International Research Journal of Pure & Applied Chemistry

21(14): 20-35, 2020; Article no.IRJPAC.60331
ISSN: 2231-3443, NLM ID: 101647669

Biostimulants: Potential and Prospects in Agriculture

Ingudam Bhupenchandra[1*], Soibam Helena Devi[2], Anjali Basumatary[3], Samiron Dutta[3], Laishram Kanta Singh[4], Prakash Kalita[2], S. S. Bora[5], S. Roma Devi[6], Amarjit Saikia[2], Priyanka Sharma[2], Seema Bhagowati[3], Babita Tamuli[3], Namita Dutta[3] and Kripal Borah[3]

[1]ICAR-KVK, Tamenglong, ICAR-RC for NEH Region, Manipur Centre, India.
[2]Department of Crop Physiology, Assam Agricultural University, Jorhat, India.
[3]Department of Soil Science, Assam Agricultural University, Jorhat, India.
[4]ICAR-KVK Imphal West, ICAR-RC for NEH Region, Manipur Centre, India.
[5]ICRI, RRS, Spices Board, Tadong, Gangtok, India.
[6]ICAR-KVK Churchandpur, ICAR-RC for NEH Region, Manipur Centre, India.

Authors' contributions

This work was carried out in collaboration among all authors. Author IB did the conceptualization, methodology formulation and implementation. Author SHD reviewed the study, did the rigorous editing of original and revised versions of the manuscript. Author AB visualized, reviewed and edited the manuscript. Authors SD and ND edited the manuscript. Author LKS prepared the original draft and revised the draft. Authors PK and SRD did the results compilation. Authors SSB, AS and KB prepared the manuscript. Authors PS, SB and BT reviewed the compilation. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/IRJPAC/2020/v21i1430244
Editor(s):
(1) Dr. Hao-Yang Wang, Shanghai Institute of Organic Chemistry, China.
Reviewers:
(1) Nain Peralta Antonio, Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias (INIFAP), Mexico.
(2) Juan Manuel Soto Parra, Autonomous University of Chihuahua, Chihuahua, Mexico.
Complete Peer review History: http://www.sdiarticle4.com/review-history/60331

Review Article

Received 19 June 2020
Accepted 24 August 2020
Published 03 September 2020

ABSTRACT

In the last thirty years, numerous scientific revolutions have been planned to improve the ecological balance of agricultural production methods, via a considerable decrease of inorganic compounds like pesticides, synthetic plant growth hormones and fertilizers. A favorable and environment sustainable modernization should be the practice of normal plant biostimulants (PBs) which

**Corresponding author: E-mail: ibhupenj@gmail.com;*

augment flowering, plant evolution, fruit formation, crop output and efficient nutrient mobilization, and ability to endure a varied array of abiotic stressors. PBs was primarily deciphered via exclusive of various functions similar to fertilizers or plant protection by-products. They are assorted constituents and microbes resorted to boost plant development. This review aimed to present the plant biostimulants definition, major classifications, and impacts on floras, soil and ecology.

Keywords: Protein; hormone; nitrogen.

1. INTRODUCTION

Biostimulants are excerpts acquired from organic raw ingredients having bioactive composites. Plant biostimulant is well-defined as components, with the exception of nutrients and pesticides, which, when subjected to plants, seeds or growing substrates in precise formulations have the ability to modify the physiological actions of plants in such way that caters prospective edge to growth, development, and/or stress response [1]. There are various classes of biostimulants, viz., peptides, amino acids, polysaccharides, humic acids, phytohormones, etc. The constituents of the biostimulants are mineral elements, humic substances (HSs), vitamins, amino acids, chitin, chitosan, and poly- and oligosaccharides [2,3,4]. The meaning of PBs has been strictly deliberated over the preceding ten years, and lately underneath the new Regulation (EU) 2019/ 1009, which directed to the following: "A plant biostimulant shall be an EU fertilizing artifact the purpose is to enhance plant nutrition courses self-reliantly of the artifact's nutrient content with the exclusive objective of refining one or more of the ensuing traits of the plant or the plant rhizosphere: i) nutrient utilization efficacy, ii) toleration to abiotic stress, iii) quality traits, or iv) accessibility of limited nutrients in the soil or rhizosphere" [5]. Established on this delineation, PBs are identified on the basis of agricultural utilities, and comprise varied bioactive inherent elements: (i) humic and fulvic acids, (ii) animal and vegetal protein hydrolysates, (iii) macroalgae seaweeds extracts, and (iv) silicon, besides beneficial microbes: (i) arbuscular mycorrhizal fungi (AMF) and (ii) N-fixing bacteria of strains belonging to the genera *Rhizobium, Azotobacter,* and *Azospirillum*.

2. CHARACTERISTICS

The genre of the biostimulant is not limiting: it can be a substance or a microbe. A substance can be a distinct chemical compound or a cluster of compounds with a firm biological origin, e.g. plant excerpts, not essentially an entirely categorised configuration. This way, it apts with the significance of the word “substance" in current regulations. It embraces the European REACH regulation (EC No1907/2006) relating to the Registration, Evaluation, Authorisation and Restriction of Chemicals, which identifies a class of substances of varied composition: 'UVCB substances (substances of anonymous or varied configuration, complex reaction outputs or biological constituents) can be enumerated as a lone substance beneath this Regulation, in spite of their variable composition, on condition that the menacing properties do not fluctuate considerably and permit the similar classification'. Another instance of complex substances possibly including various chemical components are plant extracts denoted as 'botanical active substances' and as 'basic substances' and ratified under directive (EC)No 1107/2009 on plant protection in the EU.

Biostimulants are demarcated by envisioned agricultural productivities. 'Nutrition efficiency' might include nutrient utilisation and absorption from the soil, root growth, transportation, accumalation and absorption (of nutrients in the plant. 'Abiotic stress' denotes to any physical or chemical stressor of non-biological source.'Quality traits' might be dissimilar and range from nutritional significance to shelf life or flower pigment. Biostimulants are not fertilisers in as they do not comprise nutrients envisioned to be conveyed to the plant. The target of these formulations is not to nourish with nutrients, but to support and stimulate the metabol ism of the plant, reduce plant stress, etc. They augment crop growth and yield via a sequence of differential mechanisms comprising stimulation of soil microbial activity, and expansion of the activities of soil enzymes or plant growth hormones [6]. Main categories of plant biostimulants are elucidated in Table 1.

Table 1. Main categories of biostimulants

Functions	Humic substance	Complex organic materials	Beneficial chemical elements	Inorganic salts	Seaweeds extracts	Chitin and chitosan	Anti-transparent	Free amino acid and other N containing substances
Existence of plant analogs	No	Yes	Yes	No	Yes	No	No	Yes
Action inside the plants	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Action outside the plants	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Physical or physic-chemical effects in/on the plants	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Metabolic effects	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Hormonal effects	Yes	Yes	Yes	No	Yes	Yes	Yes	Yes
Physiological effects on nutrition efficiencies	Yes	Yes	No	Yes	Yes	Yes	Yes	Yes
Physiological effects on abiotic stress response	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Physiological effects on abiotic stress response	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes

In the face of recent efforts on the regulatory status of biostimulants, except in the European Union and in the United States, there is no valid definition of plant biostimulants. This give on to a detailed listing and categorization of the substances and microorganisms covered by the concept. Being influenced by this, some major categories are universally acknowledged by scientists, regulators and stakeholders [7,1,8], covering both substances as well as microorganisms (beneficial bacteria, mainly PGPRs, and beneficial fungi). They can be free-living, rhizospheric or endosymbiotic in nature. Almost all biostimulants are applied through foliar. Although, recent research appears the benefits of applying biostimulants in-furrow. These categories are summarily established in the next section which will be further described by the accompanying papers of this special issue on plant biostimulants in horticulture.

