

## Influence of JA and CPPU on Growth, Yield and $\alpha$ - Amylase Activity in Potato Plant (*Solanum tuberosum* L.)

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**Abstract:** Field experiment was carried out in the Experimental Farm, Faculty of Agriculture, Ain Shams University, Shoubra El-Kheima, Kalubia governorate, Egypt, to investigate the influences of bioregulators; Jasmonic acid (JA) at 5 and 10 ppm and N-(2-chloro-4-pyridyl)-N'-phenylurea (CPPU) at 10 and 20 ppm and their combinations on potato (*Solanum tuberosum* L.) plant growth, yield and  $\alpha$ - amylase activity during the two successive seasons of 2007 & 2008. Bioregulators were applied as foliar spray at 55 and 75 days after planting. Application of CPPU(10 and 20 ppm) and the combination of CPPU at 20 ppm with JA at 5 and 10 ppm showed an improvement in plant growth as indicated by plant height, leaves number, branches number, plant fresh and dry weights. Application of CPPU and its combinations with JA significantly improved tubers number, tuber size, tubers weight/ plant and yield ton/fed comparing with control plants. CPPU at 20 ppm + JA at 5 ppm and CPPU at 20 ppm gave the best results of tubers number, size and the yield. These positive effects of CPPU at 20 ppm and its combination with JA at 5 ppm were correlated with significant increase in total chlorophylls in leaves and phenols, total soluble sugars and total soluble carbohydrates in tuber tissues. An enhancement in total soluble carbohydrates and  $\alpha$  amylase activity at harvest and 2 weeks later followed by reduction in these measurements at 4 weeks after harvest were observed by CPPU at 10 and 20 ppm. Excluding JA, all combination treatments showed positive increase in total carbohydrates and  $\alpha$  amylase activity at harvest, 2 and 4 weeks after harvest. While plants treated with JA showed significant increase in phenols at harvest and  $\alpha$ - amylase activity at 4 weeks after harvest.

**Key words:** Potato ( *Solanum tuberosum* L.) , bioregulators, Jasmonic acid (JA), N-(2-chloro-4-pyridyl)-N'-phenylurea (CPPU), yield,  $\alpha$ - amylase activity.

### INTRODUCTION

Potatoes are the most important kinds of vegetables which are the staple food in many parts of the world; also potato crop is one of the great economic importances as it is ranked second in export. Wherefore, potatoes cultivated widely in the world. Tuberization is a developmental process unique to some *Solanum* species , which under favorable conditions differentiate specialized underground propagation organs or tubers (Rodríguez-Falcón *et al* 2006). Tuber induction is associated with several morphological changes in the plant (Ewing and Struik 1992). Coinciding with the rapid growth phase of the tubers. Such alterations in morphology, together with transmission of the inducing signal across a graft, led to the belief that the stimulus was of hormonal nature. Several hormones, i.e., gibberellin (Railton and Wareing, 1973; Xu, *et al* 1998), cytokinin (Hussey and Stacey, 1984, Palmer and Smith, 1970), jasmonic and tuberonic acids (Koda, *et al* 1988, Van de Berg and Ewing 1991) or abscisic acid (Menzel, 1980, Xu, *et al* 1998), reportedly play a role in tuberization, and changes in the endogenous levels of these hormones could be correlated with the tuberization onset.

However, in the present study attempts were done to demonstrate a causal link between JA, CPPU and their combinations for tuberization and yield. JA derivatives in leaves turn to hydroxylated forms and these forms are then glucosylated and transported out of the leaves to initiate tuber formation (Helder *et al*, 1993). Jasmonic acid (JA) was isolated from shoots of potato plants induced to tuberize (Koda, *et al*, 1988). JA stimulates *in vitro* tuber formation when added to the agar medium (van den Berg and Ewing 1991) and increased levels of JA were detected in the stolons at tuberization onset (Abdala, *et al* 2002 ). Kris *et al* (2002) mentioned that microtubers produced either from JA preconditioned stock plants or on the JA-containing tuberization media were more uniform and larger than from other treatments. JA conditioning of stock plants

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prior to taking explants for tuberization is being proposed as a treatment enhancing the quality of microtubers. However, (Ewing 1981) suggested that the ability to produce the tuber-inducing compound, in this case jasmonates, is one of the key factors determining maturity (early- or late- maturing habit) and early maturing cultivars rapidly form the compound.

Cytokinins (CK) have long been suggested to play a prominent role in tuberization. They are predominantly used for microtuber induction in vitro (Donnelly *et al* 2003, Hussey and Stacey 1984) and promote tuberization when directly applied to isolated stolons cultured in vitro (Estrada, *et al* 1986, Palmer and Smith 1970). Attempts to induce tuber formation by applying CK to the leaves, however, have produced ambiguous results as these treatments were unable to induce tuberization in andigena plants grown under non inducing conditions. Also, even endogenous CK levels increase in stolon tips during induction (Mauk and Langille 1978) and later stages of tuber growth (Obata-Sasamoto and Suzuki 1979). Rodríguez-Falcón *et al* 2006 suggest that CK may function to control tuber enlargement and growth. Increased cytokinin levels due to antisense suppression of the potato box gene, also result in increased starch accumulation and active cell division in specific regions of the meristem and the leaves (Rosin, *et al* 2003).

