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REVIEW ARTICLE

Biostimulant for sustainable pulse production - a review

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Abstract

Globally, agriculture is facing a pressing situation, such as climate change, degradation of soil health and the necessity for sustainable food production. Biostimulants provide novel solutions to improve agricultural practices and crop productivity. Biostimulants are plant-promoting substances/microorganisms that are organic and help to increase the nutrient uptake, growth and yield of the crop, provide tolerance to abiotic and biotic challenges and improve product quality. Humic acid (HA), fulvic acid (FA), protein hydrolysates, seaweed extracts, chitosan, microbial biostimulants such as arbuscular mycorrhizal fungi and plant growth-promoting rhizobacteria (PGPR), inorganic compounds like silicon, selenium and phosphite are different types of biostimulants to enhance plant production and productivity. Unlike fertilizers, biostimulants do not pose any threat to the environment. Pulses are rich in nutrients and increasing their productivity can increase the health of the people and result in an overall increase in the growth of economy. Despite having excellent nutritional value and agricultural importance, the productivity of pulses is not up to its potential. The production and productivity are inadequate to meet the growing population's demand. Farmers growing pulses are unaware of certain package of practices and recent technological developments. Numerous studies report improvement in biomass accumulation, The present review is focused on how application of biostimulants increase the growth and yield of pulses, nutritional uptake by the plants and its potential as sustainable inputs. It aims to highlight the most significant results obtained for different pulse crops and intends to highlight the knowledge gap in biostimulants research in context of plant growth promotion. The future of widespread application of biostimulants depends on precision agriculture, market maturity and further research to harness their potential.

Keywords: biostimulants; biotic and abiotic stress; growth; pulses; sustainable agriculture; yield

Introduction

Pulses are leguminous edible dry seeds rich in protein, fibers and minerals. They play an important role in agriculture as they can be cultivated as a food crop, rotation crop, cash crop and intercrop (1). They constitute excellent sources of protein, fiber, vitamins and minerals such as iron, zinc and magnesium thereby we can conclude that they are not only economically important but also nutritionally important (2). Legumes have a balanced ratio of protein (13-15 %) to carbohydrates (4-23 %), so they can be considered a viable source of protein for the global vegetarian population (3)

In 2022-2023, pulses accounted for 30.7 mha of national crop area producing 27.3 Mt of grain with an average yield of 902 kg/ha (4). Asia holds first place in pulses production with 43 percent of the regional contribution, followed by Africa (23 %), America (21 %), Europe (11 %) and Oceania (2 %). India is one of the major pulse-producing countries, contributing about 33 percent of the global area and 25 percent of the world production of pulses (5). The main pulses grown in India include lentil (*Lens culinaris*), chickpea (*Cicer arietinum*), cowpea (*Vigna unguiculata*) and kidney bean (*Phaseolus vulgaris*), which are consumed in a variety of ways (2). Pulses require fewer natural resources, improve soil health, promote sustainability and ensure nutritional security (6). Because of the numerous benefits of pulses, the United Nations declared 2016 as the International Year of Pulses to highlight the nutritional and health benefits of pulse crops and encourage people to shift more towards this nutritional powerhouse in diets worldwide (7). Pulses pave the way for sustainable agriculture as it enhances soil fertility primarily through Biological Nitrogen Fixation (BNF) where they form symbiotic relationship with the bacteria called *Rhizobium* in root nodules and convert atmospheric nitrogen to plant usable form. Pulses also add organic matter through leaf litter and root residues and improves the soil structure by enhancing water retention and microbial activity (8).

Due to increasing population and economic activities, waste is generated in enormous amounts. Waste or the by-products produced in agriculture can be processed to anaerobic digestion, which leads to the formation of biogas and digestate. The latter is rich in micro and macro nutrients and can be used as biostimulants (9) Therefore, in recent years, there has been an increasing number of research on organic waste-derived biostimulants and their market is also increasing adding environmental safety to the agricultural production chain (10). For example, lignin salts extracted from pulp mill by-products promoted seed germination with increase in 5 % and increase in biomass of upto 22.6 % in beans. Protein hydrolysate extracted from chicken feather waste improved the soil fertility and yield of mungbean (10) Another reason for its market is the demand for chemical-free and safe food (11).

Biostimulants are natural substances that are biodegradable and safe to the environment. Humic acid (HA), fulvic acid (FA), protein hydrolysates, seaweed extracts, chitosan, arbuscular mycorrhizal fungi, phosphite and plant growth-promoting rhizobacteria (PGPR) are used as biostimulants to enhance plant production. Biostimulants are generally classified into two main groups, including non-microbial and microbial biostimulants. The non-microbial biostimulants does not contain microorganisms and are again classified as plant and seaweed extracts, protein hydrolysates, humic substances, or substances with a plant growth-promoting effect. They mostly depend on chemicals or any biological compounds that do not consider living microorganisms (12). The microbial biostimulants include phytohormones, compounds with hormonal-like activity (e.g., melatonin), or biostimulants extracted from other sources, such as organic substances from vermicompost. Chitin, chitosan, or chitosan oligosaccharides can also be used as biostimulants, as well as other natural polymers. Inorganic elements like NO and H_2S can also be considered biostimulants if they provide beneficial effects to plants. Microbial biostimulants include mycorrhizal and non-mycorrhizal fungi, *Trichoderma* and PGPR (13). Microbial biostimulants are important because the plant renders a wide range of microorganisms in their rhizosphere and phyllosphere. They help in biological nitrogen fixation, carbon and nitrogen cycling, nutrient accumulation and thereby enhance the growth and yield of pulses (14). Biostimulants significantly enhance the sustainability of the pulse production by improving the yield, nutrient uptake and stress tolerance without negative environmental impact. The purpose of this review is to systematically synthesize current knowledge on the role of biostimulants in improving pulse crop productivity, with a focus on growth performance, yield stability and environmental benefits.

Definition and types of biostimulants

There is a wide range of definitions for biostimulants; however, they are categorized into two main characteristics growth agents and plant protection as they aid in the growth of the plants as well mitigate the abiotic stress. Biostimulants are a combination of natural substances or microorganisms that enhance the condition of the crop without promoting unfavourable side effects and help in attaining sustainable agriculture (15). Farmers can either use synthetic or natural biostimulant and improve biochemical, physiological, morphological processes and ensures protection against biotic and abiotic factors in plants (16). Different kinds of biostimulants are mentioned in Fig. 1.

Challenges in pulse production

A variety of agroclimatic, infrastructural and economic constraints limit pulse output. The cultivation of key pulses, such as green gram, has declined by more than 40 % in land and almost 60 % in yield. Due to a shortage of rainfall, black gram experienced a 20 % decrease in area and productivity from the previous year in 2023-2024 (17). Although the total area under pulses reached 30.7 mha and productivity improved to 902 kg/ha in 2022 - 2023, it is still insufficient to meet national demand (17). Pulse yield is influenced mainly by two factors, biotic (i.e., insects, pests, microbial attacks) and abiotic factors (salinity, heavy metals, drought, water logging, toxicity, or deficiency of minerals). Storage losses of pulses are contributed by bacteria, fungi, rodents and insects, of which extensive loss to stored seeds is contributed by insects up to 50 % (18). Therefore, limited access to high-quality seeds is also an additional challenge.

