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Effect of crezacin and humic acid on growth and physiological aspects of tomato plants (*Lycopersicon esculentum*)

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Abstract

This study aims to enhance growth characteristics and quality parameters for tomato plants (*Lycopersicon esculentum*) by applying different doses of Crezacin (synthetic) and Energene (natural) as growth regulators instead of Chemical fertilization. A specific experiment has been carried out to study the effect of two types of plant growth regulators Crezacin: Active substances (475 g/L): Orth-o-cresoxyacetic acid triethanolammonium salt with applied doses (100, 200 and 300 ppm) and Energene**:** active substances (700 g/kg) humic acid with applied doses (250, 500 and 700 ppm) on growth and physiological characteristics of tomato plants (Kendras F1). Both Crezacin and Energene were applied three times (Soaking tomato seeds for 24 hours before sowing, spraying on plants at 35 DAP, spraying on plants at 70 DAP). Application of humic acid "Ener-700" increased plant height (78.0 cm), number of leaves/ plant (57.8), fresh weight (250.2 g), dry weight (75.1 g) of arial parts, flowers number (48.1), fruits number (35.1) and fruit weight (64.0 g). Also using " Ener-700" improved tomato fruit's quality by enhancing level of dry matter (7.6 %), sugar contents (5.8 ^{o}Brix), Ascorbic acid (26.9 mg 100 g^{-1}), Maturity Index (12.61) and Taste Index (1.09), while level of nitrate rose by increasing humic acid and the maximum level (22.0 mg kg^{-1}) obtained from Ener-700. Applying Ener-500 gave the maximum titratable acidity (0.48%) and carotenoids contents (4.89 mg 100 g^{-1})

Key words: Tomato, Crezacin, Energene, plant growth regulators, ascorbic acid, nitrate.

Introduction

In agricultural production, farmers often apply some chemical substances which have similar structure and activity with endogenous plant hormone called exogenous (Plant Growth Regulators) as a cheap alternative to enhance plant growth and increase productivity (Cai *et al.*, 2015). Application of plant growth regulators has been found very effective in improving quantity and quality of many crops (Sandeep *et al.,* 2013).

Plant Growth Regulators (PGRs) have wide category of compounds that can enhance, inhibit or change plant morphological or physiological processes at very low concentrations. Thus, the use of PGRs has become an important element of the agrotechnical procedures for most cultivated crops (Kader, 2008). The most studied PGRs include abscisic acid, indoleacetic acid, cytokinin, gibberellic acid, ethylene, jasmonic acid and salicylic acid (Santner, 2009). Also, there are new, but chemically unrelated compounds with similar hormone. Most of these substances, whether chemical or natural, have not been studied for their effect on plants, especially vegetables and fruits that are consumed as part of daily diet. such as tomatoes, which Tomato is an important condiment in most diets and a very cheap source of vitamins and nutrients that are very good for the human body and protect the body against diseases, because its vitamins and beta-carotene content work as antioxidants to neutralize harmful free radicals in the human blood (Debjit, 2012). It occupies the fourth ranks in terms of worldwide production with approximately 5 million ha as a harvested area produced 170.75 million tones according to FAO statistics' database (FAO, 2014).

The beneficial response of applying growth regulators to enhance the quantity and quality of tomato plants have been reported by many scientists (Serrani *et al.,* 2007; Batlang, 2008; Gelrnesa *et al.,* 2009; Udden *et al.,* 2009; Vikram *et al.,* 2011; Arnao and Ruiz, 2014; Rahman *et al.,* 2015). So, the present investigation was undertaken to find out the effect of different types of synthetic and natural materials as a growth regulators on tomato hybrid "Kendras F1". Also, this work can help us to expand application of organic and natural materials in agricultural fields to produce healthy products for human being.

Materials and methods

Two different types of growth regulators were applied three times on tomato plants (Kendras F1): Crezacin (Cr-100, 200 and 300 ppm): Active substances (475 g/L): Orth-o-cresoxyacetic acid triethanolammonium salt, the chemical formation is: $C_{15}H_{25}NO_6$ (Handbook, 2015); Energene (Enr-250, 500 and 700 ppm): active substances 700g / kg humic acid (Handbook, 2015). All applied doses expressed in terms of active substances. Each treatment was applied three times (Soaking tomato seeds for 24 hours before sowing, spraying on plants at 35 DAP, spraying on plants at 70 DAP) and four replicates were maintained. Soaked seeds were planted on trays and then after 30 DAP, tomatoes seedling were transplanted on pots (20-inch diameter). Average day and night temperatures in greenhouses were 27 ^{o}C and 18 ^{o}C which conformed with normal temperature ranges established for greenhouse (Maynard and Chmuth, 2007).

