

# Effect of some Simulative Compounds on Growth, Yield and Chemical Composition of Snap Bean Plants Grown under Calcareous Soil Conditions

Hanafy Ahmed, A. H.<sup>1</sup>; Nesiem, M. R.<sup>1</sup>; Hewedy, A. M.<sup>2</sup> and Sallam, H. El-S.<sup>2</sup>

<sup>1</sup>Plant Pysiology Section, Faculty of Agriculture, Cairo University, <sup>2</sup>Vegetable Research Dep., Horticulture Research Institute, Giza, Egypt

**Abstract:** Field experiments were conducted at Nubareia Agricultural Research Station Farm on a calcareous soil under drip irrigation system to study the effect of adding humic acid (20 g/l) as well as foliar application with putrescine (1.0 mg/l), novavol (2.5 ml/l) and vegimax (0.2 ml/l) on growth, yield and its components as well as chemical composition of snap bean plants cv. Paulista. The obtained results indicated that, application of humic acid, novavol or vegimax significantly increased all of the studied growth characters, i.e. plant height, number of leaves and branches/plant, leaf area as well as dry weight of shoots and roots at both samples. However, putrescine did not show a clear trend for various studied growth characters. Moreover, addition of humic acid as well as foliar application by novavol, putrescine and vegimax significantly increased yield and its components, i.e. total green pod yield, number of pods/plant, pods weights/plant, average pod weight and pod diameter at both pickings. Plant pigments, i.e. chlorophyll a, total chlorophyll and caroenoids concentrations either in leaves or pods significantly increased at both samples when the plants treated with humic acid, novavol and vegimax. In shoots, P, K, Ca, Mg, Zn and Fe concentrations significantly increased at both samples with all studied applications. Meanwhile in pods, addition of humic acid significantly affected on N, K, Fe and Cu concentrations, whereas foliar application with novavol and vegimax significantly improved N, K, Mg, Zn, Mn and Fe concentration at both pickings. On the other hand, application of any treatments significantly decreased Na and Cl concentrations in both shoots and pods at both samples. Results also showed that all studied applications significantly increased some organic compounds, i.e. total sugar, total free amino acids and total soluble phenols concentrations at both of shoots and pods in both samples as well as protein concentration in pods. Meanwhile, all studied applications significantly decreased pods bending % and fiber content in pods at both pickings as well as pod humidity % and nitrate concentrations with humic acid, putrescine and vegimax treatments. [Journal of American Science 2010;6(10):552-569]. (ISSN: 1545-1003).

**Keywords:** Field; drip irrigation system; humic acid

## 1. Introduction:

Snap bean (*Phaseolus vulgaris*, L.) is one of the most important members of leguminous crops grown in Egypt for either local consumption or exportation.

Bean plants are relatively sensitive under sandy soil conditions compared to most vegetable crops. In Egypt, the newly reclaimed land at Nubaria region reach about 900.000 feddan 290.000 feddan from its are calcareous (El-Zaher et al., 2001). Calcareous soils are considered the most important limiting factors for nutrients availability due to many problems, i.e. hardness and compaction of soil surface, less organic contents, low availability of N, P as well as micronutrients (Xudan, 1986 and Kulikova et al., 2002) and more holding water.

Many investigators reported that improving the tolerance of snap bean plants to such pure soil conditions could be achieved by application of different natural and chemical substances to enhance

its growth and maximizing the yield. It was found that, humic substances (humic and fulvic acids) are the major components of soil organic matter. The humic substances in the soil might have both direct and indirect effects on plant growth (Chen and Aviad, 1990). Indirect effects involved improvement of soil properties such as aggregation, aeration, permeability, water holding capacity, ions transport and availability through pH buffering (Tan, 2003). Direct effects are those, the uptake of humic substances into the plant tissue resulting in various biochemical effects through elvat nutrient uptake and maintaining vitamins and amino acids level in plant tissues. In addition, putrescine is considered one of the important bio-regulating growth substances affecting plant development processes under stress conditions (Galston *et al.*, 1997). The metabolic adjustment of putrescine is of adaptive significance by regulation of cellular ionic state, maintenance of membrane integrity, prevention of chlorophull loss

and cell wall stability (Evans and Malmberg, 1989) as well as shifting in hormonal balance by increasing endogenous phytohormones in plants subjected to stress (Bijay, 1999). Vitamins are among the organic or co-factor for many enzymes which the plants or all living organisms required for its. In legumes, vitamins stimulated root growth and so, they resulted higher water and nutrients uptake as well as provided larger surface area for rhizobium infection leading to increase root nodules formation. Amino acids as organic nitrogenous compounds stimulated cell growth acting as buffers maintaining favorable pH value within the plant cell as well as synthesizing other organic compounds, such as protein, amines, purines and pyrimidines, alkaloids, vitamins, enzymes, terpenoids and others (Goss, 1973). Applications of macro and micronutrients is crucial for achieving higher yields. N, P and K are essential nutrients and have prime importance in crop nutrition. Six micronutrients, i.e. Mn, Fe, Cu, Zn, B and Mo are involved in photosynthesis, N-fixation, respiration and other biochemical pathways (Marschner, 1986). Both macro and micronutrients availability is influenced by soil chemical and physical properties. Furthermore, sulphur is an

essential component of important metabolic and structural compounds. In calcareous soils, sulphur reduced soil pH values by oxidation of sulphur to sulphuric acid through species of soil microorganisms (El-Eweddy *et al.*, 2005). Decreasing soil pH improved the availability of microelements, i.e. Fe, Zn, Mn and Cu (Hetter, 1985) and improving chemical properties of alkaline soil as well as yield productivity and its related characteristics (Kineber *et al.*, 2004).

## 2. Materials and Methods:

Two field experiments were conducted during the two early summer seasons of 2006 and 2007 at El-Nubareia Horticulture Research Station (El-Behira governorate), Horticulture Research Center (A.R.C.), Giza, Egypt, to investigate the influence of soil amendment with humic acid and the foliar application with novavol, putrescine and vegimax on growth, yield and its components as well as chemical composition of snap bean plants.

The soil type under study was sandy loam, with the mechanical and chemical analysis as shown in the following Table (1) according to Jackson (1973).

**Table (1): Mechanical and chemical analysis of soil.**

Characters	Value	
	2006	2007
Particle size distribution	Sand%	68.08
	Silt%	16.00
	Clay%	15.92
	Textural class	Sandy loam
Soil chemical analysis:	pH (1:2.5)	8.50
	CaCO <sub>3</sub>	21.70
	EC mmohs/cm	1.27
	Ca <sup>++</sup>	3.38
	Mg <sup>++</sup>	3.62
Soluble cations meq/l:	Na <sup>+</sup>	3.23
	K <sup>+</sup>	0.49
	HCO <sub>3</sub> <sup>-</sup>	1.12
Soluble anions meq/l:	Cl <sup>-</sup>	1.5
	SO <sub>4</sub> <sup>-</sup>	9.1
	N	30.00
Macro-elements (ppm):	P	20.00
	K	368.00
	Zn	0.28
Micro-elements (ppm):	Mn	2.50
	Fe	3.70
	Cu	0.96
		1.00

Seeds of snap bean (*Phaseolus vulgaris*, L.) cv. Paulista were sown in 15<sup>th</sup> March in both seasons. The area of each experimental plot was 21m<sup>2</sup> and consisted of one row (35m long with 0.60m width), with distance of 25cm between plants.

Fertilization was carried out according to the recommendation of the Ministry of Agriculture as follow: 1. Organic manure in the form of poultry manure was added during soil preparation at the rate of 30 m<sup>3</sup>/feddan (Fed=4200 m<sup>2</sup>), and 2. Mineral fertilizers added in the timing and the rate as shown in Table (2).