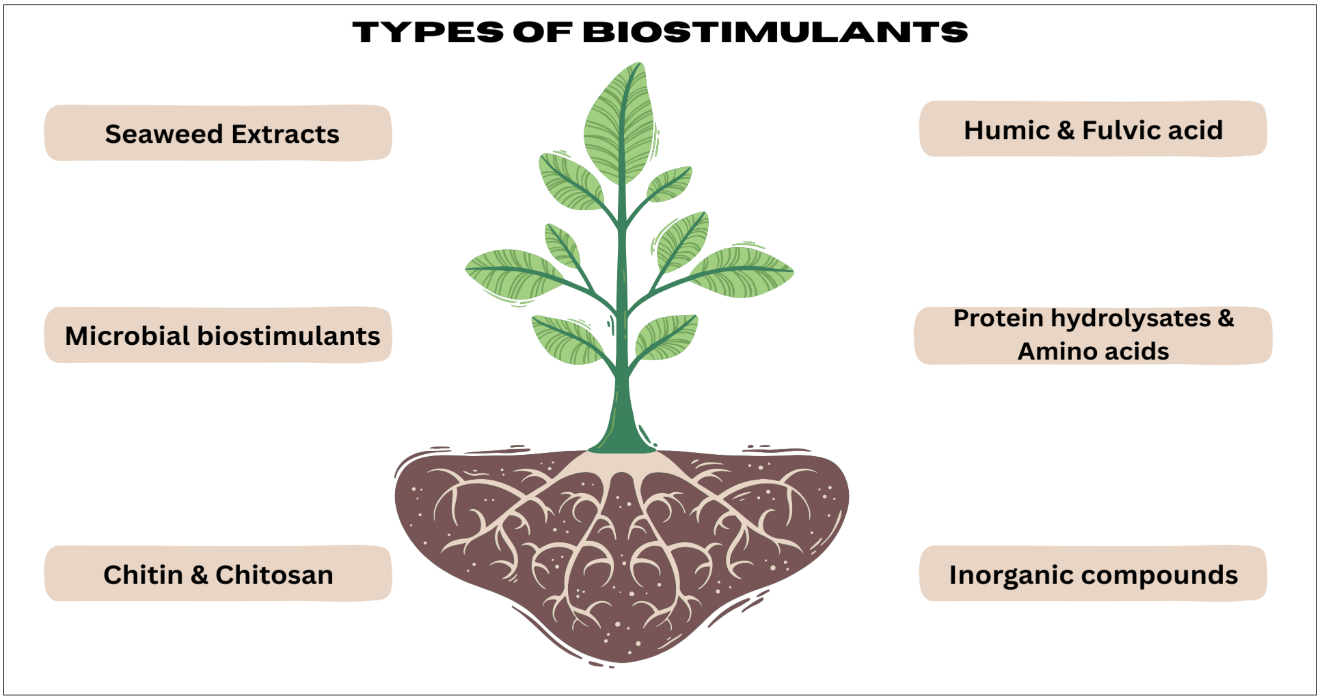

Fig. 1. Types of biostimulants.

There are several other constraints in pulse production. Almost 48.33 % of farmers face the non-availability of biofertilizers locally and 15 % of farmers face the non-availability of insecticides (19). In the current scenario, scarcity of irrigation water, only 23 % of the area cultivating pulses are under irrigation which remains the main problem for farmers as it affects crop production globally. As an alternative to mitigate this stress condition, biostimulants are increasingly used. Overall, to face these multifaceted issues urges the need for low-input, high efficiency solutions such as biostimulants, which enhance stress tolerance, nutrient uptake and crop resilience in a sustainable manner (17).

Protein-based biostimulants

Protein-based products can be divided into two major categories: firstly, protein hydrolysates which consist of mixture of peptides and amino acids of either plant or animal origin and secondly individual amino acids such as glutamine, proline, glutamate and glycine betaine. Protein hydrolysates are produced through thermal, enzymatic, or chemical hydrolysis of different variety of animal and plant residues, including animal epithelial or connective tissues, animal collagen and elastine. They are considered as biostimulants because of their minimum side effects, low cost, easy absorption, high activity and lower molecular weight (20). Amino acids used in biostimulant production are extracted from plant (corn, soyabean, etc.,) and animal proteins through chemical and enzymatic processes (21). It aids in nitrogen uptake and its assimilation by regulating enzymes. It scavenges the free radicals (reactive oxygen species) and enables the plant to tolerate environmental stress (22). In chickpea drought stress was controlled by the combined application of amino acids of commercial compound with valine (23). Increased crop yield was seen in cowpea with the application of amino acid liquid biological fertilizer (24). Protein based commercial biostimulants and their function are listed in Table 1.

Microbial biostimulants (Beneficial bacteria and fungi)

The Bacterial Plant Biostimulants (BPB) based on plant growth-promoting rhizobacteria (PGPR) have a major role in promoting/stimulating crop plant growth through a number of mechanisms. They include (i) nutrient acquisition through nitrogen (N_2) fixation and solubilization of insoluble minerals (K, P, Zn), siderophores and organic acids; (ii) antimicrobial metabolites and various lytic enzymes; (iii) induction of plant defense; (iv) mitigating abiotic stressors like high soil salinity, extreme temperatures, drought, oxidative stress and heavy metals through various modes of action; and (v) the action of growth regulators and stress-responsive phytohormones (32). In common bean, strains of *Pseudomonas. fragi*, *P. proteolytica*, *P. fluorescens*, *P. chloropaphis* and *Brevibacterium frigoritolerans* acted against cold stress by decreased ice-nucleating activity, lipid peroxidation and chill damage in proportion to ROS levels and increased apoplastic antioxidant enzyme activity (33). Commercial PGPR-based biostimulants are enlisted in Table 2.

Beneficial fungi just like beneficial bacteria, enhance plant growth, nutrient uptake and stress tolerance. They promote root growth, improve soil health and helps plants to tolerate various stress, including drought, salinity and temperature stress. Mycorrhizal fungi like *Glomus* sp. enhance water absorption and phosphorus uptake. Species like *Trichoderma* reduce disease incidence and aid in plant growth and development. In a study, a biostimulant named bioE was used in chickpea. This biostimulant is a commercial blend of endomycorrhizal fungi, such as *Glomus intraradices*, *Trichoderma virens*, *T. harzianum*, *T. viride* and *T. reesei* and plant-growth-promoting rhizobacteria (PGPB). It consists of *Azospirillum brasilense*, *Azotobacter chroococcum*, *Pseudomonas fluorescens and Bacillus megaterium* as well as vitamins, such as folic acid (B9), biotin (B7), B2, B3, B6, B12), amino acids from rotein hydrolysate, carbon (C), potassium (K) and soluble extract of *Ascophyllum nodosum* and *Yucca schidigera*. The beneficial bacteria and fungi now resulted in successful germination and increased growth attributes (37).