Cytokinins promote cell division and cell expansion in plant cell culture (Riou-Khamlichi, *et al* 1999). CPPU is a synthetic cytokinin which increased fruit set and growth in muskmelon Hayata, *et al*, 2001. Studies have shown that CPPU promotes the growth of grape berry (Nickell, 1986; Ogata *et al.*, 1988), pear (Banno *et al.*, 1986), kiwifruit (Iwahori *et al.*, 1988) and watermelon (Hayata *et al.*, 1995). The shift in cytokinin activity of the shoot is assumed to be in causal connection with an increased photosynthetic activity after the onset of tuber growth as 'sink' for assimilates (Sattelmacher and Marschner 2008).

This study aimed to improve potato plants growth and yield using JA and CPPU and their combination.

## MATERIALS AND METHODS

Field experiment was carried out at the Experimental Farm, Faculty of Agriculture, Ain Shams University, Shoubra El-Kheima, Kalubia governorate, Egypt, during the two successive seasons of 2007 and 2008 to study the effect of foliar application of jasmonic acid (JA) at 5 and 10 ppm and N-(2-chloro-4-pyridyl)-N'-phenylurea (CPPU) at 10 and 20 ppm and their combinations on growth, tubers yield and quality of potato (*Solanum tuberosum* L.) cv. Spunta. Tubers of potato were cultivated at January 15<sup>th</sup> in both seasons. Distilled water was applied as control and tween 20 at 0.05 ml/L was used as wetting agent. Plants were foliar sprayed with CPPU and JA at 55 and 75 days after sowing respectively.

The experiment was arranged in complete block design with 3 replicates, each replicate was divided into 9 plots (1.5 × 3m) with 70 cm apart rows and plants spacing 25 cm.

Two samples were taken from the fifth leaf at 60 and 80 days after planting for determination of total chlorophylls. The third sample was taken at harvest (110 days after planting) where growth measurements, plant height, leaves number, branches number, plant fresh and dry weights were recorded. As well as, yield parameters, number of tubers/plant, tubers weight, tuber size, tuber fresh and dry weights and tubers yield ton / feddan were recorded. Chemical analysis was conducted in tuber tissues at harvest time (110 days after planting) for determination of total soluble carbohydrate (TSC), total soluble sugars (TSS), reduced sugars (RS), non-reduced sugars (NRS) and phenols. Three samples were taken at harvest and 2 & 4 weeks later for determination of total soluble carbohydrates (TSC) and  $\alpha$ -amylase activity.

### **Biochemical Analyses:**

#### **Chlorophyll Determination:**

Total chlorophyll (Chl) concentration was determined in the fifth leaf. Leaf samples of 0.1 g were ground and extracted with 5 mL of 80% (v/v) acetone in the dark according to the methods described by Holder (1965). The mixture was filtered then the absorbance values at 645 and 663 nm were measured. Using Jenway 6105 UV/VIS Spectrophotometer. Concentrations of Chl a and Chl b were estimated by the equations of Arnon (1949) as follows:

$$\text{Chl a+b} = 20.2 D_{645} + 8.02 D_{663}$$

#### **Determination of Phenols:**

One gram fresh tissues of potato tuber was randomly taken and extracted with 80 % cold methanol (v/v) for three times at 0°C. The combined extracts were collected and filtered (Wt. No. 1). Then, the volume of sample was made up to 25 ml with cold methanol. Phenol determination was carried out according to Daniel and George (1972).

**Determination of Total Soluble Carbohydrate:**

Total carbohydrates was extracted according to A.O.A.C., (1990) One gram fresh tissues of potato tuber was hydrolyzed with 1 N HCl by refluxing 6 hr in boiling water bath. Resulting solution was filtered, neutralized and the total volume was made up to 100 ml with distilled water. Resulting total reducing sugars was determined colourimetrically using 1 ml of sample by alkaline potassium reagent. As for determining carbohydrates and sugars, the resultant color was measured using a spectrophotometer (Shimadzu UV-160A) at 420nm.

**Determination of Total Soluble Sugars:**

For extraction, one gram fresh tissues of potato tuber was taken and grounded in a mortar with ethanol 80% for 3 times. The extracts were combined and evaporated till dryness. The dried film was dissolved in 50 ml of 10 % aqueous isopropanol. Determination of total soluble sugars, reduced and non-reduced sugars were carried out according to the method of Shales and Schales (1945).

