

## Application of Bio-Stimulants in Comparison with Organic and Mineral N Fertilizers for Growth Promotion of Spinach and Common Bean Grown on Sandy Soil

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### ABSTRACT

Spinach and common bean plants as representatives of leafy and legume vegetables were subjected to some biostimulants to evaluate its effects, against organic and mineral fertilizers, on plant growth, nutrient uptake and nitrate accumulation under field conditions. Individual treatments of humic acid (HS) and seaweed extract (SW) as bio-stimulants were tested in comparison with organic compost and ammonium nitrate as mineral fertilizer. Fresh and dry weight of spinach and common bean plants were significantly positively affected by application of seaweed extract and humic acid, respectively. Incorporation of organic compost resulted in fresh and dry biomass yields of spinach leaves, common bean green vegetative part and pods nearly closed or slightly lower than those recorded with ammonium nitrate fertilizer. In most cases, seaweed extract was superior over other treatments. Both of humic acid and seaweed extract relatively increased fresh or dry weights of both crops by about 50% in average over the plants fertilized with ammonium nitrate or treated with organic compost. Application of seaweed extract and humic acid had reduced the nitrate content in spinach leaves as compared to plants treated with ammonium nitrate but nearly closed to those of organic compost. Either organic compost or biostimulants (SW and HS) produced edible leaves of spinach with lower nitrate content that makes it marketable according to permissible level organized by European Commission Regulation (3500 mg kg<sup>-1</sup> FW). Nitrogen, phosphorus and potassium uptake by leaves of spinach and pods of common bean were positively significantly increased with application of all tested treatments in comparison to the untreated control, but seaweed extracts and to somewhat extent the humic acid resulted in NPK values more or nearly closed to those recorded with either mineral fertilizer or organic compost additive. These results gave the chance to accept the biostimulants in addition to organic additive as alternative and sustainable tools complementing a high yield with premium quality.

**Keywords:** Biostimulants, Common bean, Mineral fertilizer, Organic compost, Spinach

### INTRODUCTION

Common bean (*Phaseolus vulgaris* L.) is one of the cash crops for many small farmers in Egypt. This crop is either consumed locally or exported in winter seasons to Arab or European countries. This vegetable crop can be used as fresh pods or as- a pulse crop for dry seed. About 16.7 million tons of dry beans were produced from about 23 million ha in addition to 4.7 million tons of green beans (FAOSTAT, 2001).

In Egypt, common bean is an important either as vegetable or dry seeds. Nine to eighteen thousand hectares were annually cultivated, yielding 15,000 tons of dry seeds and 150,000 tons of green pods (Moussa, 2016). These legume vegetable crops are a good source of protein, vitamins, dietary fiber and minerals (El-Siyad and Hassan 2014). Similarly, spinach is considered as an extremely nutritious vegetable, rich both in core nutrients and phytochemicals. In the same time, spinach has strong antioxidant activity and high levels of antioxidant compounds such as phenolics and carotenoids. These components are very important in medicine and pharmaceutical industries (Hedges and Lister 2007).

The bio-stimulants were defined as substance or microorganism applied to plants in order to improve nutrition efficiency, abiotic stress tolerance and/or crop quality traits, regardless of its nutrients content (Patrick, 2015).

As reviewed by Kocira *et al.*, (2018), bio-stimulants may be of natural or synthetic origin and contain both organic and inorganic compounds. Natural bio-stimulants are based on free amino acids, humic compounds, fruit or seaweed extracts, chitin and its derivative-chitosan, or effective microorganisms. On the other hand, synthetic bio-stimulants contains growth regulators, phenolic compounds, inorganic salts and

beneficial nutrients such as Al, Co, Na, Se, Ti, Si (Calvo *et al.*, 2014; Przybysz *et al.*, 2014; Du Jardin, 2015).

