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Review

Role of bio-stimulants on the advancement of vegetable production: A review

[1*]Bahran Knfe Yakob, EricksonTchuda Lopes Mam[2], Gins Murat Sabirovich[3]

[1,2,3] Department of Agro-biotechnology, Faculty of Agriculture, Rudn University, Moscow, Russia

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Abstract. *Since climate of the globe is changing abruptly, causing tremendous challenges (biotic and abiotic stresses) on the production of vegetable crops, it seems to be difficult to ensure the food security for the rapidly growing population of the world if sustainable production systems are adopted. Though farmers are indiscriminately applying inorganic fertilizers and plant protection chemicals to replenish the fertility of their fields and protect their plants from pests, the issue of sustainable production seems to have been forgotten. Thus, the use of organic factors of production is a must in order to overcome the challenges so that production of healthy products can be maintained in an eco-friendly manner through the utilization of Bio-stimulators. Bio-stimulants have a crucial role in enhancing the growth, development and overall performance of different vegetable crops belonging to different families; Solanaceae, Alliaceae, Amaranthaceae, and Brassicaceae. The most commonly utilized bio-stimulants in the field of agriculture worldwide are humic substances (humic acid and fulvic acid), plant or animal-based protein hydrolysates, macro and micro-algal extracts (seaweed extracts), silicon, arbuscular mycorrhizal fungi and plant growth-promoting rhizobacteria. Bio-stimulants have important role in enhancing the growth characteristics, yielding potential, biochemical compositions (concentrations of ascorbic acid, and carotenoid content) in tomato plants and in other plants belonging to Solanaceae family like eggplant. Application of different types of bio-stimulants results in the improvement of growth, yields and chlorophyll and allicin contents in the Alliaceae species (onion and garlic). In the vegetable crops belonging to the family of Amaranthaceae (Spinach and Amaranthus), bio-stimulants have important impacts in the improvement of seed germination, increasing of plant height, photosynthetic pigments, yield and nutritional composition. Increased plant growth, photosynthetic rate and stomatal conductance, yield parameters, phenolic and flavonoid compounds and seed fatty acid concentration are also the results of bio-stimulant applications in the brassica species (cabbage, broccoli, rapeseed, mustard). Thus, the sustainable agriculture systems could be guaranteed by using bio-stimulants to boost the production of vegetable crops both quantitatively and qualitatively.*

Keywords: climate, bio-stimulants, vegetable crops, growth, yield, sustainable agriculture

*e-mail: bahranknfe@gmail.com

Introduction

Climate change is one of the biggest challenges at the present times becoming a global threat to the food and nutritional security of the world. The average global temperature is increasing continuously and is predicted to rise by 2ºC until 2100, causing substantial economic losses at the global level (Malhi et al., 2021). The changes of the climatic factors expose plants to several abiotic stresses such as salinity, drought, heat stress, cold stress (Malhi et al., 2020), soil fertility loss, and pest infestations (Baul and McDonald, 2015) causing challenges on the growth and development processes of plants (Gang et al., 2013). The increase in the demand of food in 2050 is estimated to be 25 to 70% more than what is currently produced and thus, a sustainable intensification of agriculture is needed (Cristofano et al., 2021). In order to increase quality and yield of the crops, to protect from health hazards, and to sustainably maintain natural agro-ecosystems for future generations without adding more chemical fertilizers and pesticides, it is customary to increase the nutrients uptake and use efficiencies (Singh et al., 2020), on the one hand, and enhance the natural defense mechanisms of plants to face pests and diseases without resorting to chemical pesticides (Costa et al., 2019). Thus, the use of plant bio-stimulants (PBs) tends to be a valid strategy for the enhancement of sustainable practices. A plant bio-stimulant is any substance or microorganism applied to plants with the aim to enhance nutrition efficiency, abiotic stress tolerance and/or crop quality traits, regardless of its nutrients content (Jardin, 2015). According to Kauffman et al. (2007), Bio-stimulants are available in a variety of formulations and with varying ingredients but are generally classified into three major groups on the basis of their source and content. These groups include humic substances (HS), hormone containing products (HCP), and amino acid containing products (AACP), inorganic compounds, different forms of bacteria and fungi (vesicular arbuscular mycohrriza). Seaweed extract is the most commonly used product category accounting for 37% of all the Plant Bio-stimulants market share, with Plant Hormones and Humic Substances representing a 50% share combined (Rouphael et al., 2017).