3. BIOSTIMULANT CATEGORIES

Biostimulants fall into following categories:

3.1 Organic Acids

Usually humic or fulvic.

3.2 Protein Hydrolysates and Other N-containing Compounds

Amino acid based biostimulants obtained from animal or vegetable sources. The best ones are extracted from vegetable matter, usually soybeans, by enzymes.

3.3 Seaweed Extracts & Botanicals

Derived from kelp, algae or terrestrial plants.

3.4 Chitosan and Other Biopolymers

Often obtained from crustacean shells.

3.5 Inorganic Compounds

Minerals like cobalt or silicon.

3.6 Microbial Inoculants

Beneficial organisms including fungi or bacteria.

3.7 Organic Acids

3.7.1 Humic and fulvic acids

Humic substances (HS) are the innate components of the soil organic matter which is not only the consequent of the degradation of plant, animal and residue of microorganism, but the metabolic products of soil microrganisms utilizing such degraded components. HS are the assembly of some diverse substances which are initially grouped based on their atomic mass and whether they are soluble in humins, humic acids and fulvic acids. In fact they are the ultimate yield of microbial breakdowns and chemical deterioration of dead flora and fauna in soils [9] and believed to be the major organic molecules that normally occur in universe [10]. It is reported as one of the most important constituents of soil organic matter [11]. Humic substances have been acknowledged as indispensable contributors to soil fertility, acting on physical, physico-chemical, chemical and biological properties of the soil. In soil, humic substances are appeared to perform a major role in numerous soil and plant activities [12] like regulating accessibility of nutrients, diffusion of oxygen and carbon between the troposphere and the soil and lethal chemicals alteration and transport [13]. Furthermore, humic substances in soils influence the functioning of plant as well as the concentration and activities of microorganisms of the rhizosphere zone [14].

Most biostimulant effects of HS refer to the amelioration of root nutrition, via different mechanisms. One of them is the increased uptake of macro- and micronutrients, due to the increased cation exchange capacity of the soil containing the polyanionic HS, and to the increased availability of phosphorus by HS interfering with calcium phosphate precipitation. Another important contribution of HS to root nutrition is the stimulation of plasma membrane H^+-ATPases, which convert the free energy released by ATP hydrolysis into a trans-membrane electrochemical potential used for the import of nitrate and other nutrients. Besides nutrients uptake, proton pumping by plasma membrane ATPases also contributes to cell wall loosening, cell enlargement and organ growth. HS improves respiration and invertase activities by availing C substrates. Hormonal effects are also pronounced, but whether HS has functional groups documented by the reception/signalling complexes of plant hormonal pathways, liberate entrapped hormonal com-pounds, or stimulate hormone-producing microorganisms is vague [1]. The anticipated biostimulation action of HS also states to stress protection. Elevated-molecular mass HS augment the action of key enzymes of this metabolism in hydroponically-grown maize seedlings, signifying stress response modulation by HS [15].

Fulvic acid can be observed as organic portion of soil, which can dissolve in alkaline as well as in acidic medium [16]. Fulvic acid has higher total acidity, higher carboxyl groups and more adsorption and cation exchange capacities as compared to humic acid [17].

They are the main factor involve for chelation and movement of metal ions, like Fe and Al [18]. The specific roles of humic acids on plants are:

a. Plant growth, yield and nutrient uptake
b. Amelioration of abiotic stress (Salinity stress, moisture stress etc.)
c. Plant physiology and metabolism

3.7.2 Botanicals

'Botanicals' define the substances sourced from plants which are utilized in pharmaceutical and cosmetic products, as food constituents, and also in plant protection products. Their biostimulant activities, are being dedicated on their pesticidal properties. Though, there are prospects to use them as biostimulants [19]. Plant interfaces in ecosystems are [19] recognised to be facilitated by plant active compounds, stated to as allelochemicals, which are being stressed in the perspective of sustainable crop supervision. While crop rotations, intercropping, cover crops and mulching are used to estimate allelochemical relations between plants the focus on chemical interactions for the improvement of new biostimulants needs to be strengthen.

3.7.3 Protein hydrolysates and other n-containing compounds

Amino-acids and peptides mixtures are procured by the chemical and enzymatic protein degraation of by-products from agro-based industries, from both plant sources (crop residues) as well as animal wastes (e.g. collagen, epithelial tissues) [1,7,8]. Chemical synthesis could be utilized for homogenous or heterogeneous substances. Other nitrogenous compounds *viz.* betaines, polyamines and 'non-protein amino acids', which are variegated in higher plants but imperfectly classified with their physiological as well as environmental performance [20]. Glycine betaine is a unique type of amino acid derivatives having well-understood anti-stress property [21].

It plays a various roles as biostimulants of plant growth [1,8]. Direct impacts on plants comprise nodulation of N uptake and acclimatization, by the regulating of enzymes implicated in N metabolism and structural genes, and by stimulating the signalling pathway of N acquirement in roots. They regulate the TCA cycle enzymes, effects C and N metabolisms. Complex protein and tissue hydrolysates are influenced by the hormonal activities [22]. It was reported that several amino acids are involved in chelating effects protecting plants from heavy metals and enhance micronutrients mobility and acquisition. They are also implicated in antioxidant activity by the scavenging of free radicals by particular nitrogeous compounds, comprising of glycine betaine and proline, thereby mitigating environmental stress.

Furthermore, secondary effects on plant nutrition and growth are also significant when protein hydrolysates which are used to increase microbial biomass and activity, soil respiration and, overall, soil fertility are used in plants and soils. Specific amino acids and peptides with chelating and complexing activities helps in nutrients availability and acquirement. Their appliance reported to have significant developments in produce and qualities in agricultural and horticultural crops [7].

3.7.4 Seaweeds extracts & botanicals

Seaweeds or macroalgae are aquatic plants belonging to the plant kingdom Thallophyta. These organisms are often considered as an under-utilized bioresource, while many species have been utilized as food, industrial gums, and in therapeutic and botanical applications for centuries. Seaweed extracts constitutes more than 33% of the total biostimulant market worldwide and are expected to stretch a value of 894 million Euro in 2022 [23]. Seaweed extracts comprises major and minor nutrients, amino acids, vitamins, cytokinins, auxin and abscisic acid like growth promoting substances and have been informed to encourage the growth and yield of plants, improve tolerance to environment stress [24,25], increase nutrient uptake from soil [26] and enhance antioxidant properties [27]. There are many literatures available on seaweed-based inoculant used as biostimulants to enhance plant resistance against biotic and/or abiotic stress, plant growth promotion and their effects on root/microbe interactions, have demonstrated the various roles in plant health, quality/biofortification and improvement of soil. These consequences give assistance to physiological, biochemical and molecular mechanisms, as noticeable through investigations using model plants [28,29]. Any betterment in agricultural process that marks a better productivity must decrease the harmful environmental affect on agriculture and increase the self sufficiency of the system. In a move to depict the possibilities of seaweed extract acting on rhizosphere nutrients availability, investigations were conducted to describe their interaction with rhizosphere microorganisms and enzyme activity (Fig. 1). The applications of biostimulants have the capacity to improve the efficiency of regular inorganic fertilizers. It has been reported that in agricultural and horticultural crops, marine bioactive compounds isolated from marine algae are used, which have various positive effects resulting in improve productivity and quality [30,31]. The advantageous effects of seaweeds (microalgae) and their extracts on crop systems have been used for innumerable time period to upgrade the soil properties and to increase productivity and value of agri-horticulutral crops. Liquid concentrate derived from seaweeds have lately earned significance for using as foliar applications for various plant species such as cereal, flower and vegetable [see 31 for a comprehensive list]. Various poly and oligomers of naturally occurring or (semi) man-made compounds are progressively adopted in agriculture as inducers of plant defence, example seaweed polysaccharides.