In addition, bio-stimulants seem to be more effective with plants that are particularly sensitive to adverse climatic conditions and are economically important. In this regard, biostimulants seems to be effective in regulation/modification of physiological processes in plants to stimulate growth, to mitigate stress-induced limitations, and to increase yield (Yakhin *et al.*, 2017). Furthermore, organic fertilizers could be applied to avoid the toxic compounds effect such as nitrates produced by mineral fertilizers in vegetables (Masarirambi *et al.*, 2012). In the same way, lettuce grown on soil amended with organic manure combined with single super phosphate (SSP) and rock phosphate (RP) at 8 ton ha<sup>-1</sup> and 4 ton ha<sup>-1</sup> respectively exhibited significant higher yield (Sanni and Ewulo, 2015).

Considering the variable biostimulant actions depending on tested crop, two field experiments were carried out to evaluate the role of two biostimulants, i.e. humic acid and seaweed extract delivered by foliar application in addition to organic compost against the ammonium nitrate as mineral-N fertilizer on spinach (leafy) crop and common bean (vegetable) crop. The response of these crops was assessed in terms of yield, leaf nitrate content, macronutrients composition of the leaves and pods.

### MATERIALS AND METHODS

Two field experiments were carried out through the years 2016-2017 on sand soil located in Sharqia Governorate with altitudes 30°7'51" and 30°14'57.33" N, and longitudes 31°17'41.54" and 31°21'11.19" E. Common Bean (*Phaseolus vulgaris* L.) cv. Nebraska and Spinach (*Spinacia Oleracea*) cv. Balady, were used as a test crops to recognize the benefits gained from bio-stimulants,

organic and / or chemical N fertilizers. Experimental soil was subjected to physical and chemical analyses according to Carter and Gregoreish, (2008), and it has the properties listed in Table (1).

**Table 1. Some physical and chemical properties of experimental soil**

Property	Value
Particle size distribution (%)	
Coarse sand	64.1
Fine sand	26.4
Clay	6.8
Silt	2.7
Soil Texture	Sand
pH (1:2.5 paste)	7.97
E.C. (ds m <sup>-1</sup> )	0.27
Total N (%)	0.007
Cations (meq 100 g <sup>-1</sup> soil)	
Ca	1.25
Mg	1.00
Na	0.32
K	0.09
Anions (meq 100 g <sup>-1</sup> soil)	
SO <sub>4</sub> <sup>-</sup>	0.53
CO <sub>3</sub> <sup>-</sup>	0.00
HCO <sub>3</sub>	0.88
Cl <sup>-</sup>	1.25

Common bean was sown on February and harvested on May 2016, while spinach was sown on December, 2016 and harvested at January, 2017. Experimental treatments were carried out under drip irrigation system.

#### Composting manure

The organic compost was locally prepared according to Moursy, (2008). Prepared compost has the following properties; pH 6.7; EC, 12.7; total-N, 28.3 g kg<sup>-1</sup>; total-P, 8.4 g kg<sup>-1</sup>; total-K, 6.9 g kg<sup>-1</sup>; C/N 12.6; organic matter, 57%. Organic compost was applied according to its N content in equal recommended doses.

#### Mineral fertilizer

Ammonium nitrate (AN) was used as a source of chemical N fertilizer and applied at recommended rate of 75 kg N fed<sup>-1</sup> (equal to 262 kg AN fed<sup>-1</sup>) and 50 kg N fed<sup>-1</sup> (equal to 175 kg AN fed<sup>-1</sup>) for common bean and spinach crops, respectively. The recommended rates for both crops were splitted into two equal doses. The recommended rates of phosphorus (75 kg P ha<sup>-1</sup>) and potassium (54.5 kg K ha<sup>-1</sup>) were added at the initial soil before cultivation, in the form of super-phosphate and potassium sulfate, respectively. The four treatments, in addition to the untreated control, were arranged in randomized complete block factorial design and replicated three times on experimental plots of 10.5 m<sup>2</sup> (3.0 x 3.5 m). Untreated control was also included. Humic and seaweeds extract were applied as spray foliar application at 4% concentration. During the growing season the plants were sprayed three times with the bio-stimulants at monthly intervals (30 days after sowing, and the 1<sup>st</sup> of April, the 1<sup>st</sup> of May for common bean; 25 days after sowing, and the 1<sup>st</sup> of January, the mid of January for spinach). These bio-stimulants were applied in combination with quarter dose of mineral fertilizer. Some of chemical composition of humic acid and seaweeds extract are listed in Table (2).