**Determination of  $\alpha$ -amylase Activity:**

For extraction, 20 grams fresh tissues of potato tuber was taken in extraction buffer consisting of 20 mM Sodium phosphate (pH 6.0). This extract was centrifuged at 13,000 x g for 10 minutes and the supernatant removed and kept for amylase activity assays. The activity of  $\alpha$ -amylase was estimated according to the modified method of Plummer (1988) The method was based on the interaction of 3,5-dinitro-salicylic acid with reducing sugars and the amount of the resulting substance was measured colorimetrically at 530 nm.

**Statistical Analysis:**

The statistical analysis of data were carried out using the SPSS base 16.0 (SPSS Inc., Chicago, IL) packages. General linear model- Multivariate was used to test for significant main effects due to JA and CPPU treatments and their interaction.

**RESULTS AND DISCUSION**

**Growth Parameters:**

As shown in Table (1) foliar application of CPPU significantly improved most of plant growth parameters of potato plants. The highest values of plant height, leaf numbers, branches number, fresh and dry weights were obtained by CPPU at 20 ppm which recorded 123.7 cm of plant height, 124 leaves/ plant, 7.3 branches number, 107 gm fresh weight, 21.39 gm dry weight comparing with control plants which recorded 88.7 cm of plant height, 76.7 leaves/ plant, 3.7 branches number, 65 gm freash weight and 15.54 gm dry weight respectively, followed by CPPU at 20 ppm + JA at 5 ppm in both seasons. CPPU at 10 ppm and CPPU at 20 ppm +JA at 10 ppm gave significant increase in growth parameters as compared to control plants in both seasons. However, Minimum numbers of growth parameters were recorded by JA at 5 and 10 ppm and their combination with CPPU at 10 ppm.

**Table 1:** Effect of JA, CPPU and their combinations on growth parameters of potato plants during the seasons of 2007&2008.

Treatments	1 <sup>st</sup> season					2 <sup>nd</sup> season				
	Plant height (cm)	leaves no.	branches no.	Fresh Wt plant(g)	Dry Wt plant (g)	Plant height (cm)	leaves no.	branches no.	Fresh Wt plant(g)	Dry Wt plant (g)
Control	88.7	76.7	3.7	65.0	15.54	91.3	78.9	3.9	61.8	14.0
JA 5 ppm	64.0	61.7	3.0	37.3	11.67	65.6	66.7	2.7	39.4	8.9
JA 10 ppm	52.7	46.7	1.7	24.3	8.50	56.3	48.2	1.9	29.7	7.9
CPPU 10 ppm	111.7	93.3	6.3	97.3	19.44	116.3	94.8	6.8	99.5	19.7
CPPU 20 ppm	123.7	124.0	7.3	107.0	21.39	128.3	125.0	7.4	112.4	22.3
CPPU 10 ppm × JA 5 ppm	85.3	66.7	3.7	89.3	14.66	88.9	69.6	4.2	93.6	18.3
CPPU 10 ppm × JA10 ppm	80.0	53.3	4.7	74.3	12.04	83.6	57.3	5.4	85.8	16.9
CPPU 20 ppm × JA 5 ppm	118.7	119.0	5.0	109.7	22.57	122.3	120.5	6.1	117.1	23.1
CPPU 20 ppm × JA 10 ppm	107.3	71.7	4.0	94.7	17.61	109.9	73.2	5.2	98.9	19.4
LSD at 0.05%	4.65	2.24	1.87	2.65	1.24	5.03	2.67	1.94	2.81	1.03

The stimulation effect of CPPU at 20 ppm can be explained by the major physiological features of cytokinins, which is promoting cell division and elongation (Te-chato and Lim, 2000); emhancing shoot fresh and dry weights and bud sprouting (Kapchina-Toteva and Stoyanova, 2003; Kapchina-Toteva *et al* , 2005). On the other hand, Maja and Gogala ,1990 found that JA retarded callus and root formation. while, higher concentrations of JA caused growth inhibition of shoot elongation in potato plants Ulloa , *et al* (2002).

**Tuber Yield and Yield Parameters:**

Excluding individual application of JA treatment, bioregulator treatments significantly enhanced tuber size, tubers number/ plant, tubers fresh and dry weights, the yield ton / feddan in both seasons (Table 2 & 3). Application of JA at 5 and 10 ppm had negative impact for these parameters with the exception of tubers number per plant which showed significant effect. The highest values of tubers fresh and dry weights, and the lowest values of % moisture contents obtained by CPPU at 20 ppm and CPPU at 20 ppm + JA at 5 ppm treatments led to an increase in yield values in both seasons. However, the rest of bioregulator combinations gave a significant increase in yield. An increment in yield, 7.95 and 8.07 ton/fed; 6.31 and 6.92 ton/fed above control plants was gained by CPPU 20 ppm + JA 5 ppm and CPPU 20 ppm in both seasons 2007 & 2008 respectively. Similar findings were obtained by Koda and Okazawa, 1983, who mentioned that fresh weight of tubers formed was increased by addition of ZR.