The option of seaweeds' utilization in contemporary agriculture has been extensively discovered and diverse varieties of formulations of these marine algae as liquid fertilizer and either whole or finally chopped powdered algal manures are utilized. The best effects on the use of seaweed extracts were described for stimulation of seed germination and root growth, improvement of frost resistance, augmented nutrient uptake and control of phytopathogenic fungi, bacteria [32], insects or other pests [33] and also for re-establishment of plant growth under high salinity stress [34,35]. Seaweeds act on soils and on plants and can be applied on soils, in hydroponics solutions or as foliar

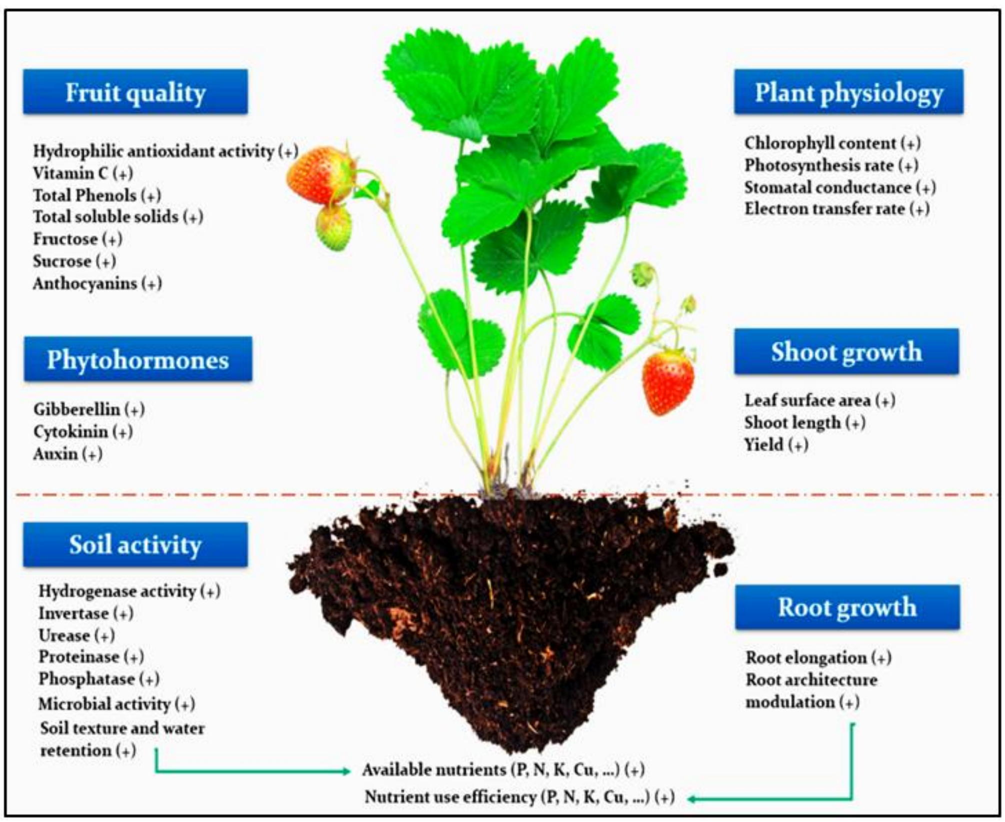

Fig. 1. Theoretical graphic emphasizing the positive effect of seaweed extracts on the whole soil–plant system. The impacts embraces refining fruit quality, and plant phytohormone content, improving soil enzymatic activity, refining the rooting system and the largely physiological features of plants

treatments [36]. In soils, their polysaccharides helps in gel formation, water retention and soil aeration. The polyanionic compounds helps in the fixation and exchange of cations, fixation of heavy metals and soil remediation. In plants, they act as fertilisers, beside their other roles on the effects on seed germination, plant establishment, growth and development which is related with hormonal effects, which is regarded as main reasons of biostimulation activity on crop plants.

3.7.5 Chitosan and other biopolymers

Chitosan, a deacetylated product of the biopolymer chitin, synthesized through both naturally and artificially. Different and controlled sizes of poly and oligomers are being utilized in various foods, cosmetic, medical and agricultural sectors. The physiological response of chitosan oligomers in crops are not only the consequent of the abilities of this polycationic substance to associate with a broad categories of cellular components, such as DNA, plasma membrane and cell wall constituents, but owing to associate with a unique receptors related to activation of gene involve defence mechanism, a way similar to plant defence elicitors [37,38,39,40]. Chitosans has been found extensively applied as a coating agent for a variety of fruits chiefly for protecting against post-harvest damages owing to pathogen infections [41]. Latest transcriptomic study highlighted that chitosans induce activities of genes responsible for varieties of events in plant life processes like SAR, plant defence system, photosynthesis, hormone metabolism , translation of HSP(s) and changing the metabolism of protein resulting in an increased in content of storage proteins [4,42]. Malerba M. and Cerana R. [43] reported *an augmentation of storage life* and maintenance of anthocyanin content in strawberry fruit encrusted with chitosan. Rahman et al. [44] study revealed that appliance of chitosan on the strawberry's leaves, notably improved the fruit growth and yield (up to 42% higher) than the control. The fruit from plants treated with chitosan show considerably high contents of carotenoids, anthocyanins, flavonoids and phenolics than the control. The antioxidant expression in fruit of chitosan sprayed plants was extensively higher than control (Fig. 2).