Table 1. Protein-based commercial biostimulants and its function

S.No	Commercial biostimulant	Function	Reference
1.	Deflan plus	Resistance to biotic and abiotic stress, increased photosynthetic rate	(25)
2.	Perfectose™	Increase the nutrient quality of the plants	(26)
3.	Phytostim®	Increase in overall growth and development of the plants	(27)
4.	Codasil®	Resistance to drought stress	(28)
5.	Bonamid®	Increased leaf water and chlorophyll content	(29)
6.	AminoPrim	Enhance the mineral component in plants	(30)
7.	Sorb®	Increased yield especially in nitrogen-limiting soil	(31)

Table 2. Commercial PGPR-based biostimulants

S.No.	Commercial biostimulants	PGPR strains	Function
1.	FZB24®fl, Rhizovital 42®	*Bacillus amyloliquefaciens* and *B.amyloliquefaciens* sp. *plantarum*	Pathogen control and Phosphate availability.
2.	Ceres®	*Pseudomonas fluorescens*	Biocontrol agent
3.	Gmax®	*P. fluorescens*, *Azotobacter* and *phosphobacteria*	Disease control, overall growth and development of plants, nutrient use efficiency of nitrogen and phosphorus.
4.	Inogro Bio Gold	*Azotobacter chroococcum* + *Pseduomonas fluorescens*	Nitrogen fixation, disease and drought resistance

Plant secondary metabolites

Plant secondary metabolites are a class of compounds produced during the growth process of plants. They are used as biostimulants because of its significant biological activity. They are classified into phenolic acids, steroids, terpenoids, flavonoids and alkaloids based on their chemical structure (38). They act as signaling molecules, protect the plants from abiotic stress and play a crucial role in plant growth. Secondary metabolites also play diverse roles such as facilitating communication, controlling enzyme functions and providing defense mechanisms. These compounds enhance the taste and olfactory characteristics of plants with their dynamic colors and aromas and they significantly contribute to plant interactions with various organisms including pollinators, pathogens and herbivores (39). To create a biostimulant, start by understanding what a plant requires during its growth stage and the specific biotic and abiotic stresses a plant may encounter. Secondary metabolites act as potent stimulators of root and shoot development (40). The root morphology influences nutrients and water content in plants which in turn, enhance plant growth and development. Heavy metals phytotoxicity is one of the major abiotic threat that affect cropping systems. Generally, cadmium (Cd), lead (Pb), arsenic (As), nickel (Ni), cobalt (Co) and mercury (Hg) are the most ample contaminants in soils (41). Plants have developed various strategies to overcome metal-induced stresses and survive healthily. These include strategies like antioxidant defense, inhibition of metal uptake by root cells and the production of stress-related compounds such as phytohormones and polyamines (42). These activities enhance the application of secondary metabolites in bioformulation to manage different abiotic stresses. Phytohormone-jasmonic acid suppressed cadmium-induced stress in *Vicia faba* when applied at a concentration of 2.1 ppm. (43). Likewise, 24 -Epibrassinolide alleviated Zn-induced stress in *Vigna radiata* when applied at the concentration of 0.0048 ppm (44).

Chitin and chitosan

Chitosan is a deacetylated form of the biopolymer chitin. It can be produced both naturally and industrially. The use of chitin and chitosan as biostimulants increased the yield and resulted in agro-environmental sustainability. They gained noticeable attention as they are biodegradable and nontoxic which is currently very much important to save the world from the environmental threat. The biological activity of chitosan and its derivatives, which mainly consists of the capacity to enhance plant tolerance to stress and activate natural defence mechanisms, is linked to the possibility for use of these substances in the elicitation process. Numerous physiological and biochemical alterations are related to this, including oxidative stress, H_2O_2 buildup, the production of secondary metabolites (phytoalexins, flavonoids, polyphenolic compounds and alkaloids), enzymes (glucanase, chitinase and protease), growth inhibitors (salicylic acid, abscisic acid, jasmonic acid) and lignin and callose accumulation (45).

Chitosan prevents harmful effect on plants and increases the yield. Several effects have been reported for chitosan application in plants. It increases the stomatal conductance and reduces transpiration and therefore can be used as a plant growth promoter. It can also be applied as a coating material in seeds. It acts as a physical barrier limiting plant contact with pathogenic microorganisms due to its film-forming properties (46). Application of a chitosan solution to soybeans has been documented to diminish the prevalence of herbivorous insects (47). Chitin prolongs the storage life through post-harvest treatments by promoting chitinolytic microorganisms and also helps in the acquisition of nutrients by the plants since it aids in the slow release of nutrients and prevents the leaching of nutrients. Since chitosan is a readily available, eco-friendly biopolymer with many advantageous biological characteristics, it finds extensive use in a wide range of industries. Furthermore, by boosting the internal potential of plants, it may be able to address a number of ecological issues, particularly those related to plant production. Benefits of chitosan as biostimulant is elucidated in Fig. 2.

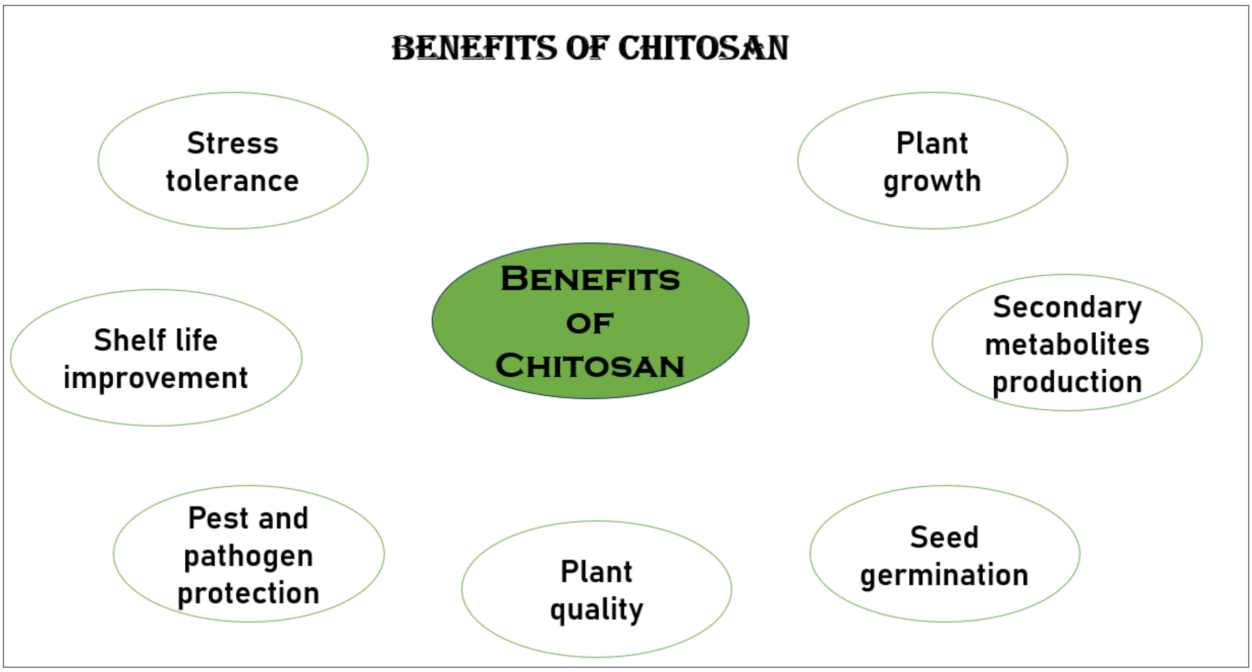

Fig. 2. Benefits of chitosan as biostimulant.