**Table 2. Chemical composition of Humic acid imported from HUMIN TECH GmbH Company, Germany and Seaweed extract has the following gradient**

Humic acid	Seaweed extract	% w/w	
Determination	Value	Total nitrogen (N) 2.5	
K- humate	97%	Organic nitrogen carbon	1.4
Humic acid	85%	Potassium (K <sub>2</sub> O)	16.0
Total K	12%	Iron (Fe)	1.6
Iron	1%	Phosphate (P <sub>2</sub> O <sub>5</sub> )	4.0
pH	9-10.5	Sulfur (S)	2.0
Density	0.55 kg L <sup>-1</sup>	Algenic acid	50.0

After harvest, spinach plants (60 days) were collected and fresh weight was recorded, and separated into leaves and roots. Both organs were oven dried on 72 °C for 48 hrs, dry weight recorded, then ground and kept for analysis. The same way was followed with common bean (120 days), but the plants were separated into green pods and shoot plus root. Samples of both crops were subjected to wet digestion with acid, diluted and kept for chemical analysis. Plant samples were analyzed according to Estefan *et al.*, (2013). Standard equations were used for estimation of nitrogen, phosphorus and potassium content in addition to nitrate content in spinach leaves.

The obtained data were subjected to ANOVA statistical analysis followed by Duncan's multiple range test (DMRT) for comparison between means using SAS software program (2002).

## RESULTS AND DISCUSSION

#### Fresh and dry weights

The fresh yield of spinach at harvest was significantly affected by the biostimulants and fertilization treatments comparing to the untreated control (Table 3). The foliar application of humic acid and seaweed extract induced a significant increase in fresh yield by 6% and 41%; 15% and 53% over mineral-N and organic compost treatments, respectively. Seaweed extract seems to be superior over humic acid in increasing spinach fresh weight. Highly significant differences were noticed between all treatments and the untreated control. Nearly closed relative increase accounted for 51.5% on average over the untreated spinach was estimated by Rouphael *et al.*, (2018) but they didn't found significant difference between the used three commercial plant biostimulants (PB) treatments (legume-derived protein hydrolysate (PH), extract of seaweed *Ecklonia maxima* (SWE) or mixture of vegetal oils, herbal and seaweed *Ascophyllum nodosum*-based extracts (VO + SWE). They explained that the highest spinach fresh yield observed in PB-treated plants comparing to the untreated control was due to an increase in the leaf area and not in the number of leaves per plant.

Similarly, the leaf dry biomass in biostimulants treated plants was significantly higher by about 10.5% and 48% on average than that obtained in mineral-N and organic compost treated spinach plants, respectively. In harmony, Halpern *et al.*, (2015) reviewed that HS have many mechanisms on enhancement of plant through a number of positive effects on plant growth, including increased biomass, increased number of fruits and flowers, and improved fruit quality. In Egypt, spinach yield (fresh and dry) grown on sandy soil located in the north region (Beheira Governorate) was enhanced with increasing mineral fertilizer-N rate from 30% up to 70% from the recommended rate (Ali *et al.*, 2013).

**Table 3. Effect of bio-stimulants, organic additives and mineral fertilizer on dry matter yield of spinach and common bean grown on sandy soil**

Treatments	Spinach		Common bean	
	Leaves	Roots	Shoot	Pods
	Fresh weight kg fed <sup>-1</sup>			
Mineral-N	5600.0 b	2625.0 b	1225.0 b	2687.5 ab
Organic compost	4225.0 c	2375.0 c	1325.0 a	2500.0 b
Humic acid	5963.0 ab	2780.0 b	1275.0 b	2637.6 ab
Seaweed extract	6475.0 a	2976.0 a	1324.9 a	2700.0 a
Untreated	609.0 d	299.0 d	134.7 e	300.0 c
	Dry weight kg fed <sup>-1</sup>			
Mineral-N	3500.0 b	1625.0 c	762.5 b	1700.0 a
Organic compost	2625.0 c	1750.0 c	825.0 a	1562.5 b
Humic acid	3750.0 ab	2000.0 b	800.0 a	1650.0 a
Seaweed extract	4000.0 a	2625.0 a	800.2 a	1675.0 a
Untreated	365.0 e	168.0 e	76.3 e	187.5 e