**Table (2.) Program of snap bean fertilization under calcareous soil (Added per feddan/week).**

Timing (dayes after sowing)	Ammonium nitrate	Calcium nitrate	Potassium sulphate	Phosphoric acid
30	15 kg	-	10 kg	5 L
45	25 kg	5 kg	25 kg	5 L
60	15 kg	5 kg	20 kg	5 L
75	10 kg	5 kg	15 kg	5 L

Humic acid was added to snap bean plants at the rate 20g/l. Novavol was a commercial fertilizers containing 6.45% N, 5.17% P<sub>2</sub>O<sub>5</sub>, 2.00% CaO and 29.7% amino acids) at the rate of 2.5ml/l. Diamine putrescine (C<sub>4</sub>H<sub>12</sub>N<sub>2</sub>) was sprayed at the rate of 1.0mg/l. Vegimax was a commercial fertilizer containing mineral elements (0.05% Zn, 0.16% Fe, 0.08% Mn, 0.03% Cu, 0.04% B, 0.34% Mg, 0.46% Ca, 0.40% S and 0.005% Co), amino acids (mg/100ml) (aspartic acid, 249; therionine, 45; serine, 56; glutamic, 55; glycine, 50; alanine, 100; proline, 38; valine, 68; cystine, 44; methionine, 18; iso-lucine, 52; tyrosine, 38; phenylalanine, 32; histidine, 12; lycine, 40; arginine, 20 and tryptophane, 20) as well as vitamins (mg/100ml) (B<sub>1</sub>, 0.08; B<sub>2</sub>, 2.4; B<sub>6</sub>, 1.2; B<sub>12</sub>, 0.82; folic acid, 4.2; pantothenic acid, 0.53 and niacin, 1.14) at the rate of 0.2ml/l, in addition to distilled water was sprayed as a control treatment. Spraying was carried out twice applications at 40 and 60 days after sowing.

Each treatment had four replicates and arranged in the field using a randomized complete block design.

#### Data recorded:

A. Plant growth: random samples of four plants were taken at 50 and 70 days after sowing from each plot to measure plant growth parameters, i.e. plant height (cm), number of leaves and branches/plant, leaf area (cm<sup>2</sup>) as well as dry weight of shoot and roots (g).

B. Pod yield and its components: two harvesting samples were taken 75 and 80 days after sowing to determine yield characters, i.e. total green pod yield (ton/fed), number of pods/plant, weight of pods/plant, average pod weight (g), pod length (cm), pod diameter (mm), pod bending (%) and pod humidity%.

C. Chemical composition: the pigments concentrations, i.e. chlorophyll a, chlorophyll b, total chlorophyll and carotenoids of leaves and pods were determined according to Nornai (1982). Determination of N, P, K, Ca, Mg, Zn, Mn, Fe, Cu, Na and Cl concentrations were carried out on the ground dry materials of shoots and pods. Nitrogen

was determined using the modified “microkjeldahl” method as described by Jackson (1973). Phosphorus was colorimetrically estimated by using chlorostannous reduced molybdophosphoric blue colour method according to Jackson (1973). Potassium and sodium were determined using the flame photometer. Calcium, magnesium, zinc, manganese, iron and copper were determined using Atomic Absorption Spectrophotometer, D. P. 3300 Parken Elemer. Chloride concentration was analyzed by potentiometric titration with AgNO<sub>3</sub> of the aqueous extract as described by Johnson *et al.* (1958). Total protein in the pods were calculated by multiplying Nitrogen% in 6.25 as described by Stewart (1989). Nitrate concentration in the pods was colorematically measured by using salicylic acid reagent according to Cataldo *et al.* (1975). Fiber content in the pods was determined according to (A.O.A.C., 2000).

Total sugar, total free amino acids and the total soluble phenols concentrations were determined in ethanol extract of shoots and pods. Total sugar concentration was determined by using phenol-sulphuric acid method according to Dubois *et al.* (1956). Total free amino acids concentration was determined using ninhydrin reagent (Moore and Stein, 1954). Total soluble phenols concentration was determined by using the folin-ciocateu colorimetric method (Swain and Hillis, 1959). Free proline concentration was colorimetrically measured in shoots and pods using ninhydrin reagent according to Bates *et al.* (1973).

All data were processed by analysis of variance according to the method described by Steel and Torrie (1960) and the means were compared using the least significant difference test (L.S.D.) at 5% (Snedecor and Cochran, 1980).

### 3. Results and Discussion:

The present experiment was twice conducted under the same conditions during two successive seasons 2006 and 2007. Therefore, the tabulated and discussed data of the various determinations represent in combined analysis system for the two seasons.

#### V.1. Vegetative growth parameters

Data showed that addition of humic acid significantly increased all the studied growth characters, i.e. plant height, number of leaves and branches/plant, leaf area as well as dry weight of shoots and roots of snap bean plants in both samples except of plant height in the first sample comparing with control plants (Table 3).

Meanwhile, foliar application of novavol significantly increased all growth characters in both

samples except of plant height at the second sample. Also, vegimax treatment significantly increased all growth characters in both samples except of plant height and dry weight of shoots in the first sample as well as number of leaves/plant in the second sample as compared to control plants. While, putrescine did not show a clear trend for various studied growth characters.

**Table (3): Effect of soil amendments with humic acid and foliar application with novavol, putrescine and vegimax on growth characters of snap bean plants**

Treatments	Plant height (cm)		No. of leaves/plant		No. of branches/plant		Leaf area (cm <sup>2</sup> )		Dry weight of shoots (g)		Dry weight of roots (g)	
	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
Control	27.4	29.5	9.3	10.8	4.3	5.4	308.5	480.4	3.27	6.87	0.52	0.8
Humic acid	28.8	33.2	11.7	13.3	4.9	5.9	395.9	648.1	4.27	7.47	0.65	1.13
Novavol	29.1	30.9	11.7	13.2	4.7	6.4	525.7	634.7	4.99	7.87	0.64	0.97
Putrescine	28.0	30.9	9.3	12.2	4.2	5.3	354.4	494.6	3.95	8.09	0.55	0.97
Vegimax	27.8	31.4	10.8	11.8	4.9	5.8	501.6	634.0	4.05	8.87	0.62	0.98
L.S.D. at 0.05	1.57	1.51	1.38	1.49	0.25	0.31	37.24	39.78	0.35	0.39	0.08	0.11

The beneficial effects of humic acid on plant growth could be referred to its acting as source of plant growth hormones. Nardi et al. (1999) mentioned that humic had a gibberellins and auxin exhibiting higher amounts of phenolic compounds and considerable amount of acids. In Egypt, the soils tended to be alkaloidy, thus the quantity of available micronutrients to plants at any one time was not only affected by its binding chelating substances, but also by transformation carried out by micro-organisms (Stevenson, 1982). Therefore, the application of humic acid through the irrigation water might increase the soil organic matter which improved retention of nutrients also increased the soil microbial activity, which convert the nutrients from organic to mineralized form as reported by Stevenson (1994). In this respect, Kaya et al. (2005) studied the effect of

foliar of humic acid (2000 ml/ha) on common bean plants and found that it significantly increased plant height. El-Bassiony et al. (2010) showed that foliar application by humic acid (at 1, 2 or 3 g/l) significantly affected on all the vegetative growth parameters, i.e. plant height, number of leaves and branches as well as fresh and dry weight of whole snap bean plants (*Phaseolus vulgaris*, L.) cv. Paulesta grown under sandy soil conditions comparing with control plants.