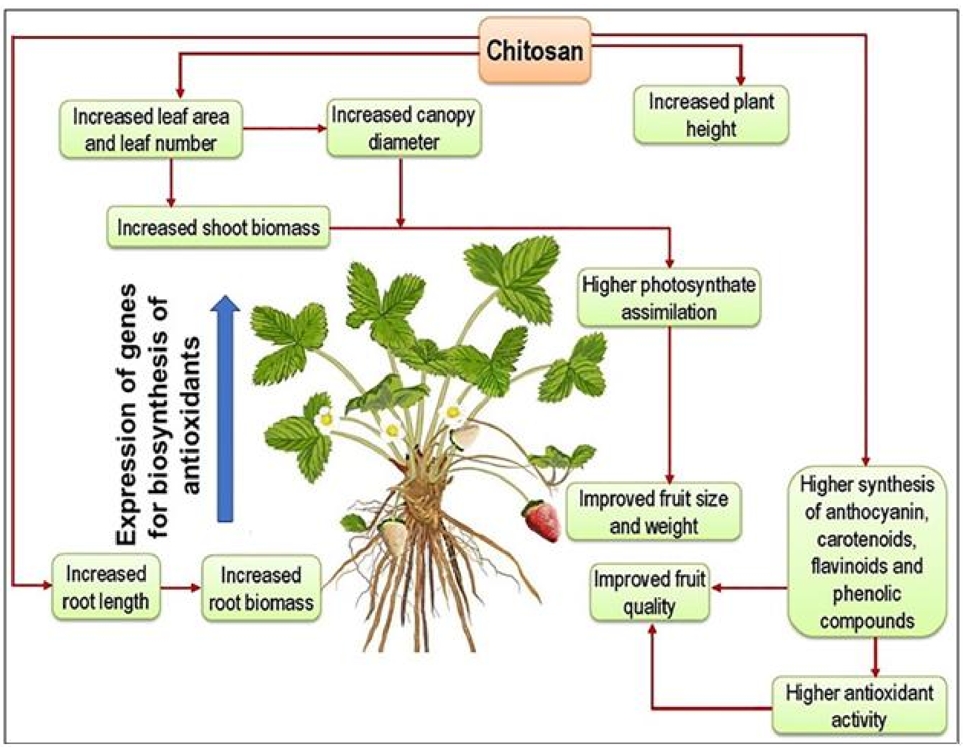

Fig. 2. A hypothetical pathway of stimulation of fruit yield and accumulation of antioxidants in strawberry fruit cv. festival by the foliar application of chitosan

(Adapted from Malerba M. and Cerana R. [43])

https://doi.org/10.1371/journal.pone.0203769.g004

Chitin and chitosan; evidently used as unique receptors and signalling pathways. They are the major players in responding to stress stimulus as well as in the developmental regulations, amongst the cellular consequences of the attaching of chitosan to approximately definite cell receptors, hydrogen peroxide aggregation and Ca^{2+} efflux into the cell were shown, seems to cause an enormous functional change. The beneficial property of chitosan (CHT) on crop response may also be attributed to the abilities CHT substances to complement plants with essential nutrients [43]. Spraying of zinc and CHT complexes competently supplement the minornutrient to wheat grown under zinc deficient situations [45]. CHT complexed with waste silica might able the farmers to decrease the application of NKP fertilizers for improving corn productivity in Indonesia with ecological and financial benefits [46]. Additionally, CHT nanoparticles may also be used as a vector for plant growth regulators. Such as CHT nanoparticles complexed with GA notably enhance the leaf area and content of chlorophylls and carotenoids in Phaseolus vulgaris [47]. Thus CHT, which are obtainable in huge amount and cheaply can be used in the development of sustainable agricultural system and in production and preservation of food.

3.7.6 Inorganic compounds

Chemical compounds which enhance plant growth and development, could be necessary to certain category of plants are known as beneficial elements [13]. Major beneficial elements *viz.* Al, Co, Na, Se and Si are generally found in soils and plant as inorganic compounds and in graminaceaous group as insoluble form (amorphous silica ($SiO_2.nH_2O$)). Such types of beneficial effects can be constitutive, like the hardening of cell walls through silica deposition, or expressed in specific environmental situations, like pathogen attack for selenium or osmotic stress for sodium. Description of beneficial elements is not restricted to their chemical natures, but essentially on their beneficial impact on plant growth and stress response. Various scientific reports are available, on the effects of beneficial elements, which increase plant growth, the quality of plant products and endurance against various abiotic stresses. These include cell wall rigidification, osmoregulation, lower the rate of transpiration by crystal deposits, thermal manipulation through radiation reflection, enzyme activity by co-factors, plant nutrition via interacting with different elements during uptake and mobility, antioxidant protection, interactions with symbionts, disease causing microbes and herbivore reaction, protection against toxic effect of heavy metals, plant hormone productions and signalling [13]. Minerals salts of beneficial and essential elements (chlorides, phosphates, phosphites, silicates and carbonates) have been used as fungicides [48].

Phosphite (reduced phosphate) is usually treated in soils in the form of phosphorus acid, phosphite salts with metal ions (e.g. K+, Na+, NH4+) and non-metallic anion [48]. Certain facts indicates the biostimulant and fungicidal properties of Phi products, however phytotoxicity risk might be associated if its rate is not maintain properly ie beyond 5 g/l or 36 kg/ha its effect is phytotoxic [49,50,48]. It can be applied to soil as a pesticide, supplemental fertilizer or biostimulant [51] even though its effect as fertilizer and growth enhancer is a matter of discussion [10]. Various evidences indicated that Phi might behave as a biostimulant by regulating sugar metabolism and intercellular hormonal changes [40], enhancing defence activities [12] and/or shifting plant P nutrition [52]. Oka et al. [11] reported that for soil saturated phosphite inhibit the establishment of the endoparasitic nematodes *Heterodera avenae* and *Melooidogyne marylandi* in both wheat and bristle oat plants. Nevertheless, even as it seems to display disease control responses in cereal crops, further effort is necessary in the field to conclude the degree and uniformity of any probable positive effects.

Silicon is a beneficial elements in some monocots, like rice, sugarcane etc. Its beneficial properties include increasing endurance to abiotic stress and resistance to pathogens and diseases. In the soil, either Si is presents as insoluble quartz or silicates which chemically bind with metals or as non-ionic silicic acid, which plant roots can take up effortlessly and translocated all over the plant system [53]. Maximum concentrations are generally located at adjoining area of the stomata. These silica deposits or phytoliths boost leaf mechanical vigour and stiffness, thus rising light absorption and photosynthesis. They also modulate nutrient and water mobility, apparently owing to silica gel form in the cell walls, which thus lessen the transpiration. Others stress-mitigating effect of Si may be attributed to its capability to halt toxic metals translocation in plant cells and the soil and to reduce the progress of plant senescence [54].

3.7.7 Microbial inoculants

Microbial inoculants are mostly free-living bacteria, fungi, and arbuscular mycorrhizal fungi (AMF) [55] which were screened from diverse situations such as soil, plants, plant residues, water and composted manures. Better assimilation of primary elements such as N, P and K has been found in the presence of PGPB (Plant growth promoting bacteria). Certain PGPB species (like *Azotobacter* and *Azospirillum*) can transform atmospheric nitrogen into a form that can used by the plants [7]. Various evidences also highlighted the phenomenon where PGPB(s) enhance the solubility of certain essential elements, thereby improving nutrient accessibility for plants. This is mainly significant for elements like P which generally not accessible to plants. Several microbes also have the ability to increase the solubility of potassium (K) from rock K minerals, by forming chelate with silicon ions, increasing its solubility and using organic acids to dissolve rock K. Therefore, application of PGPR at proper dose possibly will facilitate reduction in the rate fertilizer applications (N and P).

Some growth promoting bacteria may promote plant development through interactions with phyto-hormones. A variety of plants growth regulators like auxins, cytokinins, giberellins and ethylene are identified to be synthesized by PGPB [56], that can enhance both shoot and root growth of the plant [57]. PGPB can also enhance the ability of plant to tolerance stress by disrupting certain stress hormone (ethylene) or altering the plant hormone interaction [56]. Some species of PGPR produces an enzyme that convert the originator for ethylene into 2-oxybutanoate and NH3. Thereby, decreases both abiotic and biotic stresses like pathogenic bacteria, heavy metals, salt and drought [58,57].

