Kinetin on vegetative growth, yield and chemical composition of okra (*Abelmoschus esculentus* L.) under high temperature condition

MATERIALS AND METHODS

The current study was carried out during the two successive summer seasons of 2017 and 2018.The experiments were executed in a private farm situated at Abuo Homoss, El-Baharia Governorate, Egypt, under open field conditions. The objectives of this study are to evaluate the effect of three synthetic cytokinin types and levels (BAP, CPPU and Kinetin) each alone; on growth performance, yield and quality of okra cv "Lady's fingers".

Preceding the initiation of each experiment, soil samples of (0-30cm) depth was collected and analyzed for some physical and chemical properties of the experimental site during the both studied seasons as shown in Table (1) according to the published procedures of Black *et al.* (1965).

The experimental layout was a randomized complete block design with three replications.There were seven treatments; Benzyl amino purine (BAP) at (25, and 50 mg/l), 2-chloro-4-pyridyl-N-phenylurea (CPPU)at (10, and 20 mg/l), Kinetin at (25, and 50 mg/l) and (distilled water). Six concentrations of dissolved synthetic cytokinin types were applied as a foliar spraying until run-off using a hand sprayer. The spraying was applied twice; the first one was carried out after 30days from sowing. The second application was done 20days after the first one. The untreated control plants were sprayed with distilled water. Each sub-plot was separated by a guard row.

The minimum and maximum air temperatures per month in Abuo Homoss area during the two growing seasons are listed in Table (2).

Seeds of okra cv. "Lady's fingers" were sown on 15 March for the first and the second seasons. Each experimental unit contained 5 rows, 12 m long, 0.70 m width and the intra-row spacing was 35 cm apart at the rate of 3 seeds per hill. The experimental unit area was 42 m^2. After 21 days from seed sowing, the seedlings were thinned and the strongest one being remained in each hill. The recommended agricultural practices for commercial okra production were followed.

Data recorded:

The measured characters were divided into three groups as follows:

1- Vegetative growth characters:

Ninety days after seed sowing, five random plants were chosen and recorded for plant height of main shoot (cm), number of branches per plant, number of leaves per plant and leaves area per plant (cm^2). The leaves area per plant were determined by the disk method (Bermner and Taha, 1966).

2- Yield and its components:

After 90 days from seed sowing, the following data were recorded; number of days to the first flower, total number of edible green fruits (pods), total weight of fruits per plant as the weight of all harvested fruits $plant^{-1}$ for each treatment at edible fruit maturity stage. Ten fruits used for measuring fruit diameter (cm), fruit length (cm) and mean weight of the pod (g).

Table 1: Some physical and chemical properties of the experimental site in 2017 and 2018 seasons.

	* Soil properties	2017	2018
Physical properties	Sand (%)	30.59	29.75
	Silt (%)	9.11	10
	Clay (%)	60.3	60.25
	Textural class	Clay	Clay
chemical properties	pH (1:2)	8.12	8.05
	Ca $CO_{3\ (\%)}$	10.19	10.31
	EC (1:2, water extract) dS/m	1.82	1.87
	O.M (%)	1.38	1.4
	Ca^{2+}(meq/l)	9.72	9.02
	Mg^{2+}(meq/l)	2.82	2.91
	K^{+}(meq/l)	1.91	1.72
	HCO_3^{-}(meq/l)	10	9 9.8
	SO_4^{2-} (meq/l)	2.31	2.41
	Nitrogen (ppm)	402	413
	Phosphorus (ppm)	13.9	14.32
	Potassium (ppm)	231	226

*Physical and chemical analyses of the field tested soil were done in the laboratory of organic agriculture, Ministry of Agriculture.

Table 2: Minimum and maximum air temperature per month in Abuo Homoss area during the two growing seasons of 2017 and 2018.

