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Bio-stimulant: An innovative approach for climate smart agriculture

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Abstract
Sustainable agriculture requires innovative approaches to enhance crop productivity while minimizing environmental pollution, especially with an escalating population. Bio-stimulants are the substances or microorganisms applied to plants gives promising solution by enhancing nutrient efficiency, stress tolerance, and crop quality. These organic materials and microorganisms improve nutrient uptake, growth, and resilience to environmental stresses, offering a sustainable alternative to synthetic protectants. These are categorized as acids, microbes, plant-derived bioactive substances. Bio-stimulants support productive, resilient, and environmentally friendly farming. This review consolidates the current scientific understanding of bio-stimulants, examining their definition, modes of action, regulatory status, market dynamics, and potential to mitigate abiotic stresses such as salinity, drought, and extreme temperatures.

Keywords: Bio-stimulants, environmental impact, productivity, PGR, sustainable agriculture

Introduction
The agricultural sector has recently faced simultaneous challenges of raising productivity to feed the growing global population and increasing resource use efficiency while reducing the impact on ecosystems and human health. Fertilizers and pesticides play a vital role in agriculture, representing a powerful tool for growers to increase yield and guarantee continuous productivity but at the cost of soil deterioration. In the last few decades, several innovations have been made to enhance the sustainability of agricultural production systems by significantly reducing synthetic agrochemicals like pesticides and fertilizers. The growing need for food production through sustainable cultivation practices, without reducing crop yield and producer income, is a significant objective due to increased environmental pollution and the gradual degradation of cultivated soil. In the context of global climate change and food security, there is a need to grow crops and sustain the use of valuable and finite natural resources through biodiversity protection. Modern agriculture aims to reduce inputs without reducing the yield and quality. A promising and environmental-friendly innovation would be the use of natural plant bio-stimulants that enhance flowering, fruit set, plant growth, crop productivity, and nutrient use efficiency, and are able also to improve the tolerance against abiotic stress (Colla and Rouphael, 2015) [12]. A plant bio-stimulant is any substance or living microbe applied to plants to enhance nutrition efficiency, abiotic stress tolerance and crop quality traits. These are a diverse class of compounds that positively impact plant growth, yield and chemical composition and boost effects to biotic and abiotic stress tolerance. These are the substances or microorganisms applied to plants or soil to enhance crop quality traits, nutrient efficiency, nutrient uptake and tolerance to abiotic stress without being considered fertilizers or pesticides. Unlike fertilizers, which directly provide essential nutrients to plants, bio-stimulants enhance the plant's natural processes, improving its resilience and productivity. They are plant extracts and contain a wide range of bioactive compounds that are still unknown. The expected market growth in the bio-stimulant sector is at a compound annual growth rate of 11.24% and up to USD 4.9 billion by 2025 (Caradonia *et al.*, 2019) [9]. Therefore, there is increasing interest in the farming sector for new bio-stimulant products, and much research is carried out in this gradually evolving section of the industry.

Several commercial products are currently applied to various crops in sustainable and organic farming. Hence, bio-stimulant application can be considered an effective and sustainable nutritional crop supplementation and may alleviate the environmental problems associated with excessive fertilization. Based on this definition, PBs are classified as (i) Acids, (ii) protein hydrolysates, (iii) macroalgae seaweed extracts, and (iv) silicon, as well as beneficial microorganisms: (i) arbuscular mycorrhizal fungi (AMF) and (ii) N-fixing bacteria of strains belonging to the genera Rhizobium, *Azotobacter*, and Azospirillum (Rouphael and Colla, 2020) [34]. This review paper aims to provide an overview of the application of plant biostimulants and its effects on different crops within the framework of sustainable crop management, aiming to maximize the yield and quality of the final product. The review underscores the diverse effects of biostimulants on plant growth and stress resilience, with a particular focus on their ability to mitigate abiotic stresses such as salinity, drought and extreme temperature. Moreover, this paper explains the potential of biostimulants as a sustainable solution for enhancing agricultural productivity by exploring current research, case studies, and practical applications. Furthermore, it seeks to provide a comprehensive understanding of the role of biostimulants in modern farming practices and will showcase the efficacy of biostimulants in diverse agricultural contexts.

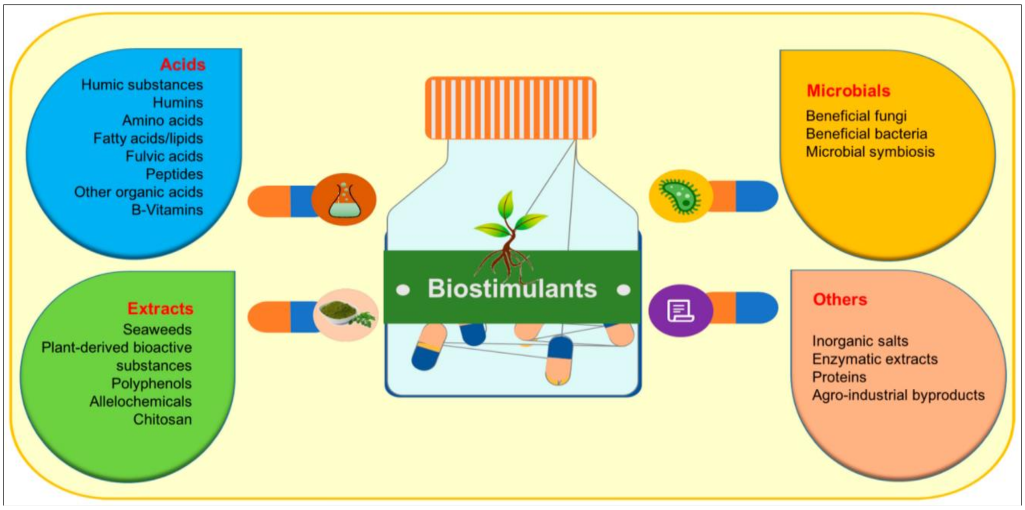

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Fig 1: Types of biostimulants

Major categories of biostimulants and their mechanism

1. Microbial Biostimulants

Microbial inoculants are commonly utilized as biostimulants because of their potential contribution to sustainable, green agriculture practices. The most prevalent types of microbial inoculants include *Trichoderma* spp., arbuscular mycorrhizal fungi (AMF), and plant growth-promoting *rhizobacteria* (PGPR). Under unfavorable environmental circumstances, the yield gap can be reduced by microbial biostimulant-induced plant growth enhancement, through improved biological N_2 fixation, solubilization of minerals and other nutrients, and increased plant access to soil resources (Pascale *et al.*, 2017) [16]. In addition, microbial inoculants produce volatile organic compounds (VOCs), while PGPR can improve plant abiotic stress tolerance by modulating different physiological processes (Hasanuzzaman *et al.*, 2021) [20].