Means in same column followed by the same letter are not significantly different at  $p \leq 0.05$

Common bean fresh green pods yield was slightly increased by application of seaweed extract and humic acid as compared to plants fertilized with ammonium nitrate or organic compost (Table 3). Highly significant increase in pods green yield affected by biostimulants and organic compost application was detected in comparison to the untreated control. Enhancement of green pods yield by biostimulant application was recorded earlier by Barakat *et al.*, (2015) who found that potassium humate applied at rate of 100 kg fed<sup>-1</sup> was sufficient to increase green pods yield fed<sup>-1</sup> by about 72.2% over the untreated control. This may be explained on the basis that potassium humate capable to increase availability, absorption and uptake of nutrients and augmented photosynthetic pigments which improve the plant growth with an eventual increase in green pods yield and its components (El-Bassiony *et al.*, 2010). In addition, results we have are in accordance with data reported by Hanafy *et al.*, (2010) who clarified that addition of humic acid, through drip irrigation system, to snap bean plants, significantly, increased number and weight of green pods plant and total green pods yield fed<sup>-1</sup>.

Likewise, El-Bassiony *et al.*, (2010) reported that increasing foliar application of humic acid up to a particular concentration accompanied with progressive increase in green pods yield of snap bean. Similar results were reported by Kaya *et al.*, (2005) on common bean. In other turn, humic acid is a component of potassium humate and the mechanism by which humic acid stimulated plant growth similar to that of plant growth regulators. Humic substances contained cytokinin (Zhang *et al.*, 2004), and auxins (Osman and Ewees 2008) and thus affect plant growth in a positive manner. Humic substances were reported to stimulate plant growth through regulating many biochemical and physiological processes as cell division, optimized uptake and assimilation of major and minor elements, enzyme activation and/or inhibition, protein synthesis and finally the activation of biomass production (McDonnell *et al.*, 2001; Atiyeh *et al.*, 2002; Chen *et al.*, 2004; Ulukon 2008). In addition, Abbas (2013) found significant increase of growth characters of shoots and roots of field bean plants treated with humic acid and algal extract and attributed that to the beneficial effect of humic acid on plant growth via promotion of nutrient uptake and nutritional status especially nitrogen, potassium and phosphorous. Furthermore, HS is considered as a source of

plant growth hormones, carbohydrates, amino acids and vitamins. Similar mechanism was also true with algae extraction promotion effects (Abd El-Moniem and Abd-Allah, 2008). French bean growth was improved by foliar application of Biomin-amino acid based fertilizer; Humifolin-humic acid based fertilizer; and DelfanPlus - only amino acid. Consequently, many parameters of vegetative growth as well as plant yield, nutrient content and fruit quality were improved by application of these three commercial organic fertilizers (Souri and Aslani 2018). Common bean grown in North West delta showed positive and significant effect of seaweed extract applied solely or in combination with amino acids where the highest yield and yield components as well as the harvest index were detected at harvest time (Zewail, 2014).

**Nitrate accumulation in spinach leaves**

Nitrate accumulated in green leaves of spinach plants was increased by addition of mineral and organic fertilizers, but it was higher in case of mineral-N than other treatments (Table 4). Although, the application of organic compost, humic acid and seaweed extract resulted in higher NO<sub>3</sub>-N than the untreated control, however it was lower than the permissible limits regulated by European commission regulation.