Humic substances

Humic substances (HSs) or humus are products of decomposing plants that have large molecular weights and complex structures (Lee et al., 2004) and comprise three types of organic acids; humin, humic acid (HA) and fulvic acid (FA) (Jan et al., 2020). Humic acid being the major constituent of humic substances contributed the main essential elements of peat, coal and soil and it is extremely soluble in water and simply absorbed by plants as compared to fertilizer (Jan et al., 2020). Humic substances encourage plant growth by the integration of major and minor elements, activator and inhibitor of several enzymes, changes in membrane penetrability, important for protein synthesis and increase of biomass production (Mackowiak et al., 2001). Humic acid and Fulvic acid have direct effect in the hormonal activity and indirect effects through the improvement of soil properties like aeration, permeability, water holding capacity, transport of ion and pH (Olaetxea et al., 2018). The most important biological effects of HA and FA include improvement of seed germination and growth, stimulation of biomass accumulation in plants, facilitation of mineral nutrient uptake, and increase of plant resistance to environmental stress (Hatami et al., 2018).

Seaweed extracts (SWEs)

Seaweed extracts (SWEs): Seaweeds are macro-algae that are a vital constituent of the marine and coastal ecosystem; enhancing biodiversity of the biosphere (Ali et al., 2021) and they can grow in extreme conditions of temperature and salinity (Reddy et al., 2023). In most cases, extracts are made by processes using water, alkalis or acids, or physically by disrupting the seaweed by low-temperature milling to give a micronized suspension of fine particles (Sharma et al., 2014). SWEs cannot be regarded as a single entity due to the fact that they are extracted from many species, the inherent variation of their constituents based on climate and season, and the plethora of extraction (Shukla et al., 2019). The extracts contain a mixture of useful biological active substances; polyphenols, polysaccharides, alginates, polyamines, pigments, free amino acids, betaines, vitamins, micro and macro-nutrients and natural phytohormones (Stirk et al., 2014; Ertani et al., 2018). Besides promoting the osmo-protection by increasing the contents of compatible solutes (proline), SWEs help in reducing oxidative stresses (reduce ion toxicity by modulating the abscisic acid and cytokinin pathways) to improve membrane stability (Cristofano et al., 2021). The effect of reduction of these stresses in the critical phases of plant biology, in turn, results in an increased plant growth, yield, and antioxidant activity (Cristofano et al., 2021).

Protein Hydrolysates (PHs)

Protein Hydrolysates (PHs): These are protein bio-stimulants originating from plants and animals in which such a protein, acting as a source of raw material forms peptides and amino acid products through hydrolysis (Reddy et al., 2023). Over 90% of PH products are produced by chemical hydrolysis of animal by-products, whereas enzymatically processed plant-based products have also been developed recently (Colla et al., 2015). It has a direct impact on N uptake and assimilation, hormonal activity, enhanced antioxidant activities, free radical scavenging control, improved chelation for plant protection against heavy metals, improves soil fertility and contributes to micronutrient mobility, thereby alleviating abiotic stresses and ultimately leading to enhanced growth, yield and quality of crops (Reddy et al., 2023).

Chitosan

Chitosan: is a deacetylated form of the biopolymer chitin, produced naturally and industrially. It has been used in protecting plants against fungal pathogens, abiotic stresses (drought, salinity, cold stress) and on quality traits related to primary and secondary metabolisms (Jardin, 2015) inducing stomatal closure via an ABA-dependent mechanism thereby protecting plants from environmental stresses (Iriti et al., 2009). The application of chitosan influences an array of protective genes in plants, which in turn encode enzymes like glucanase and chitinase, also reactive oxygen species stabilizers like super oxide dismutase (SOD), catalase, and peroxidase (Ivanova and Yaneva, 2020).