Seaweed extract

Seaweeds comprise a diverse group of macroscopic marine algae, including various multicellular algae from distinct taxonomic categories like green, brown and red algae. There are several plant growth-stimulating compounds present in seaweed extract that have been widely used as amendments in crop production. Seaweed constitutes polysaccharides, laminarin, carrageenans and alginates (48). It also contains micronutrients, macronutrients, N-containing compounds like betaines, sterols and hormones like auxins and cytokinins for plant growth promotion (49). The polyanionic group present in seaweed extract results in fixation of heavy metals and thereby aids in soil remediation. The awareness on the potential of seaweed extract is still not up to the level (50, 51). Seaweed extracts that are commercially available, frequently contain more than one species of seaweed as nonspecific selection might occur during wild collection. Compounds in seaweed extracts can influence various cellular processes by acting as signalling molecules, ultimately leading to the expression of key genes in crops that promotes enhanced growth and stress resistance (52). Seaweed production has greatly increased to 30 million metric tons yearly, also offering benefits for agriculture by enhancing crop nutritional value and stress resistance besides serving as a food source. It increased the protein content in the pulses thereby adding nutritional value to the crop (53). There is a lot of scope for exploiting seaweed extract as a biostimulant. Therefore, research is being carried out on the plant by using seaweed extract as a biostimulant and the yield is being recorded.

Humic acid

Humic acids (HA) are the primary organic components found in soil and solid fossil fuels such as peat, brown coal and sapropel. Their composition ranges from 5-7 % in humus-rich soils to more than 80 % in some forms of brown coal (leonardite) (54) Humic acid are complex supramolecular association and it is produced through humification of organic matter present in soil. Humic acid in particular improves the root growth, it focuses on growth of lateral roots, increase the contact between roots and soil and regulates nutrient transformation (55, 56). This is because humic acid have auxin-like qualities that modify plant metabolism, resulting in positive impacts on plant growth and productivity, such as better nutrient usage efficiency, increase in fresh and dry weight, photosynthesis and greater abiotic and biotic stress tolerance (57). Plants' phosphorus metabolism changes, as evidenced by an increase in the amount of organophosphorus chemicals that are involved in energy transfer and transformation reactions. Sugars build due to the intensive utilization of absorbed phosphorus and enhanced synthesis of nucleic acids (58). In *Pisum sativum* (pea), biostimulant made of humic acid resulted in increased microbial communities especially oxolobacteriaceae that increased N fixation (59). Humic acid structure contains many functional groups in which COOH and OH functional group are mainly responsible for improving soil physical and chemical properties. Increase in nutrient availability reduce the transportation of toxic heavy metals intake by plants (60). Role of humic acid is given in Fig. 3. Types of biostimulants, active ingredients, concentration and its response are given in Table 3.

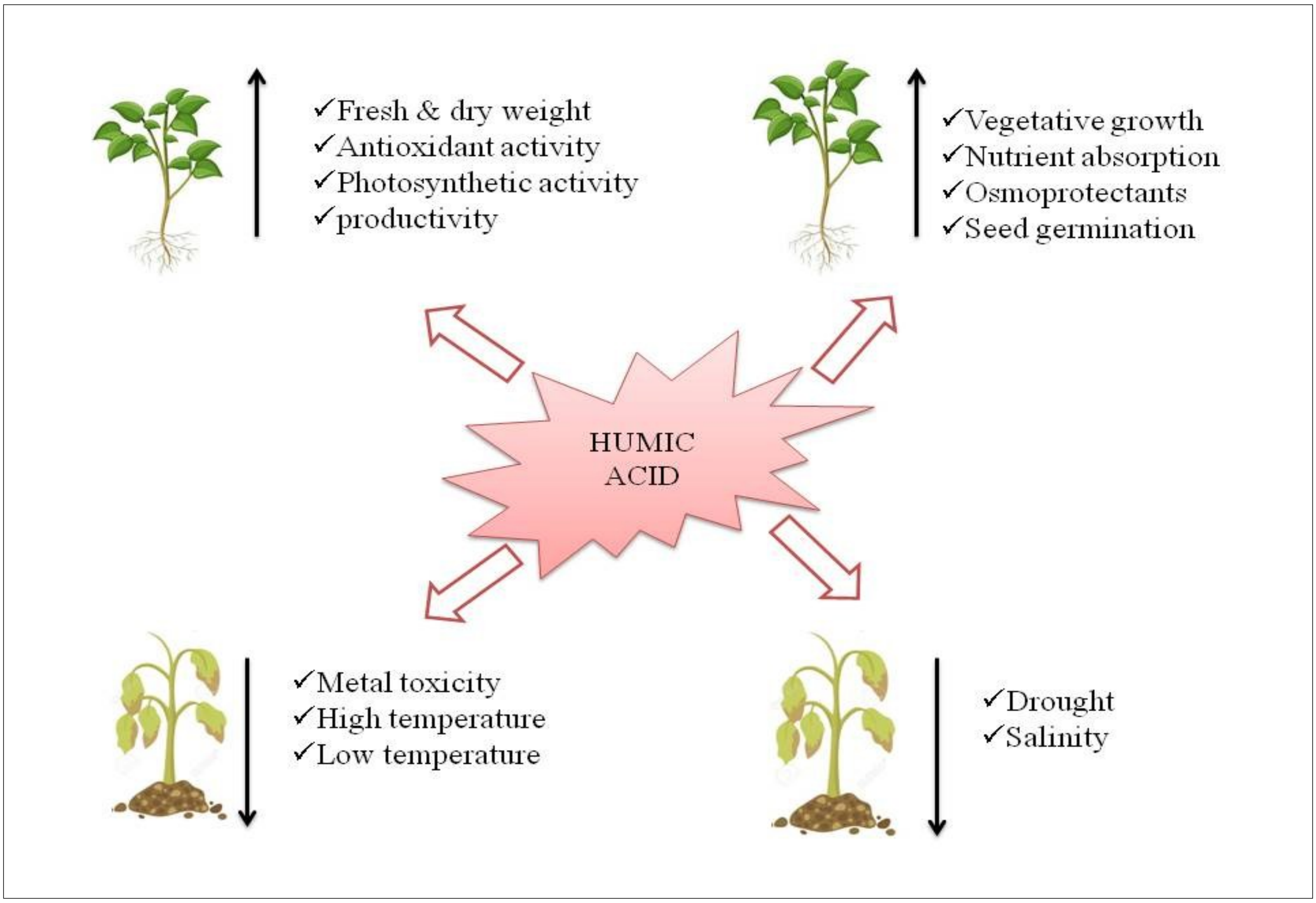

Fig. 3. Role of humic acid.

