J. Agric. Food. Tech., 4(8)1-7, 2014

ISSN 2090 – 424X
Journal of Agriculture and Food Technology
www.textroad.com

Effect of Organic Cytokinin and Nitrogen Fertilization on Wheat (*Triticumaestivum L.*) Plants

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Received: October, 22, 2014
Accepted: January 10, 2015

ABSTRACT

A field successive experiment was carried out employing sandy soil to clarify the role organic cytokines and varied levels N fertilization on some physiology properties, nutrient content yield of wheat (*Triticumaestivum L.*) plants. The experiment was carried out to evaluate the effect of three level of organic cytokinin (100, 200 and 300 ppm) as combined with different rates of nitrogen fertilization. Fertilizer nitrogen was added in four levels of100%, 75%,50% and 25% of the amount recommended by the Ministry of Agriculture. All previous treatments compared to the control treatment, which was 100% nitrogen fertilizer without adding cytokinin. The results showed that addition of cytokinin improved the yield of wheat despite the decreasing in the rate of addition of nitrogen fertilizer. Second rate of cytokinin(200 ppm) enough to obtain high value of grain and straw yield (2.81 and 6.15 ton fed^{-1}) under different rates of nitrogen fertilization. Addition first rate of cytokinin (100 ppm) under high level of nitrogen fertilization enough to obtain high value of N, P and K content of wheat plant and compared with control treatment. While it did not improve the rate of increase of cytokines from 100 ppm to 300 ppm increase in chlorophyll a and b and cartoniod under different rate of N fertilization.

KEYWORDS: Cytokinin, N fertilization, Wheat, Growth, Yield, Nutrient content, photosynthetic processes.

INTRODUCTION

The huge increase in the human population in Egypt in the last decades led to cultivate very poor soil as the sandy ones for food production. Cultivating grain crops such as corn and wheat was a must to secure bread production. Unfortunately, productivity of such newly cultivated soils is still under satisfaction; this may be due to the nature of sandy soil as well as imbalance of used fertilizers under such conditions. Wheat (*Triticumaestivum L.*) is a worldwide widespread crop by its innumerous industrialized derivatives **(Yaseen *et al.*, 2010)**.

Seaweeds form an integral part of marine coastal ecosystems. They include the macroscopic, multicellular marine algae that commonly inhabit the coastal regions of the world's oceans where suitable substrata exist. It has been estimated that there are about 9,000 species of macroalgae broadly classified into three main groups based on their pigmentation (for example, Phaeophyta, Rhodophyta, and Chlorophyta; or the brown, red, and green algae, respectively)**(Hong *et al.*, 2007)**.In order to improve the performance of agricultural crops, the use of seaweed extracts has grown, mainly because it is an environmentally friendly alternative to the use of fertilizers and plant bioregulators **(Craigie, 2011, Kumar &Sahoo, 2011 and Khan *et al.*, 2012).** *Ascophyllumnodosum* (L.) extract contains several compounds which are capable of stimulating plant growth, such as cytokinins, auxins, gibberellins, and alginates **(Khan *et al.*, 2009**). The improvement of seed germination and plant establishment, as well as an increase in crop productivity has been reported in some studies **(Craigie, 2011, Kumar &Sahoo, 2011 and Carvalho *et al.*, 2013)**. However, plant response to A. nodosum extract is highly varied **(Craigie, 2011)**.

Cytokinins are a group of plant hormones that promote celldivision and play a major role in the regulation of various biological processes associated with active growth, metabolism and plant development **(Mok&Mok 2001; Sakakibara 2006)**. They are also known to play a role in the synthesis and maintenance of chlorophyll and are known to influence chloroplast development and metabolism. As such, CKshave long been known to delay senescence **(Gan&Amasino 1995)**. CKs also impact plant nutrient translocation by converting source tissues into active sinks **(Mok&Mok 2001)**. Finally, they are also known to play a role in integrating diverse environmental stress responses **(Hare *et al.*, 1997)**.

The objective of this work was study the effects organic cytokinin under different levels of nitrogen fertilization on yield, nutrients content and physiology properties of wheat (*Triticumaestivum L.*) plants.