Inorganic compounds

Inorganic compounds: Chemical elements like aluminum, cobalt, sodium, selenium, and silicon help in promoting plant growth, the quality of plant products and tolerance to abiotic stresses (Jardin, 2015) though they are not required by plants as nutrients. They are present in soils and in plants as different inorganic salts and as insoluble forms like amorphous silica ($SiO_2.nH_2O$) in graminaceaous species. The mechanisms by which the benefits of these inorganic compounds get achieved are through cell wall rigidification, osmoregulation, reduced transpiration by crystal deposits, thermal regulation via radiation reflection, enzyme activity by co-factors, plant nutrition via interactions with other elements during uptake and mobility, antioxidant protection, interactions with symbionts, pathogen and herbivore response, protection against heavy metals toxicity, plant hormone synthesis and signaling (Pilon-Smits et al., 2009).

Beneficial microorganisms

Beneficial microorganisms: Fungi interact with plant roots in a symbiotic relation through direct contact (Behie and Bidochka, 2014). The Arbuscular-mycorrhizal fungi (AMF) are a widespread type of endomycorrhiza associated with many crops including horticultural plants, where fungal hyphae of Glomeromycota species penetrate root cortical cells and form branched structures (Behie and Bidochka, 2014). Such a symbiotic interaction with the plants helps to improve the nutrition efficiency (for both macronutrients, especially P, and micronutrients), water balance, biotic and abiotic stress protection of plants (Siddiqui et al., 2008; Gianinazzi et al., 2010).

The purpose of this review article is to introduce the beneficiaries about the great advancements that have been achieved regarding the utilization of bio-stimulants by forwarding scientific results obtained from diversified vegetable crops. The content of this manuscript is believed to have a good contribution to the clients due to the fact that application of bio-stimulants have the capacity to compensate the loss of production potentials caused by diversified forms of stresses majorly caused by the ongoing climate changes of our globe. Farmers who will adopt new production systems through the use of bio-stimulants are not only expected to boost their production potentials but they will be able to produce healthy organic produce that will ensure the health condition and also the sustainability of their production systems.

