

## **RESPONSE OF COWPEA PLANTS (*Vigna unguiculata* (L.) WALP.) TO SOME BIOSTIMULANTS AND ORGANIC NUTRIENTS DURING LATE SUMMER SEASON.**

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### **ABSTRACT**

*Two field experiments were conducted during 2005 and 2006 at El-Baramon Experimental Farm, El-Dakahlia Governorate, Hort. Research Institute. In order to study response of cowpea plants El-Balady cv. grown during late summer season to foliar feeding with some biostimulants (amino acids– seaweed extracts) and organic nutrients,(potassium borate citrate, organic boron,zinc citrate, calcium citrate, NPK-Humate)besides to NAA, on growth, mineral composition and yield and its components towards, maximizing its productivity under such season condition.*

***The results could be summarized as follows:***

*The applied treatments were significantly differed among them and were superior the control in all growth parameters, mineral concentration (NPK and Ca) in leaves, seed yield and its components during the two seasons.*

*The best ones in diminished order were KB-citrate NPK-humate, NAA, amino acids, B-EA (organic boron), seaweed extracts, Zn-citrate and at least Ca-citrate, those increased their plants, dry weight and total seed yield increased over the control by (35.67%,43.9%), (26.79% ,35.01%), (26.36% ,35.1%), (24.66% ,30.8%), (24.63% ,32.3%), (17.98% ,27.5%) ,(16.64%,25.8%) and (5.73%,20.5%) means of the two seasons, respectively. Also, these treatments resulted in significant increase in concentration of N,P,K and Ca in leaves compared with the control one.*

*It could be sprayed late summer grown cowpea plants with potassium borate citrate (3ml/L) or NPK-humate (2.5ml/L) to obtain the best growth and mineral concentration and the maximum total seed yield.*

**Key words:** Cowpea plants (*Vigna unguiculata* (L.) WALP.), biostimulants, organic nutrients, summer season.

### **INTRODUCTION**

In recent years, stressful high temperature prevails in Egypt during late summer season. This frequently cause severe depression in different performances of vegetable crops including cowpea (Wein *et al.*, 1993; Fathy and Farid, 2000; Fathy and Khedr, 2005 b).

At the physiological and metabolic basis high temperature induces serious internal disordered case and drastic events of bioassimilates, hormones and nutrients shortage and imbalances and lacking of their supply into reproductive sinks; as well as induces serious injurious oxidative stress. This case lead to serious agronomical problems of poor fruit setting, abscission of fruits and low productivity (Wien and Zhang, 1991, Huberman *et al.*, 1997; Fathy and Farid, 2000, Khedr *et al.*, 2004).

In addition, thermotolerance and thermoinjury known to be associated with the internal ions level that play a role in the activation of the transporter enzymatic system (H-ATP-ase) membrane pump and certain  $H^+$ ,  $K^+$  and  $Ca^{++}$  balance leading to stress protection processes and responses (Palta, 1990).

Also, with the capability to activate the antioxidantal defensive enzymatic systems against high temperature inducable degradable oxidative stress (Dicknson *et al.*, 1991 and Cakmak and Marschner, 1992).

Based on the above mentioned information and the cited literature, the present work aimed to ameliorate the propable destructive effects of high temperature condition, restore the plants into normal function and enhanced productivity case. This suggested to be achieved by application and then identification the effect of promising natural biostimulants and organic nutrients, i.e. amino acids, seaweed extracts, NPK- humate complex, K-B citrate, Ca-citrate, Zn-citrate, and B-EA (boron organocomplex) as well as NAA on different performance of cowpea plants.

Herein of value to cited the relevant information about these treatments Franco *et al.* (1998) showed that foliar spraying of amino acids improved fruit yield and quality of tomato. Fathy and Khedr (2005a) illustrated that the frequent foliar feeding with amino acids, Zn-citrate and phytoextract (yeast) individually or in combination was considerably recovered the biotic stressed tomatoes due to their beneficial effects on the metabolic potential for synthesis of amino acids, protein, sugars, carbohydrates also to their antioxidant defensive function including synthesis of carotenoids and phenols also enhancing efficient on uptake of N, P and K and at least corresponding these responses with normal growth and high fruit yielding capacity.