Table 3. Types of biostimulants, active ingredients, concentration and its response

S. No	Biostimulant	Crop	Active ingredients	Concentration	Response	Reference
1.	Bio E (Commercial Biostimulant)	Chickpea	•Endomycorrhizal fungi - *Glomus intraradices* •Other fungi including *Trichoderma virens*, *T. harzianum*, *T. viride and T. reesei* •Plant-growth-promoting rhizobacteria (PGPB) - *Azospirillum brasilense*, *Pseudomonas fluorescens,* •*Azotobacter chroococcum* and *Bacillus megaterium* •vitamins- folic acid (B9), - biotin (B7), B2, B3, B6, B12 •Carbon (C), potassium (K), •Amino acids derived from protein hydrolysate, •Soluble extract of *Yucca schidigera and Ascophyllum nodosum*,	•1 mL of liquid biostimulant for 100 g of seed 24 hr before sowing (BioE).	•Increase in germination percentage, plant height and number of other growth attributes	(61)
2.	Microalgal suspension	Common bean	*Chlorella vulgaris*, *Tetradesmus dimorphus* and *Arthrospira platensis*	•Concentration of 10 g 100 mL^{-1} as foliar spray	•Increase in plant height •Increase in dry weight of the plant •Increase in protein content •Number of pods/plant and seeds/pod is increased •Increase in dry weight of the pods	(62)
3.	Natural plant extracts	Faba bean	Extracts derived from raw propolis material and maize grains	•Foliar application - Maize grain extract - 40 g L^{-1} •Raw propolis material extract - 60 g L^{-1}	•Tolerance against salinity, drought and cadmium stress.	(63)
4.	Quantis (Commercial Biostimulant)	Black gram	Organic carbon, amino acid and micronutrients.	•Foliar spray at the concentration of 5 mL L^{-1}	•Increase in growth and yield attributes and overall production efficiency.	(64)

Mode of application of biostimulants

Depending on the type of biostimulant and crop stage, biostimulant can be sprayed on pulse crops using several techniques. Seed treatment, in which seeds are covered using biostimulant before sowing to boost early growth and root development. Commerical biostimulant BioE increased the growth attribes of chickpea through seed treatment (61). Foliar application where the biostimulants are applied straight to the leaves for rapid absorption during crucial development phases. A microalgal suspension was applied to common bean by foliar spray resulting in enhanced plant development (62). Soil application, either drip irrigation or direct incorporation into the soil to stimulate microbial activity and improve nutrient availability is another mode of application (61). These techniques guarantee focused distribution and maximize the biostimulant efficacy in enhancing plant health and output. Different mode of application of biostimulants are given in Table 4 and Fig. 4.

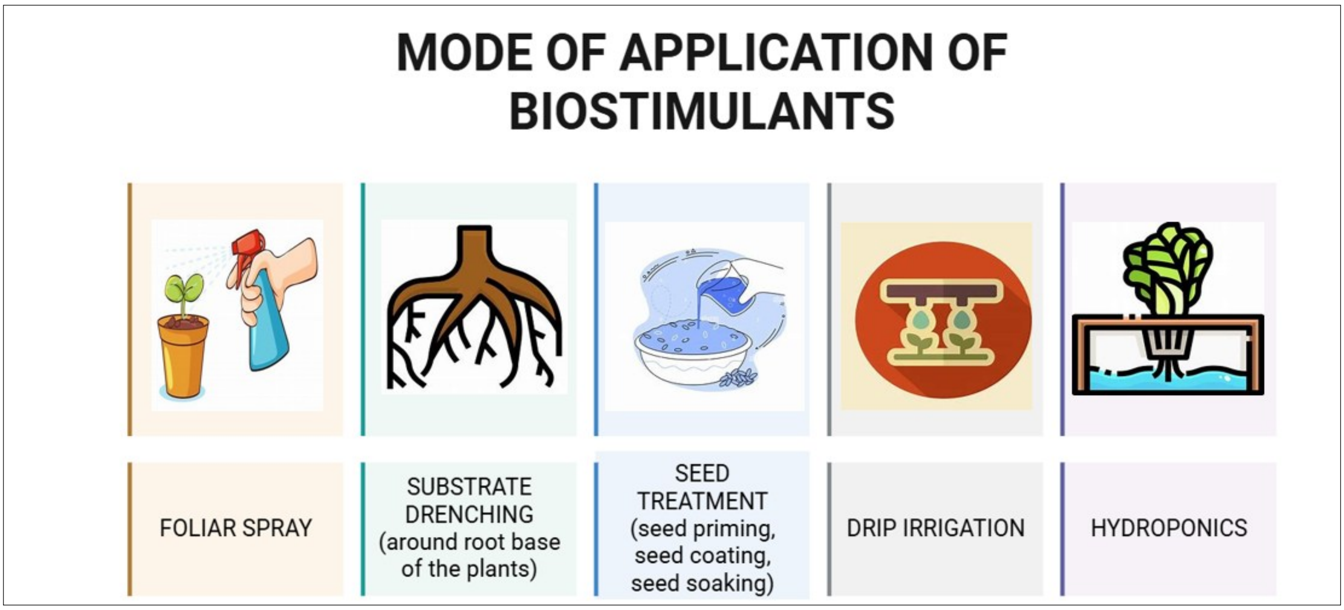

Fig. 4. Mode of application of biostimulants (Created in BioRender.com).

Table 4. Commercial biostimulant, active ingredients, mode of application and its response

S. No	Commercial biostimulant	Active ingredients	Mode of application	Response
1	Acadian Goldstar	*Ascophyllum nodosum*	Foliar Drip drenching	•Mitigates abiotic stress. •Tillering and branching is increased. •Improve nutrient and water use efficiency •Improves the health of both soil and the plant. •Overall yield and the quality of the crop is optimized.
2	Biozyme	78 % Natural plant extracts, 0.4 % sulphur, 0.4 % sulphur, 0.4 % iron, 0.3 % zinc, 0.3 % boron, 0.1 % manganese, 0.1 % magnesium	Foliar drenching	•Development of root and shoot •Tolerance to stress •Increase in nutrient uptake •Soil health is improved
3	Abda gold	Humic and fulvic acids	•Should be applied immediately after sowing •Can be applied with basal fertilise •Can be applied at the initial stages like earthing up or top dressing •Ring application is suitable for tree crops.	•Gives robust and uniform root growth •Produces balanced root: shoot ratio •Economic yield of the crop is increased •Increases in cost benefit ratio
4	Spic em Power - I	16 % Humic, 8 % Fulvic and 9 % Amino acids	Foliar spray	•Promotes photosynthesis •It improves the overall physical, chemical and biological activities of the plant

Effect of biostimulants on productivity of pulses

Biostimulants alleviate the negative effects of environmental stresses to which plants are exposed including both biotic and abiotic factors. They act through multiple mechanisms, gene expression, modifying metabolism and phytohormone production, promoting the accumulation of compatible solutes and scavenges the Reactive oxygen species thereby mitigating oxidative stress (65).