Beneficial fungi having biostimulant activity such as symbiotic fungi, principally arbuscular mycorrhizal fungi (AMF) in the genus *Glomus*, (forms a well branched network of roots and hyphae) enable the plants to widen their root arrangement further than the exhaustion region, thus allow them to enhance the uptake of minerals and moisture improving their stress tolerance capacity. Other strains of non-pathogenic fungi having potential biostimilants activities that associate with roots include *Aspergillus*, *Trichoderma*, *Penicillium*, *Saccharomycetes*, *Mortierella* and *Mucor*. They have been established to enhance plant growth, increase plant nutrition, improve tolerance to abiotic and biotic stress [59].

Table 2. Common modes of action for biocontrol of plant diseases by plant growth promoting rhizobacteria (PGPR)

Mode of action	Mode of action description
Antagonism	Bacteria produce antibiotics that are delivered in sufficient quantities to the correct areas of the rhizosphere and therefore directly kill pathogens. A range of antibiotics can be produced, including hydrogen cyanide (HCN).
Signal interference	Some PGPR can degrade homoserine lactones (AHL) which are thought to be used as signalling molecules by certain pathogenic bacteria, and may also be involved in biofilm production. Thus by degrading AHL, certain PGPRs can affect the ability of bacteria to act as pathogens.
Induced systemic resistance	Presence of certain PGPR in the rhizosphere may result in the plant developing resistance to certain pests and pathogens. In contrast to antagonism, induced systemic resistance doesn't require the PGPR to colonise a large proportion of the root system, and therefore may have a faster and more consistent action against pests.
Siderophore production	In soils where Fe^{3+} is low, the production of siderophores may mean that the PGPR make Fe unavailable for phytopathogens resulting in improved plant health.
Competition for nutrients and niches	It is possible that certain PGPR strains can colonise the entire plant root and outcompete pathogens for both nutrients and space. Whether this is consistent under field conditions is unclear
Interference with pathogen activity and competition	By growing on or in the pathogen, it interferes with its fundamental growth and activity.

Summarised from [57]

Trichoderma having symbiotic relations with crops (promoting branching of roots) are found to secrete active solubilising agent to the rhizosphere and thus promoting translocation of essential elements [60] and capability to infect other fungi, they are frequently used as biocontrol agents for controlling fungal diseases of crops.

Bio control microorganisms (BCMs) can be employed as activator of SAR. Simultaneously, fungal BCMs have the ability to enhance crop growth and development thus responding as plant growth-promoting microorganisms (PGPMs), sequentially govern an increase tolerance against abiotic stresses viz. drought, salinity etc. The capability of BCMs and PGPMs of regulating plant defence mechanisms, like SAR, was revealed, but the overall information of this BCM-plant molecular cross talks are not understood properly and many defensive substances may present but need to be recognised and isolated. Consequently, BCMs and PGPMs can be classified as “biostimulant microorganisms”, which has the capacity to promote crop growth and protect against pathogens throughout the plant life process i.e. seed germination to plant maturity.

This chapter target to provide an advanced information on the recent developments in utilizing biostimulant microorganisms on crops for enhancing crop vigour, yields and quality and for enhancing plant survivability against both biotic and environmental stresses [61]. Plant growth-promoting rhizobacteria (PGPR) formulations are one of the important biostimulant classes due to their ability to improve the root growth, mineral availability, and nutrient use efficiency in crop rhizosphere [62]. Microbial soil balance has often been ignored by modern agriculture, however biostimulation though soil microorganisms are now gaining momentum.

3.7.8 Chitosan and other biopolymers

Chitosan, a deacetylated product of the biopolymer chitin, synthesized through both naturally and artificially. Different and controlled sizes of poly and oligomers are being utilized in various foods, cosmetic, medical and agricultural sectors. The physiological response of chitosan oligomers in crops are not only the consequent of the abilities of this polycationic substance to associate with a broad categories of cellular components, such as DNA, plasma membrane and cell wall constituents, but owing to associate with a unique receptors related to activation of gene involve defence mechanism, a way similar to plant defence elicitors [37,38,39,63]. Chitin and chitosan; evidently used as unique receptors and signalling pathways. They are the major players in responding to stress stimulus as well as in the developmental regulations, amongst the cellular consequences of the attaching of chitosan to approximately definite cell receptors, hydrogen peroxide aggregation and Ca^{2+} efflux into the cell were shown, seems to cause an enormous functional change.

3.7.9 Microbial inoculants

Microbial inoculants are mostly free-living bacteria, fungi, and arbuscular mycorrhizal fungi (AMF) [55] which were screened from diverse situations such as soil, plants, plant residues, water, and composted manures. Particularly, bio control microorganisms (BCMs) can be employed as activator of SAR. Simultaneously, fungal BCMs have the ability to enhance crop growth and development thus responding as plant growth-promoting microorganisms (PGPMs), sequentially govern an increase tolerance against abiotic stresses viz. drought, salinity etc. The capability of BCMs and PGPMs of regulating plant defence mechanisms, like SAR, was revealed, but the overall information of this BCM-plant molecular cross talks are not understood properly and many defensive substances may present but need to be recognised and isolated. Consequently, BCMs and PGPMs can be classified as “biostimulant microorganisms”, which has the capacity to promote crop growth and protect against pathogens throughout the plant life process i.e. seed germination to plant maturity. This chapter target to provide an advanced information on the recent developments in utilizing biostimulant microorganisms on crops for enhancing crop vigour, yields and quality and for enhancing plant survivability against both biotic and environmental stresses [61]. Plant growth-promoting rhizobacteria (PGPR) formulations are one of the important biostimulant classes due to their ability to improve the root growth, mineral availability, and nutrient use efficiency in crop rhizosphere [62]. Microbial soil balance has often been ignored by modern agriculture, however biostimulation though soil microorganisms are now gaining momentum.

3.7.10 Modes of action/mechanisms of action

The mechanism/ course of action signify “a specific effect on a discrete biochemical or regulatory process”. The course of action of all but a few biostimulants continues basically unnoticed. According to Paraąikovic et al. (2011) this is chiefly owing to the diversified property of natural resources utilized for production and the composite nature of the constituent confined in biostimulant, a product that makes it more or less incredible to extract the exact factor(s) accountable for biological activity and to conclude the concerned mechanism(s) of action. Thus, importance must be given on the recognition of the "mechanisms of action" of biostimulants as shown by a natural specific outcome on plant yield through improvement in mechanisms such as photosynthesis, senescence, modulation of phytohormones, uptake of minerals and water, and regulation of genes involve for tolerance to environmental stresses and transformed crop canopy structure and phenology. A model of this development is the approaches which imply in the use of protein-based biostimulants for which advance studies have recognized the specific metabolic process and some of the system over which they exercise their affect on crops. In order to strengthen our knowledge on course of action of biostimulants the following steps of biostimulants activities, after their application on plant have been established: (1) diffusion into plant cells, translocation and transformation in plants, (2) gene regulation, plant signalling and control of hormonal status, (3) metabolic activities and consolidated whole plant reaction.

4. APPLICATION OF BIOSTIMULANT

4.1 Agronomic and Physiological Traits of Crops

The enhancement of plant development process along with crop yield as a result of application of PBs has been generally correlated to the activity of signalling biologically active elements in the primary and secondary metabolisms [7]. Diverse types of hydrolyzed collagen, like granulated gelatin, gelatin hydrolysate and amino acid mixtures resembling gelatin composition were determined in relation to plant growth on cucumber [64]. In their experiment, they revealed that application of gelatin hydrolysate enhanced the activity of genes encoding for amino acid permeases (AAP3, AAP6) and transporters of amino acids and nitrogen. Thus, they inferred that gelatin hydrolysate served as a continuous resource of N and also functioned as a biostimulant. The application of this natural elements or microorganism not only improves the reactions of microbial and non microbial PBs, but also enhances tolerance to environmental and biotic stresses.