Collecting data: Ten plants from each replicate (4 replicates) were selected to measure the following parameters: Plant height (cm), number of branches per plant, number of leaves per plant, leaf area index (LAI), fruit set (%), days to flowering and weight of fruit (g). LAI was measured by (non-destructive method). Biochemical properties were evaluated at the fruit ripening stage. The dry matter was calculated by weighting fruits and after being oven dried at 105 °C. The titratable acidity (TTA) of tomato was expressed as percentage citric acid by the methods described by (Seyoum *et al.,* 2009). Ascorbic acid (AA) was determined by the 2, 6- dichlorophenol indophenol method (AOAC, 1990). Carotenoids and nitrate were determined based on standards of the association of analytical communities (Horwitz, 2006). Sugar-acid ratio (Maturity index) was calculated by dividing total soluble solid by the titratable acidity of the given sample under analysis as described by (Mohammed *et al.,* 1999). The taste index was calculated using the equation given by Navez *et al.* (1999) and Hernandez *et al. (*2007).

The results were analyzed using one-way analysis of variance (ANOVA) followed by Tukey's HSD test with $\alpha = 0.05$ using MINITAB (v. 19.0) program.

Results and discussions

Plant height (cm): Statistically significant variation was observed on tomato plants height due to application of plant growth regulators. Generally, the highest plants were obtained when Energene (humic acid) was applied at 30, 60 and 90 Days after planting (22.1, 56.5 and 78.0 cm respectively). At 60 DAP, application of Energene doses and Crez-300 had the same results and they significantly differed from control. Also, at 90 DAP, application of humic acid increased plant height compared to control. The shortest plant (71.8 cm) was recorded when Crez-200 was applied (Fig. 1).

Our results agree with Valdrighi *et al.* (1996), who reported that humic acid (Energene) increased absorption of nutrients by plants and improved the permeability of membranes of root cells to tomato cultures. Also, (Chen and Aviad, 1990; Varanini and Pinton, 1995) proved that humic acid has a positive effect on seed germination, seedling growth, root initiation, root growth, shoot development and the uptake nutrient elements.

Number of leaves per plant: Data in Fig. 2 showed the effect of different doses from Crezacin and Energene on number of leaves per tomato plant. At 30 DAP, data showed a non-significant differences between Crezacin and Energene treatments, which may be due to the age of plants at the time of measurement. At 60 DAP Crezacin and Energene doses were not statistically different, but they both produced more number of leaves per plant (from 36 – to 39.3) comparing to control (35.4), and the treatment Ener-700 gave the maximum number of leaves per plant (39.3). At 90 days from planting, impact of applying Crezacin and Energene was visible. Ener –700 and Crezacin – 300 increased leaves number per plant (57.8 and 56.9 respectively) compared to other doses and control.

Our results are consistent with those obtained from Gabal *et al.* (1999). The number of leaves in tomatoes increased with the use of plant growth regulators, which may be attributed to the fact that plant growth regulators improved cellular nutrition and division with a significant elongation of the stem. The results also corroborate with the results of (Ibrahim *et al.,* 2000; Sajjan *et al.,* 2002), who reported that the varieties affected the nature of the growth process.

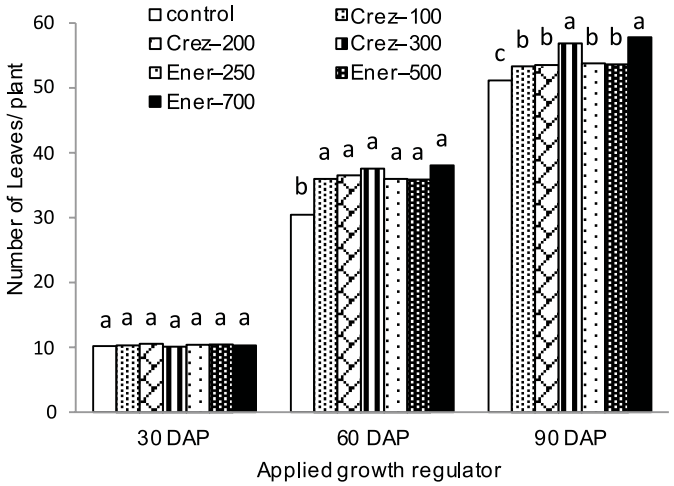

Fig. 2. Effect of applying Crezacin (Crez-100, Crez-200 and Crez-300 ppm),Energene (Ener-250, Ener-500 and Ener-700 ppm) as growth regulators, additional to control treatment on number of leaves/tomato plant in 30, 60 and 90 day after planting, Hybrid Kendras Fl.