The stimulative effects induced by putrescine application on vegetative growth characters could be explained by Popvic et al. (1979) and Besford et al. (1993), they found that putrescine inhibited stress-induced senescence of barley leaf through

stabilization of thylakoid membranes. They added that putrescine effectively protected D1 and D2 proteins, and cytochrome F of the thylakoid membranes as well as rubisco large subunit and chlorophyll from osmotically stressed leaf tissue. It is probably due its binding to thylakoid membranes, thus preventing them from lipid peroxidation and proteolytic action. Consequently, the anti-senescence properties of polyamines and their correlation to cell proliferation and differentiation tended to support their action as growth factors, especially under stress conditions. In several dicotyledonous plants, putrescine retarded senescence of leaves by preventing chlorophyll, protein and RNA breakdown (Kaur- Sawhney and Galston, 1979) and by increasing mitotic activity in protoplasts (Kaur-Sawhney et al., 1980). In this concern, Gharib and Hanafy Ahmed (2005) postulated that spraying pea plants with putrescine at 1 or 2 ppm significantly increased plant height and shoot dry weight.

On the contrary, Zeid (2004) reported that foliar application of putrescine at (10-2mM) did not affect growth characters, i.e. shoot length, leaf area, fresh and dry weight of leaves on bean plants grown under salinity stress conditions.

Concerning the effect of amino acids which it is the main component of novavol and vegimax, Bidwell (1980) stated that, the importance of amino acids came from their widely uses for the biosynthesis of a large variety of non-protein nitrogenous materials, i.e. pigment, vitamins, coenzymes, purine and pyrimidine bases. Amino

acids could directly or indirectly influence the physiological activities of plant growth and development, through their regulatory effects on production of gibberellins in plant tissues (Waller and Nowaki, 1978). In this concern, El-Ghamry et al. (2009) cleared that foliar application of amino acids significantly increased plant height as well as number of leaves and branches/plant of faba bean plants.

Regarding to the effects of vitamins as a main constituent of vegimax were discussed by Kodandaramaiah and Gopala Rao (1985) who stated that vitamins compounds act as co-enzymes in a number of enzyme systems and thus take part in the regulation of metabolism, participated in plant growth and development indirectly by enhancing the endogenous levels of various growth factors such as cytokinins and gibberellins. Treating legume crops with B-vitamins could increase Rhizobium infection leading to increase root nodules formation and this in turn manifested in better growth and higher productivity (Simiullah and Afridi, 1988). In this respect, El-Tohamy and El-Greadly (2007) indicated that foliar application by Vit. E (0.1 or 0.3 ml/l) significantly increased plant height, number of leaves as well as plant fresh weight of snap bean plants. Omaira Mohamed et al. (2009) found that spraying snap bean plants with Vit. B1 at 25 ppm significantly increased number of branches, dry weight of shoots and total dry weight in both seasons.

## V.2. Yield and its components

Data in Table 4 revealed that addition of humic acid significantly increased total pod yield, number of

pods/plant, pods weight /plant, average pod weight as well as pod diameter in both pickings of snap bean plants comparing with control plants.

Meanwhile, foliar application of novavol and vegimax significantly increased total pod yield, number of pods/plant, pods weight/plant, average pod weight as well as pod diameter in both pickings except of total pod yield with vegimax and pod diameter with novavol in the first picking as well as average pod weight in the first picking with both foliar treatments. Furthermore, putrescine treatment significantly increased pods weight/plant in both pickings, whereas total pod yield and average pod weight were significantly increased in the first picking as well as number of pods/plant and pods diameter in the second picking as compared with control plants. On the other hand, all studied applications did not affect on pod length in snap bean plants at both pickings, except of vegimax in the second picking which showed a significant increase as compared with control plants.

Humic acids are considered as a important source of organic matter and their effects on yield and its components could be through their enhancing effect on increase soil moisture holding capacity, improve soil texture as well as promote the uptake of nutrients leading to stimulation of plant growth (higher biomass production) and consequently on total pods yield and its components (Zhang et al., 2003). Schnitzer and Skinner (1962) stated that humic acids increased yield and its components by increasing the availability of nutrients through its chelating capacity with micronutrients.

**Table (4): Effect of soil amendments with humic acid and foliar application with novavol, putrescine and vegimax on yield and its componenets**

Treatments	Total pod yield (ton/fed)		No. of pods/plant		pods weight/plant (g)		Average pod weight (g)		Pod length (cm)		Pod diameter (mm)	
	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>
Control	0.635	0.737	9.7	11.1	22.69	26.31	2.30	2.34	12.0	11.9	6.9	7.0
Humic acid	1.219	1.100	15.3	13.5	42.16	39.28	2.80	2.93	11.9	12.1	7.1	7.4
Novavol	1.029	1.067	13.7	14.1	31.88	38.08	2.39	2.76	12.2	11.8	7.0	7.2
Putrescine	0.801	0.905	11.6	13.7	28.59	32.30	2.57	2.37	12.0	11.6	6.9	7.3
Vegimax	0.780	1.048	12.4	14.4	27.83	37.40	2.39	2.59	12.1	12.4	7.2	7.4
L.S.D. at 0.05	0.16	0.27	2.20	2.40	3.30	3.41	0.23	0.19	0.28	0.22	0.20	0.17

Additionally, humic acid could improve the chemical properties of soil by counteracting soil alkalinity (Ghabbour and Davies, 1998). Furthermore, humic acid could increase dry matter of foliage, lateral root growth and N uptake under saline irrigation water (Tattini et al., 1991). This might contribute to regulate the nutritional and the adaptability state of stressed plants (Jianguo et al.,

1998). In this respect, Kaya et al. (2005) mentioned that foliar application of common bean by humic acid at 2000 ml/ha significantly increased number of seeds/plant and seed weight/plant. Zaky et al. (2006) found that application of humic acids as foliar application (1 g/l), injection application (50 g/m<sup>3</sup>) through the irrigation water or their combined application gave a significant superior effect over

non-treated plant on number of pods/plant, total pod yield/plant and average pod fresh weight of common bean. El-Bassiony et al. (2010) stated that green pod yield of snap bean plants (*Phaseolus vulgaris*, L.) cv. Paulesta grown under sandy soil conditions significantly increased by increasing the spray of humic acid from 0 to 1 up to 2 g/l.

The effects of putrescine application on yield and its components could be explained by Galston (1983) who indicated that the diamine Put and PAs, Spd and Spm have been frequently described as endogenous plant growth regulators or intracellular second messengers mediating the effects of phytohormones. Therefore, PAs have been implicated to affect a variety of molecular and cellular functions (Tiburcio et al., 1993) thereby influencing various physiological and developmental processes (Galston and Kaur-Sawhney, 1990 and Rajam, 1993) including pollen fertility (Martin-Tanguy et al., 1982) and in flowering and fruit ripening (Kakkar and Rai, 1993). Kaur-Sawhney et al. (1990) found that putrescine seem to play a regulatory role in morphogenetic processes preceding fruit set; that is, in the formation of the flowers. Galston et al. (1994) proposed that increasing in putrescine immediately proceeded the activation of cell division in meristems passing from the vegetative to the floral stage. In addition, Crisosto et al. (1988) reported that putrescine was found to improve pears fruit set, delayed senescence of the ovules and enhanced pollen germination and fertilization by two days. Iannotta et al. (1996) revealed that putrescine foliar application (10-4 mM) on olive trees reduced ovary abortion and improved fruit set. In this concern, Gharib and Hanafy Ahmed (2005) recorded that foliar spraying of putrescine (1 or 2 ppm) significantly increased weight and number of pods/plant as well as total fresh yield/fed of pea plants.

Concerning the effects of amino acids as a main component of novavol and vegimax, Waller and Nowaki (1978) suggested that the pronounced effects of amino acids on plant development and yield might be due to the regulatory influence on enhancing

production of gibberellins in plant tissues. In this respect, Melouk (2007) showed that spraying Thompson seedless grapevine with phenylalanine at 400 ppm significantly increased the vine yield. Amira Abdul Qados (2009) revealed that pretreatment of wheat grains with 1.25 mM arginine significantly increased the most of yield components of wheat plant irrigated with different salinity levels (from 2000 up to 8000 ppm).