Biostimulants like cow urine, panchagavya, jeevamrutham, vermiwash and seaweed extract have been used all over the world to improve crop yields. They are natural substances derived from plants and animals that stimulate plant processes at very low concentrations. These can be given to plants in various forms like seedling dip, soil application and foliar application and when applied to the plants, it modifies the metabolic processes of plants such as photosynthesis, respiration, ion uptake and nucleic acid synthesis. These biostimulants are rich sources of macro and micronutrients that are required by plants in different amounts for the better growth of plants. Biostimulants also constitute naturally occurring plant growth promoters like Gibberellic acid, NAA, cytokinin, etc. in good concentration (66).

Effect of biostimulants on plant growth

Experiment was conducted in black gram to study the increase in growth attributes of Black gram by the application of PEPTO (plant based biostimulant at the rate of at the rate of 5l/ha on 20-25 and 40-45 days after sowing (DAS). The maximum plant height was recorded at 45 DAS (28.57 cm), at harvest (40.62 cm). Maximum leaf index at flowering (1.79) and dry matter production (20.51g/plant) was observed. The increased LAI is because of presence of certain metabolites in the biostimulant which made the stomata to open for a longer period both during favourable and adverse conditions (67). Different biostimulants such as humic acid, moringa leaf extract, vermicompost tea and yeast extract were used to increase the growth parameters in *Phaseolus vulgaris* L. (common bean) in which moringa leaf extract resulted in maximum plant height (84.56 cm), number of leaves (24.11), number of branches/plant (7.0), total fresh weight (157.89 g), dry weight (31.14 g), stem diameter (1.10 cm) (68).

Biostimulants such as seaweed extracts are that are obtained from marine brown algae - *Sargassum plagiophyllum, Turbinaria conoides, Padina tetrastromatica, Dictyota dichotama.* Use of biostimulants stimulated the growth parameter of green gram. The maximum root length (8.8 cm) and shoot length (16.4 cm) was observed in green gram with the treatment with *Turbinaria conoides* at the concentration of 6.3 % out of all six seaweeds tested (69). The genotypes kabuli & desi chickpea treated with 2.0 % *Ascophyllum nodosum* (biostimulant). They were treated for 6 hours and resulted in maximum number of branches at 25 DAS (17.93 & 16.60) and 50 DAS (36.70 & 33.57) (70). Liquid biostimulant extracted from *Chlorella vulgaris* and *Spirulina platensis* resulted in maximum shoot and root length in green gram (71). Another study in common bean using the biostimulant extracted from chlorella vulgaris resulted in increased plant height and dry weight (62). Application of natural extracts of propolis, a biostimulant increased the plant height upto 100 cm in faba bean (63). Use of biostimulants such as fulvic and ascorbic acid at the rate of 200 ppm in *Pisum sativum* resulted in increased shoot length (109 %) when compared to control (72). Similar increase in growth parameter was observed in chickpea when a blend of biostimulants were used such as *Trichoderma viride*, plant growth promoting rhizobacteria (*Azospirillum brasilense)* and soluble extract of *Ascophyllum nodosum* (61).

Effect of biostimulants on yield attributes of plant

Application of organic biostimulants in black gram along with liquid *Rhizobium* resulted in increased yield and yield attributes. Maximum number of pods plant^{-1} (37.7), number of seed pod^{-1} (9.2), grain yield (1063 kg/ha) and haulm yield (2108 kg/ha) was recorded by the application of biostimulants (73). Quantis (biostimulant) was applied to black gram at 5mL L^{-1} during the pre-flowering stage recorded a higher number of pods/plant (49.38), pod weight/plant (18.45 g/plant), seed yield

(922 kg/ha), haulm yield (323 kg/ha) and biological yield (1245 kg/ha) (74). When chickpea was treated with 2 % of *Ascophyllum nodosum*, for six hours increased the number of pods plant^{-1} (61.52 & 60.24), number of pods plot^{-1}(1025.67 & 922.00), number of seeds plant^{-1} (102.5 & 100.4), number of pods plant^{-1} (14.25 & 1525), seed yield plant^{-1} (28.30 & 25.35), seed yield plot-1 (358.44 & 328.86), biological yield (3187.42 & 3061.24), economical yield (1341.67 & 1231.33) in both kharif and rabi season (70).

Application of Novel prime in cowpea at the rate of 2 % resulted in maximum number of pods/cluster (3.22), average pod length at second and fourth picking (14.14 cm and 14.17 cm respectively), number of clusters (35.07 plant^{-1}), pod yield (0.22kg/ plant), total pod yield (11.25 t/ha), marketable pod yield (10.52 t/ha), average pod volume at second and fourth picking (3.73g and 3.00g respectively) (75). Similar results were seen in the experiment conducted in black gram with the application of *Rhizobium* at 1 % with organic biostimulants (seaweed extract at the concentration of 1 % and 3 %) and pink pigmented facultative methylotrophs. The study resulted in the highest yield attributes such as number of pods/plant (37.7), number of seeds/pod (9.2), grain yield (1063 kg/ha) and haulm yield (2108 kg/ha) when compared to control with grain yield (936 kg/ha) and haulm yield (1666) (76). Maximum number of pods per plant (30.33), grain per pod (2.33) and grain production (2535.8 kg/ha) was recorded in chickpea with the application of Potassium Sulfate (5 %) 120 L of water/acre and the least number of number of pods per plant (29.11), grain per pod (2.01) and grain production (2353.1 kg/ha) was recorded in control (77).

To enhance the yield and yield attributes of bush bean, biostimulants such as seaweed extract, panchagavya, chitosan and effective microorganism were used at different concentration. Foliar application of biostimulants was carried on 30, 45 and 60 DAS. Treatment with seaweed extract at the rate of 5 mL/litre proved to be superior compared to all other biostimulants and resulted in increased length of raceme (51.2 cm), racemes/plant (8.5), flowers/raceme (26.4), flowers/plant (221.5), number of pods/plant (41.6), pod length (10.6 cm), pod width (3.2cm), single pod weight (4.6g), single pod weight (3.0 g), number of seeds/pod (5.2) and B:C ratio (3.14), whereas, control resulted in minimum number of pods/plant (32.1), pod length (6.0 cm), pod width (2.3 cm), number of seeds/pod (4.2) and B:C ratio (1.78) (75). Application of mixture of biostimulant protein hydrolysate resulted in increased pod weight (17.00 g) compared to control (14.00 g) when applied at the rate of 5l/ha each 7 days (78).