Role of Bio-stimulants on Solanaceae family

Experimental results of Azarpour et al. (2012), showed that foliar spraying of humic acid had significant effect on all vegetative and yield parameters and increased concentration of humic acid foliar spraying up to 50 mg/L led to improved plant growth and yield of eggplant. Applying Ener850 humic acid caused the highest significant plant height (264.6 cm), number of leaves/plant (45), stem diameter (1.9 cm) and fruit weight (137 g) in addition to its positive impact on increasing the dry weight (75.1 g) of arial parts with improved tomato fruit quality through enhancing the concentrations of ascorbic acid, level and carotenoid content (Ali et al., 2020). As per the findings of Yildirim (2007), shoot and root weights were increased by 22% through the use of Humic Acid in tomato plants. Humic acid with concentration of 1000 mg/L led to the production of significantly higher values of the chemical parameters such as: N, P, K, total soluble carbohydrate and total Chlorophyll in Leaves and N, P, K, TSS and amount of ascorbic acid in the fruits as compared to the control treatment which gave the least values for the above-mentioned parameters (Aboohanah et al., 2021). Selim et al. (2009) found that application of humic substances via drip irrigation enhanced potato tuber yield, starch content and total soluble solids and its application associated with the decrease of nutrients leaching, led to increased concentration of macro and micronutrients in potato tubers. In addition to the chlorophyll content of tomato leaves being increased from 13.12 (control) to 22.41 (30 ppm Humic acid and 15 mM Calcium application) (Kazemi, 2014), growth parameters (number of branches, plant height and dry weight) and yield and fruit quality were significantly increased by Humic acid or Ca foliar applications. Yildirim (2007) reported a significant enhancement of fruit quality as a result of exogenous Humic acid application in tomato. The bio-stimulant (*Azospirillum brasilense*) significantly enhanced the following parameters of eggplant: plant height, total yield, fruit dry matter, total anthocyanins concentration of fruits, nitrogen use efficiency and protein concentration, but its effect on the concentration of Potassium and Phosphorous were insignificant (Consentino et al., 2022). Gravel et al (2007) reported tomato plants inoculated with plant growth promoting bacteria showed higher performance in terms of the plant vigor parameters. Ordookhani and Zare (2011), upon studying the effect of plant growth promoting bacteria on the tomato fruit quality, found out an encouraging influence of the microorganism (*Azospirillum brasilense*) on fruit Soluble sugar concentration. Eggplants treated with a microbial bio-stimulant (arbuscular mycorrhiza fungi) had the highest fruit protein concentrations and decreased values of glycoalkaloids (Sabatino et al., 2020). According to Mystkowska (2019), potato plants treated with bio-stimulants had a lower glycoalkaloids concentration than the control plants. Bio-stimulated eggplant roots effectively penetrated deeper in the soil and were able to absorb more nutrients (Pohl et al., 2019). Increase in root length, shoot length values were observed in tomatoes treated with *Sargassum johnstonii* extracts (Kumari et al., 2011; Zodape et al., 2011) and with *Ascopyllum nodosum* extracts (Stępowska, 2008). Tomato plants treated with Seaweed extract showed more numerous and bigger cells of vascular bundles, with thicker and stronger lignified xylem and phloem secondary walls than untreated control plants (Koyama et al., 2012). Seaweed extracts (SWEs) enhanced the plant height, leaf area, nitrogen, phosphorous, potassium, crude protein, carbohydrates, and leaf chlorophyll of eggplant due to the presence of growth regulators, amino acids, and vitamins in the SWEs that could control cell division, maintain photosynthetic rates, improve plant