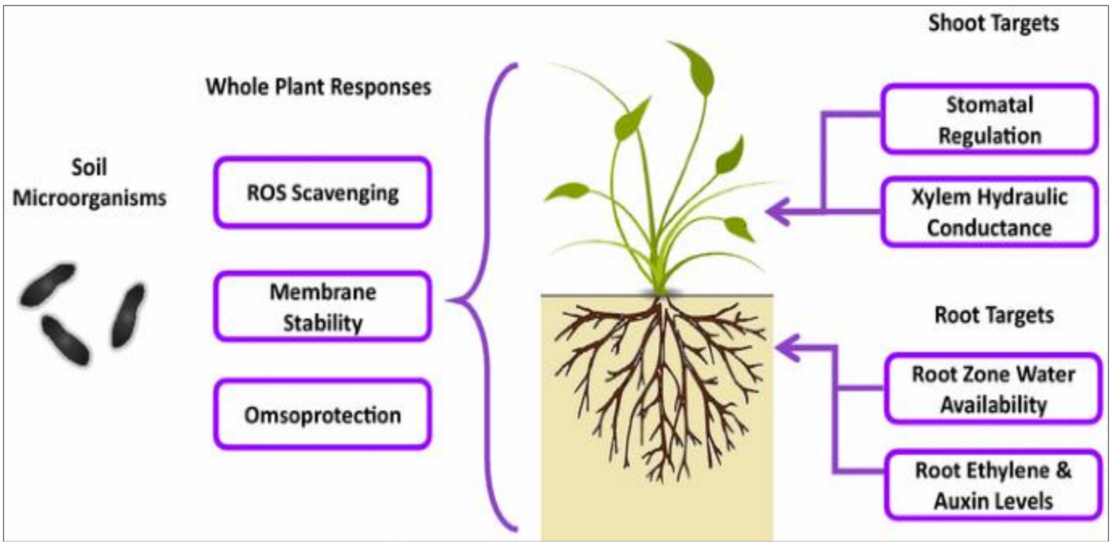

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Fig 2: Key mechanism targeted by microbial inoculation based biostimulants

Moreover, PGPR also induce the biosynthesis of plant hormones including auxins, ethylene, gibberellins, cytokinins, and ABA and thus contribute to stimulating growth, nutrient uptake, delayed leaf senescence, fruit and flower formation, seed maturation, and dormancy regulation. Bacteria with potential to act as biostimulants have been isolated from a number of ecosystems such as saline, alkaline, acidic, and arid soils. (Oosten *et al*., 2017) [40]. Members of these genera have developed strategies to adapt and thrive under adverse conditions (Upadhyay *et al*., 2009) [39]. Inoculation of maize with *Azotobacter* strains has been shown to have general positive effects under saline stress by facilitating uptake of K+ and exclusion of Na+ as well as increasing phosphorous and nitrogen availability (Rojas-Tapias *et al*., 2012) [33]. In wheat, inoculation of salt tolerance *Azobacter* strains increased biomass, nitrogen content, and grain yield under salt stress (Chaudhary *et al*., 2013) [10].

2. Acid based biostimulants

Humic substances (HS) are organic matter that naturally occurs in soil formed by decomposition of plant, animal and microbial residues and also from the metabolic activity of soil microbes. Humic substances (humic acid (HA); fulvic acid (FA); humins, fatty acids, amino acids, and organic acids comes under this group of biostimulants. Humic substances are mainly produced from soil organic matter not only from the decomposition process but also from microbial activity (Hasanuzzaman *et al*., 2021) [20]. Humic substances improve plant uptake of nutrients and water, which promotes plant growth and stress tolerance. These chemicals are useful at increasing plant abiotic stress tolerance because they boost water status, antioxidant capacity, and endogenous cytokinin levels. Humic materials lead to greater concentrations of Ca, Mg, P,N, K, Fe, S, Mn, and Cu, which are linked to improved salt tolerance, and lower concentrations of harmful Na. (Çimrin *et al*., 2010) [11]. Fulvic acids are the humic acids containing higher oxygen content and lower molecular weight (Bulgari *et al*., 2015) [8]. (Arjumend *et al*., 2015) [4] stated humic acid applied to wheat increased stem length by 18%, root length by 29%, leaf chlorophyll content by 96%, and soil organic matter content by 9%. In line with it, applying humic acid to durum wheat enhanced grain production by 21% when compared to the control. Extracts from vermicompost applied to rice (Oryza sativa L.) helps in activating anti-oxidative enzymatic function and increased ROS scavenging enzymes (Van Oosten *et al*., 2017) [40].

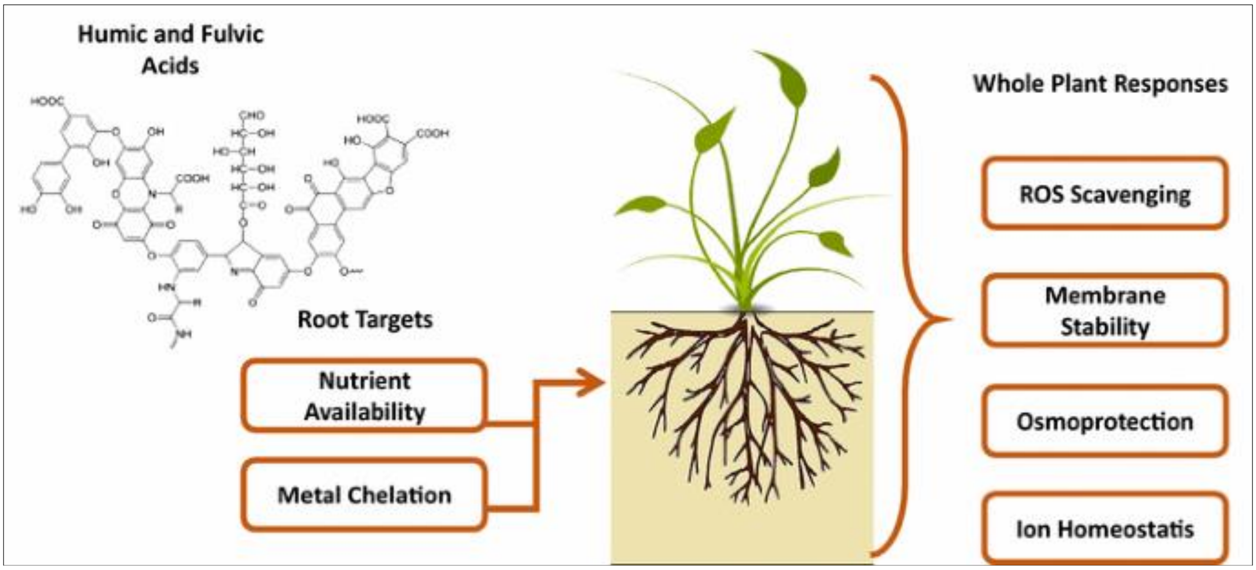

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Fig. 2.2: Key mechanism targeted by Humic and Fulvic acid based biostimulants.