4.2 Implications of Biostimulants for Abiotic Stresses Tolerance

Wang et al. [65] reported that global climate change such as adverse ecological condition and edaphic factors viz. drought, salinity and extreme weather, imposed about 70% yield gap. In accordance with substantial changes in the global climatic condition, these environmental stresses are likely to have a high harmful impact, imparting serious effect on crop production, and hence global food safety [66]. Therefore, the utilization of non-microbial and microbial PBs has been proposed, as an important and efficient mechanism to achieve stability in crop productivity. Also non-microbial and microbial PBs can be treated as a feasible mechanism to increase endurance to salinity. These positive consequences may be correlated to various physio-chemical processes such as (i) reduced membrane lipid peroxidation, (ii) enhanced chlorophyll content, (iii) better antioxidant activities and (iv) an improved afflux and compartmenlizing the intracellular ions.

4.3 Biostimulants for Improving Nutrient Use Efficiency

The utilization of biologically active native elements and microbial extracts can signify an important means to augment nutrient status the soil, plant nutrient absorption, translocation, transformation and metabolism [67]. Enhancing nutrient utilization efficiency in particular N and P is essential for both economic and environmental point of views. At both ideal and low N regimens (112 and 7 mg L^{-1}, respectively) the use of legume based PH specifically such a as substrate drench increases the number of leaf, SPAD (Soil Plant Analysis Development) index and biomass production of greenhouse tomato [53]. The improved agronomic feedbacks of PH applied tomato were related to the improvements of root architecture that resulted in better N uptake and translocation. Furthermore, in suboptimal N level, PH treatment enhanced the expression of genes encoding for amino acid transporter and ferredoxin-glutamate synthases and glutamine synthetase in roots, which are reported to be necessary in N metabolism.

4.4 Biostimulants for Enhancing Produce Quality

The microbial and non-microbial plant biostimulants application have the capacity to alter plant's metabolism both primary and secondary [68] which results in the synthesis as well as build up of antioxidant compounds (i.e., secondary metabolites) which are vital for human nutrition. This treatment also cause a considerable boost in tomato fruit quality such as antioxidant capacity, total soluble sugars, carotenoids (lycopene, lutein, and b-carotene), total polyphenols and flavonoids contents along with mineral composition (P, K, Ca, Mg, Fe, Mn, and Zn). Furthermore, Haplern et al. [69] conducted an experiment on two Brassica species: *Brassica campestris* and *Brassica juncea* and studied the effect of PAR (photosynthetically active radiation) (low or high), phosphate (low or high), and phosphite (low, optimal or high), and their interlinkage on the content of glucosinolates, flavonoids, and nitrate. They highlighted that the addition of phosphite in the nutrient medium may cause an enhancement in phosphate deficiency; thus it stimulates the biosynthesis as well as aggregation of several target flavonoids and glucosinolates as a probable defence system to cope with nutrient stress.

4.5 Con's of Biostimulants Science and Practice

The scientific community and commercial enterprise are having immense interest in identification of bioactive components of PBs and explicating the stimulation mechanisms at molecular and physiological levels. The most proficient technology to develop novel biostimulants is the use of small/medium/large high throughput phenotyping, because of complex matrices having diverse groups of bioactive and signalling molecules [66]. The niche of biostimulants is a baffling concept [70,71] and it has ensured that biostimulant market is not on the basis of science or efficacy and that many products are not much more than recycled waste products and its selling is based on pseudoscience and marketing. On biostimulant products several researches has been conducted and it has founded to be futile or to have inactive, unstable or inconsistent properties and some are having negative effects when compared with well-designed controls [72,73,74,14,75,76,77,78]. It has been reported that "none of the biostimulant products tested achieved a sufficient degree of pathogen control to warrant replacement of or supplementation with conventional synthetic fungicides" [75] and it poses positive and negative impacts and as a concern for the economic viability, the use of humic substances improves the crop yield [79]. The discovery of novel pharmaceuticals is done through expedition of marine organisms, plants and microorganism from diverse ecosystems, so too development of biostimulants from the different resources holds a symbolic discovery. Still there are two major inconvenience within the industry such as: (1) the identification of a primary mode of action is extremely hard due to highly comlex multicomponent preparation of products and incomplete identified composition and (2) the existing classification and legislation/legal framework for regulation of biostimulants is mainly on the basis of source material but not on biological mode of action. Therefore there is inadequate ability to distinguish products and there are the potential to demostrate a single product within a biostimulant category successfully, to incorrectly indicate the worthiness of the group as whole.