resistance, and delay plant senescence (Abd El-Gawad and Osman, 2014). Manna et al. (2012) associated the increased growth of pepper to the presence of cytokinin and auxin precursors in *Ascopyllum nodosum* extract, which increase the cell division as well as optimal partitioning and utilization of metabolics. SWEs lowered salt stress in pepper plants through an increase of proline content (Yildiztekin et al., 2018). Application of *Melanothamnus afaqhusainii*, and Halimeda tuna extracts can suppress rotting fungi *Rhizoctonia solani* and *Fusarium solani* on tomato roots (Sultana et al., 2012). SWE, applied as a soil drench to pepper plants, reduced Verticillium wilt of pepper through improving plant fitness and increasing resistance to pathogens (Rekanović et al., 2010). A mixture of four bio-stimulants containing amino acids, polysaccharides, vitamins, humic acids, organic carbon, and enzymatic proteins caused a comparable increase in the yield of two varieties of yellow pepper with the peptides and amino acids possessing a protective action against high temperatures in the summer season and also induced root growth, while vitamins and humic acids resulted in fruit growth (Parađiković et al., 2011). Protein hydrolysate at a rate of 2 ml/L, increased the lycopene and ascorbic acid of tomato fruits by 18% and 27.3%, respectively, as compared to control (Møller and Laursen, 2015). The use of bio-stimulants and herbicides (Zarzecka et al., 2019) increased the content of polyphenols in the leaves of the potato to an average of 289.2–291.2 mg/kg compared to the control (287.8 mg/kg). Tomato plants treated with one of the tested bio-stimulators (arbuscular mycorrhizal fungi + Pseudomonas sp. 19 Fv1T and Pseudomonas fluorescens C7) showed an increase in the concentration of glucose in tomatoes at 11.83 g/kg, while in controls the content of glucose was 10.45–11.0 g/kg (Drobek et al., 2019). Significant effect of Aqueous Garlic extract was observed on the growth of tomato seedlings both in terms of application method and concentrations used with the highest plant height being recorded from plants fertigated with 100 µg/mL followed by 200 µg/mL foliar spray, whereas the lowest values were observed in control seedlings (Hayat et al., 2018). Foliar-applied silicon reduced the Fe concentration and increased Cu and Mn concentrations in early crop potato tubers under water deficit conditions, but did not affect the Zn, B, or Si concentrations (Wadas and Kondraciuk, 2023). The most significant increase in yield was observed when chitosan was used in the treatment of early maturing cultivar of potato 'Snegir' with an increase of 1.96 t/ha or 1.96 Mg/ha (Shitikova et al., 2022). Mahmoud and Hafez (2010) reported that the vegetative growth parameters, potato yield and tuber size, weight, and quality as well as nutritive value of potato tuber were significantly increased with increasing the level of Humic acid application from 0 up to 2 kg/ha. In the experiment conducted by Majkowska-Gadomska et al. (2021), the leaf greenness index (SPAD) in *Capsicum annuum* L. was similar in plants sprayed with bio-stimulants and in control plants. Upon studying about the application of an animal-based protein hydrolysate (Pepton) in tomato under water stress, Casadesús et al. (2019) observed a significant increase in the hormone profile in the plants with increased levels of auxin, cytokinin and gibberellin concentrations, ultimately resulting in better plant growth under drought stress conditions. Application of *Kappaphycus alvarezii* (SWE) was observed to contribute to positive results in the improvement of plant growth and yield in potato (Garai et al., 2021). In the study conducted on the influence of *Bacillus pumillus* on tomato (He et al., 2019), a significant increase of nutrient content in fruit and fruit yield was reported and when combined with *Pseudomonas putida* there was an increase in healthy fruit yield.