3. Algal Extracts

This is a large category that includes many items made from many species, such as polyphenols, allelochemicals, seaweed, chitosan, and bioactive molecules obtained from plants. Seaweeds have long been employed as a source of organic matter and fertilizers, but more recently, their efficacy as biostimulants has been acknowledged. In commercial formulations, seaweed extracts (SWE) are being used as biostimulants to promote plant growth and enhance tolerance to heat, salinity, and drought. Seaweed extracts (SWE) offer a variety of application methods, such as foliar treatments and root zone application, and are rich in active minerals and organic compounds that effectively promote plant growth, photosynthetic activity, and abiotic stress tolerance. Therefore, they contribute to water retention and soil aeration, heavy metal fixation, and soil remediation. Plant-extracted materials have the potential to be employed in plant protection in addition to being used as food additives. (Seiber *et al*., 2014) [37] stated that wheat production metrics, biochemical parameters, and biometrics were all significantly impacted by *Ascophyllum nodosum* extract. The height, dry mass of shoots, and number of spikes in plants watered with *Ascophyllum nodosum* extract increased by 11.72%, 22.22%, and 13.19%, respectively. Similarly, (Al- Hasany *et al*., 2019) [3] explained that using seaweed extracts up to 2 g.l-1 increased the number of spikes/m2, grain production, and biological yield by 31.85%, 39.05%, and 39.79%, respectively.

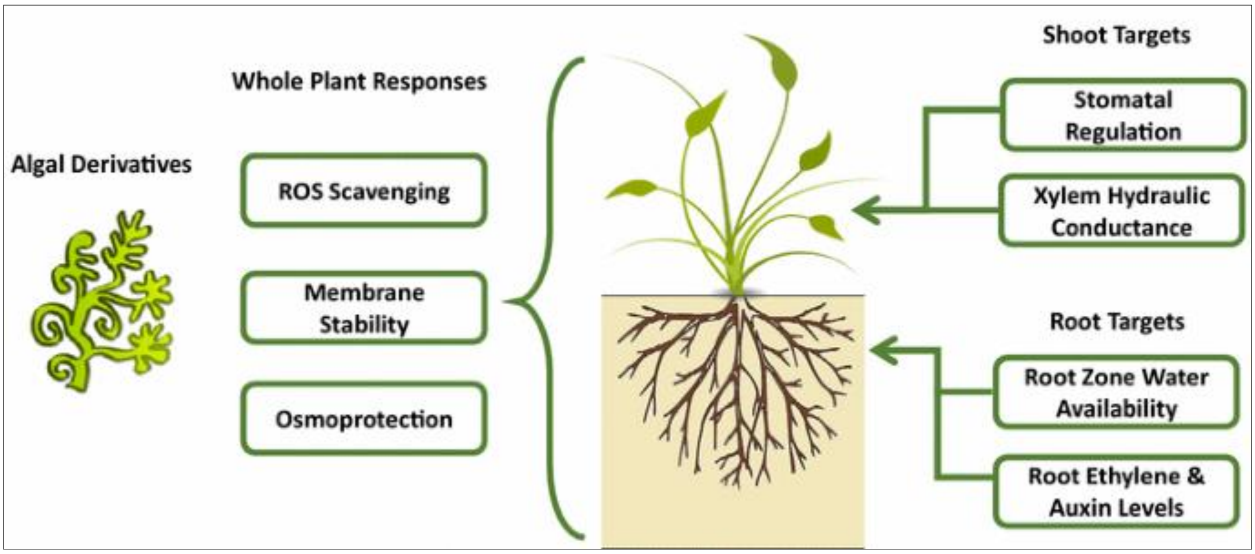

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Fig 2.3: Key mechanism targeted by algal-based biostimulants

4. Others (Inorganic-, carbohydrate-, protein-, amino acid-, and lipid-based biostimulants)

Lipids, proteins, amino acids, and carbohydrates can all improve stress tolerance. Plant growth regulators are frequently included in the formulations of protein hydrolysates (PH) that are commercialized. Over 90%, are produced from chemical hydrolysis of animal by-products while enzymatically processed plant-based products are a recent development (Colla *et al*., 2015) [12, 13]. Amino-acids and peptides mixtures are obtained by chemical and enzymatic protein hydrolysis from agroindustrial by-products, from animal wastesand plant sources (crop residues) (e.g. collagen, epithelial tissues) (du Jardin, 2012) [18]. The five main beneficial elements are Na, Se, Al, Co, and Si, present in soils and in plants as different inorganic salts and as insoluble forms lik amorphous silica ($SiO_2.nH_2O$) in graminaceaous species. These beneficial functions can be constitutive, like the strengthening of cell walls by silica deposits, or expressed in defined environmental conditions, like pathogen attack for selenium and osmotic stress for sodium. Indirect effects on plant nutrition and growth are also important in the agricultural practice when protein hydrolysates are applied to plants and soils. Protein hydrolysates are known to increase microbial biomass and activity, soil respiration and, overall, soil fertility. Chelating and complexing activities of amino acids and peptides contribute to nutrients availability and acquisition by roots.