5. CONCLUSION

Recent biostimulants are hetergenous mixtures obtained from natural resources of highly varied origin and manufacture using highly diverse techniques thus it can be expected to have a wide-range of potential biological activity and security. Numerous experiments have suggested that linking numerous biostimulants could offer steady effects as compared with their individual tratment. As biostimulating effects are distinctly species-specific and product-specific and our knowledge of one biostimulant or about single plant species can't be sprightly transfered to another biostimulant or another plant species. In order to expand both basic and applied science regarding the efficacy of biostimulants for a particular plant species, it is necessary to perform a broad spectrum research on this species, with a variety of products, treatments, growth stages, etc. PBs such as natural substances and microbial inoculants seems as a novel and possible group of agricultural inputs, complementing agrochemicals including synthetic fertilizers, enhancing tolerance to abiotic stresses and along with this increasing the quality of agricultural and horticultural commodities.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Du Jardin P. The science of plant biostimulants- A bibliographic analysis. Adhoc Study Report to the European Commission DGENTR; 2012.
2. Berlyn GP, Russo RO. The use of organic biostimulants to promote root growth. Belowground Ecol. 1990;2:12-13.
3. Hamza B, Suggars A. Biostimulants: Myths and realities. Turfgrass Trends. 2001;10:6-10.
4. Landi L, De Miccolis Angelini RM, Pollastro S, Feliziani E, Faretra F, Romanazzi G. Global transcriptome analysis and identification of differentially expressed genes in strawberry after preharvest application of Benzothiadiazole and chitosan. Front Plant Sci. 2017;8:235.
5. EU. Regulation of the European parliament and of the council laying down rules on the making available on the market of EU fertilising products and amending Regulations (EC) No 1069/2009 and (EC) No 1107/2009 and Repealing Regulation (EC) No 2003/2003; 2019.
6. Piccolo A, Spaccini R, Savy D, Drosos M, Cozzolino V. The soil humeome: Chemical structure, functions and technological perspectives. Sustain. Agrochemi. 2019;183–222.
7. Calvo P, Nelson L, Kloepper JW. Agricultural uses of plant biostimulants. Plant Soil. 2014;383:3-41.
8. Halpern M, Bar-Tal A, Ofek M, Minz D, Muller T, Yermiyahu U. The use of biostimulants for enhancing nutrient uptake. In: Sparks, D.L. (Ed.). Adv Agron. 2015;129:141–174.
9. Asli S, Neumann Peter M. Rhizosphere humic acid interacts with root cell walls to reduce hydraulic conductivity and plant development. Plant Soil. 2010;336:313–322.
10. Thao HTB, Yamakawa T. Phosphite (phosphorous acid): Fungicide, fertilizer or biostimulator? Soil Sci. Plant Nutr. 2009;55:228-234.
11. Oka Y, Tkachi N, Mor M. Phosphite inhibits development of the nematodes *Heterodera avenae* and *Meloidogyne marylandi* in cereals. Phytopathol. 2007;97:396-404.
12. Olivieri F, Feldman M, Machinandiarena M, Lobato M, Caldiz D, Daleo G, Andreu A. Phosphite applications induce molecular modifications in potato tuber periderm and cortex that enhance resistance to pathogens. Crop Prot. 2012;32:1-6.
13. Pilon-Smits EAH, Quinn CF, Tapken W, Malagoli M, Schiavon M. Physiological functions of beneficial elements. Curr. Opin. Plant Biol. 2009;12:267-274.
14. Vasconcelos ACF, Zhang X, Ervin EH, Kiehl JC. Enzymatic antioxidant responses to biostimulants in maize and soybean subjected to drought. Sci. Agric. 2009;66:395-402.
15. Olivares FL, Aguiar NO, Rosa RCC, Canellas LP. Substrate biofortification in combination with foliar sprays of plant growth promoting bacteria and humic substances boosts production of organic tomatoes. Sci. Hortic. 2015;183:100–108.
16. Schiavon M, Pizzeghello D, Muscolo A, Vaccaro S, Francioso O, Nardi S. High molecular size humic substances enhance phenylpropanoidmetabolismin maize (*Zea mays* L.). J. Chem. Ecol. 2010;36:662–669.
17. Bocanegra A, Benedi J, Sanchez-Muniz F. Differential effects of konbu and nori seaweed dietary supplementation on liver glutathione status in normo- and hypercholesterolaemic growing rats. Bri. J Nutr. 2006;95:696–702.
18. Esteves da Silva J. Fluorescence quenching of anthropogenic fulvic acids by Cu(II), Fe(III) and UO22+. Talanta, 1998;45(6):1155–1165.
19. Ertani A, Schiavon M, Muscolo A, Nardi S. Alfalfa plant-derived biostimulant stimulate short-term growth of salt stressed *Zea mays* L. plants. Plant Soil. 2013;364:145–158.
20. Vranova V, Rejsek K, Skene KR, Formanek P. Non-protein amino acids: Plant, soil and ecosystem interactions. Plant Soil. 2011;342:31-48.
21. Chen THH, Murata N. Glycinebetain protects plants against abiotic stress: Mechanism and biotechnological applications. Plant Cell Environ. 2011;34:1-20.
22. Colla G, Rouphael Y, Canaguier R, Svecova E, Cardarelli M. Biostimulant action of a plant-derived protein hydrolysate produced through enzymatic hydrolysis. Front. Plant Sci. 2014;5:1–6.
23. Eef B, Marlies D, Van Swam K, Veen A, Burger L. Identification of the seaweed biostimulant market (Phase1); The North Sea Farm Foundation: AD Den Haag, The Netherlands; 2018.

24. Zhang X, Schmidt RE. Hormone-containing products' impact on antioxidant status of tall fescue and creeping bentgrass subjected to drought. Crop Sci. 2000;40:1344–1349.
25. Zhang X, Ervin EH, Schmidt ER. Plant growth regulators can enhance the recovery of Kentucky bluegrass sod from heat injury. Crop Sci. 2003;43:952–956.
26. Turan M, Kose C. Seaweed extracts improve copper uptake of grapevine. Acta Agriculturae Scandinavica. Section B, Soil Plant Sci. 2004;54:213–220.
27. Verkleij FN. Seaweed extracts in agriculture and horticulture: A review. Biol. Agric. Hortic. 1992;8:309–324.
28. Wajahatullah K, Usha PR, Sowmyalakshmi S, Mundaya NJ, Prasanth R, Mark HD, Alan TC, James SC, Jeff N, Balakrishan P. Seaweed extracts as biostimulants of plant growth and development. J. Plant Growth Regul. 2009;28:386-399.
29. Pamela C, Louise N, Joseph WK. Agricultural uses of plant biostimulants. Plant Soil; 2014.
30. Blunden G. Agricultural uses of seaweeds and seaweed products. In: Guiry, M.D., Blunden, G. (Eds.), Seaweed Resources in Europe: Uses and Potential. John Wiley and Sons, Chichester. 1991;65-81.
31. Crouch IJ, Van Staden J. Commercial seaweed products as biostimulants in horticulture. J. Home Consumer Hort. 1994;1:19-76.
32. Alves RC, Merces PFF, Souza IRA, Alves CMA, Silva APSA, Lima VLM, Correia MTS, Silva MV, Silva AG. Antimicrobial activity of seaweeds of Pernambuco, Northeastern Coast of Brazil. Afr. J. Microbiol. Res. 2016;10:312–318.
33. Asha A, Rathi JM, Raja PD, Sahayaraj K. Biocidal activity of two marine algal extracts against third instar nymph of *Dysdrcus cingulatus* (Fab.) (Hemiptera, Pyrrhocoridae). Biocididal activity of marine algae. J. Biopestici. 2012;5:129–134.
34. Nabti E, Sahnoune M, Ghoul M, Fischer D, Hofmann A, Rothballer M, Schmid M, Hartmann M. Restoration of growth of durum wheat (*Triticum durum* var. waha) under saline conditions due to inoculation with the rhizosphere bacterium *Azospirillum brasilense* NH and extracts of the marine alga *Ulva lactuca*. J. Plant Growth Regul. 2010;29:6–22.
35. Pacholczak A, Szydlo W, Jacygrad E, Federowicz M. Effect of auxins and the biostimulator algamino plant on rhizogenesis in stem cuttings of two dogwood cultivars (*Cornus alba* 'AUREA' and 'Elegantissima'). Acta Sci. Pol. Hortoru. 2016;11:93–103.
36. Craigie JS. Seaweed extracts stimuli in plant science and agriculture. J. Appl. Phycol. 2011;23:371–393.
37. El Hadrami A, Adam LR, El Hadrami I, Daayf F. Chitosan in plant protection. Mar Drugs. 2010;8:968-987.
38. Hadwiger LA. Multiple effects of chitosan on plant systems: Solid science or hype. Plant Sci. 2013;208:42-49.
39. Katiyar D, Hemantaranjan A, Singh B. Chitosan as a promising natural compound to enhance potential physiological responses in plant: A review. Indian J. Plant Physiol. 2015;20:1-9.
40. Ávila W, Faquin V, Araújo JL, Marques DJ, Júnior PMR, Da Silva Lobato AK, Ramos SJ, Baliza DP. Phosphite supply affects phosphorus nutrition and biochemical responses in maize plants. Aust. J. Crop Sci. 2011;5:646.
41. El-Sawy NM, El-Rehim HA, Elbarbary AM, Hegazy ES. Radiation-induced degradation of chitosan for possible use as a growth promoter in agricultural purposes. Carbohydr. Polym.; 2010.
42. Xoca-Orozco LAA, Cuellar-Torres EA, GonzaAlez-Morales S, GutieÂrrez-MartoAnez P, LoApez-GarcoAa U, Herrera- Estrella L. Transcriptomic analysis of avocado hass (*Persea americana* Mill) in the interaction system fruit-chitosan-Colletotrichum. Front Plant Sci. 2017;8:956.
43. Malerba M, Cerana R. Recent advances of chitosan applications in plants. Polymers. 2018;10:118.
44. Rahman M, Mukta JA, Sabir AA, Gupta DR, Mohi-Ud-Din M, Hasanuzzaman M. Chitosan biopolymer promotes yield and stimulates accumulation of antioxidants in strawberry fruit. PLoS ONE. 2018;13(9).
45. Deshpande P, Dapkekar A, Oak MD, Paknikar KM, Rajwade JM. Zinc complexed chitosan/TPP nanoparticles: Promising micronutrient nanocarrier suited for foliar application. Carbohydr. Polym. 2017;165:394–401.
46. Gumilar TA, Prihastanti E, Haryanti S, Subagio A, Ngadiwiyana A. Utilization of waste silica and chitosan as fertilizer nano

chisil to improve corn production in Indonesia. Adv. Sci. Lett. 2017;23:2447–2449.