Many different effects have been reported as a result of the application of seaweed extracts, including enhanced crop yield, increased uptake of inorganic nutrients from the soil, improved different growth aspects, induced resistance to stress conditions (biotic and a biotic ones). Such extracts know to exhibit different regulatory, and defensive effects through elicitation and singling of different physiological and metabolic processes (Metting, 1990; Blunden, 1991). Whapham *et al.* (1993) used alkaline extract of *Ascophyllum nodsum* either as foliar or soil application, this result in improving growth and chlorophyll content of treated plants El-Aidy *et al.* (2002) found that, foliar spraying of different commercial formulations of seaweed extracts (Maxicrop and ASCO) considerably improved growth, N and P content of leaves, early and total yield of sweet pepper. Similar results were obtained by using foliar feeding of brown algae (*Ascophyllum nodsum*) (Goemar) (Klarzynski *et al.*, 2006 and Zdravkovic *et al.*, 2006) on bean.

Meanwhile it was demonstrated that, humic acid, alone and/or its, formulations in form of mineral organocomplex applied through foliar or root feeding resulted in

considerable improvement in different crops growth, mineral contents and bioconstituents and productivity.

On the other hand, it was well known that, some nutrients (K, Ca, Zn and B) specially in organic chelated form are efficiently participated in the internal processes associated with protection against high temperature stress condition also, that their foliar spraying is considerably improved different performances of the stressed plants (Franco *et al.*, 1998; Wenyi *et al.*, 1999; Khedr *et al.*, 2004 ; Fathy and Khedr, 2005 a and b).

In addition, it was found that foliar application of NAA especially during high temperature condition is used as a standard practice for reducing abscission of reproductive organs, improving fruit set and productivity also enhancing growth and mineral content of different crops (Pressman *et al.*, 1998 with pepper; Singh and Awasthi, 1998 with *Vigna radiate*; Resmi and Gopalakrishnan, 2004 with *Vigna unguiculata* (L.) Walp .

Therefore, the aim of the present study to investigate the response of cowpea plants (*Vigna unguiculata* (L.) Walp.) to some biostimulants and organic nutrients during late summer season.

## MATERIALS AND METHODS

Two field experiments were performed at El-Baramon Experimental Farm, El-Dakahlia governorate, Horticulture Research Institute during two summer successive seasons of 2005 and 2006 on cowpea plants. This work aimed to study the effect of some biostimulants and organic nutrients on different performances of this crops under high temperature stressful conditions of late summer season towards maximizing its productivity under such condition.

May, 2<sup>nd</sup> seeds of cowpea El-Balady cv. were sown in hills on one side ridge of 5m long and 0.6m wide and 0.25m between hills. After emergence the plants thinned leaving one plant per hill. The plot area was 9m<sup>2</sup> with planting density of about 26600 plant /fed.

These plants were sprayed 3 times (20 day after sowing and again every 20 day) with the solution of the following treatment :-

- 1-Amino acids (35.0% w/v total amino acids) from biological origin, commercially named Mëgfol introduced from Valagro Comp., Italy., registered in Egypt as biostimulants with number 4827, applied at concentration of 1.5ml/L.
- 2-Seaweeds, natural phytoextract of *Ascophyllum nodosum* a brown algae introduced from Goëmar Laboratoires, France, registered in Egypt as biostimulant (Goëmar BM86) with No. 5468. It contains more than 17 essential amino acids (5738 $\mu$  mol / L), vitamins (40 mg / kg) phytohormons (200 mg / L), Betaines (140 mg / L), polysaccharides (50% DM), also enriched with N, Mg, S, B and Mo, applied at concentration of 2.5ml/L.
- 3-NPK-Humate: 10% humic acids + 8% (N) + 8%( P<sub>2</sub>O<sub>5</sub>) + 8%( K<sub>2</sub>O) registered with NO. 5709 Horticulture. Research Institute, applied at concentration of 2.5 ml/L.