Effect of biostimulants on nutrient accumulation

Humic substances positively influence plant nutrition by affecting soil processes and it directly affect plant physiology. The mechanism with which the biostimulants affect the soil processes includes 1) improvement of the soil structure, 2) enhancing the micronutrient solubility in soil, 3) changes in root morphology, 4) increase in the root activity of H^+ ATPase and 5) an increase in the activity of NO_3. Studies in lentils and peas proved that co-inoculation of two biostimulants increased the nitrogen content of the plants. Harvested grains show enhanced micronutrients in the biostimulant inoculated plot - Fe (6.1 and 3.9 mg 100 g^{-1}), Zn (3.9 and 4.0 mg 100 g^{-1}), Mn (3.1 and 1.6 mg 100 g^{-1}), Cu (0.7 and 1.4 mg 100 g^{-1}) and Ca (155 and 141 mg 100 g^{-1}) over the un-inoculated control plots with Fe (5.0 and 3.0 mg 100 g^{-1}), Zn (3.4 and 3.7 mg 100 g^{-1}), Mn (3.0 and 1.4 mg 100 g^{-1}), Cu (0.7 and 1.1 mg 100 g^{-1}) and Ca (122 and 120 mg 100 g^{-1}) for chickpea and pigeon pea respectively (79). Mung bean inoculated with *Bacillus aryabhattai* S10 and *Bacillus subtilis* ZM63 resulted in maximum nitrogen concentration (2.71 %), phosphorus concentration (0.59 %), potassium concentration (2.15 %) and protein contents (16.96) in shoot over uninoculated control with minimum nitrogen (1.38 %), phosphorus (0.30 %), potassium (1.63 %) and protein content (8.65) (80). Seaweed fertilizer of *Caulerpa peltate* at the concentration of 10 % resulted in increased protein content (256 mg/g) when compared to control with the least protein content (128 mg/g) in green gram (81).

Bacterial biostimulants can be categorized into two types: endosymbionts mutualistic *Rhizobium* and Rhizospheric mutualistic plant growth-promoting rhizobacteria (PGPR). *Rhizobium* bacteria aids in the acquisition of nutrients, especially nitrogen whereas plant growth-promoting rhizobacteria contributes to multifunctional benefits including improvement of growth and nutrition, tolerance to abiotic and biotic stress, development and morphogenesis and interactions with other agrosystem organisms (82). Examples of PGPR include some strains of the genus *Pseudomonas*, *Bacillus* and *Glomus*. Bacteria perform certain mechanisms for the enhancement of nutrient uptake such as nitrogen fixation, phosphorous solubilization, enhancement of root surface area and siderophore production. During a symbiotic relationship both rhizobia and legume undergoes transformation. Several PGPRs does not help in fixing nitrogen but enhance the uptake of nitrogen by increasing the growth of roots thereby large amount of soil comes in contact with plants. This is how *Rhizobium* activity increase yield and quality and enhanced the NPK uptake (83).

Fulvic acid and amino acid at 200 ppm in combination resulted in excellent nutrient accumulation in pea seeds i.e., N (16.17 %), P (40.47 %) and K (39.96 %) (72). Foliar application of organic biostimulant Panchagavya @ 3 % solution increased the nitrogen (65.72 kg/ha), phosphorus (11 kg/ha), potassium (44.52 kg/ha) and protein uptake (20.89 %) in chickpea (84). An experiment was conducted in *Vicia faba* with the foliar application of zinc sulphate (0.3 %), ammonium molybdate (0.05 %) and iron sulphate (0.6 %) along with biostimulants – inoculation with *Rhizobium* and 10kg humic acid/feddan. It resulted in increased N (4.65 %), P (0.47 %), K (3.76 %). Therefore, there was increased N, P and K content in seed in biostimulant treated faba bean than control (85). Effects of biostimulants on cellular, biochemical and physiological characters are elucidated on Fig. 5.

Effect of biostimulants on quality parameters

Application of Novel prime 2 % in cowpea resulted in increased protein content (6.13 %) which was on par with the application of seaweed extract at the concentration of 15 %. Crude fibre content (14.25 %) was highest in the treatment with application of cow urine. Total soluble solids (7.77 brix) were highest in the treatment with application of Novel prime 2 % (75). Addition of vermicompost tea as biostimulant in common bean increased the crude protein content (32.07g/100g) and the crude fibre content (3.14g/100g) (68).

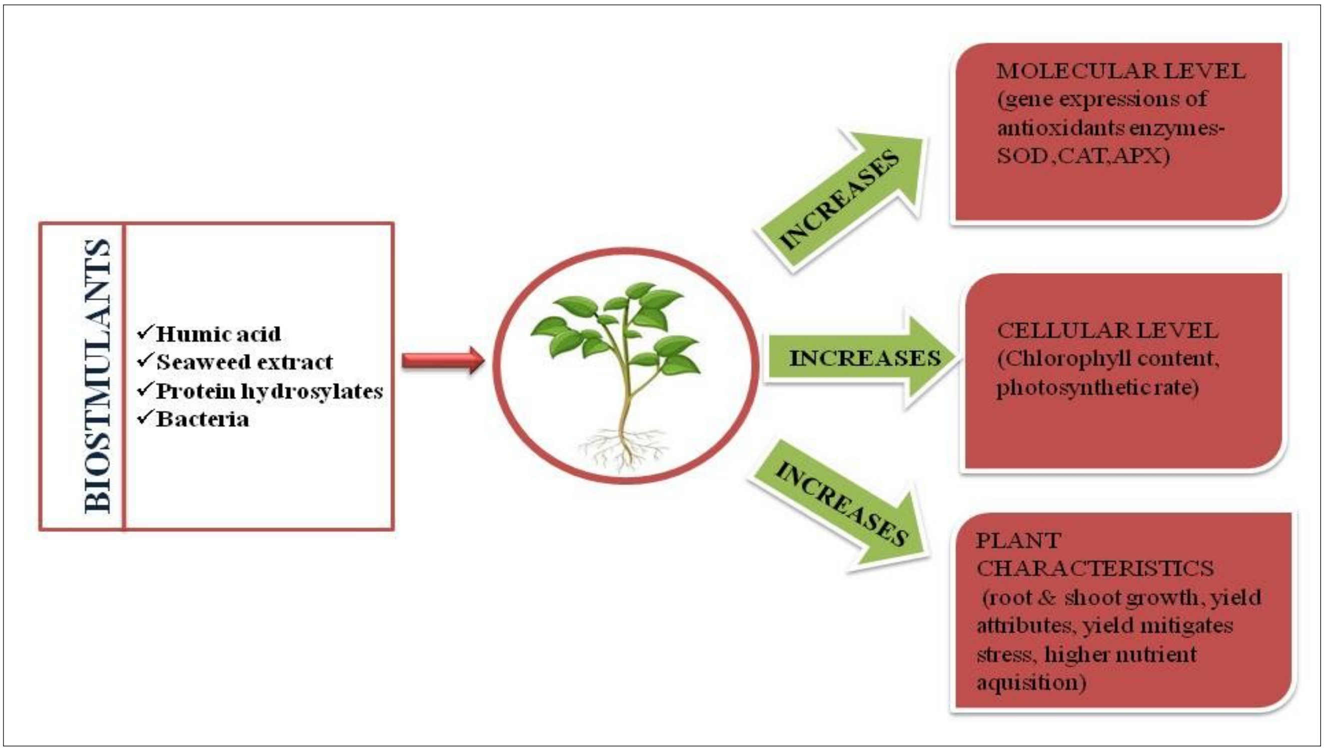

Fig. 5. Biostimulants and its effect on cellular, biochemical and physiological characters.