The effects of vitamins as a main constituent of vegimax were discussed by Alscher et al. (1997) who suggested that vitamins could represent a natural and safety antioxidants substances, having the ability to quench free radicals and thereby form a protective screen around plant cells and hence increasing plant resistance to stress. It was suggested that B-vitamins participated in plant growth and development indirectly by enhancing the endogenous levels of various growth factors such as cytokinins and gibberellins (Kodandaramaiah and Gopala Rao, 1985). In this respect, El-Tohamy and El-Greadly (2007) showed that foliar application of snap bean plants with vitamin E at 0.1 or 0.3 ml/l significantly increased number of pods and fresh weight of pods. Omaira Mohammed et al. (2009) mentioned that spraying snap bean plants with Vit. B1 at 25ppm increased number of pods/plant and total pod yield/fed in both seasons.

In addition, data in Table 5 recorded that all studied applications significantly decreased pods bending % and fiber % of snap bean plants at both pickings as compared to control plants. Moreover, addition of humic acid or vegimax treatments significantly decreased pods humidity percentage of snap bean plants in the first picking. However, vegimax significantly increased pods humidity percentage in the second picking comparing with control plants.

It was found that humic substances possess auxin-like activity, leading to increase water permeability through plant membranes and hence could indirectly increased the water holding capacity for harvested pods (Pascual et al., 1999).

**Table (5): Effect of soil amendments with humic acid and foliar application with novavol, putrescine and vegimax on bending pods%, humidity and fiber %**

Treatments	Bending pods%		Humidity in pods%		Fiber %	
	1 st	2 nd	1 st	2 nd	1 st	2 nd
Control	74.08	55.37	89.48	85.76	11.22	11.51
Humic acid	66.64	48.11	88.43	86.28	9.19	9.65
Novavol	63.59	50.29	89.40	86.07	9.74	10.13
Putrescine	64.49	52.11	89.14	85.92	10.10	10.33
Vegimax	66.80	51.63	88.80	86.57	9.94	10.35
L.S.D. at 0.05	3.07	2.99	0.63	0.68	0.30	0.28

The effect of putrescine on decreasing pod bending % might be due to their effect as endogenous plant growth regulators. Therefore, polyamins have been implicated to affect a variety of molecular and cellular functions inducing cell elongation (Tiburcio et al., 1993).

The effect of amino acids as a main component of novavol on pod bending % might be referred to their role as chelating agent for micronutrients. Some of these micronutrients could play a role in plant resistance by regulating the levels of auxin in plant tissues, thus affecting cell elongation and pod bending.

In this respect, Szot and Tys (1979) reported that an important problem in the cultivation of leguminous plants is the tendency of pods to convexing and bursting, leading to considerable seed losses. The susceptibility of pods to bursting is conditioned by their anatomic-morphological structure (Esau, 1973 and Hejnowicz, 1985). The main characteristic causing pod bursting is the structure of the endocarp cell wall, which consists of fiber layers with stringly thickened sclerenchymatic cells obliquely arranged to the fruit axis. As a result of various micro-fibril arrangement in cell wall, these layers of thick-walled parchment stratum shrink in various directions during drying and this causes pod cracking along the "abdominal" seam (at carpel fusion) and dorsal seam (the main vein/rib).

Other significant factors determining the pod strength are its moisture content (Kuzniar and Sosnowski, 2000) and shape (Szwed et al. 1999). Furthermore, Kuzniar and Sosnowski (2000) stated that the dependence of the susceptibility of bean pods to cracking on their moisture content resulted from the pericarp structure, which inner surface (endocarp) consists of two parts of thick-walled ligneous cells. As a result of different alignment of the micro-fibrils in the cell walls, the two parts of endocarp shrank in different directions as they dried out, thus creating internal forces tending to open the pod (Esau, 1973). Tomaszewska (1954) revealed that the lupine pods which shells have thicker endocarp and thinner mesocarp (parenchyma cells) exhibited higher tendency to cracking.

Moreover, Kuzniar and Sosnowski (2002) showed that an increase in the shape factor with a rising moisture content of pods was recorded for all studied bean cultivars. It means that internal stress on sclerenchyma, which increased with pod drying, tended to open the pod and increased the convexity of the shells of those fruits, and the more convex the pod the less force is required for its opening. The reduction in the shape factor, recorded for studied cultivars as they were drying, which means that

internal stress in fiber layer, which increased with pod drying and increased its convexity.

Szwed et al. (1999) suggested that the opening of more convex pods requires smaller force, because smaller curve radii increase the arm of the pod shell bending moment in the perpendicular plane to the direction of the parchment layer fibers.

The results of humic acid, novavol and vegimax on pod humidity % are in conformity with those obtained from Faten Abd El-Aal et al. (2005) revealed that application of potassium humate with irrigation water (6 L/fed) significantly increased onion bulb dry weight. Also, Salman et al. (2005) stated that applied humic acid with water irrigation (2, 4 or 6 L/fed) significantly increased dry matter of watermelon fruits.

Al-Said and Kamal (2008) who found that individually foliar application of folic acid (50 or 100 ppm), methionine (25 ppm), lysine (50 ppm) or cysteine (50 ppm) did not significantly affect sweet pepper fruit dry weight. However, the mixtures between them significantly increased fruit dry weight as compared to control plants. Dawa et al. (2003) reported that combination between Vit. B1 (50 ppm) and yeast extract (25 mg/l) gave the highest total dry seed of pea yield/fed. Amer (2004) mentioned that yeast extract (1 or 2 g/l) significantly increased common bean dry seed yield. Awad et al. (2007) showed that foliar application of glycine and lysine (100 ppm) either alone or in combination significantly increased dry matter content in potato plants comparing with control plants.

Concerning the effect of different treatments on decreasing fiber content in snap bean pods, these results are in harmony with those reported by El-Tohamy and El-Greadly (2007) who showed that foliar application by vitamin E (at 0.1 or 0.3 g/l) significantly decreased fiber content of snap bean pods. Furthermore, Neveen Turkey (2007) pointed out that spraying snap bean plants either with Ascopen (ascorbic acid plus citric acid 38%) at 2 g/l or Delfan (11% free amino acid) at 2 cm<sup>3</sup>/l significantly decreased fiber content of snap bean pods. Shereen El-Sayed (2007) stated that spraying snap bean plants either with Faster (contains 0.1% ascorbic acid, 1% citric acid, 30% K<sub>2</sub>O and 5% P<sub>2</sub>O<sub>5</sub>) at 250 cm<sup>3</sup>/100 l water or Setter-2 ( contains 5000 ppm ascorbic acid, 5000 ppm citric acid, 5000 ppm nitrogen, 1000 ppm copper, 90000 ppm chelated calcium, 15000 ppm chelated boron and 1000 ppm manganese) at 500 cm<sup>3</sup>/200 l water significantly decreased in snap bean pods. Moreover, El-Bassiony et al (2010) revealed that fiber content significantly decreased in snap bean pods with increasing the level of humic acid from 0 to 1, 2 up to 3 g/l.

### V.3. Chemical composition

#### V.3.1. Plant pigments

##### V.3.1.1. Leaves

Data in Table 6 indicated that addition of humic acid significantly increased chlorophyll a, total chlorophylls and carotenoids concentrations in leaves of snap bean plants of both samples except of chlorophyll a in the second sample comparing with control plants.

Meanwhile, foliar application of novavol and vegimax significantly increased chlorophyll a, total chlorophylls and carotenoids concentrations in leaves of both samples as compared to control plants. However, putrescine treatment significantly increased

chlorophyll a, total chlorophyll and carotenoids concentrations but only in the first sample as compared to control plants.