- 4-Potassium borate citrate (35% K<sub>2</sub>O , 5% B), formulation of Hort. Res. Inst. applied at concentration of 3.0ml/L.
- 5-Calcium citrate (Ca-cit.) 25% Ca, registered No. 5713 Hort. Res Inst., applied at concentration of 5ml/L.
- 6-Zinc citrate (Zn cit.) 12% Zn, registered No. 5714, Hort. Res Inst., applied at concentration of 1.5ml/L.
- 7-Organic boron (B-EA) (Solbor) 8% B, from Hort. Res Inst., applied at concentration of 1ml/L.
- 8-NAA, from Alfrut commercial formulation (3.5% pure NAA w/v), from SIFO, Italy applied at concentration of 10mg/L.
- 9-Control, only sprayed with water.

All cultural practices were performed as recommended by Hort. Res Inst.

Randomized complete block design was adopted and the treatments were replicated three times.

At 75 days from sowing 5 plants from each plot were taken and No. of leaves, No. of shoots, fresh and dry weight (gm) were recorded and calculated per plant. Samples of fully expanded mature leaves were taken from each plot 10 days after the last spray for chemical analysis of N, P, K and Calcium. Nitrogen, P and K were analyzed and determined according to methods of Horneck and Miller (1998), Sandell (1950) and Horneck and Hanson (1998), respectively. Calcium was analyzed using atomic absorption method.

Yield and its components: number of pods/plant and number of seeds/pod were recorded from the cumulative harvestings of ten marked plants of each plot. The dried seeds weight determined on plot basis then, seed yield (gm)/plant and total seed yield (ton)/fed. were calculated.

All data were statistically analyzed according to (Snedecor and Cochran ,1980). Also, data of air temperature(C<sup>o</sup>) and relative humidity(%) as well as of soil analysis during 2005 and 2006 are presented herein in Tables 1 and 2.

**Table (1): Means monthly air temperature(C<sup>o</sup>) and relative humidity (%) at El-Dakahlia Governorate, during 2005 and 2006.**

Months	Air temperature (C <sup>o</sup> )				Relative Humidity (%)	
	2005		2006		2005	2006
	Max.	Min.	Max.	Min.		
<b>May</b>	30.37	15.95	29.52	15.13	61.10	61.81
<b>June</b>	31.66	19.87	32.85	19.80	65.67	65.97
<b>July</b>	33.55	20.30	32.69	21.61	69.87	70.55
<b>August</b>	35.68	24.39	34.47	22.55	71.00	71.26
<b>September</b>	33.45	20.75	33.65	19.82	68.10	65.67

Source: Mansoura Meteorological Station.

**Table 2 : Mechanical and chemical analysis of the experimental soil during 2005 and 2006.**

Soil properties	2005	2006
Clay (%)	59.95	58.62
Silt (%)	24.94	26.50
Sand (%)	15.11	14.88
Texture	Clayey	Clayey
Organic matter % (O.M.,%)	1.95	1.89
Ca CO <sub>3</sub> (%)	1.99	2.35
Available N ( ppm)	21.12	22.2
Available P ( ppm)	11.72	9.36
Available K ( ppm)	391	399
EC m. mhos/cm (25°C)	1.50	1.48
pH	7.9	8.1

The experimental soil was analyzed by using standard method described by Page (1982).

## RESULTS AND DISCUSSION

### 1- Vegetative Growth:

The data in Table 3 prove that, all the applied foliar sprays of biostimulants and minerals in organic form were significantly increased number of leaves and shoots, fresh and dry weight of their plants in comparison with the control one during the two seasons. The most stimulatory and potent effects in diminished order were of KB-citrate followed by NPK-humate and NAA, then B-EA and amino acids, Zn-citrate and seaweed extracts and at least Ca-citrate respectively.

These treatments increased dry weight of plants relative to the control one by 35.67%, 26.79%, 26.36%, 24.66%, 24.63%, 17.98%, 16.64%, and 5.73% (mean value of the two seasons), respectively.