Year	*Temp °C	Month							
		March	April	May	June	July	August	September	October
2017	Min	9.1	12.2	17.4	20.9	23	22.8	19.1	15.2
	Mix	27.7	32.2	37.4	39.8	41.4	40.5	38.4	32.5
2018	Min	11.7	16.1	19.7	21.4	22	22.6	20.1	16.6
	Mix	32.7	36.7	39.5	40.6	41.6	41.2	38.4	34.2

*These data were obtained from the Meteoblue site

3- Chemical composition:

The leaf and fruit samples were taken after 90 days from seed sowing for determining the chemical composition of N, P, K. Nitrogen was determined using micro-kjeldahal method (Chapman and Pratt, 1961). Each of potassium and phosphorus were determined by flame photometer according to the method of Temminghoff and Houba (2004). Moreover, total protein (%) was calculated by multiplying Nitrogen% by 6.25. Also, mucilage content in fruits was estimated according to the method of (Rao and Sulladurath, 1977).

Total chlorophyll (SPAD) was determined as SPAD units using "Minolta (chlorophyll meter) SPAD-502 (Yadava, 1986).

All obtained data were statistically analyzed according the design using CoStat program (Version 6.4, Co Hort, USA, 1998–2008). Revised least significant difference test (LSD) was applied at 0.05 probability level to compare means of different treatments as illustrated by Williams and Abdi (2010).

RESULTS AND DISCUSSION

Results presented in Table 3, generally, revealed that foliar application of the three synthetic cytokinin: BAP, CPPU and kinetin, significantly, enhanced plant height, number of leaves and leaf area of okra plants compared to the control treatment. However, number of branches in the second seasons was not significant, affected. The highest mean values for the previously mentioned traits were found to be associated with the addition of BAP at 50 ppm , in general even if the difference is not so high to be significant, compared to other treatments. Spraying BAP at 50 ppm recorded the highest averages of plant height (162.03 and 161.67 cm), number of leaves plant^{-1} (44.67 and 46.33), branch number plant^{-1} (5.33 and 5.67) and leaf area plant^{-1} (2688.73 and 3034.30 cm) in comparison to control plants in the first and second season, respectively. On the other side, control plants recorded the lowest averages of plant height (137.13 and 142.20 cm), number of leaves (32 and 33.67), branches number plant^{-1} (4 and 5) and leaf area plant^{-1} (1854.73 and 1952.7cm^2) in the first and second season respectively. Similar results were obtained by Taheri and Haghighi (2018), who found that foliar application of BAP increased plant height, shoot and root dry weight in bell pepper. Kinetin increased stem thickness and the fresh weight by increasing stem diameter, but reduced shoot length in morning glory (Kaul and Farooq, 1994), in okra (Chaudhry and Khan, 2000) in cucumber, tomato and pepper (Papadopoulos *et al* ., 2006). Khandaker *et al*., (2018) found that kinetin increased the plant height, number of leaves, branch number and leaf area of stevia plants. The increment in the plant height, number of leaves, branch number and leaf area of stevia plants may be the results of stimulating activity of apical and lateral meristems. Application of CPPU increased plant height, leaves number, branch number, plant fresh and dry weights potato plant (El-Shraiy and Hegazi, 2010).

Table 3: Plant height, number of leaves, number of branches and leaf area of okra plants as affected by foliar application of BAP, CPPU and kinetin during 2017 and 2018 seasons.

Treatments	Plant height (cm)		Number of leaves/plant		Number of branches		Leaf area (cm^2/plant)	
	2017	2018	2017	2018	2017	2018	2017	2018
Control	137.13	142.20	32.00	33.67	4.00	5.00	1854.73	1952.70
BAP25ppm	145.73	150.17	34.33	36.67	5.33	5.67	2499.30	2773.93
BAP50ppm	162.03	161.67	44.67	46.33	5.33	5.67	2688.73	3034.30
CPPU10ppm	152.03	150.00	41.00	43.00	5.00	5.33	2302.37	2563.00
CPPU20ppm	157.60	150.83	42.33	43.33	5.00	5.33	2433.17	2699.15
Kinetin 25 ppm	157.20	150.73	41.33	42.00	5.00	5.33	2348.43	2578.97
Kinetin 50 ppm	155.37	148.67	40.33	42.33	4.67	5.00	2169.40	2268.60
LSD at 0.05	14.49	17.16	4.532	4.30	0.85	0.43	509.15	615.44