HS have a positive effect on nutrient uptake whereas it increases NO<sub>3</sub> uptake (Quaggiotti *et al.*, 2004), but this was not the case in the present study. In fact, spinach is normally considered a nitrate-accumulating leafy vegetable species (Colla *et al.*, 2018), and this phenomenon was recorded with mineral-N and the untreated control where nitrate content was significantly ( $p < 0.05$ ) higher than those resulted in by application of biostimulant or organic compost. In reverse, our results of organic compost and biostimulants refute those reported by Rouphael *et al.*, (2018) where they recorded the highest values of nitrate content with application of seaweed (SWE) and volatile oil (VO) + SWE treatments (3227 and 3485mg kg<sup>-1</sup> fw, respectively comparing to the control (2040 mg kg<sup>-1</sup> fw). In the present study, either application of organic compost, seaweed extract or humic acid reduced the accumulation of nitrate in leaves. The recorded values were lower than the permissible level regulated by European Commission Regulation (3500 mg kg<sup>-1</sup> fw).

Interestingly, the nitrate content in leaf of spinach treated with biostimulants make it safely marketable. This

phenomenon may be attributed to the ability of these biostimulants to avoid nitrate accumulation by regulation of nitrate reductase, which responsible for higher assimilation of nitrates into amino acids (Tsouvaltzis *et al.*, 2014).

In the same way, Souri and Aslani (2018) concluded that application of organic chelate fertilizers can avoid all negative effects of routine chemical salt fertilizers such as urea or ammonium nitrate, including leaching, volatilization and nitrate accumulation in vegetable tissues.

Therefore, they represent modern and suitable alternatives to simple salts, and even to commercial synthetic chelates such as EDTA due to their cheaper price.

**Table 4. Nitrate content in leaves and roots of spinach plants treated with bio-stimulants, organic and mineral fertilizers under sandy soil conditions.**

Treatments	Nitrate-N			
	Plant organs			
	Leaves		Root	
	kg fed <sup>-1</sup>	mg kg <sup>-1</sup> FW	kg fed <sup>-1</sup>	mg kg <sup>-1</sup> FW
Mineral-N	23.3 a	4160.0 a	5.3 ab	2019.0 a
Organic compost	10.0 c	2367.0 b	3.9 b	1642.0 b
Humic acid	11.0 c	1845.0 c	4.2 b	1511.0 b
Seaweed extract	16.9 b	2610.0 b	6.4 a	2150.0 a
Untreated	8.9 c	1461.0 d	3.4 b	1371.0 c

\*Maximum NO<sub>3</sub> level according to EC regulation no 1258/2011 is 3500 mg kg<sup>-1</sup>fresh weight (Rouphael *et al.*, 2018)

On sandy clay loam soil under Egyptian conditions, Ghaly *et al.*, (2017) concluded that nitrate accumulated in leaves of spinach was severely increased with increasing nitrogen fertilizer rates up to 150% of the recommended rate while the highest safe yield of spinach plant and the best quality parameters were realized for the plants treated with N-fertilization at the rate of 100% of the recommended rate.

**Nitrogen, phosphorus and potassium uptake**

Macronutrients (NPK) uptake by common bean (Table 5), were significantly affected by application of either mineral-N or organic compost as well as humic acid and seaweed extract. The highest nitrogen content in shoots was recorded with organic compost and humic acid followed by those of mineral-N and seaweed extract. All treatments caused very significant increases in shoot nitrogen uptake over the untreated control. Phosphorus uptake by shoot didn't reflect any significant difference between mineral-N, humic acid and seaweed extract while it was the highest with organic compost. However, all treatments enhanced P uptake by shoot comparing to the untreated control. Potassium uptake by shoot showed no remarkable difference between either mineral-N and organic compost or humic acid and seaweed extract. In this respect, the highest values of K uptake by shoot was recorded with seaweed extract which slightly increased K content, but not significant, over humic acid.

In general, NPK content in pods were significantly higher than those recorded with shoot. Nitrogen content in pods recorded the highest value with application of seaweed extract, but there was no significant difference between organic compost and humic acid followed by mineral-N treatment. All treated plants uptake more nitrogen in pods than the untreated one. Pods accumulated more phosphorus when treated with ammonium nitrate comparing to those treated with organic compost or biostimulants but all were higher than the untreated plants.