**Table 2:** Effect of JA, CPPU and their combinations on the yield and its parameters of potato tubers at harvest during the season of 2007.

Treatments	1 <sup>st</sup> season					
	No. of tuber / plant	Tuber size (cm <sup>3</sup> )	Yield / toon	Fresh Wt tuber (g)	Dry Wt tuber (g)	% moisture content
Control	3.7	173.7	7.71	87.7	12.3	86.0
JA 5 ppm	6.7	94.7	7.53	47.0	9.8	79.3
JA 10 ppm	5.7	80.0	4.55	33.5	12.7	61.9
CPPU 10 ppm	4.3	213.0	10.98	105.6	21.6	79.6
CPPU 20 ppm	5.3	227.3	14.02	109.5	26.0	76.3
CPPU 10 ppm × JA 5 ppm	4.3	145.3	9.49	91.2	13.5	85.2
CPPU 10 ppm × JA10 ppm	4.7	187.0	9.86	87.9	18.0	79.6
CPPU 20 ppm × JA 5 ppm	6.0	266.7	15.66	108.8	25.9	76.2
CPPU 20 ppm × JA 10 ppm	4.3	245.3	9.70	93.2	18.6	80.1
LSD at 0.05%	2.23	3.07	1.75	4.14	2.46	

**Table 3:** Effect of JA, CPPU and their combinations on the yield and its parameters of potato tubers at harvest during the season of 2008.

Treatments	2 <sup>nd</sup> season					
	No. of tuber / plant	Tuber size (cm <sup>3</sup> )	Yield / toon	Fresh Wt tuber (g)	Dry Wt tuber (g)	% moisture content
Control	3.5	165.8	7.19	85.6	11.7	86.3
JA 5 ppm	5.7	91.6	5.84	42.7	9.4	78.0
JA 10 ppm	4.8	79.8	4.10	35.6	11.6	67.4
CPPU 10 ppm	4.9	199.9	11.91	101.3	19.8	80.5
CPPU 20 ppm	5.4	210.6	14.11	108.9	23.4	78.5
CPPU 10 ppm × JA 5 ppm	5.2	139.8	11.19	89.7	12.6	86.0
CPPU 10 ppm × JA10 ppm	5.4	181.3	10.42	80.4	15.7	80.5
CPPU 20 ppm × JA 5 ppm	5.9	229.3	15.26	107.8	23.1	78.6
CPPU 20 ppm × JA 10 ppm	4.6	218.4	10.10	91.5	15.9	82.6
LSD at 0.05%	2.12	2.98	1.65	3.87	2.07	

The stimulatory effect of CPPU can be explained by cytokinin activity was low in the stolon tips prior to tuberization, but increased considerably in both stolon tips and young tubers during tuberization (Sattelmacher and Marschner 2006). Despite the close correlation between tuberization and cytokinin activity, it is assumed that cytokinins are not directly responsible for the onset of tuberization, although they play an important role in tuber growth. On the other hand, JA caused a four and six fold increase in cell area in both zones in hooked apex stolons and initial swelling stolons, tuber formation is concluded to occur as a consequence of increased cell expansion, a reduction in the length of leaf primordia, enlargement of meristems, and early vascular tissue differentiation (Cenzano. *et al*, 2003). This explains the results of combination CPPU at 20 ppm + JA at 5 ppm which increased yield. On the other hand, JA treatments at 5 and 10 ppm didn't reach the values of control plants. That result may be due to the reduction in vegetative growth caused by this treatment. Fan *et al*. 1997, Koda 1997 mentioned that jasmonic acid affects plant development and physiology which could in turn affect plant yield. Also, exogenous jasmonic acid decreases the photosynthetic rate of plants (Metodieva, *et al* 1996), causes a reduction in bud formation (Barendse *et al*. 1985), and causes an increase in ethylene, a hormone involved in fruit ripening (Saniewski, *et al* 1987). However, when the low concentration of JA at 5 ppm was applied in combination with the high concentration of CPPU at 20 ppm increase in tubers yield over control and individual application of cppu at 20 ppm was observed. Evidence for jasmonic acid playing a role in tuberization is provided by experiments on transgenic potato plants expressing a tuber-specific lipoxygenase, which acts in jasmonic acid biosynthesis, in an antisense orientation. The transgenic plants exhibited decreases in tuber yield, possibly via an effect on tuber enlargement (Kolomiets *et al.*, 2001).