Role of Bio-stimulants on Alliaceae family

Onion plants treated with 1 kg/fed of humic acid foliar application resulted in a significantly superior marketable yield recorded in two successive growing years (14.03 t/fed and 13.04 t/fed) as compared to those inoculated with *Azotobacter chroococum* spp. and *Azospirilium* spp, foliar spraying with active dry yeast at rates of 6 g/L, foliar spraying with compost tea at the rate of 20 L/fed compost tea, also control (Hafez and Geries, 2019). Foliar application of algal bio-extract resulted in significant positive effect on chlorophyll a and b and carotene content of onion as compared with control treatment with 1.5 g/L resulting in the highest pigment contents as compared to the control (Yassen et al., 2018). Results of an experiment conducted by Mahmoud et al. (2020) indicated that plants that were treated with all stimulants (seaweed extracts, amino acids as well as combinations of (whey + molasses), (whey + yeast) and (whey + molasses + yeast)) gave significantly higher values of vegetative growth parameters, photosynthetic pigments, yield and characteristics of bulb as well as some phytochemical compounds than plants that did not receive any stimulants. Out of all these individual bio-stimulants and their combinations, (whey + molasses + yeast) recorded the highest significant values of yield, chlorophyll a, b and carotenoid content of the onion bulbs. According to (Sundharaiya et al., 2016), foliar application of seaweed extract@ 2% registered significantly higher plant height, number of leaves, leaf length and leaf breadth at 30 DAP, 60 DAP, 90 DAP and at the time of harvest and yield traits; bulb length, bulb girth, number of bulblets per clump, individual bulb weight, yield per plot and yield per hectare. Results of the experiment conducted by El-Shaboury and Sakara, (2021), indicated that soil addition of 4 kg/fed of potassium humate recorded the highest significant values for growth traits, yield and N, P, K, Fe, Mn and Zn contents in the leaves and bulbs as compared to other bio-stimulants (Garlic and onion extracts) as well as control. Sunflower derived humic acid (SDHSs) and maize derived humic acid (MDHSs), and coal-derived humic acid (CDHSs) yielded the highest onion bulb yields of 25.1 t/ha and 24.5 t/ha and 24.7 t/ha, respectively (Khan et al., 2017). The bio-stimulant treatment of shallots with effective microorganisms (EM) and Goëmar Goteo (GG) seaweed extract, lowered the yields of the bulbs by 14% and 4% as compared to the control, respectively (Francke et al., 2022). A positive effect of seaweed extracts on the biometric parameters of plants was reported by Abbas et al. (2020) in common onions and by Sulakhudin et al. (2019) in shallots. Trisula variety of shallot treated with *Trichoderma* species showed a significant difference in plant height measured at 40 and 60 days after planting (DAP) when compared with the control (Hadiawati et al., 2020). Results of the research conducted by Rokhminarsi et al. (2019) stated that utilization of *Azola mycorrhiza* on shallots can reduce chemical fertilizers requirement by 40%. According to experimental findings of Jędrszczyk et al. (2019), the use of growth and development bio-stimulants (oligosaccharide granule, *Bacillus subtilis* and concentrate of humic and fulvic acids supplemented with macro- and microelements) influenced the chemical composition of garlic, though the effect was modified by the course of weather in different years of the research and also differed in individual edible organs. Besides the mean value for years and organs, all the bio-stimulants increased the level of protein, minerals expressed as ash and enhanced antioxidant activity, whereas decreased the level of total carbohydrates. On the basis of the morpho-physiological attributes and biochemical properties, a low dose (2.0 g/L) was more effective than a high dose (4.0 g/L) of *Trichoderma album* (microbial bio-stimulant) in promoting onion growth (Younes et al., 2023). Besides the enhancement of plant growth, nutrition and Nitrogen metabolism, Humic substances stimulated Sulphur assimilation by promoting the enrichment of garlic plantlets with the Sulphur based amino acid “alliin”, which has been recognized to have beneficial properties in human health (Pizzeghello et al., 2020). According to Ghasemi et al. (2015), higher rate of antioxidant activity was observed when selenium was used in low concentrations (0 and 10 μg/mL) and also by applying humic acid at the rate of 10 kg/ha whereas in high concentrations of Se (20 and 30 μg/mL) and humic acid, even at the rate of 10 kg/ha, caused a decrease in antioxidant activity. In addition to this, the sulfur-uptake by garlic plants was found to be the highest when the plants are supplied with 20 kg/ha of humic acid coupled with 0 μg/mL of selenium resulting in the increased production of allicin. Application of humic acid had significant effect on total chlorophyll content of garlic increasing the total chlorophyll from 0.74 mg/g fresh weight (control treatment) to 1.01 mg/g fresh weight at 3000 mg/l humic acid. Besides this, humic acid also significantly increased essential oil and allicin compositions of garlic with the essential oil and allicin percentages being increased from 0.55%