Benzyl amino purine (BAP), 2-chloro-4-pyridyl-N-phenylurea (CPPU) and 6-furfuryl-aminopurin (Kinetin)

Table 4: Number of days to flowering, fruit length, fruit diameter, fruit diameter, number of fruits and weight of fruits per plant of okra plants as affected by foliar application of BAP, CPPU and kinetin during 2017 and 2018 seasons.

Treatments	Number of days to flowering		Fruit length (cm)		Fruit diameter (cm)		Fruit weight (gm)		Number of fruits/plant		Weight of fruits/plant (g/plant)	
	2017	2018	2017	2018	2017	2018	2017	2018	2017	2018	2017	2018
Control	41.67	41.00	4.00	4.33	1.45	1.43	5.67	5.67	27.00	24.67	153.09	139.88
BAP 25 ppm	50.67	48.33	4.33	4.30	1.59	1.68	6.17	6.41	32.00	34.33	197.44	220.06
BAP 50 ppm	52.00	54.67	5.47	5.37	1.77	1.83	7.13	7.07	39.00	40.67	278.07	287.54
CPPU 10 ppm	50.00	50.67	5.47	5.97	1.77	1.94	7.20	7.16	35.67	36.33	256.82	260.12
CPPU20 ppm	48.67	48.33	6.17	6.03	1.85	1.87	7.24	7.16	37.67	37.00	272.73	264.92
Kinetin 25 ppm	51.33	53.00	6.30	6.10	2.03	2.17	7.82	7.97	38.33	37.00	299.74	294.89
Kinetin 50 ppm	51.00	48.33	6.03	6.07	2.06	1.94	7.69	7.62	34.67	34.33	266.61	261.59
LSD at 0.05	2.58	3.43	0.906	0.877	0.167	0.205	0.719	0.65	2.59	3.43	28.85	30.96

Benzyl amino purine (BAP) , 2-chloro-4-pyridyl-N-phenylurea (CPPU) and 6-furfuryl-aminopurin (Kinetin)

Yield and yield components:

The Data listed in Table (4) clearly showed that the effect of application of cytokinin foliar spray of BAP at 25, BAP at 50, CPPU at 10, CPPU at 20, Kinetin at 25 and Kinetin at 50 ppm on the number of days to the first flower, fruit length, fruit diameter, average fruit weight, number of fruit plant^{-1} and total yield plant^{-1} were significant, in both seasons compared to control treatment. The statistical comparisons between the mean values of the different treatments illustrated that the highest mean values of the fruit length, fruit diameter, average fruit weight and total yield plant^{-1} were obtained from the treatment sprayed with kintein at 25 ppm, even if the difference is not reaching the significant level. However, the highest values of the number of days to flowering and the number of fruit plant^{-1} reached to the maximum with BAP at 50 ppm compared to other treatments. The result, also, revealed that every cytokinin treatment used was associated with a marked increase in total yield plant^{-1}.The increase in total yield plant^{-1} was appeared to be, in descending order, kinetine at 25 ppm (95.78 and 110.81 %), BAP at 50 ppm (81.63 and 105.56%), CPPU at 20 ppm (78.15and 89.39%), kinetine at 50 ppm (74.15 and 87.01%) CPPU CPPU at 10 ppm (67.75 and 85.95%) and BAP at 25 ppm (28.96 and 57.32%) compared to control treatment for the first and second seasons, respectively. These results are in agreement with those of (Vijay and Laxmi, 2001; Cho *et al*., 2002 and Patil *et al*., 2002) on soybean, (Janowska, 2014) on Zantedeschia spreng and (Zaki *et al*., 2014) on snap bean. Application of CPPU, significantly, increased fruit set and growth of muskmelon (Hayata, *et al*, 2001). Also, using CPPU on potato plants improved tuber number, tuber size, tuber weight plant^{-1} and yield ton fed^{-1} compared with control plants (El-Shraiy and Hegazi 2010). Kinetin increased number of flowers as well as fruit yield per plant of the lentil plant (Khalil *et al*. 2006) and early and total yield production of cucumber, tomato and pepper (Papadopoulos *et al*., 2006). 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.