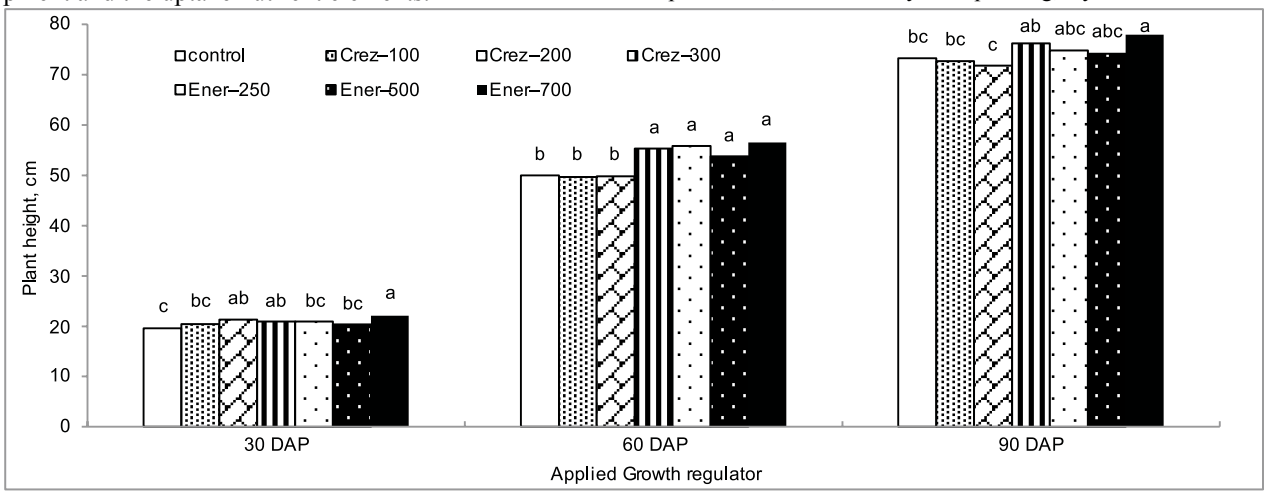

Fig. 1. Effect of applying Crezacin (Crez-100, Crez-200 and Crez-300 ppm),Energene (Ener-250, Ener-500 and Ener-700 ppm) as growth regulators, additional to Control treatment on tomato plant height (cm) in 30,60 and 90 day after planting, Hybrid Kendras Fl.

Fresh and dry arial parts: Development of arial parts of tomato plants (weight of leaves and branches) under the effect of different doses of Crezacin and Energene is shown in Figs. 3 and 4. Data showed that, applying both Cezacin and Energene enhanced the accumulation of nutritional elements and formation of dry matters (metabolism) in tomato plants compared to control. Applying Ener-700 (HA) had the maximum fresh and dry weights of Arial parts compared to other doses. Results, obtained from Ener-250 and Ener-500 were not statistically different in all stages. The maximum dose of Orth-o-cresoxyacetic acid triethanolammonium salt (Crez-300) gave the highest weights of arial parts compared to least doses of Crezacin (Crez-100 and Crez- 200). No significant effect was observed on weight of arial parts when Crez-100 was applied, and the results didn't differ from control's treatments. Our results agree with Valdrighi *et al.* (1996), who reported that HA enhanced mineral nutrients absorption by plants and increased the permeability of membranes of root cells to tomato plants. Also, shoots and root weights were increased by 22% as a result of using HA on tomato plants (Yildirim, 2007).