**Chlorophyll Concentrations:**

As shown in Table (4), the highest values of chlorophyll concentration were observed with CPPU at 20 ppm and CPPU at 20 ppm + JA (5 and 10 ppm) at 60 and 80 days after planting in the two seasons. Meanwhile, CPPU at 10 ppm, CPPU at 10 ppm + JA at 5 ppm and CPPU at 10 ppm + JA at 10 ppm didn't give any significant increase comparing with control plants. On the other hand, JA treatments at 5 and 10 ppm significantly decreased chlorophyll levels at 60 and 80 days after planting in both seasons. The stimulation effect of CPPU on chlorophyll concentrations was confirmed by Fletcher and Arnold (2008) on cucumber plants, Cherniadev and Manakova (2003) on wheat plants; Antognozzi, *et al* 1996 on *Actinidia deliciosa* and Kishorekumar, *et al* 2007 on *Solenostemon rotundifolius*. Regarding JA, Kumari and Sudhakar, 2004 found reduction of chlorophyll concentration and chlorophyll stability index in groundnut seedling. Ueda *et al.*, 1981; Watanabe, *et al* 2000 reported that jasmonyl acid, and its related compounds, promote the degradation of chlorophylls or carotenoids, followed by senescence, in detached green or etiolated plant tissues, respectively.

**Table 4:** Effect of JA, CPPU and their combinations on total chlorophyll contents (mg/g) in potato leaves at 60 and 80 days after planting during the seasons of 2007 & 2008.

Treatments	Total chlorophyll (mg/g)			
	1 <sup>st</sup> season		2 <sup>nd</sup> season	
	60 days after planting	80 days after planting	60 days after planting	80 days after planting
Control	7.4	10.7	7.9	10.5
JA 5 ppm	5.3	8.1	6.8	8.4
JA 10 ppm	4.8	6.9	5.5	6.4
CPPU 10 ppm	8.9	10.9	9.4	11.4
CPPU 20 ppm	10.7	12.9	11.2	13.3
CPPU 10 ppm × JA 5 ppm	9.5	10.8	10.2	11.4
CPPU 10 ppm × JA 10 ppm	9.4	11.3	10.4	11.9
CPPU 20 ppm × JA 5 ppm	10.9	13.6	11.8	13.9
CPPU 20 ppm × JA 10 ppm	10.9	11.1	11.9	12.4
LSD at 0.05%	2.65	2.78	2.45	2.63

**Phenol and Total Soluble Sugars Concentrations:**

It was clearly shown that CPPU at 10 and 20 ppm treatments significantly decreased phenol concentration and reduced sugar as compared with control plants (Table 5). Application of JA showed significant increase in phenols concentration and significant decrease in total soluble sugars in tuber tissues in the two seasons. However, an opposite trend was noticed with CPPU at 10 and 20 ppm treatments as compared with control plants. Significant increase in total soluble sugars and phenols was noticed by combination treatments and the superiority was due to CPPU at 20 ppm + JA at 10 ppm with phenols and CPPU at 20 ppm + JA at 5 ppm with total soluble sugars in both seasons. Parthasarathy *et al* 2002 and Tsui, *et al* 1980 found that the high concentration of cytokinins reduced the amount of total phenol and increased soluble and reducing sugars simultaneously.

**Table 5:** Effect of JA, CPPU and their combinations on phenols, reduced, non-reduced and total soluble sugars (mg/g) in tuber tissues of potato at harvest during the seasons of 2007 & 2008.