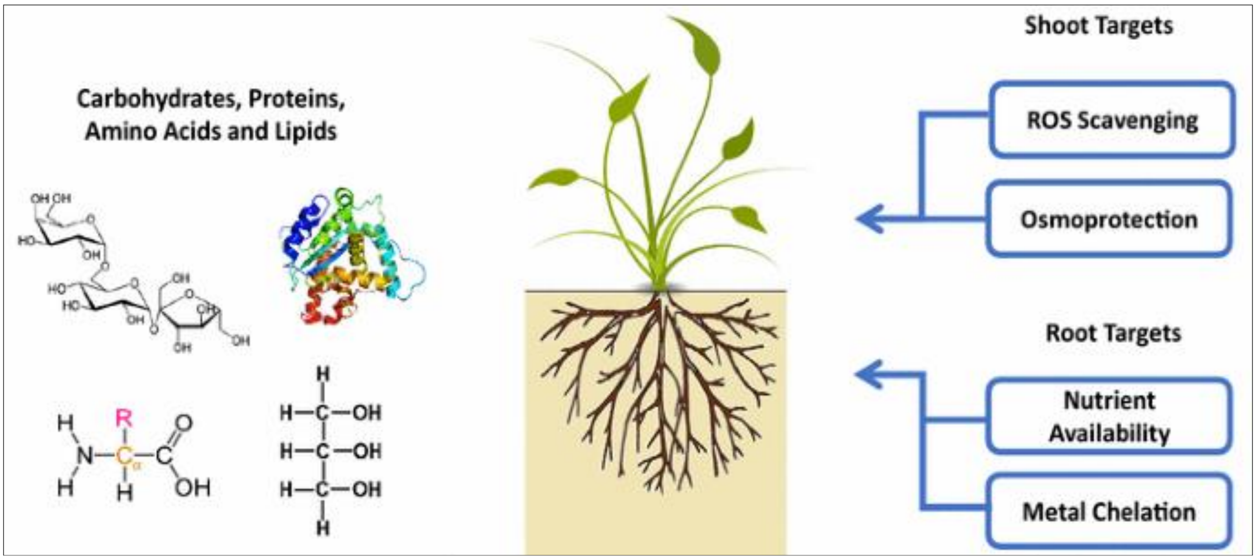

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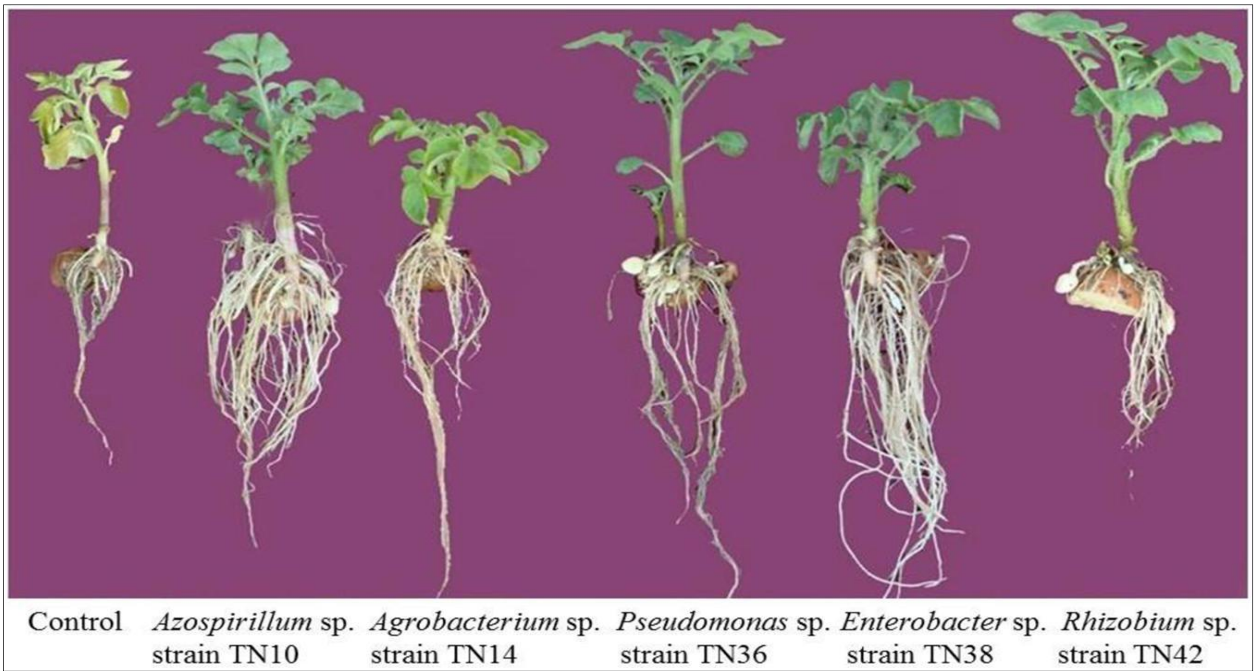
Fig 2.4: Key mechanism targeted by carbohydrate-, protein-, amino acid-, and lipid-based biostimulants.

Effects of different biostimulants on various crops

1. Microbial inoculants

(Ram *et al*., 2005) [31] stated that the inoculation with *Azotobacter* strains had significant effect on growth parameters of wheat. The inoculation with Mac-27 significantly increased plant height to maximum i.e. 79.7 cm and gave highest leaf area index to 4.40 than control at all the stages of crop growth. The highest dry matter was produced with the inoculation of seed with Mac-27. In line with it (Baral & Adhikari, 2013) [6] explained that inoculation of *Azotobacter* increased 15 to 35% grain yield over non inoculated treatments. Similarly, (Katiyar *et al*., 2011) [25], stated that if the wheat seed was inoculated with *Azotobacter* it increases the yield up to 1.92 – 2.0% as compared to non-inoculated seed. (Jaga *et al*., 2017) [22] concluded that with *Azotobacter* inoculation, significant increase was seen for protein output (632.7 to 678.0 kg ha-1) and grain protein content (12.95 to 13.22%). *Azotobacter* nitrogen uptake by the crop and the amount of available nitrogen in post harvest soil increased significantly with *Azotobacter* inoculation over no inoculation. (Kaur *et al*., 2018) [26] described that application of *Azotobacter* gave the maximum net returns of Rs. 277684.1 ha-1 and maximum benefit: cost ratio 1.65 is also observed in T8. (Mehboob *et al*., 2011) [21] isolates some strains of rhizobium from chickpea, lentil and mung been nodules. The results indicated improved plant height (up to 18.66%), tillers per plant (up to 68.76%), straw yield (up to 35.14%), grain yield (up to 30.29%), 1000-grain weight (up to 28.40%), root length (up to 51.72%),% N in grains and straw (up to 15.07 and 33.16%),% P in grains and straw (up to 23.39 and 66.66%) and% K in grains and straw (up to 51.72 and 21.80%) compared with un-inoculated control. (Sajid *et al*., 2011) [35] concluded that the maximum plant height (88.43 cm), number of shoots (19.9 plant^{-1}), number of leaves (173.27 plant^{-1}), number of pods (79.8 plant^{-1}), number of nodules (156.27 plant^{-1}), yield (252.66 g plant^{-1}) and production (1856 kg ha^{-1}) was observed in synthetic rhizobium inoculated seeds. (Ojha *et al*., 2008) [30] concluded that the symbiotic relationship between the mycorrhizal fungus and the VAM-treated plants results in higher phosphorus uptake and higher levels of chlorophyll content, which are subsequently maintained throughout the plants' developmental stages. Several growth characteristics, such as the plant's height and the fresh and dry weight the roots and shoots were observed to be significantly high in VAM (*G. fasciculatum*) treated plants compared to the respective controls. In a pot experiment carried out by (Reddy *et al*., 2018) [32], concluded that the strains of *Azotobacter* sp. and *Azospirillum* sp. possess great potential to be developed as biofertilizer to enhance soil fertility and plant growth. Maximum germination (%) 90, plant height 51cm, Leaf area 59cm^2, Branches per plant 8.66 and Leaf per branch 17.33cm were obtained by inoculation with these strains as compared with control.

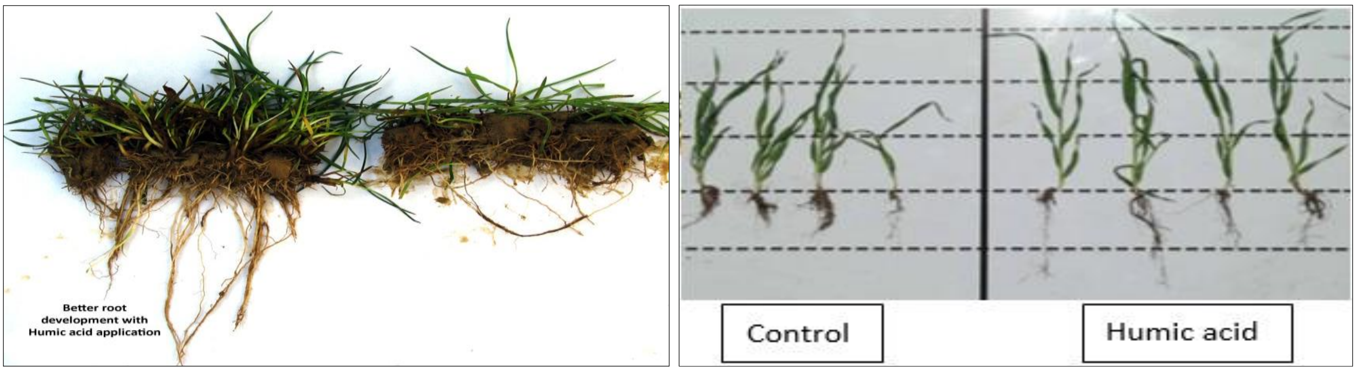


(Source: https://www.frontiersin.org/journals/plant-science/articles/10.3389/fpls.2016.00144/full**)**