##### V.3.1.2. Pods

Data in Table 7 revealed that all studied applications significantly increased chlorophyll a, total chlorophyll and carotenoids concentrations in snap bean pods of both pickings comparing with control pods, except of chlorophyll a and total chlorophyll with putrescine in the second picking as well as carotenoids with novavol and putrescine in the first picking as compared with control plants.

**Table (6): Effect of soil amendments with humic acid and foliar application with novavol, putrescine and vegimax on leave pigments concentration (mg/g F.W.)**

Treatments	Chlorophyll a		Chlorophyll b		Total chlorophyll		Carotenoids	
	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
Control	0.871	0.962	0.199	0.451	1.070	1.413	0.617	0.927
Humic acid	1.080	1.129	0.245	0.453	1.325	1.581	0.843	1.028
Novavol	1.116	1.179	0.261	0.415	1.377	1.593	0.873	1.019
Putrescine	1.100	1.129	0.272	0.398	1.372	1.526	0.925	1.004
Vegimax	1.358	1.379	0.336	0.396	1.694	1.775	1.042	1.145
L.S.D. at 0.05	0.18	0.18	0.09	0.11	0.18	0.15	0.110	0.090

**Table (7): Effect of soil amendments with humic acid and foliar application with novavol, putrescine and vegimax on pod pigments concentration (mg/g F.W.)**

Treatments	Chlorophyll a		Chlorophyll b		Total chlorophyll		Carotenoids	
	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
Control	0.343	0.531	0.167	0.199	0.509	0.731	0.310	0.390
Humic acid	0.442	0.653	0.123	0.21	0.564	0.862	0.492	0.709
Novavol	0.502	0.738	0.157	0.210	0.659	0.947	0.462	0.805
Putrescine	0.434	0.541	0.131	0.203	0.564	0.744	0.437	0.753
Vegimax	0.539	0.686	0.232	0.259	0.77	0.945	0.591	0.803
L.S.D. at 0.05	0.06	0.08	0.07	0.04	0.06	0.09	0.16	0.12

The stimulatory effects of humic acid on increasing chlorophyll concentration in leaves and pods might be attributed to the lowering of pH value through yielding intermediate organic acids as well as increasing the activity of soil organisms to liberate more nutrients from the unavailable reserves, i.e. correcting iron deficiency of chlorotic sensitive to plant species when grown in calcareous soil (Wallace and Khadr, 1996). Moreover, Dekock (1955) reported that humic substances prevented immobilization of Fe and P and facilitated their translocation from roots to shoots. In addition, Schnitzer and Khan (1972) suggested that humic substances exert two types of effects in relation to plants, a) indirect effects through acting as suppliers and regulators of plant nutrients similar to synthetic ion exchangers and b) direct effects through uptake of humic substances by plant roots.

In this respect, Zaky et al. (2006) reported that application of humate acid either foliar (at 1g/l every seven days interval until the first harvest) or injection application (at 50 g/m<sup>3</sup> through the irrigation water seven days interval until the first harvest) gave a significant increase in total chlorophyll of pods of common bean plants (*Phaseolus vulgaris*, L.) cv. Xera. On the other hand, HA application and control treatment exhibited a similar effect on total protein and carbohydrate in pods under low tunnels and open field during both seasons. Habashy et al. (2008) revealed that applied of micronutrients as chelating compounds for humic acid significantly increased total chlorophyll of tomato plants grown under salinity conditions. El-Ghamry et al. (2009) cleared that foliar application by humic acid and amino acids either alone or in combination significantly increased



chlorophyll a, b and carotenoids concentrations of faba bean leaves.

The effects of putrescine are explained by Kaur-Sawhney and Galston (1991) who reported that polyamines retarded senescence, regulated cell proliferation and differentiation. Also, polyamines were important factor for growth regulation, protein biosynthesis as well as stabilizing chloroplasts thylakoid membranes and retarding chlorophyll degradation. In this respect, Zeid (2004) found that application of putrescine at 10-2mM markedly increased chlorophyll a, b and carotenoids content in bean leaves of stressed seedlings.

Concerning the stimulative effects of novavol and vegimax on plant pigments might be due to introducing amino acids into biosynthesis of large group of nonproteinic nitrogenous compounds, i.e. pigments, vitamins, Co-enzymes, purine and pyrimidine bases (Strove, 1989).

The enhancing effect of vitamins as a main constituent of vagimax on plant pigments might be attributed to their role in improve mineral uptake by plant shoots and hence improved plant growth, accordingly this was reflected on plant productivity and pod quality. In this respect, Hess (1993) stated that vitamin E played unique role as an antioxidant and a stabilizer for biological membranes as well as protects chlorophyll. El-Tohamy and El-Greadly (2007) indicated that foliar application with vitamin E (0.1 or 0.3ml/l) significantly increased total chlorophyll content of snap bean leaves. Sharaf et al. (2009) noticed that contents of chlorophyll a, b, and total chlorophyll of broad bean and lupin leaves were significantly increased in response to the application of vit. B (75ppm).

### V.3.2. Macro-elements

#### V.3.2.1. Shoots

Data in Table 8 recorded that all treatments did not show any significant differences among nitrogen concentration in snap bean shoots of both samples except of vegimax treatment which showed a significant increase in the first sample. On the other hand, phosphorus and calcium concentrations increased only in the first sample by all applications comparing with the control.

Meanwhile, potassium concentration was increased in both samples by addition novavol or vegimax as compared to control plants. However, humic acid and putrescine increased K concentration in leaves of second and first sample, respectively as compared to control plants. In addition, all studied treatments significantly increased magnesium concentration in the second sample as well as with novavol in the first sample as compared with control untreated plants.

#### V.3.2.2. Pods

Data in Table 9 indicated that humic acid, novavol and vegimax applications significantly increased nitrogen, potassium and magnesium concentrations in both pickings of snap bean pods except of humic acid on magnesium in the first picking as well as novavol on potassium in the second picking comparing with control plants.

The stimulative effect of humic acid on minerals uptake might be due to their effect on stability of membrane permeability (Zientara, 1983), and this is related to the surface activity of humic substances containing both hydrophilic and hydrophobic sites (Chen and Schnitzer, 1978). Moreover, humic acid appeared to be associated with its chelating power of nutrients along with its impact on physicochemical and biological properties of soil.

In addition, humic acid as a good manure state causing more availability for the nutrients in the soil (Taha and Modaihsh, 2003 and Modaihsh et al., 2005) by lowering soil pH value through yielding intermediate organic acid as well as increasing the activity of soil organisms to liberate more nutrients from the unavailable reserves.

In related to this results, Sharif (2002) revealed that addition of humic acid (0.5 or 1 kg/ha) significantly improved N and P concentrations in wheat and maize plants grown under alkaline soil.

Akinci et al. (2009) showed that addition of humic acid to broad bean plants significantly increased K content, whereas Ca content was not significantly increased. In addition, El-Ghamry et al. (2009) found significant increases of N, P and K content in seed and straw of faba bean plants as response to humic acid added (at 1000, 2000 or 3000 ppm). Katkat et al. (2009) reported that soil application with humus (1 g/kg) limited the decrease in negative effects of lime levels on mineral elements uptake, i.e. N, P, K, Ca and Mg of wheat plants grown under calcareous soil.

The results of putrescine on macro-elements concentrations in snap bean shoots are in harmony with those obtained from Manoj-Sharma et al. (1997) working on chickpea, as well as Gharib and Hanafy Ahmed (2005) working on pea plants. In this concern, El-Tohamy et al. (2008) showed that foliar application with putrescine (at 25 or 50 ppm) significantly increased N, P and K contents in eggplant leaves.