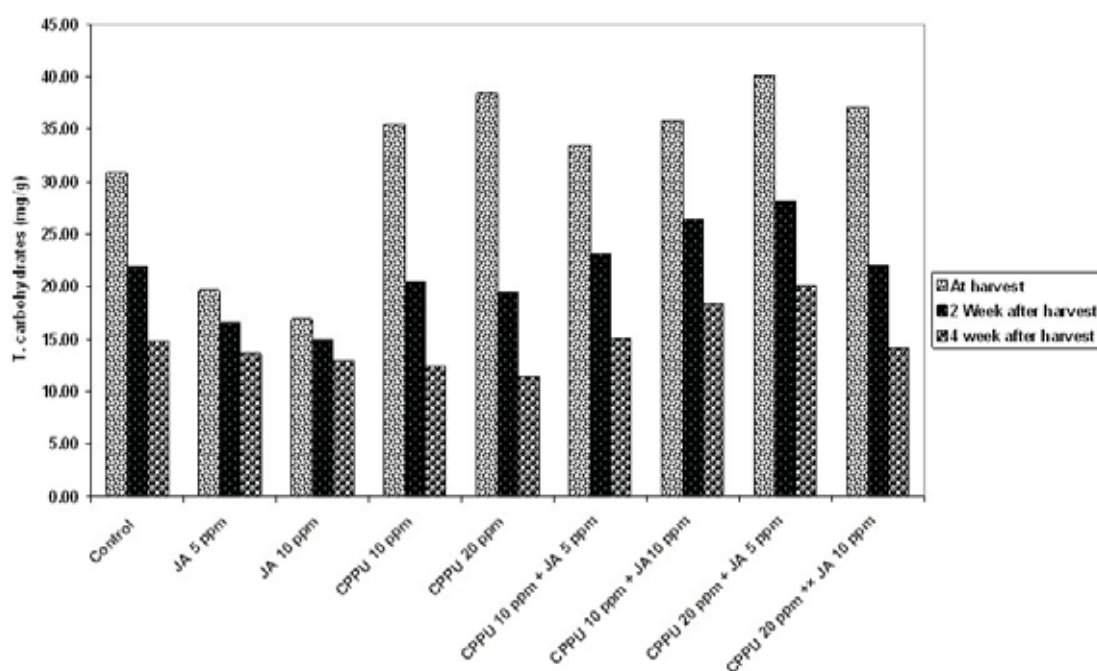
Treatments	Phenols (mg/g)		Total soluble sugars (mg/g)		Reduced sugar (mg/g)		Non-reduced sugar (mg/g)	
	1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season
Control	15.7	17.9	4.65	3.10	1.65	1.10	3.00	2.00
JA 5 ppm	25.8	25.3	3.14	2.09	1.03	0.69	2.11	1.41
JA 10 ppm	31.5	33.7	3.07	2.05	0.87	0.58	2.20	1.47
CPPU 10 ppm	16.5	18.4	7.25	5.83	2.14	1.43	5.11	4.40
CPPU 20 ppm	18.8	21.3	8.65	7.42	2.98	1.99	5.67	5.43
CPPU 10 ppm × JA 5 ppm	21.4	22.7	4.88	3.25	1.53	1.02	3.35	2.23
CPPU 10 ppm × JA 10 ppm	23.5	23.4	5.12	4.41	1.14	0.76	3.98	3.65
CPPU 20 ppm × JA 5 ppm	28.7	29.8	7.89	5.26	1.98	1.32	5.91	3.94
CPPU 20 ppm × JA 10 ppm	32.4	33.9	5.25	4.50	1.87	1.25	3.38	3.25
LSD at 0.05%	1.06	1.12	0.87	1.01	0.97	0.81	1.34	1.12

**Total Soluble Carbohydrates Concentrations:**

As shown in Table (6) and Figure (1 and 2), CPPU and all combination treatments significantly increased total soluble carbohydrate concentrations in tuber tissue at harvest in both seasons. The highest values were

**Table 6:** Effect of JA, CPPU and their combinations on total soluble carbohydrates (mg/g) in tuber tissues of potato at harvest, 2 and 4 weeks (wk) after harvest during the seasons of 2007 &2008.

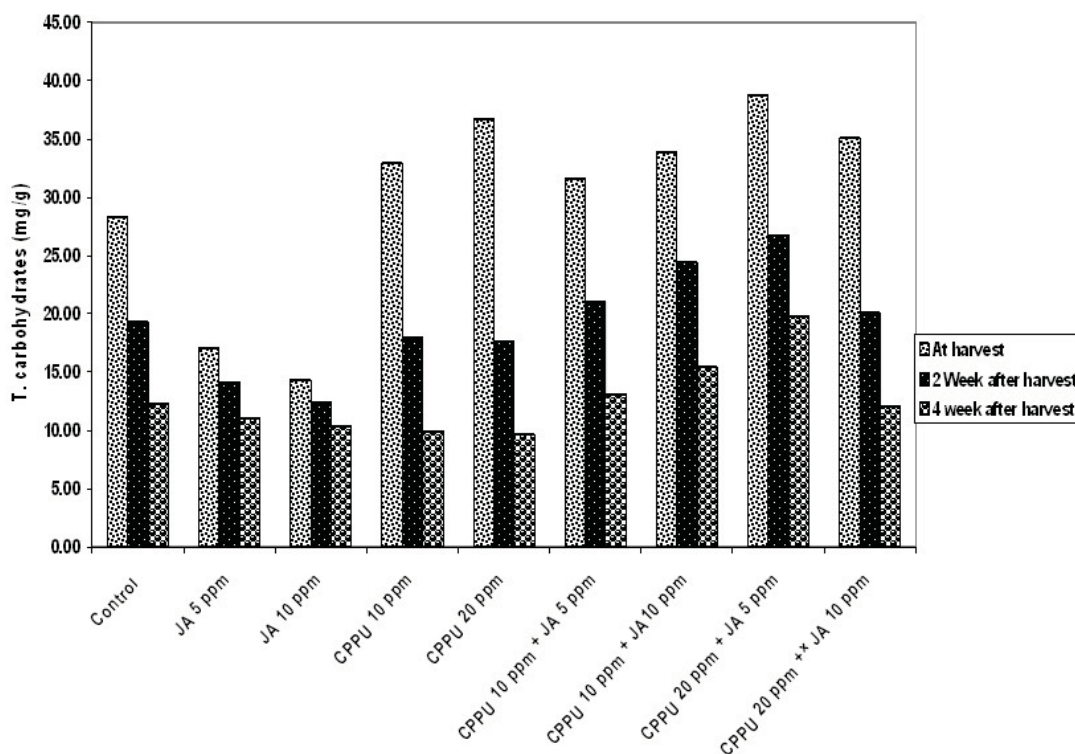
Treatments	Total soluble carbohydrates mg/g					
	1 <sup>st</sup> season			2 <sup>nd</sup> season		
	At harvest	2 Wk after harvest	4 Wk after harvest	At harvest	2 Wk after harvest	4 wk after harvest
Control	30.86	21.86	14.86	28.30	19.30	12.30
JA 5 ppm	19.62	16.62	13.62	17.06	14.06	11.06
JA 10 ppm	16.91	14.93	12.93	14.35	12.37	10.37
CPPU 10 ppm	35.51	20.51	12.51	32.95	17.95	9.95
CPPU 20 ppm	38.41	19.41	11.41	36.67	17.67	9.67
CPPU 10 ppm × JA 5 ppm	33.55	23.07	15.07	31.55	21.07	13.07
CPPU 10 ppm × JA10 ppm	35.88	26.40	18.40	33.88	24.40	15.40
CPPU 20 ppm × JA 5 ppm	40.18	28.18	20.18	38.73	26.73	19.73
CPPU 20 ppm × JA 10 ppm	37.07	22.07	14.07	35.07	20.07	12.07
LSD at 0.05%	3.02	1.45	2.34	2.76	2.01	1.65