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MATERIAL AND METHODS

A field trial was successively conducted on a loamy sand soil at Ismailia Agricultural Research Station cultivated with wheat (*Triticumaestivum L.*, cvSakha 69) at winter 2013. Some physical and chemical properties of the cultivated soil were evaluated in samples taken before wheat planting according to standard procedures reported by **Cotteine (1980)** to be presented in (Table, 1).

Three level of organic cytokinin n_1, n_2, n_3 (100, 200 and 300 ppm, respectively) as combined with different rates of nitrogen fertilization. Seaweed (*Ascphyllumnodosum*) extracts source of organic cytokinin, Which contains a concentration of cytokines equal to 0.02%; Seaweed has been sprayed on the plants two weeks after germination, was repeated three times spraying. Fertilizernitrogen (in the form of NH_4 (SO4)$_2$ was added in four levels of100%, 75%,50% and 25%of the amount recommended by the Ministry of Agriculture. All previous treatments compared to the control treatment, which was 100% nitrogen fertilizer without adding organic cytokinin.

Plant samples were dried at 65C° for 48 hrs, ground and wet digested using H_2SO_4: H_2O_2 method **(Cottenie, 1980)**. The digests samples were then subjected to measurement of N using Micro-Kjeldahle method; P was assayed using molybdenum blue method and determined by spectrophotometer and K was determined by Flame Photometer (**Chapman and Pratt, 1961)**. Leaf chlorophyll reading (mg/100 mg DW) according to **(black 1965)**.

Table (1): Some physical and chemical properties of soil before wheatcultivation.

Soil property	Value	Soil property	Value
		pH (1:2.5 soil suspension)	8.1
Sand	84.1	EC (dS m^{-1}), soil paste extract	1.20
Silt	5.70	Soluble ions (mmol L^{-1})	
Clay	10.2	Ca^{++}	6.12
Texture	Loamy sand	Mg^{++}	4.60
		Na^{+}	1.54
$CaCO_3$ %	2.00	K^{+}	0.52
Organic matter%	0.03	CO_3^{--}	nd.
Available N (mg kg^{-1})	61.4	HCO_3^{-}	1.10
Available P (mg kg^{-1})	3.15	Cl^{-}	0.96
Available K (mg kg^{-1})	**6.10**	SO_4^{--}	9.60

nd: not detected

RESULTS AND DISCUSSIONS

Data presented in table(2) show the effect of N fertilization and organic cytokinin rates on yield and growth of wheat plants. Increasing cytokinin rate under different levels of nitrogen fertilization increasing grain, straw yield and some growth properties (such as leaves – and length and height plants) of wheat plants. Second rate of cytokinin (200 ppm) enough to obtain high value of grain and straw yield (2.81 and 6.15 ton fed^{-1}) under different rates of nitrogen fertilization. As well as ctyokinin enhancing nitrogen fertilizer efficiency, it was found that with the addition of cytokinin improved the yield of wheat despite the decreasing in the rate of addition of nitrogen fertilizer; where the yield has not decreased much in the low rates on nitrogen fertilizer by him in high rates. Cytokinins have been shown to participate in the regulation of numerous aspects of plant development including initiation of buds, flowering, abscission and yield by enhancing the cell expansion **(Morris *et al*., 1990)**. Cytokinins are a group of mobile phytohormones that play a critical role in plant growth and development by regulating leaf senescence **(Kim *et al*., 2006)**, apical dominance **(Tanaka *et al*., 2006)**, root proliferation **(Werner *et al*., 2001)**, phyllotaxis **(Giulini *et al*., 2004)**, reproductive competence **(Ashikari *et al*., 2005)**, and nutritional signaling **(Takei *et al*., 2002)**. Recent studies oncytokinin metabolism and signal transduction have identified a series of genes involved in these processes and a model for the regulation of developmental processes by cytokinin-related genes has been proposed **(Ferreira and Kieber, 2005 and Sakakibara, 2010)**. It is also now known that cytokinins participate in the maintenance of meristem function **(Leibfried *et al*., 2005 and Kurakawa *et al*., 2007)** and in the modulation of metabolism and morphogenesis in response to environmental stimuli **(Werner *et al*., 2006)**

(Table, 2): Effect of N fertilization and organic cytokinin rates on yield and growth of wheat plants.