These results were inconsistent with those obtained by Wenyi *et al.* (1999); Fathy *et al.* (2003) and Khedr *et al.* (2004) for foliar feeding with K, Ca, Zn, and B under stress condition; Neri *et al.*, 2005, Padem *et al.*, 2007, El-Aidy *et al.*, 2002 (humic acids and seaweed extracts), Franco *et al.*, 1998 (amino acids); Resmi and Gopalakrishnan, 2004 (NAA). Seemingly the applied treatments achieved growth increments in dry weight ranged from 16.6 to 35.6%, except that of Ca-citrate 5.7%, also, some treatments such as KB-cit. were superior NAA (the standard synthetic phytohormone) or some what equal it NPK-humate, B-EA and amino acids. This to far extent proved high stimulatory effects for these treatments).

Herein, the lowest growth behavior of untreated cowpea plants proved that, these plants, were to certain extent stressed by high temperature. This could be due to



the inhibitory effect of high temp. condition on uptake and in turn concentration of N, P, K and Ca in control plant leaves (Table 4). Also may be due to the inducible stress related internal disturbances and shortage of bioconstituents, hormones and nutrients (Wien and Zhang, 1991 and Huberman *et al.*, 1997). High temperature known to induce serious physiological oxidative stress of elevated toxic level of ROS, thereby more destructive and degradable effects on the whole biosynthetic machinery (Dickson *et al.*, 1991 and Cakmak and Marschner, 1992). So it is not surprising to hold great responses and mitigation for the treated cowpea plants under present work climatic condition (Table 2).

The most growth encourages effect of KB-citrate may be due to that this formulation contain K and B of the similar and harmonic synergetic action, complexes with organic moiety of citrate, ensuring potent K and B nutrition for more enhancement of carbohydrates, proteins, enzymes, phenols and energy synthesis and saving as well as more osmoregulation and protection, lignification and stress protection. At the same basis the beneficial effect of B organo complex B-EA may be explained (Marschner, 1995, Khedr *et al.*, 2004 and Fathy *et al.*, 2003). The organo / mineral complex of NPK-humate is greatly induced high balanced N, P, K and Ca concentration within cowpea leaves (Table 4), besides to the known encouraging action of humic acids, for protein synthesis, nutrients translocation, antioxidantal enzymes, root proliferation and foliar growth (Sun *et al.*, 2004 and Trkmen *et al.*, 2005).

NAA as a commonly used synthetic auxin, known to stimulate cell division and differentiation process of root and tip, activate oxidative stress protective peroxidases (Hess, 1981).

Amino acids and seaweeds extracts via their amino acids and other constituents enhanced biosynthesis of protein / enzymes (antioxidantal and transporter ones) and/or compensated it under stress condition, improve the capacity of photosynthesis, carbohydrate and phenol content as well as the biochemical defensive responses against environmental stresses (Franco *et al.*, 1998, Fathy and Khedr, (2005a) on amino acids, (Zdravkovic *et al.*, 2006, Klarzynski *et al.*, 2006) on seaweed extracts.

Lastly, the beneficial influence of foliar applied organometal complexes of Zn and Ca (citrate form) this form suggested to be due to their growth related efficient involvements and utilization, i.e. cell division, hormone synthesis and balance, stress signaling and regulatory roles (Marschner, 1995).

## **2- Mineral Composition:**

From data presented in Table 4, it was recognized that, all foliar spray treatments significantly differed among them and were highly superior to the control in concentration of N, P, K and Ca in leaves during the two seasons. The same data cleared that the highest N and P concentration recorded with NPK-humate followed by KB-citrate and seaweed extracts. Where as, the highest K concentration was of KB-citrate followed by NPK-humate, NAA and B-EA treatments. Ca-citrate followed by KB-citrate and NAA showed the considerable highest Ca in leaves of their plants. The results, were in the same trends in both seasons.