and 4.91 mg/g at control treatment to 0.64% and 5.31 mg/g at 3000 mg/l humic acid, respectively (Zeinali and Moradi, 2015).

Role of Bio-stimulants on Amaranthaceae family

Humic acid application at medium levels (250 g/m^2) increased yield and nutrient content of spinach (Ayas and Gulser, 2005). According to Drobek et al. (2019), application of SWE increased the surface area of spinach leaves by 16% (foliar spray), 21% (bio-stimulant in the irrigation system), and 38% (bio-stimulant in the irrigation system and in a spray). A study carried out by Lestari and Dewi (2020) on the effect of humic acid on amaranthus, indicated that humic acid (20 mg/L) applied either through foliage spay or soil surface increased plant height, yield and betacyanin content, but it reduced doxalate content of red amaranth. Research results showed that the application of microorganisms (*Bacillus licheniformis* and *Pseudomonas fluorescens*) alone had a deleterious effect on the growth of *Amaranthus hybridus* (leaf number and area, shoot and root length, and fresh weight) as compared to the control plants (Ngoroyemoto et al., 2020) and in contrast to this, the combined application of the microorganisms and Kelpak (seaweed extract) overcame the negative effect of the microorganisms with comparable growth achieved as with the Kelpak treatment lone effect. Pyakurel et al. (2019) reported that spraying of molasses boosted the photosynthetic pigments content in spinach leaves. Foliar application of humic acid on amaranthus at a concentration of 20 mg/L, significantly increased plant height, leaf number, the fresh and dry weight of shoot and betacyanin content (Lestari and Dewi, 2020). Rouphael et al. (2018) reported that spinach leaves treated with seaweed-based extracts (*Ascophyllum nodosum*) accumulated 35% more nitrates than the leaves of control plants. Çakmakçi et al. (2007) reported a higher level of antioxidants in spinach following the application of *Bacillus subtilis*. The results of the experiment carried out by Devi et al. (2016) revealed that the yield parameters of beetroot (*Beta vulgaris* L.); root length, root girth, root weight, shoot:root ratio plant^{-1}, yield plot^{-1} and yield hectare^{-1} were recorded maximum in the treatment which received the soil application of vermicompost @ 5 t ha^{-1} and CBF @ 2 kg ha^{-1}, combined with foliar application of effective microorganism @ 1:1000 dilution ha^{-1}, PK @ 3% and HA @ 2%. In accordance with the experimental results of Khodadadi et al. (2020), foliar application of humic acid, Salycilic acid, GA_3 and Ascorbic acid significantly increased the activities/levels of enzymatic and non-enzymatic antioxidants and vital organic osmolytes of sugar beet. Although amaranth plants treated with Stimplex® did not show any significant difference in weight when compared with the untreated plants (Sandhu et al., 2021), the Stimplex®-treated Swiss chard plants did exhibit a significantly increased leaf weight values with the treated plants producing 445.7 g and the control plants (401.33 g). On the basis of the research results of Redondo-Gómez et al. (2022), a positive effect of plant growth promoting rhizobacteria (PGPR) consortia was observed in halophytes on the growth of Swiss chard under salt stress improving the concentrations of chlorophylls and carotenoids, the photosynthetic capacity, and reduced the ionic toxicity ultimately resulting in the alleviation of stress from the plants which is mediated by the acquisition of nutrients and the enhancement of antioxidant capacity. The results obtained from the experiment conducted by El-Nakhel et al. (2022) proved that legume-derived protein hydrolysates (LDPH) reduced the detrimental effect of saline irrigation water on baby spinach plants for indeed under all the salinity treatment levels, the bio-stimulant-treated plants showed higher yields than the untreated ones, even at the highest saline treatment when the irrigation water reached 9 dS m^{-1}. The fact that freshwater alga (*Chlorella fusca*) is an efficient and economical bio-stimulant in improving plant growth and quality of spinach was demonstrated by Kim et al. (2018), in which the thickness and number of spinach leaves, fresh weight and yield of the spinach treated with chlorella were 27.9% and 41.8%, 63.6% and 31.5%, respectively, higher than those of the untreated. Moreover, the mineral content of K, Ca, Mg, P, Fe, and Mn were recorded higher in the spinach treated with chlorella compared with that of the untreated. The outcomes of a study performed by Braziene et al. (2021) indicated that fulvic acid bio-stimulants were found to improve germination in sugar beet (*Beta vulgaris*) and also increased the length of shoots and dry weight of shoots and roots.