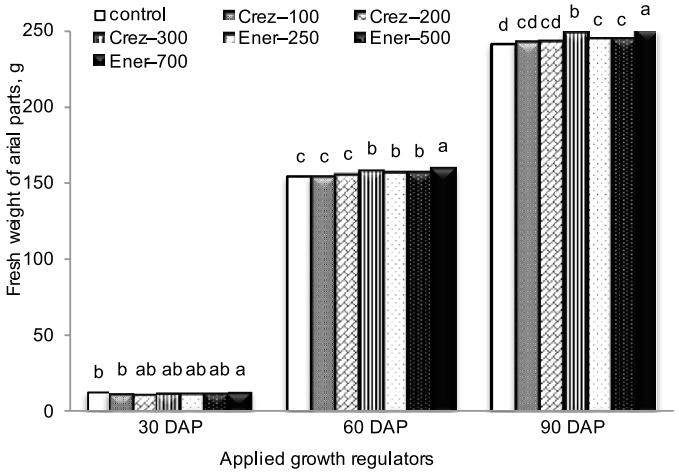

Fig. 3. Effect of applying Crezacin (Crez-100, Crez-200 and Crez-300 ppm),Energene (Ener-250, Ener-500 and Ener-700 ppm) as growth regulators, additional to Control treatment on Fresh weight (g) of Arial parts (leaves and branches) of tomato plant in 30, 60 and 90 day after

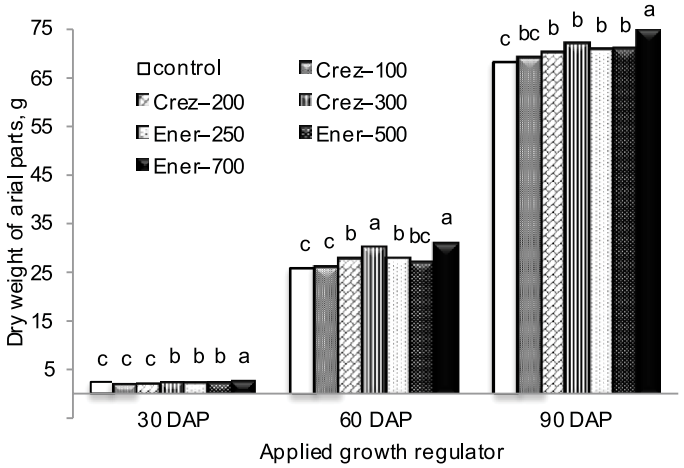

Fig. 4. Effect of applying Crezacin (Crez-100, Crez-200 and Crez-300 ppm),Energene (Ener-250, Ener-500 and Ener-700 ppm) as growth regulators, additional to Control treatment on Dry weight (g) of Arial parts (leaves and branches) of tomato plant in 30, 60 and 90 day after planting, Hybrid Kendras Fl.

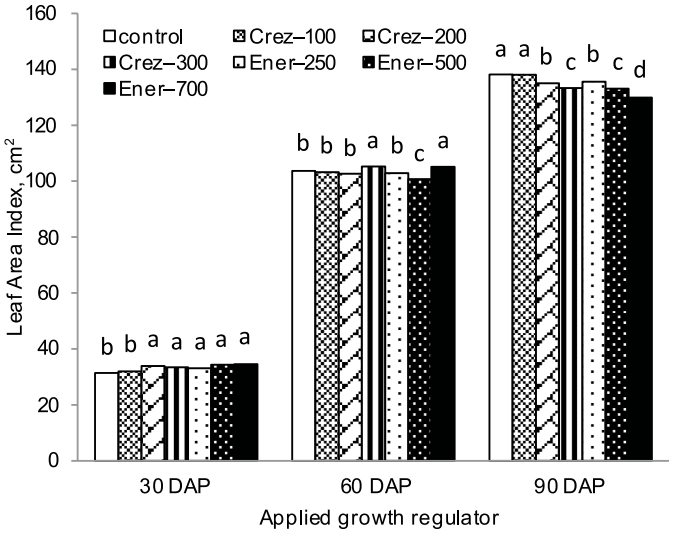

Fig. 5. Effect of applying Crezacin (Crez-100, Crez-200 and Crez-300 ppm), Energene (Ener-250, Ener-500 and Ener-700 ppm) as growth regulators, additional to Control treatment on Leaf Area Index (cm2) of tomato plant in 30, 60 and 90 day after planting, Hybrid Kendras Fl.