**Table 4: Effect of some biostimulants and organic nutrients on mineral composition of cowpea leaves during 2005 and 2006.**

Treatments	N%		P%		K%		Ca%	
	2005	2006	2005	2006	2005	2006	2005	2006
<b>Amino acids</b>	3.53	3.59	0.261	0.253	3.08	3.01	2.30	2.28
<b>Seaweed extracts</b>	3.43	3.49	0.264	0.261	2.90	2.84	2.21	2.14
<b>NPK-Humate</b>	3.75	3.93	0.342	0.337	3.50	3.51	2.31	2.31
<b>KB-cit.</b>	3.50	3.60	0.326	0.321	3.93	3.84	2.42	2.39
<b>Ca-cit.</b>	3.05	3.17	0.243	0.242	2.40	2.45	2.91	2.75
<b>Zn-cit.</b>	3.07	3.17	0.263	0.258	2.85	2.79	2.22	2.12
<b>B-EA</b>	3.35	3.44	0.259	0.248	3.20	3.16	2.18	2.09
<b>NAA</b>	3.27	3.47	0.295	0.282	3.26	3.26	2.40	2.33
<b>Control</b>	2.69	2.88	0.243	0.233	2.75	2.68	2.06	2.00
<b>L.S.D (0.05)</b>	<b>0.08</b>	<b>0.05</b>	<b>0.006</b>	<b>0.004</b>	<b>0.03</b>	<b>0.10</b>	<b>0.08</b>	<b>0.07</b>

The present significant increments in mineral concentration values of all treatments over the control may be explained based on the degradable effect of high temperature condition (Table 1) and its paralleled inducible oxidative stress on plasma membrane and its binding transporter enzymes and then the consequential inhibition for, mineral, uptake and translocation processes (Pallta, 1990 and Dicknson *et al.*,1991).

Meanwhile, the direct or indirect involvement of the applied treatments, either by feeding with certain minerals or to protect and recovered specific transporter enzymes and/or the whole metabolic machinery under such conditions is documented (Palta, 1990 and Marschner, 1995) on K, Ca, Zn; (Klanzynski *et al.*, 2006) on seaweed, ex.; (Fathy and Khedr, 2005a) on amino acids and Zn-citrate; (Neri *et al.*, 2005, Padem *et al.*, 2007) on humic acids, (Singh and Awasthi, 1998; Resmi and Gopalakrishnan, 2004) on NAA.

### 3- Yield and its Components:

From the data illustrated in Table 5, it was shown that, all treatments were greatly increased yield and its components over the control in the two seasons and differed among them in most cases. The significant highest total seed yield/fed. was of KB-citrate treatment, its increment reach 43.9% over the control, this considerably was due to the superiority of this treatment in number of seeds/pod and seed yield/plant but not in number of pods/plant, during the two seasons.

Also due to its noticable enhancable effect on the accumulation of dry matter and concentration of minerals in their plants foliage (Tables 3 and 4). So the seed yield enhancement of this treatment suggested to be mainly via the synergetic efficient role of its K and B together in photoassimilate formation and transportation into the reproductive sinks, in turn increasing-yield mass, besides to their other known regulatory and protective functions (Marschner, 1995). Also in similar to results of





(Fathy *et al.*, 2003 and Khedr *et al.*, 2004). The same data proved that, the subsequent effective treatments were, NAA and NPK-humate those which equally increased total yield of seeds by 35.0% and greatly improved all yield components specially the number of pods/plants with less or no differences between them in most cases.

This was in similar to the findings obtained by Samet (2004), Sun *et al.*, (2004) for humic acids, Singh and Awasthi (1998); Resmi and Gopalakrishnan (2004) for NAA.

It was also obvious that the significantly following seed yield encouraging treatments were B-EA, amino acids, seaweed extracts, Zn-citrate and at least Ca-citrate. Those increased total seed yield of cowpea plants over the control by 32.3%, 30.8%, 27.5%, 25.8% and 20.0% (mean of the two seasons) respectively. This in the same line was due to their enhanceable effects either on number or mass of the reproductive organs as shown by their effect on number of seeds / pod, number of pods/plant and seed weight/plant in both seasons. Also, due to their advantageous effect on growth and mineral / concentration of their plants in similar fashion. (Tables 3 and 4).