47. Espirito Santo Pereira A, Mayara Silva P, Oliveira JL, Oliveira HC, Fernandes Fraceto L. Chitosan nanoparticles as carrier systems for the plant growth hormone gibberellic acid. Colloids Surf. B. Biointerfaces. 2017;150:141–152.
48. Deliopoulos T, Kettlewell PS, Hare MC. Fungal disease suppression by inorganic salts: A review. Crop Prot. 2010;29:1059-1075.
49. Hardy GSJ, Barrett S, Shearer B. The future of phosphite as a fungicide to control the soilborne plant pathogen *Phytophthora cinnamomi* in natural ecosystems. Australas Plant Path. 2001;30:133-139.
50. Barrett S, Shearer B, Hardy GSJ. The efficacy of phosphite applied after inoculation on the colonisation of *Banksia brownii* sterns by *Phytophthora cinnamomi*. Australasi Plant Path. 2003;32:1-7.
51. Gómez-Merino FC, Trejo-Téllez LI. Biostimulant activity of phosphite in horticulture. Sci. Hortic. 2015;196:82-90.
52. Varadarajan DK, Karthikeyan AS, Matilda PD, Raghothama KG. Phosphite, an analog of phosphate, suppresses the coordinated expression of genes under phosphate starvation. Plant Physiol. 2002;129:1232-1240.
53. Savvas D, Ntatsi G. Biostimulant activity of silicon in horticulture. Sci. Hortic. 2015;196:66-81.
54. Albrecht Ute. Plant biostimulants: Definiition and overview of categories and effects. HS1330, One of a Series of the Horticultural Sciences Department, UF/IFAS Extension; 2019.
55. Vessey JK. Plant growth promoting rhizobacteria as biofertilizers. Plant Soil. 2003;255:571-586.
56. Dodd I, Zinovkina N, Safronova V, Belimov A. Rhizobacterial mediation of planthormone status. Ann. Appl. Biol. 2010;157:361-379.
57. Lugtenberg B, Kamilova F. Plant-growth-promoting rhizobacteria. Annu. Rev. Microbiol. 2009;63:541-556.
58. Ahmad F, Ahmad I, Khan MS. Screening of free-living rhizospheric bacteria for their multiple plant growth promoting activities. Microbiol Res. 2008;163:173-181.
59. Owen D, Williams A, Griffith G, Withers P. Use of commercial bio-inoculants to increase agricultural production through improved phosphrous acquisition. Appl. Soil Ecol. 2015;86:41-54.
60. López-Cervantes JR, Karl FR. Microbial process and composition for agricultural use; 2012.
61. Adrian S, Maria N, Antonella V, Giuseppe T, Antonio S. Control of biotic and abiotic stresses in cultivated plants by the use of biostimulant microorganisms. Improvement of crops in the era of climatic changes. 2014;1:107-117.
62. Minh Luan N, Bernard B, Marc O, Gilles C. Impacts of organic matter type and biostimulant products on the growth of wheat and the microbial communities of its rhizosphere under contrasted production systems. NSABS; 2014.
63. Yin H, Zhao XM, Du YG. Oligochitosan: A plant disease vaccine- a review. Carbohyd. Polym. 2010;82:1-8.
64. Yakhin OI, Lubyanov AA, Yakhin IA, Brown PH. Biostimulants in plant science: A global perspective. Front. Plant Sci. 2017;7:2049.
65. Wang W, Vinocur B, Altman A. Plant responses to drought, salinity and extreme temperatures: Towards genetic engineering for stress tolerance. Planta. 2003;218:1-14.
66. Rouphael Y, Kyriacou MC, Colla G. Vegetable grafting: A toolbox for securing yield stability under multiple stress conditions. Front. Plant Sci. 2018a;8: 2255.
67. De Pascale S, Rouphael Y, Colla G. Plant biostimulants: Innovative tool for enhancing plant nutrition in organic farming. Eur. J. Hortic. Sci. 2017;82:277-285.
68. Colla G, Nardi S, Cardarelli M, Ertani A, Lucini L, Canaguier R, Rouphael Y. Protein hydrolysates as biostimulants in horticulture. Sci. Hortic. 2015;196:28-38.
69. Haplern M, Bar-Tal A, Ofek M, Minz D, Muller T, Yermiyahu U. The use of biostimulants for enhancing nutrient uptake. Adv. Agron. 2015;130:141-174.
70. Torre LA, Battaglia V, Caradonia F. Legal aspects of the use of plant strengtheners (biostimulants) in Europe. Bulg J. Agric .Sci. 2013;19:1183-1189.
71. Traon D, Amat L, Zotz F, du Jardin P. A legal framework for plant biostimulants and agronomic fertiliser additives in the EU. Report to the European Commission, DG Enterprise & Industry, Arcadia International. 2014;115.

72. Csizinszky AA. Response of tomatoes to seaweed based nutrient sprays. Proc Fla State Hort Sco. 1984;97:151-157.
73. Albregts EE, Howard CM, Chandler C, Mitchell RL. Effect of biostimulants on fruiting of strawberry. Proc. Fla. State Hort. Soc. 1988;101:370-372.
74. Di Marco S, Osti F. Effect of biostimulant sprays on *Phaeomoniella chlamydospora* and esca proper infected vines under greenhouse and field conditions. Phytopathol. Mediterr. 2009;48:47-58.
75. Banks J, Percival GC. Evaluation of biostimulants to control Guignardia leaf blotch (*Guignardia aesculi*) of horsechestnut and black spot (*Diplocarpon rosae*) of roses. Arboric. Urban Forest. 2012;38:258-261.
76. Cerdan M, Sanchez-Sanchez A, Jorda JD, Juarez M, Sanchez-Andreu J. Effect of commercial amino acids on iron nutrition of tomato plants grown under lime-induced iron deficiency. J. Plant Nutr. Soil Sci. 2013;176:859-866.
77. De Oliveira FDAD, Medeiros JFD, Oliveira MKTD, Souza AAT, Ferreira JA, Souza MS. Interaction between water salinity and biostimulant in the cowpea plants. Rev. Brasil. de Eng Agric. Ambien. 2013;17:465-471.
78. Carvalho MEA, Castro PRDCE, Gallo LA, Ferraz MVDC Jr. Seaweed extract provides development and production of wheat. Rev. Agrarian. 2014;7:166-170.
79. Rose MT, Patti AF, Little KR, Brown AL, Jackson WR, Cavagnaro TR. A meta-analysis and review of plant-growth response to humic substances: Practical implications for agriculture. Adv. Agron. 2014;124:37-89.

Peer-review history:
The peer review history for this paper can be accessed here:
http://www.sdiarticle4.com/review-history/60331