Leaf Area Index (LAI): Data in Fig. 5 explains changes in leaf area measurements as a response of the application of different growth regulators on tomato plants at various times. At 30 DAP, applying Crezacin and Energene increased LAI comparing to control, except (Crez – 100) which was non-significant. The maximum area (34.6 cm^2) was obtained from Ener-700, while the minimum (31.5 cm^2) was from control. At 60 DAP, we observed that vegetative growth of tomato plants duplicated two times compared to earlier stage (30 DAP), which may be due to the fact that in that stage plants try to build a good vegetative growth, as a preparation for transition to the next stage (flowering and fruiting). At 90 DAP, The rate of LAI began to decline as a sign of transition to fruiting stage. From the figure we can notice that control treatment obtained the highest LAI (138.1 cm^2) whereas, applying Crezacin and Energene decreased vegetative growth except Crez-100. The minimum LAI (129.9 cm^2) recorded from plants which were treated with Ener-700.Our results were in harmony with those observed previously (Ayas and Gluser, 2005; Celik *et al.,* 2008) where it was reported that application of HA (Energene) significantly increased minerals nutrients uptake, stimulated chlorophyll content and accumulation of minerals in leaves that lead to healthy production of leaves.

Productive parameters: Data in Table 1 show statistically non-significant variation due to the application of plant growth regulators on number of flowers in tomato plant. Number of fruits per plant was statistically significant due to application of plant growth regulators (Table 1). The maximum number of fruits (35.7) was recorded from (Ener-700), while the minimum number of fruits per plant (32.0) was recorded from (Crez-100) which was close to control treatments. It might be due to enhanced fruit setting in tomato plants by application of HA. In respect of fruit set per plant, it was found that the response of hybrid Kendras F1 was in agreement with previous studies (Olaniyi *et al.,* 2010), where it was reported that about 55% of the flowers were pollinated and bore fruits. They associated this with the genetic effects of the formation of tomato fruits. Reportedly, a high number of fruits per plant was found after application

Table 1. Effect of application different doses from Crezacin and Energene on flowering and crop tomato parameters.

Treatment	Number of flowers/ plant	Number of fruits/plant	Days to fruits maturity (50%)	Individual fruit weight, g
Control	48.6 a	32.1 c	109.9 a	62.0 b
Crezcin-100 ppm	48.4 a	32.0 c	110.3 a	62.3 b
Crezcin-200 ppm	48.2 a	34.5 b	106.0 b	63.9 ab
Crezcin-300 ppm	48.9 a	33.6 b	109.1 a	63.5 ab
Energene-250 ppm	47.9 a	33.9 b	109.4 a	63.6 ab
Energene-500 ppm	48.9 a	34.1 b	109.6 a	63.2 ab
Energene-700 ppm	48.1 a	35.7 a	105.5 b	64.8 a

Means within the same column that do not share a letter (s) are significantly different at $P \leq 0.05$.

of Energene which might be due to apparently reduced heat effect by growth regulators and thus increased the number of fruits set in tomatoes (Rahman, 2015). Some synthetic growth regulators have often been used to increase fruit set in vegetables including tomatoes (Serrani *et al.,* 2007; Batlang, 2008), where application of Crezacin (Crez- 200) and Energene (Ener- 700) has accelerated the maturity of tomato fruits compared to other doses. The shortest period of tomato fruits' maturity (105.5 days) was obtained from (Ener-700), while the longest (110.3 days) was recorded from Crez-100. Using Crezacin and humic acid as growth regulators on tomato plants increased individual weight of tomato plant compared to that of control.The maximum applied dose of HA (Ener-700) produced the heaviest fruit weight (64.8 g) whereas, the minimum weight (62.0 g) of tomato fruits obtained from non-treated plants (control) which did not differ statistically from the weight resulting from application of Crez- 100. These results corresponded with Enujeke and Emuh (2015) where they mentioned that a higher weight of fresh fruit may be associated with the possibility of possessing higher stomata conductivity, better genetic structure and a higher potential for the transport of photosynthetic materials within plants.

The outcomes of present investigations are in harmony with the previous findings (Singh and Singh, 1993; Singh and Lal, 2002). The other possible reasons for the enhancement in the yield by applications of growth regulators may be due to optimal utilization of nutrients and photosynthates for enhanced fruits formation as a result of reduced vegetative processes. These results may be attributed to enhanced plant growth and faster rate of plant development due to the action of applied growth regulators on elongation, division and differentiation of plant cells which in turn resulted into increase in fruit set and weight of fruits (Rodrigues *et al.,* 2001). Similar increase in yield was also reported in preceding studies (Kishan *et al.*, 2001; Rai *et al.*, 2002; Nibhavanti *et al.*, 2004; Singh and Sant, 2005; Bokade, 2006).