These results were in agreement with those obtained by Wenyi *et al.*, 1999; Plese *et al.*, 1998; Khedr *et al.*, 2004 for Zn, B and Ca; Franco *et al.*, 1998 for amino acids; Blunden *et al.*, 1991 for seaweed extracts.

*Conclusively*, of interest to be considered and give more attention for the following point :

- 1-The essentiality of foliar feeding with certain one or more elements of similar and synergistic roles such as K and B in metal-organo complex from to exert satisfactory nutritional, regulatory and stress defensive effects .
- 2-Using minerals / humic acids complex as new and advanced nutrition tool.
- 3-Under high temperature condition cowpea plants is exhibit certain sensitivity level and in turn it required some encouraging treatments for mitigation and maximizing its productivity .

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## إستجابة نباتات اللوبيا لبعض المنشطات الحيوية والمغذيات العضوية خلال العروة الصيفية المتأخرة

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أجريت تجربتان حقليتان عامي ٢٠٠٥ ، ٢٠٠٦ بالمزرعة البحثية التابعة لمعهد بحوث البساتين – البرامون دقهلية لدراسة تأثير التغذية الورقية لبعض المغذيات في صورة بعض المنشطات الحيوية (الأحماض الأمينية – مستخلصات الأعشاب البحرية) أو في صورة عضوية (سترات و بورات البوتاسيوم ، بورون مخلي ، سترات زنك ، سترات كالسيوم ، هيومات النيتروجين و الفوسفور و البوتاسيوم) إضافة إلى نقتالين حمض الخليك على النمو والتركيب المعدني والمحصول ومكوناته لنباتات اللوبيا صنف البلدي تحت ظروف العروة الصيفية المتأخرة بهدف تعظيم الإنتاجية تحت تلك الظروف .

وكانت أهم النتائج كما يلي :

اختلفت المعاملات فيما بينها وتوقفت جميعاً معنوياً على معاملة المقارنة في تحسين جميع صفات النمو الخضري تحت الدراسة (عدد الأوراق وعدد الأفرع والوزن الطازج والجاف/ النبات) في موسمي التجربة . وكانت أفضل المعاملات على التوالي كما يلي :  
سترات و بورات البوتاسيوم ، هيومات النيتروجين و الفوسفور و البوتاسيوم ، نقتالين حمض الخليك ، الأحماض الأمينية ، البورون المخلي ، مستخلصات الأعشاب البحرية ، سترات زنك ، سترات كالسيوم .

حيث أدت جميع هذه المعاملات إلى زيادة الوزن الجاف للنبات بمقدار ٣٥.٦٧% ، ٢٦.٧٩% ، ٢٦.٣٦% ، ٢٤.٦٦% ، ٢٤.٦٣% ، ١٧.٩٨% ، ١٦.٦٤% ، ٥.٧٣% على التوالي مقارنة بمعاملة الكنترول .

كما أدت هذه المعاملات إلى زيادة معنوية في تركيز النيتروجين والفوسفور والبوتاسيوم والكالسيوم بأوراق نباتات اللوبيا مقارنة بتلك النباتات غير المعاملة .

في نفس الوقت اختلفت تلك المعاملات معنوياً فيما بينها وتوقفت على معاملة المقارنة في التأثير على المحصول ومكوناته وأدت تلك المعاملات بنفس الترتيب إلى زيادة المحصول الكلي للبذور بمقدار ٤٣.٩% ، ٣٥% ، ٣٥.١% ، ٣٠.٨% ، ٣٢.٣% ، ٢٧.٥% ، ٢٥.٨% ، ٢٠.٥% .

وتوصي الدراسة برش نباتات اللوبيا النامية خلال العروة الصيفية المتأخرة بالمغذيات العضوية الآمنة سترات و بورات البوتاسيوم بتركيز (٣مل/لتر) أو هيومات النيتروجين والفوسفور والبوتاسيوم بتركيز (٢.٥مل/لتر) ثلاث مرات خلال موسم النمو للحصول على أفضل نمو ومحتوى كيماوي وأعلى محصول كلي للبذور .