The effect of amino acids and vitamins as the main components of novavol and vegimax might be due to their roles in enhancing many physiological processes including nutrients uptake by roots and their metabolism in treated plants. In this respect, El-Tohamy et al. (2008) reported that foliar application

with vitamin C (at 100 or 200ppm) increased the concentration of N, P and K in eggplant leaves. The influence of riboflavin (vit. B2) on the mechanism of ions uptake might be related to their effect on membrane permeability and thus on ion entry through the membrane. On the other hand, increasing of Ca could maintaining the turgor of tissues under stress conditions, thus ameliorating the deleterious effects of stress on growth and yield (Sivritepe et al., 2003).

Furthermore, Awad et al. (2007) revealed that foliar application with mixture of glycine plus lysine each at 100 ppm gave the highest values of N, P, and K contents in leaves of potato plants. In this concern, El-Ghamry et al. (2009) stated that foliar application by peptone (6% free amino acid + 12% organic nitrogen + 3.5% K<sub>2</sub>O) at the rate of 1000, 2000 or 3000 ppm significantly increased N, P and K contents in seeds and straw of faba bean.

**Table (8): Effect of soil amendments with humic acid and foliar application with novavol, putrescine and vegimax on macro-elements (N, P, K, Ca and Mg) concentrations (mg/g D.W.) in shoots**

Treatments	Nitrogen		Phosphorus		Potassium		Calcium		Magnesium	
	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
Control	14.88	20.00	3.12	3.46	12.58	12.48	8.63	16.63	9.93	8.58
Humic acid	14.63	20.00	5.28	2.94	13.83	17.36	13.38	14.38	10.15	13.02
Novavol	14.13	20.38	5.57	3.56	25.72	14.49	12.75	16.00	11.80	13.62
Putrescine	15.88	19.00	4.55	2.39	14.31	12.48	13.00	17.00	10.68	12.63
Vegimax	22.00	21.00	6.16	5.01	19.01	17.70	13.13	16.13	10.68	13.55
L.S.D. at 0.05	1.73	1.50	1.34	0.99	1.53	1.45	1.20	0.96	1.36	1.32

**Table (9): Effect of soil amendments with humic acid and foliar application with novavol, putrescine and vegimax on macro-elements (N, P, K, Ca and Mg) concentrations (mg/g D.W.) in pods**

Treatments	Nitrogen		Phosphorus		Potassium		Calcium		Magnesium	
	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
Control	17.75	17.25	6.57	7.64	19.23	17.29	5.50	3.75	8.80	8.95
Humic acid	19.75	18.88	6.75	8.17	22.04	18.94	5.13	3.63	9.78	11.50
Novavol	20.50	19.88	7.18	7.51	22.49	18.24	4.63	3.63	11.13	11.05
Putrescine	18.63	17.88	5.43	7.63	20.72	17.86	5.00	4.25	9.18	9.70
Vegimax	20.38	19.75	5.70	7.68	23.02	19.00	4.50	3.75	11.28	13.02
L.S.D. at 0.05	1.47	1.44	1.23	1.24	1.15	1.20	1.11	1.31	1.35	1.47

### V.3.3. Micro-elements

#### V.3.3.1. Shoots

Data in Table 10 revealed that all compounds used in this study significantly increased zinc concentration in both samples in shoots of snap bean plants except of vegimax in the second sample. Meanwhile, manganese concentration was significantly increased only in the first sample by putrescine and vegimax treatment as compared with control plants.

Moreover, iron concentration was significantly increased in both samples with novavol or vegimax treatments as well as humic acid at the second sample of snap bean shoots. In addition, copper concentration was significantly increased only in leaves of the second sample by humic acid treatment comparing with control plants.

On the other hand, addition of humic acid, putrescine or vegimax significantly decreased sodium concentration in shoots of the first sample. However, chloride concentration was not affected in both

samples by any studied applications as compared to control plants.

#### V.3.3.2. Pods

Data in Table 11 showed that addition of humic acid significantly increased iron and copper concentrations in the pods of both pickings except of copper in the second picking. Meanwhile, foliar application of novavol, putrescine or vegimax significantly increased zinc concentration in pods of both pickings except of vegimax in the second picking. Manganese and iron concentrations in pods were significantly increased in both pickings by spraying the plants with novavol or vegimax except of manganese in the second picking. Moreover, it was found that novavol or putrescine significantly increased copper concentration in pods but only in the first picking. In addition, all studied applications significantly decreased chloride concentration in pods but only in the first picking as compared with control plants.

**Table (10): Effect of soil amendments with humic acid and foliar application with novavol, putrescine and vegimax on micro-elements (Zn, Mn, Fe and Cu) concentrations (ppm) as well as ionic salts (Na<sup>+</sup> and Cl<sup>-</sup>) concentrations (mg/g D.W.) of snap bean shoots**

Treatments	Zinc		Manganese		Iron		Copper		Sodium		Chloride	
	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
Control	63.75	75.00	89.07	103.14	68.75	68.75	23.75	53.45	0.74	0.53	3.57	4.12
Humic acid	75.00	86.25	80.63	98.43	70.00	86.25	16.25	57.22	0.62	0.50	3.48	4.03
Novavol	72.50	93.75	91.88	98.47	81.25	93.75	18.75	51.59	0.70	0.56	3.66	3.39
Putrescine	73.75	93.75	94.69	90.94	70.00	63.75	25.00	47.84	0.63	0.47	3.57	4.21
Vegimax	85.00	80.00	98.43	103.47	87.50	93.75	15.00	49.72	0.62	0.56	3.66	4.12
L.S.D. at 0.05	6.30	6.02	5.59	6.06	3.02	1.52	3.08	2.78	0.09	0.07	0.21	0.19

**Table (11): Effect of soil amendments with humic acid and foliar application with novavol, putrescine and vegimax on micro-elements (Zn, Mn, Fe and Cu) concentrations (ppm) as well as ionic salts (Na<sup>+</sup> and Cl<sup>-</sup>) concentrations (mg/g D.W.) of snap bean pods**

Treatments	Zinc		Manganese		Iron		Copper		Sodium		Chloride	
	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
Control	60.00	63.75	63.76	60.94	65.00	68.75	31.25	32.50	0.69	0.59	2.84	2.38
Humic acid	50.00	67.50	65.63	61.88	82.50	77.50	35.00	23.75	0.66	0.54	2.01	2.19
Novavol	76.25	72.50	73.13	57.82	75.00	77.50	36.25	25.00	0.68	0.58	2.29	2.38
Putrescine	67.50	73.75	61.88	56.25	63.75	62.50	36.25	33.75	0.66	0.55	2.20	2.20
Vegimax	66.25	62.50	84.38	64.69	75.00	82.50	26.25	26.25	0.64	0.60	2.01	2.38
L.S.D. at 0.05	5.31	5.09	5.33	5.43	3.32	2.93	3.36	3.39	0.07	0.06	0.25	0.24

The stimulative effect of humic acid on micronutrients concentrations might be explained by David et al. (1994) who indicated that humic acid enhanced cell permeability, which in turn made more rapid entry of minerals into root cells and so resulted in higher uptake of plant nutrients. In addition, the application of soil humus has a number of potential benefits for plants, i.e. increased water and nutritional capacity, increased reserve of slow release nutrients, enhanced solubility of zinc, iron, manganese and copper as well as increased resistance to soil pH change.

In addition, humic acid is the most significant component of organic substances (Mecan and Petrovic, 1995), so, their application particularly as soil treatment, effectively minimized the negative effects of salinity. It also found that humic acid substances promoted lateral growth and N uptake rate (Tattini et al., 1991).