There was no significant difference between organic compost, humic acid and seaweed extract.

Potassium uptake by pods was not significantly varied among the different treatments but all of them induced higher K uptake than those recorded with the untreated plants.

**Table 5. Nitrogen, phosphorus and potassium uptake by common bean organs as affected by bio-stimulants, organic and mineral fertilizers under sandy soil conditions.**

Treatments	Nutrients uptake kg fed <sup>-1</sup>					
	Plant organs					
	Shoot			Pods		
	N	P	K	N	P	K
Mineral-N	22.6 b	2.2 b	20.0 b	42.1 c	6.6 a	46.1 a
Organic compost	28.3 a	3.6 a	21.0 b	50.6 b	4.4 b	45.8 a
Humic acid	28.4 a	2.3 b	25.4 a	49.5 b	4.8 b	44.6 a
Seaweed extract	21.3 b	2.2 b	25.9 a	54.7 a	4.7 b	46.5 a
Untreated	3.6 d	0.78 d	1.8 d	3.9 d	0.97 d	4.7 d

Means in same column followed by the same letter are not significantly different at p ≤ 0.05

Earlier, faba bean treated with humic acid and algae extract individually significantly increased nitrogen, phosphorus and potassium concentration as compared with the untreated control plants. However, algae extraction treatment recorded the highest values of phosphorus (Abbas 2013). These results proved, more or less, the obtained results in the present study.

Halpern *et al.*, (2015) reported many mechanisms responsible for role of HS in improving plant nutrition through affecting soil processes and directly affecting the plant's physiology. Furthermore, they added that seaweed extract (SE) improves plant nutrition by affecting soil processes and by affecting the plant's physiology directly.

Under Egyptian conditions, Zewail, (2014) found that nitrogen, phosphorus and potassium concentrations in common bean leaves were highly significantly increased with addition of seaweed extract combined with amino acids (2 m l<sup>-1</sup> seaweed + 4 ml l<sup>-1</sup> amino acids).

Like common bean, spinach leaves accumulated higher nitrogen when treated with seaweed extract than those treated with all other biostimulant or organo/mineral fertilizers. Humic acid treatment came to the next followed by mineral fertilizer, then the organic additive. In general, all treatments enhanced the uptake of nitrogen as compared to the untreated one. Similar trends with varied values were noticed with phosphorus and potassium content in the spinach leaves. Similarly, was the case of NPK uptake by root but in lower extent comparing to those of spinach leaves.

**Table 6. Nutrients (NPK) uptake by spinach organs as affected by bio-stimulants, organic and mineral fertilizers under sandy soil conditions.**

Treatments	Nutrients uptake kg fed <sup>-1</sup>					
	Plant organs					
	Leaves			Root		
	N	P	K	N	P	K
Mineral-N	44.8 b	32.2 a	77.0 a	8.8 b	1.3 b	17.2 b
Organic compost	27.8 c	21.5 b	46.2 b	8.1 b	1.6 b	14.7 b
Humic acid	46.9 b	31.5 a	71.6 a	13.2 b	1.4 b	17.0 b
Seaweed extract	72.8 a	34.4 a	81.6 a	22.8 a	3.1 a	28.1 a
Untreated	3.4 d	3.0 c	4.2 c	0.4 c	0.1 c	1.3 c

Means in same column followed by the same letter are not significantly different at p ≤ 0.05