Chemical composition:

The effects of foliar application of the three synthetic cytokinins (BAP, CPPU and kinetin) on okra leaves and fruits are presented in Tables (5 and 6). The obtained results, generally, reflected significant differences among cytokinin treatments on chemical compositions, compared to control treatment. Concerning the leaves chemical concentrations, the BAP at 50 ppm showed the highest mean values of nitrogen %, phosphour %, potassium %, protein% and total chlorophyll compared to control and BAP at 25 ppm treatments in both seasons. With regard to, fruit chemical compositions, the highest nitrogen %, protein%, phosphour %, potassium % and mucilage % were recorded by Kinetin at 25 ppm compared to other treatments in both seasons. Exogenous application of kinetin increased photosynthetic pigments contents in the leaves of wheat (Malibari, 1993) and corn (Kaya *et al*. 2010).Kinetin protect chlorophylls against the photo - oxidation process by enhancing the concentration of carotenoids (Petrenko and Biryukova, 1977). Application of cytokinin may increase the content of chlorophyll in leaf tissues, because it reduced chlorophyll degradation and delays the aging process (Fu *et al*. 2000; Costa *et al*., 2005; Xu *et al*., 2011). Chlorophyll formation and protein synthesis within tissues of shoot senescence were enhanced by application of synthetic cytokinins on that shoots (George *et al*., 2008). Moatshe *et al*., (2011) found that leaf mineral content of Morula tree sprayed with BA had significantly higher than control trees.

Table 5: Leaves nitrogen, protein, phosphorus, potassium and total chlorophyll contents in okra plants as affected by foliar application of BAP, CPPU and kinetin during 2017 and 2018 seasons.

	Concentrations in leaves									
	N (%)		P (%)		K (%)		Protein %		Total chlorophyll (unit SPAD)	
Treatments	2017	2018	2017	2018	2017	2018	2017	2018	2017	2018
Control	2.99	3.16	0.41	0.42	2.51	2.64	18.69	19.75	46.00	43.67
BAP 25 ppm	3.19	3.23	0.46	0.44	2.82	2.81	19.94	20.19	47.00	42.33
BAP 50 ppm	3.45	3.56	0.49	0.48	3.07	2.97	21.56	22.25	51.33	51.33
CPPU 10 ppm	3.34	3.29	0.42	0.44	2.82	2.80	20.88	20.56	47.67	46.67
CPPU 20 ppm	3.35	3.45	0.44	0.44	2.90	2.86	20.94	21.56	44.67	45.67
Kinetin 25 ppm	3.37	3.49	0.44	0.46	2.83	2.74	21.06	21.81	48.00	50.33
Kinetin 50 ppm	3.28	3.43	0.48	0.47	2.96	2.89	20.50	21.44	50.67	48.00
LSD at 0.05	0.162	0.131	0.048	0.026	0.153	0.127	1.012	0.818	3.32	4.03

Benzyl amino purine (BAP), 2-chloro-4-pyridyl-N-phenylurea (CPPU) and 6-furfuryl-aminopurin (Kinetin)

Table 6: Nitrogen, protein, phosphorus, potassium and mucilage concentrations in okra edible fruits as affected by foliar application of BAP, CPPU and kinetin during 2017 and 2018 seasons