According to that, the effects of humic acid on increasing Fe and Zn concentrations in the shoots might be due to their effect on the reduction of Fe<sup>3+</sup> to Fe<sup>2+</sup>, making iron chelates are readily available to the plants (Gregor and PowerII, 1988). Moreover, humic acid might prevent the formation of insoluble complexes of zinc and facilitated their uptake by plants. The obtained results of humic acid on micro elements concentrations of snap bean pods were in conformity with those obtained by Katkat et al. (2009) working on wheat plants grown under calcareous soil, who found that addition of humic acid (1 g/kg) increased Fe, Cu and Mn uptake. Taha et al. (2006) reported that humic acid significantly

increased N, P, K, Fe, Mn, Zn and Cu uptake by wheat plants grown under calcareous soil conditions. Moreover, Faten Abd El-Aal et al. (2005) mentioned that humic acid (12 l/fed) increased the contents of Fe, Mn, Cu and Zn in onion bulbs. Furthermore, El-Naggar and El-Ghamry (2007) showed that foliar application of humic acid (1500 ppm) increased Fe, Zn, Mn and Cu concentrations in wheat grains and straw. Asik et al. (2009) indicated that addition of humus at 1 or 2 g/kg enhanced wheat uptake of N, P, K, Ca, Mg, Na, Fe, Cu, Mn and Cu, especially under 60 mM NaCl treatment.

The effects of humic acid on reducing the concentrations of Na and Cl in pods of snap bean plants might be due to their benefit effects on soil (Tan, 1998; Nardi et al., 2002; Cimrin and Yilmaz, 2005; Sangeetha et al., 2006), which might reflect on nutrient uptake then on plant growth and productivity (Chen and Aviad, 1990).

The effects of novavol and vegimax treatments as a source of amino acids on micro elements of snap bean plants might be due to their enhancing effect on hormones and vitamins produced by plants, their regulatory roles in plant development (Waller and Nowaki, 1978) as well as synthesis of alkaloids and various secondary metabolites which could affect pod quality, which indirectly improve minerals compositions of the shoots.

The effects of putrescine on decreasing Na and Cl concentrations in pods of snap bean plants could reflect better selectivity under salt and osmotic stress (Erdei et al., 1996), and better adaptability to salt stress. In this respect, Hanafy Ahmed et al. (2002)

stated that putrescine foliar application reduced Na value in stressed *Myrtus communis* plants. Again, putrescine effect could be explained by its ability to stabilize membrane and maintaining cation-anion balance in plant tissues.

The effects of amino acids on reducing concentrations of Na and Cl in pods of snap bean plants might be due to its role as components of salt tolerance mechanism build up a favorable osmotic potential materials inside the cell. In this concern, Amira Abdul Qados (2009) reported that spraying arginine (1.25 mM) decreased Na and Cl concentrations in wheat grains obtained from plants irrigated with salinized water (2000-8000 ppm).

The effect of vitamins as a main component of vegimax on microelements concentration might be referred to their ability to quench free radicals and thereby form a protective screen around plant cells and hence increasing plant resistance against the deleterious effects of abiotic stresses (Alscher et al., 1997). The resistance to abiotic stress might depend partially at least on the inhibition of ROS (reactive oxygen species) production and/or the enhancement of antioxidant level as well as the osmotic adjustment mechanism during plant development. So, the strategy of osmotic adjustment in salinized seedlings treated with vitamins might be mediated by accumulation of some compatible solutes (amino acids, sugars or proline) acting as osmolytes.

### V.3.4. Organic compounds

#### V.3.4.1. Shoots

A studied application significantly increased total sugar and total free amino acids concentrations in shoots of snap bean plants during both samples except of total free amino acids with humic acid and putrescine in the second sample as well as total sugar in the second sample with putrescine comparing with control plants (Table 12). As well as, significantly increased total soluble phenols and proline concentrations in snap bean shoots of both samples except of proline concentration in the second sample with novavol or vegimax treatments comparing with control plants (Table 12).

#### V.3.4.2. Pods

Data in Table 13 showed that all applications significantly increased total sugars, total free amino acids, total soluble phenols and protein concentrations in snap bean pods during both pickings except of humic acid with total sugar in the second picking, putrescine and vegimax with total soluble phenols in the first picking as well as putrescine with protein in the first picking comparing with control plants. Moreover, application of putrescine in the first picking as well as vegimax in the second picking significantly increased proline concentration in snap bean pods comparing with control plants.

**Table (12): Effect of soil amendments with humic acid and foliar application with novavol, putrescine and vegimax on total sugars, total free amino acids and total soluble phenols concentrations (mg/g F.W.) as well as proline concentration (mg/g D.W.) of snap bean shoots**

Treatments	Total sugars		Total free amino acids		Total soluble phenoles		Proline	
	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
Control	3.99	2.96	3.61	3.66	0.43	0.51	0.80	0.59
Humic acid	4.19	3.49	4.51	3.87	0.60	0.59	1.07	0.74
Novavol	4.77	3.41	4.43	4.85	0.58	0.59	0.95	0.59
Putrescine	4.21	3.04	4.70	3.97	0.64	0.63	0.94	0.81
Vegimax	4.68	4.35	4.86	4.52	0.59	0.64	0.97	0.52
L.S.D. at 0.05	0.16	0.17	0.60	0.52	0.11	0.07	0.10	0.10

**Table (13): Effect of soil amendments with humic acid and foliar application with novavol, putrescine and vegimax on total sugars, total free amino acids and total soluble phenols concentrations (mg/g F.W.) as well as proline (mg/g D.W.), protein (%) and nitrate (mg/g D.W.) concentrations of snap bean pods**

Treatments	Total sugars		Total free amino acids		Total soluble phenoles		Proline		Protein		Nitrate	
	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
Control	7.19	5.79	3.86	3.62	0.12	0.10	0.40	0.24	11.10	10.10	7.04	3.36
Humic acid	9.46	5.88	5.67	5.31	0.18	0.17	0.42	0.26	12.35	11.80	6.12	2.40
Novavol	8.63	6.62	5.16	5.00	0.24	0.13	0.40	0.24	12.82	12.43	6.72	2.67
Putrescine	8.80	6.55	4.66	4.67	0.14	0.13	0.46	0.26	11.64	11.17	5.98	1.94
Vegimax	8.18	7.07	5.49	5.09	0.15	0.18	0.40	0.30	12.74	12.35	6.62	3.13
L.S.D. at 0.05	0.37	0.28	0.72	0.62	0.05	0.02	0.05	0.05	0.57	0.57	0.92	0.88

The effects of humic acid on increasing some organic compounds, i.e. total sugars, total free amino acids, total soluble phenols and proline in snap bean plants might be directly related to their role in increase plant tolerance against a biotic stress or indirectly through improvement of soil properties such as aggregation, aeration, permeability, water holding capacity, and hence increased the activity of immune plant system, i.e. total soluble phenols against stress conditions. Adani et al. (1998) stated that the use of humic substances has often been proposed as a method to improve crop production as well as soil amendment for plant protection against various a biotic stresses. Moreover, Kulikova et al. (2005) and Xudan (1986) pointed out that humic substances might show anti-stress effects under a biotic stress conditions. In addition, Randhawa and Broadbent (1965) reported that humic acid produced ligands capable of complexing nutrient elements and these complexes elements remained more available to plants roots as complexation shields them against immobilization in soil. Inhibition of urease activity by humic acid (Vaughan and Ord, 1991), led to reduce loss of N by volatilization, as described by Flaig (1984) thus could increase availability of nitrogen to plants, leading to the enhancement of free amino acids and protein synthesis (Guminiski, 1968).

Similarly, Sahar Zagloul et al. (2009) revealed that the highest shoots sugar content of *Thuya orientalis*, L. was obtained from plants treated with 2.0 or 2.5% potassium humate. Also, Chen and Aviad (1990) added that humic acid correcting plant chlorosis and thus enhanced photosynthetic potential as well as increasing total sugar content in plants. Omar and Abd El-Aal (2005) revealed that addition of humic acid (6, 9 or 12 l/fed) significantly decreased proline concentration shoots of superior seedless vines grown under saline conditions.