Role of Bio-stimulants on Brassicaceae family

Foliar spraying of seaweed extract (Stimplex®) resulted in a significantly increased weight of mustard leaves (500.77 g) when compared to the untreated (368.63 g) and leaves of Stimplex®-treated mustard plants appeared to be smoother, dark-colored and portrayed more red coloration on their leaves in comparison to the untreated plants (Sandhu et al., 2021). Moreover, foliar application of seaweed extract (Stimplex®) also resulted in a significant weight increase in collard (423 g) and kale (456.2 g) plants as compared to the untreated collard (354.43 g) and kale (388.4 g) plants. Jannin et al. (2012) reported that treating rape seed (*Brassica napus*) plants with Humic substances positively impacted the Carbon, Nitrogen and Sulphur metabolism. Experimental findings of Toscano et al. (2021) demonstrated significant effects of Moringa leaf extract (MLE) on nitrate content in the baby leaves of kale and broccoli where MLE significantly reduced nitrate content (−70%) in kale, while it increased by 60% in broccoli. The effects of Humic acid in the photosynthetic mechanism of *Brassica napus* under water deficit was reported to positively affect the rate of the electron transporter chain, thus exhibiting higher net photosynthesis rates (Lotfi et al., 2018). Foliar application of protein hydrolysates was observed to cause amelioration in the gas exchange and transpiration rates, as well as an increase in photosynthetic rate and stomatal conductance of broccoli (Kałużewicz et al., 2017). Increased values of yield, chlorophyll content; improved oil quality, plant net photosynthesis, gas exchange rate and electron transport flux; decrease in soluble carbohydrates, linolenic and erucic acid of *Brassica napus* as influenced by humic acid were reported from the experiment conducted by Nasiri et al. (2021). Experimental results of Lee et al. (2019) indicated that foliar application of bio-stimulants; brassinosteroid (BR), methyl jasmonate (JA), salicylic acid (SA), and sodium nitroprusside (SNP) reduced physiological damage and enhanced the activity of the antioxidant enzymes, thereby improving heat stress tolerance in Kimchi cabbage. In accordance with the research outcomes of Sujata et al. (2023), Foliar application of Orthosilicic acid (OSA) and Seaweed extract (*Ascophyllum nodosum*) had a positive impact on the growth, physiological, and biochemical processes of the brown mustard crop under normal and stressed conditions. Moreover, increased physiological efficiency, photosynthetic rate, stomatal conductance, transpiration rate, relative water content, water potential, osmotic potential, chlorophyll fluorescence, chlorophyll stability index, total soluble sugars, total protein content, and antioxidant enzyme activity; and decreased canopy temperature depression, proline, glycine-betaine, H_2O_2 content, and MDA content were observed in OSA and SWE treated-plants, under water stress conditions. Hashemi et al. (2010), reported that application of Silicon (2 mM Na_2SiO_3) increased Catalase(CAT) and Peroxidase (POD) activity to reduce the overproduction of Reactive oxygen scavenger (ROS) in hydroponically grown *Brassica napus* spp, thus helped to maintain membrane integrity by removing negative effects of salinity under salinity (150 mM NaCl). Lola-Luz et al. (2013) found that cabbage plants (grown under normoxic conditions) and treated with foliar application of two *Ascophyllum nodosum* extracts (Algae Green™ and XT) at the rates of 3.5 L ha^{-1} (containing 2.6 mL seaweed extract) and 5 L ha^{-1} (containing 3.7 mL seaweed extract) tended to have increased contents of phenolic and flavonoid compounds. However, this increase in phenolic and flavonoid compounds did not lead to a significant increase in total yield compared to the untreated control.

Conclusion

Currently, heavy use of chemical fertilizers and plant protection chemicals caused the reduction of soil productivity and also resulted in deleterious effects on human health. The use of organic inputs like bio-stimulants in agriculture preserved not only the fertility of soils, but also the sustainability of the eco-system in general and the health of humans in particular. Various experiments conducted on the application of bio-stimulants through seed treatments, foliar applications or soil applications proved its effectiveness in overcoming the stresses (biotic and abiotic) thereby increased the growth, yield and quality parameters of different vegetable crops. Humic acids contributed to the improvement of the following parameters: seed germination, growth parameters (eggplant and red amaranthus), yield parameters (potato, onion, spinach eggplant and red amaranthus), Chlorophyll content and net photosynthesis (Garlic and rapeseed), concentrations of ascorbic acid and carotenoid content (tomato), production of allicin (Garlic), starch content (potato), Carbon, Nitrogen and Sulphur metabolism (Rapeseed). Application of seaweed extracts also tended to have positive impacts in improving growth characteristics (onion, potato, eggplant, mustard, spinach and Swiss chard), yield (onion, potato, mustard, spinach and Swiss chard), chlorophyll composition of leaves and nutrient content (eggplant, onion, spinach and cabbage) and biotic and abiotic stresses (Pepper). The beneficial microorganisms including bacteria and fungi were also found to have good results in ameliorating the following characters: the growth (eggplant, tomato and rapeseed), yield (eggplant, tomato and rapeseed), and nutrient composition of the produce (eggplant, spinach, tomato, and rapeseed). Lastly, the protein hydrolysates also had a significant importance in increasing the photosynthetic rate and stomatal conductance (broccoli), Increased the lycopene and ascorbic acid (tomato). Having known all this, application of bio-stimulants should go hand in hand in the olericulture industry.

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