Treatments		Yield ton fed^{-1}		Leaves	Leaves length	Plant height
N fertilization rate	Cytokinin rate	Grain	Straw		cm	
100%	n_1 (100 ppm)	2.79	6.09	17.84	22.80	79
	n_2 (200 ppm)	2.81	6.15	19.26	23.45	80.2
	n_3 (300 ppm)	2.69	5.85	18.65	22.2	79.2
75%	n_1 (100 ppm)	2.66	5.83	18.79	20.1	79.1
	n_2 (200 ppm)	2.68	5.85	19.0	21.2	80
	n_3 (300 ppm)	2.59	5.80	19.04	21.0	80
50%	n_1 (100 ppm)	2.62	4.95	18.60	19.1	79.8
	n_2 (200 ppm)	2.61	4.94	17.49	19.4	78.5
	n_3 (300 ppm)	2.60	4.90	17.46	19.0	78.4
25%	n_1 (100 ppm)	1.44	3.81	15.70	18.40	79.0
	n_2 (200 ppm)	1.46	3.79	14.72	18.41	68.3
	n_3 (300 ppm)	1.41	3.79	14.72	18.40	67.9
Control		2.72	6.10	17.88	22.78	80.1

Data presented in table (3) show the effect of N fertilization and organic cytokinin rates on N, P and K in the flag leaf of wheat plants. Decreasing N fertilization rates decreasing N content but P and K content don't affect when N fertilization rate increased.

(Table, 3): Effect of N fertilization and organic cytokinin rates on N, P and K content of wheat plants.

Treatments		Nutrient content in the flag leaf, (%)		
N fertilization rate	Cytokinin rate	N	P	K
100%	n_1 (100 ppm)	3.13	0.39	2.91
	n_2 (200 ppm)	2.94	0.34	2.87
	n_3 (300 ppm)	2.95	0.35	2.87
75%	n_1 (100 ppm)	2.94	0.42	2.76
	n_2 (200 ppm)	2.93	0.39	2.70
	n_3 (300 ppm)	2.98	0.45	2.82
50%	n_1 (100 ppm)	2.89	0.47	2.76
	n_2 (200 ppm)	2.85	0.41	2.77
	n_3 (300 ppm)	2.85	0.39	2.76
25%	n_1 (100 ppm)	2.08	0.42	1.99
	n_2 (200 ppm)	2.09	0.40	2.02
	n_3 (300 ppm)	2.02	0.39	2.10
Control		2.98	0.45	2.90

Addition first rate of cytokinin (100 ppm) under high level of nitrogen fertilization enough to obtain high valueof N, P and K content of wheat plant and compared with control treatment.

Many studies highlighted the close correlation between nitrogen and cytokinin in several plant species **(Takei *et al*., 2002 and Cline *et al*., 2006)**. The interactions between N and cytokinin in the regulation of metabolism and development revealed that cytokinin biosynthesis is up-regulated by nitrate and the hormone in turn was proposed as a regulator of the morphological response, protein synthesis capacity and macronutrient acquisition **(Sakakibara *et al*. 2010)**. These findings unequivocally indicate modulation of CK metabolism and translocation by the N nutritional status. The results of **Garnica *et al*. (2010)** showed that the presence of NO3– was associated with clear increases in the active forms of cytokinin.