Fig 3.1. Effect of different microbial biostimulants on root of potato

2. Acids

Kandil *et al*., (2016) [23] highlighted that when humic acid was applied topically to wheat grains, the protein and starch ratios increased by 7-8% and 2-3%, respectively, in comparison to the control whereas humic acid used as foliar spray improves plant height, grain yield, protein and gluten content, and 1000-grain weight Kara, (2015) [24]. According to Elshabrawi *et al*., (2015) [19], increasing the dose of foliar humic acid spray resulted in an increase in the grain yield from 2990.0 to 3414.1 kg ha^{-1} and an increase in the grain protein ratio of wheat from 10.1 to 11.41%. Similarly Merwad, (2019) [27] showed that humic acid applied to durum wheat significantly increased the plant height, grain yield, and grain weight. In contrary Arjumend *et al*., (2015) [4] also pointed out that humic acid applied to wheat increased stem length by 18%, root length by 29%, leaf chlorophyll content by 96%, and soil organic matter content by 9%. (Braziene *et al*., 2021) [7] concluded that the length and air-dry weight of spring wheat and barley shoots, as well as their roots, were both boosted by Fulvic acid. Using Fulvic acid during plant vegetation consistently enhanced the yield of sugar beet roots and spring wheat grains while also improving the quality of the harvest. Fulvic acid applied as a seed dressing in spring wheat, spring barley, and sugar beet consistently increased the percentage of final germination and decreased the mean germination time. (Suh *et al*., 2014) [38] stated that fulvic acid could be used to promote plant growth and increase marketable yield in tomato production. (Al-Haidary *et al*., 2020) [2] concluded that the spraying of fulvic acid at 6 mg L-1 was significantly superior and had the highest means of plant height (80.89 cm), number of grain (44.09 grain spike-1), spike weight (3.50 g spike-1), 1000 grain weight (33.60 g), grain yield (4.162 ton ha1) and biological yield (17.14 ton ha-1).

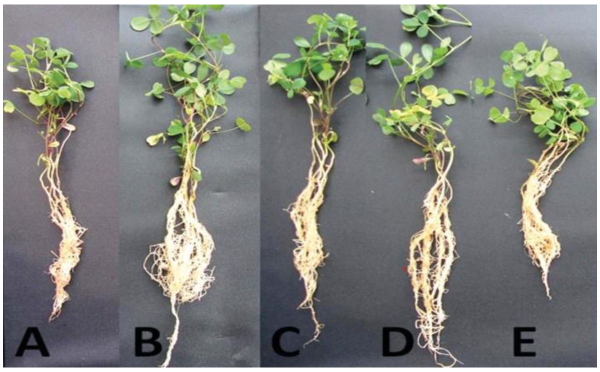


(Source: https://www.humicchina.com/blog/humic-acid-can-stimulate-the-plants-brain.html), (https://www.google.com/url?sa=i&url=https%3A%2F%2Fcanariashorizontedesalud.itop.es%2F%3Fm%3Dthe-benefits-of-humic-acid-to-turfgrass-soil-environments-qq-Pk3aAQFd&psig=AOvVaw3mFLQ4S32HS7DIdvDheTZ&ust=1713163796966000&source=images&cd=vfe&opi=89978449&ved=0CAQQjB1qFwoTCOCYmM6OwYUDFQAAAAAdAAAAABAs)

Fig 3.2: Effect of humic acid biostimulants on root and shoot of tuffgrass

3. Algal extracts

Carvalho *et al.*, (2013) [45] stated that wheat growth and yield, biochemical parameters, and biometrics were all significantly impacted by *Ascophyllum nodosum* extract. The plant height, dry mass of shoots, and number of spikes in plants watered with *Ascophyllum nodosum* extract increased by 11.72%, 22.22%, and 13.19%, respectively. Hasany *et al.*, (2019) [3] explained that compared to the control treatment, seaweed extracts up to 2 g/l increased the number of spikes/m^2, grain production, and biological yield by 31.85%, 39.05%, and 39.79%, respectively. Deepana *et al.*, (2021) [17] on rice explained that higher plant height (121.1 cm), dry matter production (11390 kg/ha), number of grains per panicle (166), panicle length (21.8 cm), thousand-grain weight (14.7g), number of productive tillers per m^2 (275), grain yield (5612 kg/ha), and straw yield (7829 kg/ha) were observed when Sea weed extract was applied at a rate of 12.5 kg/ha in combination with a foliar spray of seaweed liquid. Abdel *et al.*, (2023) came to the conclusion that the tallest barley plants (78.48 cm), most spikes/m^2 (282.42), most spikelets /spike (46.09), heaviest 1000-grain weight (35.82 g), highest grain and biological yields (3.87 and 9.06 t/fed., respectively), highest harvest index, and highest grain protein content (42.72% and 8.88%, respectively) were all produced by applying the foliar spray of highest seaweed level (6 l/fed). Although the use of seaweed extracts can be very positive, it is also worth mentioning that it is very dependent on the quality and consistency of the extract being produced.



(Source: https://scienceinhydroponics.com/2020/11/the-effect-of-seaweed-kelp-extracts-in-plants.html?print=print)

Fig 3.3: Effect of algal extracts biostimulants on root and shoot A (Control), B (Ascophyllum nodosum), C (Methanol extract), D (Chloroform), E (Ethyl acetate)

4. Others (Inorganic-,carbohydrate-, protein-, amino acid-, and lipid-based biostimulants)

(Zhang *et al.*, 2023) [42] explained that foliar biostimulants (Protein hydrolysate (PH), seaweed extracts (SW)) reduce the detrimental effects of salinity stress on tomato plants at both the physiological and agronomic levels. They varied the buildup of organic acids and the mineral makeup of tomato leaves. When used, it actively changed key metabolic pathways that are involved in the stress response. (Popko *et al.*, 2018) [43] The application of products based on amino acids influenced the increase of grain yield of winter wheat (5.4% and 11%), respectively for the application of Amino Prim at a dose 1.0 L/ha and Amino Hort at dose 1.25 L/ha) when compared to the control group without biostimulant. It also showed an increase of technological characteristics of grain such as ash content, Zeleny sedimentation index and content of protein. (Ertani *et al.*, 2017) [44] concluded that applying protein hydrolysate improved root growth, photosynthetic rate, and the concentration of soluble sugars and chlorophyll and phenolic content. It also regulated genes linked to antioxidant activity, photosynthesis, nutrient uptake, and primary metabolisms.