	Concentrations in edible fruits								Mucilage (%)	
	N (%)		P (%)		K (%)		Protein (%)			
Treatments	2017	2018	2017	2018	2017	2018	2017	2018	2017	2018
Control	2.21	2.32	0.40	0.43	2.52	2.61	13.81	14.50	3.55	3.61
BAP 25ppm	2.59	2.68	0.47	0.50	2.80	2.78	16.19	16.75	4.18	4.33
BAP 50ppm	2.74	2.77	0.57	0.60	2.87	2.88	17.13	17.31	4.45	4.59
CPPU10ppm	2.75	2.81	0.60	0.67	2.84	2.86	17.19	17.56	4.70	4.68
CPPU20ppm	2.79	2.82	0.63	0.63	2.84	2.89	17.44	17.63	4.80	4.90
Kinetin 25 ppm	2.90	2.87	0.74	0.72	2.91	2.93	18.13	17.94	4.93	4.90
Kinetin 50ppm	2.87	2.84	0.66	0.70	2.81	2.92	17.94	17.75	4.71	4.55
LSD at 0.05	0.121	0.155	0.196	0.122	0.239	0.149	0.756	0.971	0.366	0.369

Benzyl amino purine (BAP), 2-chloro-4-pyridyl-N-phenylurea (CPPU) and 6-furfuryl-aminopurin (Kinetin)

Where,the fruit, mineral content such as nitrogen, potassium, magnesium and calcium was increased significantly with increasing BA concentrations. Application kinetin increased phosphorus, nitrogen and protein content in okra fruit, may be due to plant senescence (Singh and Paliwal, 2017). Protein content of muskmelon leaf was increased by increasing the application of CPPU (Sindhuja *et al*., 2017).

Applying the three synthetic cytokinins (BAP, CPPU and Kinetin) increased the growth and yield and mineral contents of both leaves and fruits of okra under high temperature conditions. The study provides an evidence about the possibility of using cytokinin type, especially, BAP at 50 ppm, CPPU at 10 ppm and kinetine at 25 ppm to increase the productivity with good quality of okra under high temperature stress conditions.

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الملخص العربى

تأثير ثلاث انواع من السيتوكينينات المخلقة ومستوياتهم علي النمو والمحصول والتركيب الكيماوي لنبات الباميا تحت ظروف الحرارة المرتفعة

ساري حسن مصطفي أحمد برنجي

قسم البساتين – كلية الزراعة – جامعة دمنهور – مصر

اجريت تجربتين حقليتين في مزرعة خاصة في منطقة ابوحمص – محافظة البحيرة – جمهورية مصر العربية خلال موسمي الصيف لعام ٢٠١٧ و٢٠١٨ لتقييم تأثير ثلاثة انواع من السيتوكينين المخلقة وهي بنزيل امينو بيورين بتركيز (٢٥ و٥٠ ملجرام/ لتر) وسي بي بي يو بتركيز (١٠ و٢٠ ملجرام / لتر) وكينتين بتركيز (٢٥ و٥٠ ملجرام/ لتر)علي نبات الباميا صنف اصابع الست. أوضحت النتائج ان الرش الورقي بالأنواع المختلفة من السيتوكينين ادت الي زيادة النمو الخضري متمثلا في ارتفاع النبات، عدد الاوراق، عدد الافرع، المساحة الورقية للنبات، كذلك ادت لزيادة التركيب الكيماوي للأوراق متمثلا في محتوي الاوراق من النتروجين والفوسفور والبوتاسيوم والكلورفيل الكلي مقارنة بمعاملة الكنترول وسجلت اعلي القيم تحت معاملة بنزيل امينو بيورين (٥٠ ملجرام/ لتر)، كما ادت المعاملة بالأنواع المختلفة من السيتوكينين لزيادة المحصول وتحسين خصائصه وزيادة محتوي الثمار منالنتروجين والفوسفور والبوتاسيوم والمادة اللزجة في الثمار مقارنة بمعاملة الكنترول وكانت اعلي القيم تحت معاملة الكينتين بتركيز (٢٥ملجرام/ لتر) تحت ظروف اجهاد الحرارة المرتفعة بينما ادي الرش بالأنواع المختلفة من السيتوكينين وبالتركيزات المختلفة لتأخير الازهار مقارنة بالكنترول.