The results of putrescine on some organic compounds, i.e. total sugars, total free amino acids, total soluble phenols and proline in snap bean pods were similar to those obtained by Hanafy Ahmed et al. (2002) working on *Myrtus communis* plants found that putrescine (0.1 ppm) enhanced amino acids, especially proline concentration in both shoots and roots. They suggested that putrescine is involved in ionic balance and DNA, RNA and protein stabilization, hence leading to the enhancement of free amino acids and protein synthesis. Zeid (2004) mentioned that exogenous putrescine treatment (10-2 mM) significantly increased proline content of bean seedlings grown under salinity stress comparing with control plants. Gharib and Hanafy Ahmed (2005) stated that putrescine foliar application (1 or 2 ppm) increased total soluble phenols of pea shoots. Moreover, Neveen (2003) mentioned that putrescine

foliar application 1 and 2 ppm enhanced total soluble phenols in sweet pepper shoots.

The effects of novavol and vegimax on increasing the organic compounds, i.e. total sugars, total free amino acids, total soluble phenols and proline in snap bean pods were agreement with those obtained by Winicov (1998) demonstrated that the relationship between proline biosynthesis and increasing in concentrations of related metabolites such as polyamines, ammonia, arginine, ornithine, glutamine and glutamate. Thus, the increase in proline concentration might be related to the applied foliar amino acids, leading to proline accumulation in plant tissues exposed to environmental stress (Silveira et al., 2002). In this respect, Azooz (2009) showed that foliar application of riboflavin (100 ppm) exhibited stimulatory effect on the accumulation of proline in *Hibiscus sabdariffa* plants grown under salinity stress.

Nahed Abd El-Aziz et al. (2006) reported that foliar application of ascorbic acid (200 or 400 ppm) significantly increased total sugar percentage of *Khaya senegalensis* under salinized water irrigation plant. Moreover, Nahed Abd El-Aziz and Laila Balbaa (2007) showed that application of tyrosine (100 ppm) significantly increased sugar and free amino acids content in cuttings of *Salvia farinacea*. Furthermore, Al-Said and Kamal (2008) mentioned that foliar application of methionine, lysine and cysteine mixture at 25, 50 and 50 ppm, respectively resulted in the highest significant total sugar in pepper leaves comparing with control (tap water).

Emam and Helal (2008) revealed that foliar application of folic acid (20  $\mu\text{M}$ ), ascorbic acid (0.5  $\mu\text{M}$ ) or coblamin (2  $\mu\text{M}$ ) accumulated total carbohydrate and free amino acids contents at the expense of the soluble one in flax plants grown under salt conditions. Whereas, coblamine treatment (2  $\mu\text{M}$ ) resulted a pronounced increase in proline content of salinized flax seedlings. Nahed Abd El-Aziz et al. (2009b) noticed that foliar application of phenylalanine or tryptophan (50 or 100 ppm) significantly increased total sugar and total free amino acids contents in *Antirrhinum majus* plants.

El-Desuki and Nadia El-Greadly (2006) stated that foliar application with yeast extract increased the concentration of free amino acid in pea plants comparing with control plants. Karima Gamal El-Din and Abd El-Wahed (2005), stated that both foliar application of proline and phenylalanine at 150 mg/l significantly increased total phenol contents in vegetative parts of chamomile plants. Furthermore, Nahed Abd El-Aziz et al. (2009a) noticed that foliar application of ascorbic acid or thiamine, each at 50, 100 or 200 ppm increased total phenol contents in gladiolus plants. Moreover, Azooz (2009) showed

that foliar application of riboflavin (100 ppm) accumulated proline in *Hibiscus sabdariffa* plants grown under salinity stress.

### V.3.5. Protein

Protein was significantly increased concentration in snap bean pods during both pickings except of putrescine in the first picking comparing with control plants (Table 13). Similarly results obtained using humic acid treatment enhancing protein concentration in pods of snap bean plants (Zaky et al., 2006). Moreover, Atak et al. (2005) found that, humic acids as foliar treatment significantly increased yield and protein contents of common bean.

The favorable effects of humic acid on increasing protein concentration in pods might be due to their effect on improving soil nitrogen uptake and encourage potassium, calcium, magnesium and phosphorus availability to plant root system (Singer et al., 1998; Piccolo et al., 1997; Pascual et al., 1999). Furthermore, McDonnell et al. (2001) reported that humic substances represented an important soil component because they constituted a stable fraction of carbon and improved water holding capacity, pH buffering and thermal insulation.

The favorable effects of putrescine on increasing protein concentration in pods of snap bean plants was similar to that obtained from Gharib and Hanafy Ahmed (2005) working on pea plants.

Improving total protein in seeds by putrescine application might be due to the role of putrescine on improving plant growth, photosynthetic pigments content and endogenous phytohormones especially under stress. In this regard, Sawsan-Suleiman and Grieve (2002) and Zeid (2004) mentioned that putrescine application resulted an increase in the biosynthesis of nucleic acids, synthesis of macromolecules particularly protein and photosynthesis pigments.

Concerning the effects of amino acids and vitamins on increasing protein concentration in pods of snap bean plants were found to be similar to that obtained by El-Desuki and Nadia El-Geready (2006) on pea. In this respect, El-Tohamy and El-Geready (2007) stated that foliar application with vitamin E (0.1 or 0.3 m/l) or yeast extract (5 or 10 g/l) significantly improved protein of snap bean pods comparing with control plants.

### V.3.6. Nitrate

The addition of humic acid or putrescine treatment showed significantly decreased nitrate concentration in snap bean pods of both pickings. On the other hand, novavol and vegimax treatments did not show any obvious trend for nitrate concentration

in pods of both pickings comparing with control plants (Table 13).

The reduction effect of humic acid on nitrate concentration of pods are in agreement with those obtained by Hassanpanah et al. (2007) who mentioned that potassium humates decreased nitrate accumulation in potato tubers. Humic acid might activated nitrate reductase leading to reducing nitrate concentration in the pods (Gadimov et al., 2007). On the contrary, Awad and El-Ghamry (2007) reported that addition of humic acid (100 ml/plant) significantly increased nitrate concentration in potato tubers of both seasons.

The results of putrescine on reducing nitrate concentration in pods are in harmony with those obtained by Zeid (2004) who found that nitrate reductase activity increased in roots of bean plants under 10-2 mM putrescine. Moreover, Slocum and Flores (1991) and Cohen (1998) showed the diverse roles of polyamines in plants which include storing excess N and reducing  $\text{NH}_4/\text{NO}_3$  toxicity.

In this respect, it can be suggested that, the reduction in nitrate concentration might be due to increasing the level of organic compounds, i.e. total sugar, total free amino acids, total soluble phenols and proline. In this respect, Seginer et al. (1998) working on lettuce plants concluded that there is a negative correlation between the concentrations of soluble carbohydrates and nitrate in the cell sap. This correlation reflects the equivalent roles of nitrate and organic solutes in the maintenance of cell turgor. In addition, Kirkby (1981) mentioned that plants contain adequate concentrations of sugars are able to assimilate nitrate at a faster rate than comparable plants containing lower concentrations of sugars. Moreover, Aslam and Huffaker (1984) reported that the rate of nitrate in the primary leaves of barley was much more affected by the concentration of sugars than the rate of nitrate uptake into the plants. Moreover, Hanafy Ahmed (1996) suggested that increasing sugars and free amino acids concentrations under high supply of calcium (as calcium chloride) may replace nitrate in the vacuole of lettuce cell. In this connection, Blom-Zandstra and Lampe (1985) and Blom-Zandstra et al. (1988) pointed out that  $\text{NO}_3$  accumulation was inversely related to accumulation of sugars and organic acids. They also mentioned that the availability of sugars might affect the need for nitrate as an osmoticum.

### Corresponding author

Sallam, H. El-S

Vegetable Research Dep., Horticulture Research Institute, Giza, Egypt

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