Data presented in Figure (1, 2 and 3) show the influence of different levels of N fertilization and different concentration of organic cytokinin on some physiology properties such as chlorophyll a and b and cartoniod of wheat leaves. Increasing the rate of decline in N fertilization rate from 100% to 25% lead to a decrease in chlorophyll a and b and cartoniodin wheat leaves. Nitrogen is a macronutrient present in many key biological molecules. It is available for plants predominantly as $NO_3^-$and NH_4^+. It controls many aspects of plant life, and has a strong impact on plant development. In response to changes in nitrogen supply, plants display elaborate responses at both physiological and morphological levels to adjust their growth and development **(Vidal and Gutierrez 2008, Lošák *et al*. 2010)**. The reduction of $NO_3^-$to ammonium (NH_4^+) involves the sequential action of nitrate reductase and nitrite reductase. The resulting $NH_4^+$is then assimilated by glutamine synthetase and glutamate synthase to organic forms such as glutamate (Glu) and glutamine (Gln). These amino acids (AAs) are precursors in the synthesis of other AAs, nucleic acids, chlorophylls or hormones (**Pavlíková *et al*., 2012**).While it did not improve the rate of increase of cytokines from 100 ppm to 300 ppm increase in chlorophyll a and b and cartoniod, but it was the first rate (100 ppm) sufficient to obtain a higher concentration of green matter. **Pazurkiewicz-Kocot *et al*.,(2011)** reported that plant metabolism may be affected by kinetin in different ways and particularly processes of biosynthesis of chlorophylls in higher plants are susceptible to kinetin. Under reduced chlorophyll synthesis the photosynthesis is reduced. The results of chlorophyll content indicated a decrease with increasing concentration of kinetin in the culture medium. On the other hand, photosynthesisis one of the most important studied physiological processes in plant sciences. Plant metabolism is affected by photosynthesis and particularly photosynthetic processes in higher plants decided about plant growth and development. Kinetinpenetrates into chloroplast seems to be a very important factor protecting photochemical activity. The photosynthetic activity of chloroplasts is related to the presence of many factors. One of them is chlorophyll pigments.

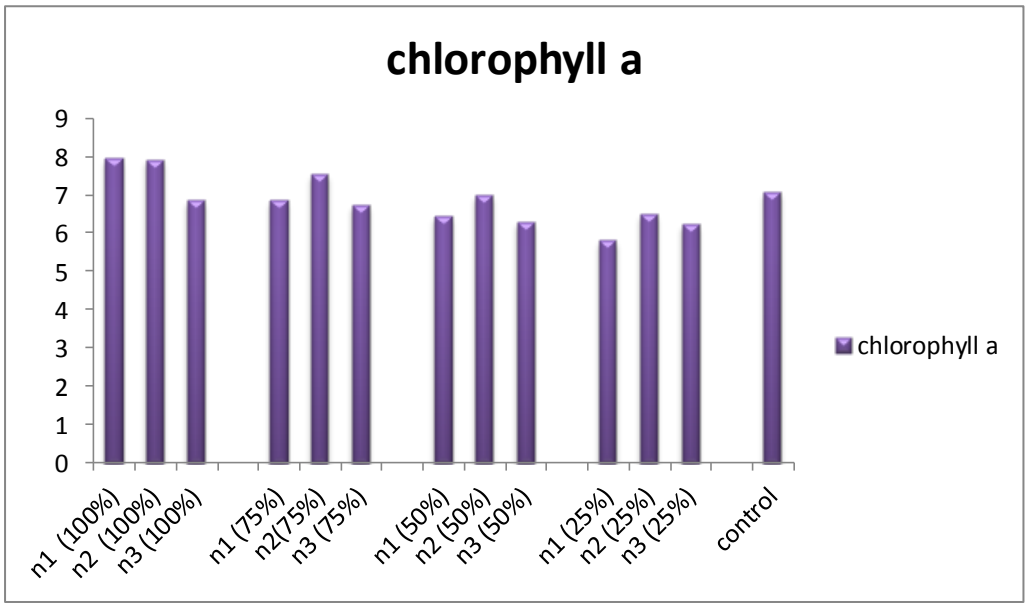

Fig. 1: Effect organic cytokinin and nitrogen fertilization levels on leaves chlorophyll a of wheat plants.