These results were recently confirmed by Galal *et al.*, (2018) who found that spinach plants treated with organic compost, chemical-N, chem+humic acid and chem+proline or combined treatment of them reflected high N uptake comparing to the untreated control. In accordance, they reported that plant treated with compost has gained more P than the untreated control but there was no remarkable difference among chemical-N or chem+humic acid and chem+proline. In addition, K uptake by spinach plants affected by organic additives and biostimulants was enhanced comparing to the untreated control. Under compost treatment, K uptake by spinach fertilized with only chemical-N achieved the highest value followed by those treated with combined treatment of chem+humic acid plus proline, chem+humic then chem+proline. In other study with lettuce crop, Shahein *et al.*, (2015) indicated the superiority of 50% mineral recommended NPK with humic substances extracted from biogas manure as foliar treatment (HBF), 50% NPK+HBS+HCF, 50% NPK with humic substances of compost as drench soil treatment (HCS) and 50% NPK+HBF+HCS resulted in the highest NPK contents in lettuce leaves. In addition, NPK uptake by spinach grown in sandy soil was increased with application of 70% of chemical NPK fertilizers recommended by Ministry of Agriculture of Egypt either applied solely or in combination with bio-fertilizer (Ali *et al.*, 2013).

## CONCLUSION

Either organic fertilizer or biostimulants like humic acid and seaweed extract could be safely used as alternative innovative tools to avoid toxic effects of some chemical derivatives such as nitrate produced from the excessive application of mineral fertilizers. It is very important to the producers of vegetable to keep their production in safe category. This may help them to sell their production as a marketable product. Leafy and vegetables should be kept with minimum content of nitrates lies within the permissible limits regulated by international and local organization. Biostimulants and organic compost used in this study achieved this goal.

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## إضافة المحفزات الحيوية مقارنة بالأسمدة العضوية والمعدنية لتحسين السبانخ والفاصوليا النامية على الأرض الرملية

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تم معاملة نباتات السبانخ والفاصوليا ببعض المحفزات الحيوية لتقييم تأثيرها، مقارنة بالأسمدة العضوية والمعدنية، على نمو وامتصاص المغذيات وتراكم النترات تحت الظروف الحقلية. اختبرت المعاملات المنفردة من حمض الهيوميك ومستخلص عشب البحر كمحفزات حيوية مقارنة بسماد نترات النشادر والكمبوست العضوي. تأثرت إنتاجية المادة الطازجة والجافة لكل من السبانخ والفاصوليا معنويا وإيجابيا بإضافة مستخلص عشب البحر وحمض الهيوميك على الترتيب. المعاملة بالكمبوست العضوي أنتجت مادة طازجة وجافة من أوراق السبانخ والمجموع الخضري لنبات الفاصوليا وكذلك القرون قريبة جدا أو تقل قليلا عن تلك المسجلة مع معاملة نترات النشادر. تفوقت المعاملة بمستخلص عشب البحر في معظم الحالات على المعاملات الأخرى. المعاملة بكل من مستخلص عشب البحر وحمض الهيوميك أدت الى زيادة نسبية في محصول المادة الطازجة والجافة لكلا المحصولين حوالي 50% كمتوسط أعلى من النباتات المسمدة بنترات النشادر أو تلك المعاملة بالكمبوست العضوي. أدت إضافة مستخلص عشب البحر وحمض الهيوميك الى تقليص محتوى النترات في أوراق السبانخ مقارنة مع النباتات المعاملة بنترات النشادر ولكنها كانت قريبة من النباتات المعاملة بالكمبوست العضوي. سواء النباتات المعاملة بالمحفزات الحيوية أو الكمبوست العضوي أنتجت أوراق سبانخ ذات محتوى قليل من النترات ما يجعلها مرغوبة للعرض في السوق طبقا للمواصفات المأخوذ بها في الاتحاد الأوروبي (3500 ملجم نترات لكل كجم مادة طازجة). امتصاص كل من النيتروجين والفوسفور والبوتاسيوم بواسطة أوراق السبانخ وقرون الفاصوليا زاد معنويا وإيجابيا مع كل المعاملات مقارنة بالكونترول الغير معامل ولكن إضافة مستخلص عشب البحر ، والى حد ما حمض الهيوميك أدت الى زيادة في محتوى النباتات من تلك العناصر المغذية أكبر أو قريبة من تلك المسجلة مع السماد المعدني أو الكمبوست العضوي. هذه النتائج اتاحة الفرصة لقبول المحفزات الحيوية الى جانب الإضافات العضوية كوسائل بديلة ومستدامة محققة إنتاجية عالية مع جودة مباشرة.