**Fig. 1:** Effect of JA and CPPU and their combination on total soluble carbohydrates (mg/g) in tuber tissues of potato at harvest, 2 and 4 weeks after harvest during the season of 2007.

obtained by CPPU at 20 ppm + JA at 5 ppm and CPPU at 20 ppm which recorded 40.18 mg/g and 38.41 mg/g fresh weight comparing with control plant which recorded 30.86 mg/g fresh weight. JA treatments showed a significant reduction in carbohydrate concentrations at harvest.

At 2 weeks after harvest, significant reduction in total soluble carbohydrates was noticed by all bioregulator treatments in both season. However, carbohydrate reduction was more obvious with CPPU at 20 ppm followed by CPPU at 10 ppm and CPPU at 20 ppm+ JA at 10 ppm as compared with control plants. While at 4 weeks after harvest, carbohydrates reduction increased with all bioregulators and their combinations in the two seasons. The effect of CPPU on carbohydrate concentrations has been mentioned by several workers (Day *et al* 1995; Engels and. Marschner 1986). Ron'zhina, (2007) suggested that Cytokinins stimulated growth, activated source function of leaves, owing to the stimulation of leaf expansion, increasing of photosynthesis, changing a balance between transportable and storage forms of photoassimilates (increasing sucrose and reducing starch synthesis) and activate the genetic program of the development and functioning both of source and sink organs.

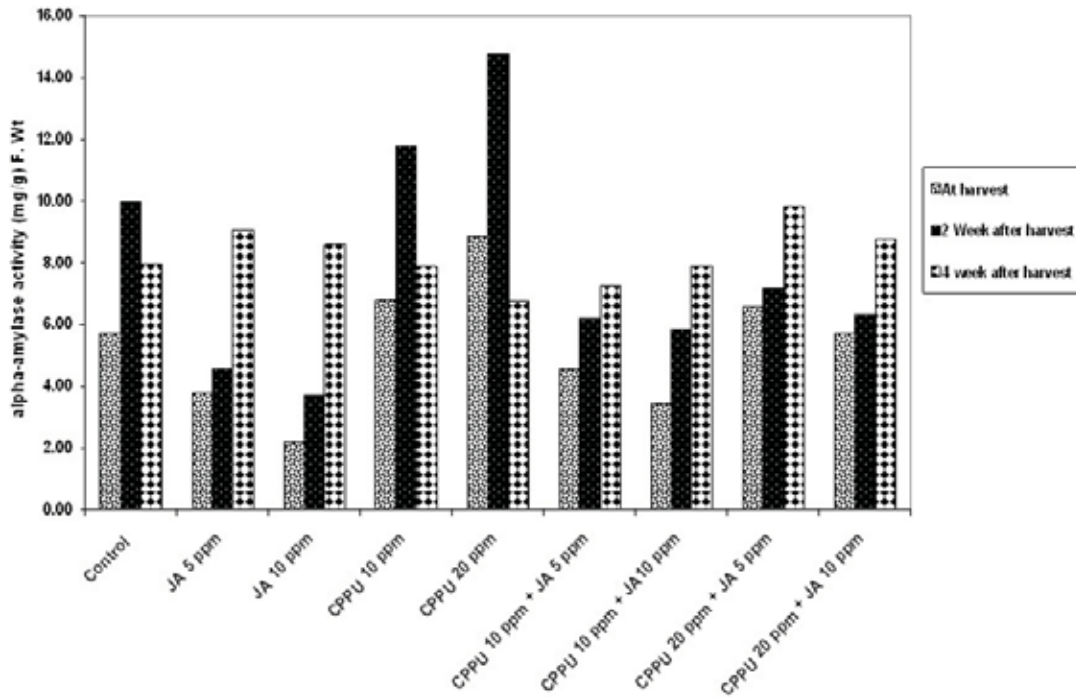


**Fig. 2:** Effect of JA and CPPU and their combination on total soluble carbohydrates (mg/g) in tuber tissues of potato at harvest, 2 and 4 weeks after harvest during the season of 2008.