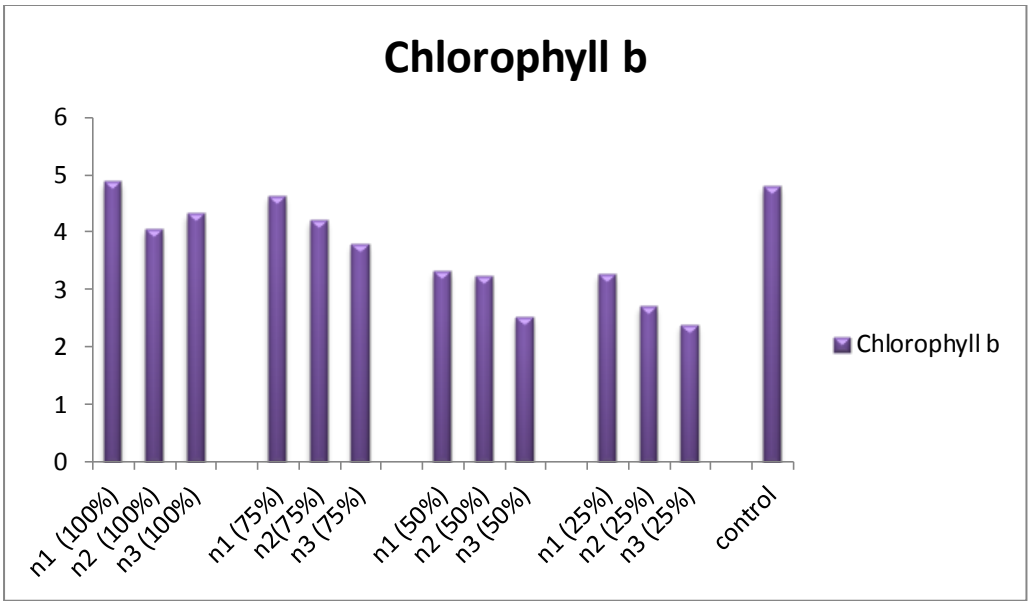

Fig. 2: Effect organic cytokinin and nitrogen fertilization levels on leaves chlorophyll b of wheat plants.

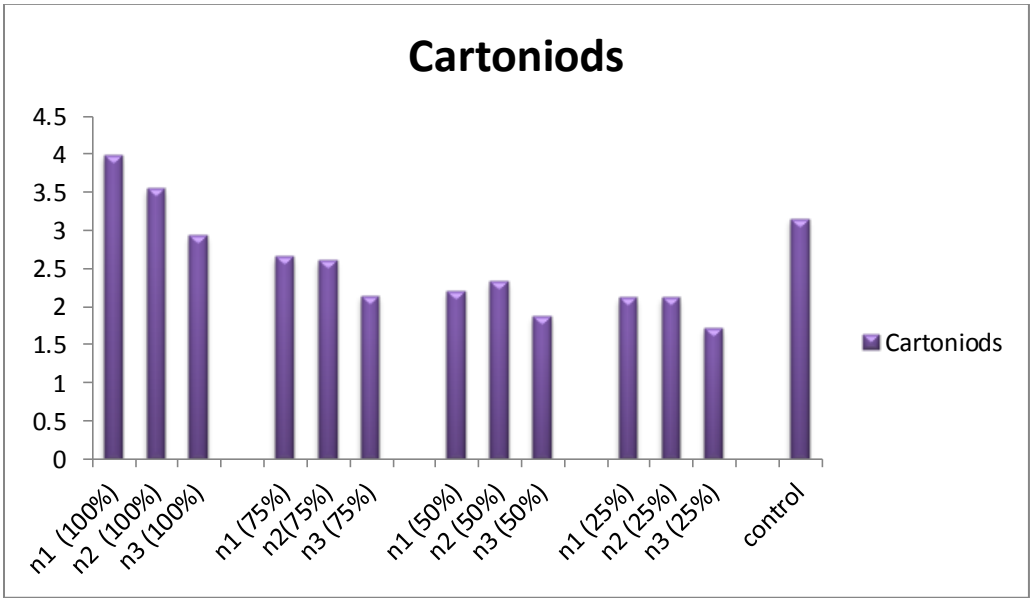

Fig. 3: Effect organic cytokinin and nitrogen fertilization levels on leaves cartoniods of wheat plants.

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