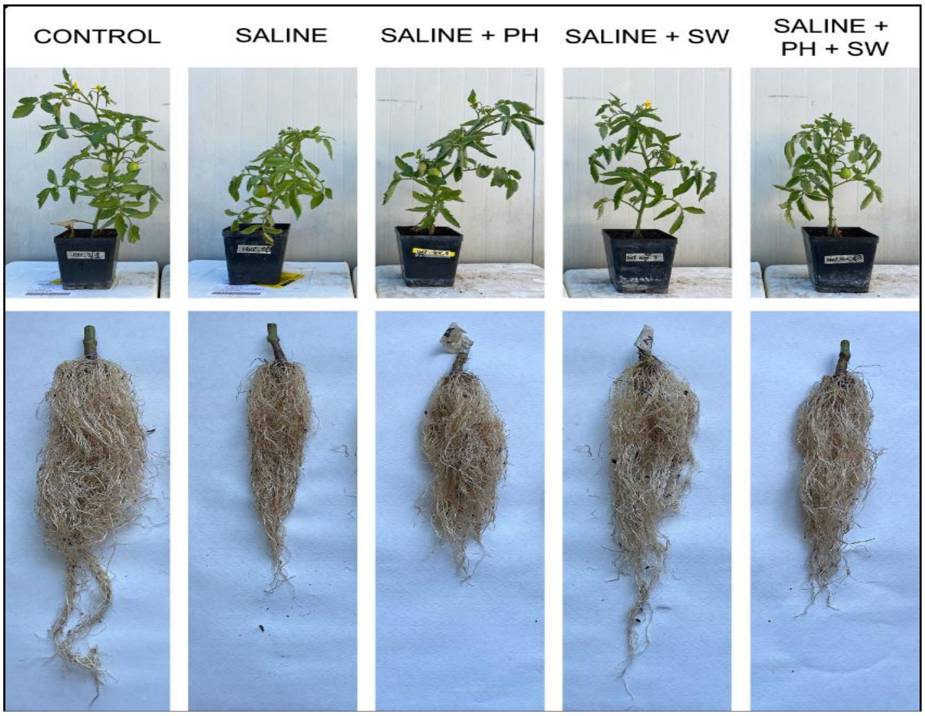


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Fig 3.4: Effect of Protein hydrolysate (PH), Seaweed extracts (SW) biostimulants on root and shoot

Applications and Advantages of biostimulants

- **Increased Crop Yield:** Studies have shown that the application of biostimulants can lead to significant improvements in crop yield, often surpassing the effects of traditional fertilizers alone.
- **Reduced Environmental Impact:** Biostimulants are derived from natural sources and generally have minimal adverse effects on the environment compared to synthetic chemicals. They promote sustainable agricultural practices by reducing the reliance on chemical inputs.
- **Enhanced Soil Health:** Many biostimulants contribute to the enrichment of soil microbiota and organic matter, thereby improving soil structure, fertility, and long-term productivity.
- **Improved Nutrient Uptake:** Biostimulants can enhance the plant's ability to absorb and assimilate nutrients from the soil, thereby optimizing nutrient utilization efficiency.
- **Stress Tolerance:** Biostimulants contain compounds that help plants cope with environmental stresses such as salinity, drought,and extreme temperatures. They stimulate the production of stress-related proteins and metabolites, enabling plants to thrive under adverse conditions.

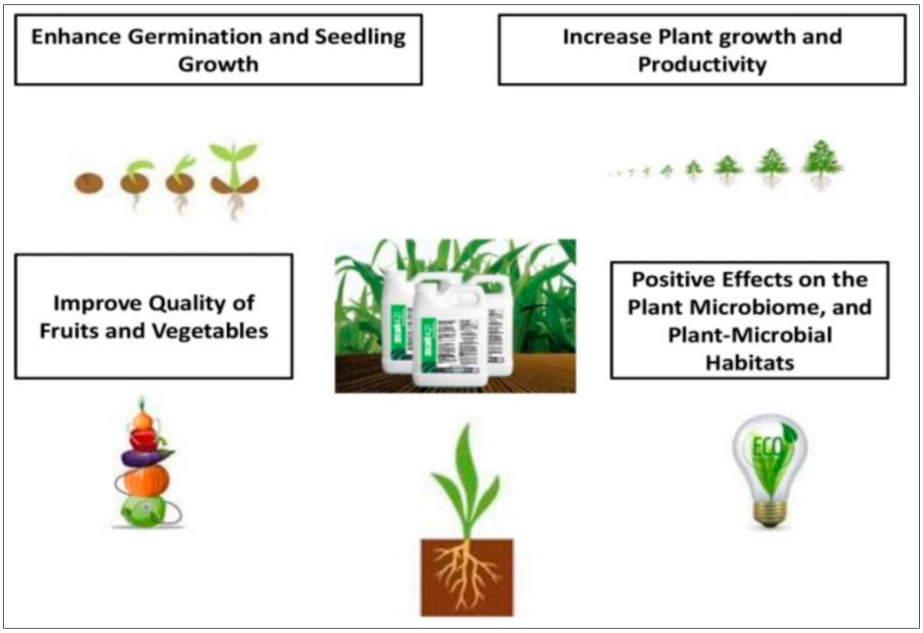

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Fig 4.1: Advantages of biostimulants

- **Root Development:** Certain biostimulants promote root growth and development, thereby enhancing the plant's capacity to access water and nutrients from the soil.
- **Plant Metabolism:** Biostimulants can influence various metabolic processes within plants, leading to improved photosynthesis, hormone regulation, and overall growth.
- **Resilience to Climate Change:** By enhancing stress tolerance and promoting robust growth, biostimulants help mitigate the negative impacts of climate change on crop production.
- **Compatibility with Organic Farming:** Biostimulants are often approved for use in organic farming systems, providing organic growers with effective tools for optimizing crop performance while adhering to organic standards.

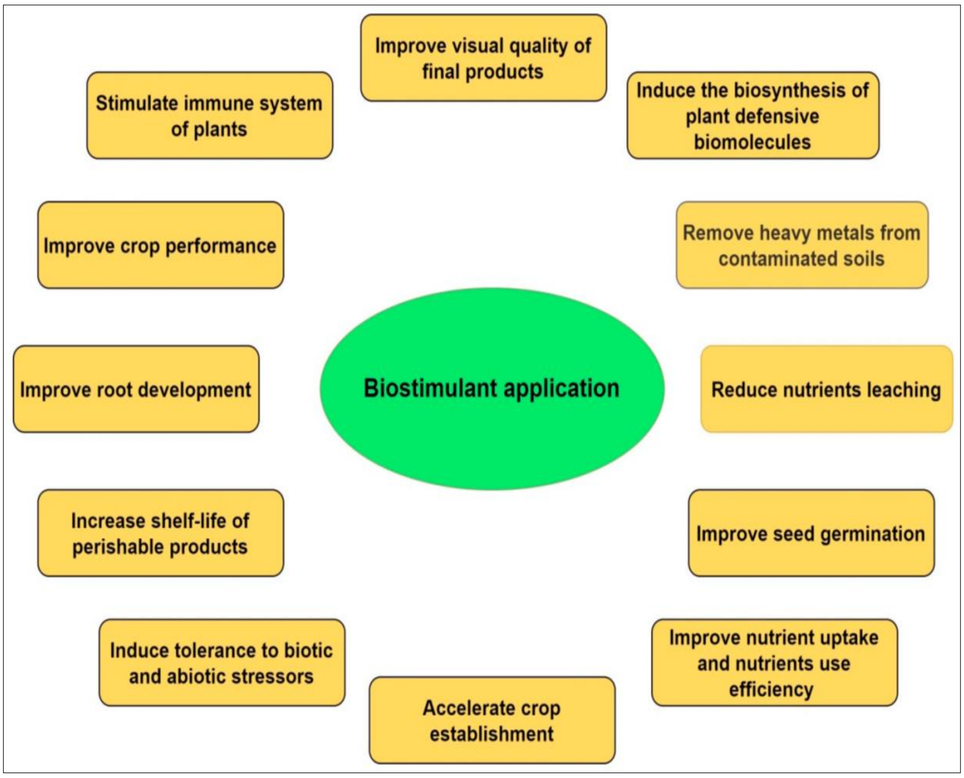

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Fig 4.2: Applications of biostimulants.