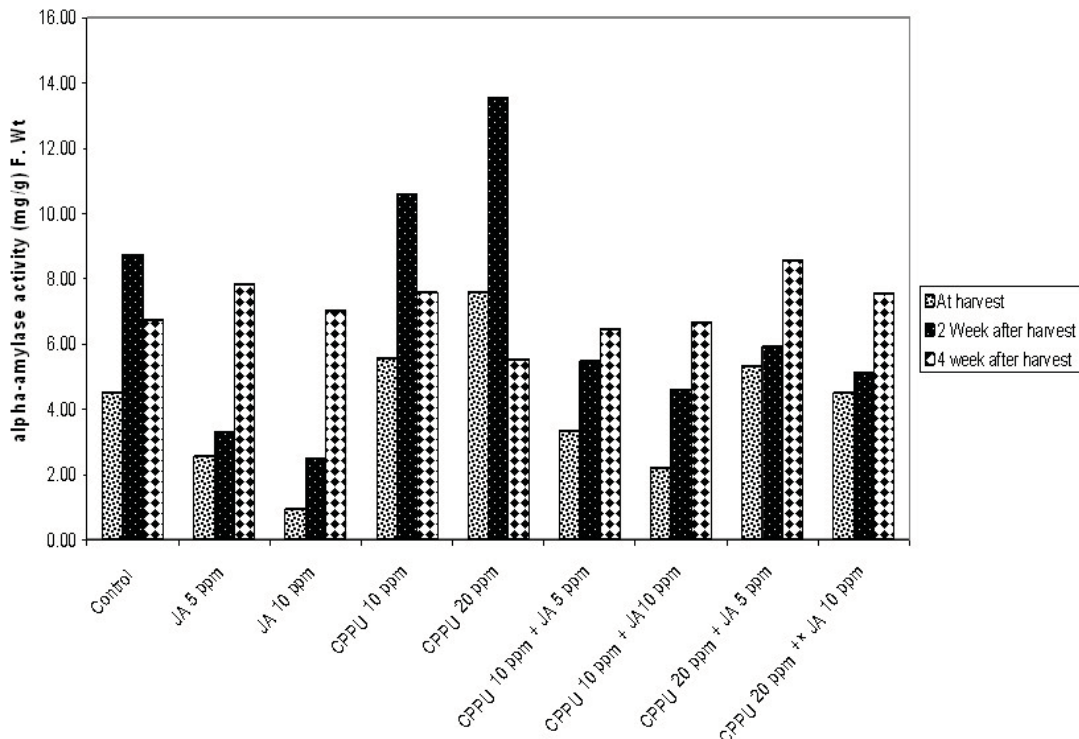
***α- amylase Activity:***

Plants treated with CPPU at 10 and 20 ppm, as well as, control plants showed high  $\alpha$ - amylase activity at harvest and at 2 week after harvest followed by reduction in this parameter was observed at 4 weeks after harvest. An opposite trend was noticed by plants treated with JA at 5 and 10 ppm and bioregulator combinations where  $\alpha$ - amylase activity increased gradually through the three sampling dates, at harvest, 2 and 4 week after harvest respectively. These results in agreement with Häggman and Haapala 2008, who mentioned that 6-benzylaminopurine (BAP) caused a great increase in amylase activity which was accompanied by a relatively high concentration of starch in the chloroplasts and decreased amylase activity in weak-growing plants.

It could be concluded from the present study that application of CPPU improved growth and tuber yield. Although, individual application of JA did not improve tubers yield. The combination of CPPU at 20 ppm and JA at 5 ppm was better in all aspects, plant growth, yield and alpha amylase activity. CPPU is recommended as the best treatment when dealing with tuber formation (tuberization) while JA is recommended as the best treatment for initiation flow of potato tuber. This means that the overlap between CPPU and JA lead to get the best yield of potato tubers.



**Fig. 3:** Effect of JA and CPPU and their combination on alpha-amylase activity (mg/g) in tuber tissues of potato at harvest, 2 and 4 weeks after harvest during the season of 2007.



**Fig. 4:** Effect of JA and CPPU and their combination on alpha -amylase activity (mg/g) in tuber tissues of potato at harvest, 2 and 4 weeks after harvest during the season of 2008.

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