Effect of biostimulants in mitigating abiotic stresses, pest and diseases

Abiotic stress in plants include drought, salinity, high and cold temperature and metal toxicity (86). Abiotic stress almost contributes 50-70 % of yield losses (87). To overcome this problem, biostimulants act as excellent agricultural tool and reduces the stress thereby increasing the yield and productivity (88). For example, seaweed extract application in chickpea under salinity stress, the yield recovery was 20-30 % and inoculation of *Rhizobium* and *Azospirillum* in faba bean under salinity and drought stress resulted upto 20-28 % yield recovery. In general, biostimulants can partially recover 20-40 % losses caused by abiotic stress in pulses (89). Biostimulants, both microbial and non microbial enhances stress tolerance by various mechanism such as scavenging reactive oxygen species, metal chelation, phytohormonal signaling in plants (90). The mechanism of action of biostimulants vary from one biostimulant to another. Protein hydrolysate enter plants through diffusion process via membrane openings whereas microbial biostimulants enter the plants by the process of penetration of hyphae into plant tissues and forms a symbiotic association with plants (91).

Seaweed biostimulants based on *Ascophyllum nodosum* increase the activity of antioxidant enzymes (e.g., Catalse, Superoxide dismutase and Ascorbate peroxidase) and the production of antioxidants (e.g., ascorbate), resulting in lower Reactive oxygen species (ROS) accumulation and less membrane damage in *Phaseolus vulgaris* (53) under drought and salt stress. Application of propolis extract (biostimulant) in faba bean enhanced stress tolerance such drought and salinity by altering the antioxidant system and by increasing the enzymatic activities of superoxide dismutase, catalase, ascorbate peroxidase etc., (62). Application of vegetal protein hydolysates at the concentration of 2 mL L^{-1} under water deficit conditions resulted in membrane stability index, total chlorophyll and proline content of leaves and relative water content (92). When chickpea was treated with a commercial biostimulant (Regoplant), increased resistance to pathogenic micromycetes of *Fusarium* L. genus was seen (93). Commercial biostimulant (EM Bokashi) contains a mixture of fungi and bacteria, which on application on bean plants controlled *Rhizoctonia solani* disease (94). Type of biostimulants used in target crop, its effect on stress tolerance and its mechanism is given in Table 5.

India biostimulants market

Between 2017 and 2025, the global biostimulants market is projected to expand at a Compound Annual Growth Rate (CAGR) of 10.2 %, increasing from its 2016 size of USD 1.74 billion to USD 4.14 billion. The market is expected to be propelled over the forecast period by a heightened focus on enhanced productivity alongside escalating soil degradation. Seaweed extract-based biostimulants predominate in the Indian plant biostimulants industry. It constitutes over 39 % of the overall market value in 2024 (100).

Way forward

In the current climatic conditions, pulses can tolerate a wide range of adverse conditions, as they require less resources to produce potential yield. Biostimulants are regarded as novel and safe with the utmost potential to not only increase the productivity of crop but to alleviate stress. Even then farmers intend to use conventional fertilizers rather than biostimulants.

1. Enhancement of farmer awareness and training Demonstration on the application of biostimulants should be made and more research should be carried out in this field to boost the productivity of pulses. When compared to cereals & vegetable crops, use of biostimulants in pulses is not much in practice. Establishing standard protocols and framework will build trust among farmers. Therefore, research and experimentation should be done in pulse crop, to increase the annual global pulse productivity.

Table 5. Type of biostimulants, target crop, stress tolerance and its mechanism

S. No.	Environmental stresses	Biostimulants	Crop plants	Mechanism	Reference
1.	Salinity	*Azospirillum brasilense* (Seaweed extract)	*Vicia faba*	salinity stress tolerance	(95)
2.	Salinity	Humic acid (0.05 % and 0.1 %)	*Phaseolus vulgaris*	Increased uptake of nitrogen, phosphorus and nitrate	(96)
3.	Drought	*Pseudomonas putida, P. fluorescens* (microorganism)	*Pisum sativum*	Increase in overall growth and development	(97)
4.	Salinity	*L. leguminosarum* (microorganism)	*Pisum sativum*	Stress tolerance and plant growth	(98)
5.	Abiotic stresses	PGPR + Trichoderma	Pulse crops	Increases in activity of peroxidase and phenylalanine ammonia lyase	(99)

2. Development of crop and stress specific biostimulants: Tailoring biostimulants to targeted stress and crops can optimize efficacy. Custom formulation addresses the specific physiological needs of different pulse crops and different stress conditions.
3. Additionally, formulation of nano biostimulants, applying AI based tools leads to precise application.
4. Finally, disseminating biostimulants into national pulse development programs such as the National Food Security Mission (NFSM) can institutionalize their use and promote their use across India and beyond. The development of biostimulants holds promise for food security and sustainable agriculture.

Conclusion

In the last decade the major focus is towards sustainable agriculture considering the climate change. The increasing need to attain sustainable agriculture paves the way for the utilization of biostimulants to its maximum potential. Plant biostimulants are microorganisms that improves the plant growth, soil health, social health, increase the climate resilience and play a key role in environmental preservation. Declining soil fertility and increasing population are the two major problems in agriculture. We must produce food for the population without affecting the soil health. Therefore, biostimulants can be used as an alternative to increase productivity by enhancing soil health. Pulses are rich sources of protein, fibre and vitamins and they form a complete balanced diet, especially for vegetarians. Despite all the benefits of pulses, there is a yield gap because of improper package of practices. The use of biostimulants increases the growth, yield attributes, nutrient use efficiency, overall yield and mitigates the stress. Since biostimulants increase the chlorophyll content, increase in photosynthetic rate is seen. Good vegetative phase contributes to a good reproductive phase and results in increased yield and yield attributes of the crop. It helps in the fixation of atmospheric nitrogen and lead to enhanced nutrient accumulation. By this way biostimulants naturally reduces the amount of chemicals introduced in the environment, thereby reducing the pollution.

Biostimulants have the natural ability to biostimulants are of natural origin and is different from conventional inputs like fertilizers and pesticides. It contributes great to the environmental protection and sustainable agriculture. The main advantage of using biostimulants includes the positive impact on the crop growth and development without any negative impact. To attain the food security, the production of biostimulant products is needed in much higher quantity. Farmers are still unaware of technology like biostimulant. Different plant requires different biostimulant and all this nature of biostimulants make it less possible to use biostimulants in farmer's field. Easy mode of application can be introduced for its commercial usage. This review gathered the information on how the biostimulants contribute towards increase in the growth and yield of crop. Therefore, prospects of biostimulants in sustainable pulse production are highly promising as it offers eco-friendly solutions to increase crop yield, mitigate stress and significantly reduce chemical inputs.

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Authors' contributions

RB gathered the literature and structured the article's framework; APS formulated the study and authored the manuscript; MR evaluated and provided recommendations for enhancement; AS contributed to the study's design and revised the article; CB was involved in its design and coordination.

Compliance with ethical standards

Conflict of interest: Authors do not have any conflict of interest to declare.

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