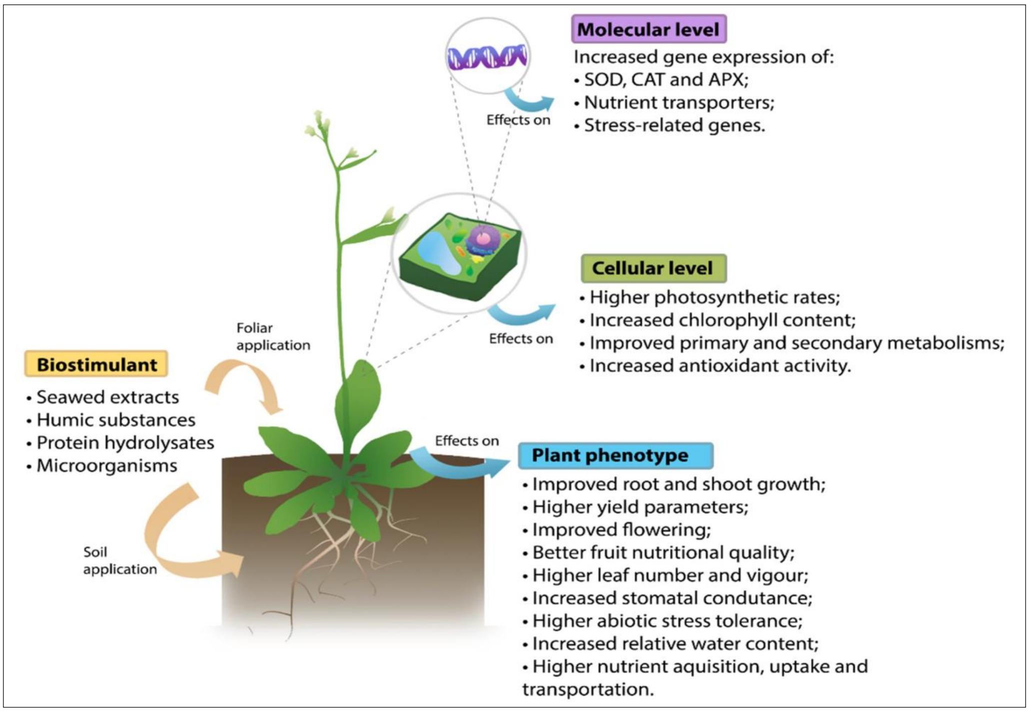

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Fig 5.1: Summarized effects of biostimulants in plants at the molecular and cellular level, and in the plant phenotype.

Conclusion

The integration of plant biostimulants (PBs) into modern agricultural practices offers a promising pathway towards sustainable and resilient crop production systems. PBs, defined as substances or microorganisms aimed at enhancing crop quality traits, nutrition efficiency and abiotic stress tolerance, emerged as a key solution. Biostimulants hold immense promise for enhancing the resilience of agricultural crops to environmental stresses. They have the capability to increase the yield of produce along with maintaining soil properties without harming the environment. Unlike conventional agricultural inputs such as synthetic fertilizers and pesticides, biostimulants offer a sustainable alternative that aligns with the principles of environmental management and resource efficiency. Moreover, the growing market for biostimulants reflects increasing interest and recognition within the farming sector of their potential benefits. Also, PBs demonstrate efficacy not only in enhancing yield and quality but also in mitigating abiotic stresses such as salinity, drought and extreme temperature. This resilience capacity is crucial due to climate change causing environmental uncertainties, where maintaining crop productivity under adverse conditions becomes vital.

This review paper presented here highlights the effects, potential applications and advantages of biostimulants in agriculture. Moreover, this paper underscores the significant potential of plant biostimulants as a sustainable solution for enhancing agricultural productivity and resilience. By embracing biostimulants along with sustainable crop management, it can be used for maximizing yields, improving product quality, and safeguarding the environment for future generations.

References

1. Abate T, Dessie A, Mekie T. Technical efficiency of smallholder farms in red pepper production in North Gondar Zone Amhara Regional State, Ethiopia. J Econ Struct. 2019;8:18-32.
2. Al-Haidary HKMA, Al-Zubaidy SAA. Behavior of growth and yield bread wheat by the influence of fulvic acid and seeding rate. Systematic Reviews in Pharmacy. 2020;11(9):606-612.
3. Al-Hasany AR, Aljaberi MA, Alhilfi SK. Effect of spraying with seaweed extract on growth and yield of two varieties of wheat (*Triticum aestivum* L.). Basrah J Agric Sci. 2019;32:124-134.
4. Arjumend T, Abbasi MK, Rafique E. Effects of lignite-derived humic acid on some selected soil properties, growth and nutrient uptake of wheat (*Triticum aestivum* L.) grown under greenhouse conditions. Pak J Bot. 2015;47(6):2231-2238.
5. AS AA, NA S, AA M. Effect of humic acid and seaweed extract rates on yield and yield components of barley (*Hordeum vulgare* L.). Alex Sci Exch J. 2023;44(3):459-463.
6. Baral BR, Adhikari P. Effect of Azotobacter on growth and yield of maize. Annals of Plant and Soil Research. 2013;19(1):141-147.
7. Braziene Z, Paltanavicius V, Avizienytė D. The influence of fulvic acid on spring cereals and sugar beets seed germination and plant productivity. Environ Res. 2021;195:110824.
8. Bulgari R, Cocetta G, Trivellini A, Vernieri PAOLO, Ferrante A. Biostimulants and crop responses: a review. Biol Agric Hortic. 2015;31(1):1-17.
9. Caradonia F, Battaglia V, Righi L, Pascali G, La Torre A. Plant biostimulant regulatory framework: prospects in Europe and current situation at international level. J Plant Growth Regul. 2019;38:438-448.
10. Chaudhary D, Narula N, Sindhu SS, Behl RK. Plant growth stimulation of wheat (*Triticum aestivum* L.) by inoculation of salinity tolerant Azotobacter strains. Physiol Mol Biol Plants. 2013;19:515-519.
11. Çimrin KM, Türkmen Ö, Turan M, Tuncer B. Phosphorus and humic acid application alleviate salinity stress of pepper seedling. Afr J Biotechnol. 2010;9(36):5913-5920.
12. Colla G, Rouphael Y. Biostimulants in horticulture. Scientia Horticulturae. 2015;196:1-134.
13. Colla G, Nardi S, Cardarelli M, Ertani A, Lucini L, Canaguier R, *et al*. Protein hydrolysates as biostimulants in horticulture. Scientia Horticulturae. 2015;196:28-38.
14. Cozzolino E, Di Mola I, Ottaiano L, Nocerino S, Sifola M, El-Nakhel C, *et al*. Can seaweed extract improve yield and quality of brewing barley subjected to different levels of nitrogen fertilization? Agronomy. 2021;11(12):2481.
15. de Carvalho MEA, de Camargo PR, Gallo LA, Junior MVCF. Seaweed extract provides development and production of wheat. Agrarian. 2014;7(23):166-170.
16. De Pascale S, Rouphael Y, Colla G. Plant biostimulants: Innovative tool for enhancing plant nutrition in organic farming. Eur J Hortic Sci. 2017;82(6):277-285.
17. Deepana P, Bama KS, Santhy P, Devi TS. Effect of seaweed extract on rice (*Oryza sativa* var. ADT53) productivity and soil fertility in Cauvery delta zone of Tamil Nadu, India. J Appl Nat Sci. 2021;13(3):1111-1120.
18. du Jardin P. The Science of Plant Biostimulants–A bibliographic analysis. Ad hoc study report; c2012.
19. El-Shabrawi HM, Bakry BA, Ahmed MA, Abou-El-Lail M. Humic and oxalic acid stimulates grain yield and induces accumulation of plastidial carbohydrate metabolism enzymes in wheat grown under sandy soil conditions. Agric Sci. 2015;6(1):175-184.
20. Hasanuzzaman M, Parvin K, Bardhan K, Nahar K, Anee TI, Masud AAC, Fotopoulos V. Biostimulants for the regulation of reactive oxygen species metabolism in plants under abiotic stress. Cells. 2021;10(10):2537.
21. Ijaz Mehboob IM, Zahir ZA, Muhammad Arshad MA, Asif Tanveer AT, Farooq-e-Azam FEA. Growth promoting activities of different *Rhizobium* spp. in wheat; c2011.
22. Jaga PK, Sharma S, Patel Y. Response of wheat (*Triticum aestivum*) to Azotobacter inoculation and nitrogen in soils of Vidisha, Madhya Pradesh. Ann Plant Soil Res. 2017;19(1):42-45.
23. Kandil AA, Sharief AEM, Seadh SE, Altai DSK. Role of humic acid and amino acids in limiting loss of nitrogen fertilizer and increasing productivity of some wheat cultivars grown under newly reclaimed sandy soil. Int J Adv Res Biol Sci. 2016;3(4):123-136.
24. Kara B. Effect of alternative fertilizer on some physicochemical characteristics in wheat flour. Ziraat