Chemical composition: Results of the chemical composition of tomato fruits as affected by different doses of studied growth regulators are shown in Table 2. Dry matter ranged between 5.7 g/100 g in control samples and 7.6 g/100 g in the fruits treated by HA(Ener-700). Applying HA enhanced dry matter contents (7.1-7.6 g/100 g) compared to Crezacin (6.4-6.6 g/100 g). The same trend observed on T S S contents, where applying Ener- 700 on tomato registered the maximum level of sugar contents (5.8 °Brix) and the minimum (4.9 °Brix) obtained from control. Our results agree with those obtained from Cramer *et al.* (2001) who observed that the values commonly obtained for soluble solids of different cultivars of tomato fruit ranged from (4 to 6 ºBrix). Also agreed with (Hernandez *et al.*, 2008) who noticed that the main soluble sugars in tomato fruit are glucose and fructose which make up 47% of the fruit dry matter. Ener-500 gave the highest values of Titratable Acidity (0.48 %) and Carotenoids (4.89 mg 100 g^{-1}) whereas, the minimum results recorded from control (0.41% and 3.09 mg 100 g^{-1} respectively). Generally, using HA increased contents of tomato fruit from vitamin C (Ascorbic acid) and nitrate compared to Crezacin, and the highest level of Ascorbic acid (26.9 mg 100 g^{-1}) and nitrate contents (22.0 mg kg^{-1}) observed when Ener – 700 was applied, while the samples treated by Crezacin gave the minimum results for vitamin C (18.0-18.2 mg 100 g^{-1}) and nitrate (18.1-18.9 mg kg^{-1}).

These results are consistent with (Yildirim, 2007), who reported a significant improvement in Ascorbic acid content as a result of applying Energene on tomato plants. Applying growth regulators also enhanced carotenoids contents compared to control except (crez-100). Our results are consistent with (Dumas *et al.*, 2003) who found that all the differences observed in the antioxidant content of tomato cultivars such as carotene were associated with the genotype, as well as with growth regulators. As in the case of sugars, acids are crucial for the taste of the tomato fruits, so maturity index is the best predictor of tomato fruits quality than Brix level or acidity (Nielsen, 2003; Hernandez *et al.*, 2007). Our results showed significant differences among studied parameters, the maximum result of maturity index (12.61) was recorded from Ener-700, which was statistically similar to that obtained from Crezacin (12.38-12.56). Values of taste index in all variants (Table 2) were higher than 0.85, which indicated that the fruits were

Table 2. Effect of application different doses from Crezacin and Energene on tomato quality characteristics.

Treatment	Dry matter, (g 100 g^{-1})	TSS (°Brix)	Titratable Acidity (%)	Caroten-oids (mg 100 g^{-1})	Ascorbic acid (mg 100 g^{-1})	Nitrate contents (mg kg^{-1})	Maturity index	Taste index
Control	5.7 d	4.9 c	0.41 b	3.09 c	19.2 c	19.6 c	11.65 b	1.008 c
Crezacin-100 ppm	6.4 c	5.2 b	0.42 b	3.12 c	18.0 d	18.9 cd	12.38 a	1.039 b
Crezacin-200 ppm	6.6 c	5.7 a	0.46 ab	4.14 b	18.2 d	18.8 cd	12.39 a	1.080 a
Crezacin-300 ppm	6.6 c	5.4 ab	0.43 b	4.10 b	18.0 d	18.1 d	12.56 a	1.058 ab
Energene-250 ppm	7.1 ab	5.2 b	0.46 ab	4.10 b	22.3 b	20.9 b	11.30 b	1.025 bc
Energene-500 ppm	7.3 a	5.6 a	0.48 a	4.89 a	25.8 a	21.2 b	11.43 b	1.061 ab
Energene-700 ppm	7.6 a	5.8 a	0.47 a	4.30 b	26.9 a	22.0 a	12.61 a	1.090 a

Means within the same column that do not share a letter(s) are significantly different at $P \leq 0.05$.

delicious; if value of the taste index is below 0.7, tomato fruits is not considered tasty (Navez *et al.*, 1999).

Our study was concentrated to the effect of various plant growth regulators, synthetic (Crezacin) and natural (Energene) with different doses on tomato plant (hybrid Kendras F1).Results concluded that, plant growth regulators had significant influence on growth and quality parameters of tomato plants especially the treatment with Energene (700 ppm). It may be useful in reducing impact of abiotic stresses such as heat, salt, drought and light intensity by compensating for shortage in growth, or reducing the direct effect of stress potency or improvement in the micro-environmental conditions of the plant, and may result in plant access to optimum conditions and accelerated rate of transportation of nutrients from soil to plant.

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