Fakültesi Dergisi-Süleyman Demirel Üniversitesi. 2015;10(2):34-39.
25. Katiyar NK, Rarawat S, Pathak RK, Kumar A. Effect of Azotobacter and nitrogen levels on yield and quality of wheat. Ann Plant Soil Res. 2011;13(2):152-155.
26. Kaur R, Kumar S, Kaur R, Kaur J. Effect of integrated nutrient management on growth and yield of wheat (*Triticum aestivum* L.) under irrigated conditions. Int J Chem Stud. 2018;6(4):1800-1803.
27. Merwad ARM. Using humic substances and foliar spray with moringa leaf extract to alleviate salinity stress on wheat. Sustainability of Agricultural Environment in Egypt: Part II: Soil-Water-Plant Nexus. 2019, 265-286.
28. Tufail MMT, Nawaz KKN, Muhammad Usman MU. Impact of humic acid on the morphology and yield of wheat (*Triticum aestivum* L.). [Journal Name Missing]
29. Naqqash T, Hameed S, Imran A, Hanif MK, Majeed A, van Elsas JD. Differential response of potato toward inoculation with taxonomically diverse plant growth promoting rhizobacteria. Front Plant Sci. 2016;7:164522.
30. Ojha S, Chakraborty MR, Dutta S, Chatterjee NC. Influence of VAM on nutrient uptake and growth of custard-apple. Asian J Exp Sci. 2008;22(3):221-224.
31. Ram T, Yadav SK, Sheoran RS. Growth analysis of wheat (*Triticum aestivum* L.) under varying fertility levels and Azotobacter strains. Indian J Agric Res. 2005;39(4):295-298.
32. Reddy S, Singh AK, Masih H, Benjamin JC, Ojha SK, Ramteke PW, *et al*. Effect of *Azotobacter* sp and *Azospirillum* sp on vegetative growth of tomato (*Lycopersicon esculentum*). J Pharmacogn Phytochem. 2018;7(4):2130-2137.
33. Rojas-Tapias D, Moreno-Galván A, Pardo-Díaz S, Obando M, Rivera D, Bonilla R. Effect of inoculation with plant growth-promoting bacteria (PGPB) on amelioration of saline stress in maize (*Zea mays*). Appl Soil Ecol. 2012;61:264-272.
34. Rouphael Y, Colla G. Biostimulants in agriculture. Front Plant Sci. 2020;11:511937.
35. Sajid M, Rab A, Wahid F, Shah SNM, Jan I, Khan MA, *et al*.. Influence of rhizobium inoculation on growth and yield of groundnut cultivars. Sarhad J Agric. 2011;27(4):573-576.
36. Sayed WH, Dawood RA, Abd El-Rahman KA, El-Morshidy MA, Galal AH. Impact of humic acid and nitrogen fertilization on productivity of some bread wheat cultivars. Assiut J Agric Sci. 2019;50(4):22-34.
37. Seiber JN, Coats J, Duke SO, Gross AD. Biopesticides: state of the art and future opportunities. J Agric Food Chem. 2014;62(48):11613-11619.
38. Suh HY, Yoo KS, Suh SG. Effect of foliar application of fulvic acid on plant growth and fruit quality of tomato (*Lycopersicon esculentum* L.). Hort Environ Biotechnol. 2014;55:455-461.
39. Upadhyay SK, Singh DP, Saikia R. Genetic diversity of plant growth promoting rhizobacteria isolated from rhizospheric soil of wheat under saline condition. Curr Microbiol. 2009;59:489-496.
40. Van Oosten MJ, Pepe O, De Pascale S, Silletti S, Maggio A. The role of biostimulants and bioeffectors as alleviators of abiotic stress in crop plants. Chem Biol Technol Agric. 2017;4:1-12.
41. Yakhin OI, Lubyanov AA, Yakhin IA, Brown PH. Biostimulants in plant science: A global perspective. Front Plant Sci. 2017;7:238366.
42. Zhang L, Freschi G, Rouphael Y, De Pascale S, Lucini L. The differential modulation of secondary metabolism induced by a protein hydrolysate and a seaweed extract in tomato plants under salinity. Front Plant Sci. 2023;13:1072782.
43. Popko M, Michalak I, Wilk R, Gramza M, Chojnacka K, Górecki H. Effect of the new plant growth biostimulants based on amino acids on yield and grain quality of winter wheat. Molecules. 2018;23(2):470.
44. Ertani A, Schiavon M, Nardi S. Transcriptome-wide identification of differentially expressed genes in *Solanum lycopersicon* L. in response to an alfalfa-protein hydrolysate using microarrays. Front Plant Sci. 2017;8:260167.
45. Carvalho CM, Pehlivan D, Ramocki MB, Fang P, Alleva B, Franco LM, *et al*. Replicative mechanisms for CNV formation are error prone. Nature genetics. 2013 Nov;45(11):1319-1326.