

## Bio-stimulants and their role in fruits production

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### Abstract

The term bio-stimulants, refers to a group of compounds that act neither as fertilizers nor as pesticides, but have a positive impact on plant performance when applied in small quantities. Some major categories of plant bio-stimulants are humic and fulvic acids, protein hydrosylates and other N-containing compounds, seaweed extract, botanicals, chitosan and other biopolymers, inorganic compounds, beneficial fungi and beneficial bacteria. Major role of plant bio-stimulants is to increase soil fertility, nitrogen assimilation, antioxidant activity, plant nutrient uptake, abiotic stress tolerance, induction of plant defense enzymes, disease resistance and to improve the product quality and shelf life of fruits. Climate change affects horticulture production through the occurrence of late spring frosts. The bio-stimulants containing amino acids of animal origin (porcine blood) has shown to increase frost resistance, early flowering and yield in several strawberry cultivars. The application of bio-stimulants in fruit crop cultivation allows higher levels of sustainability by the reduction of fertilizers and environmental contamination and, at the same time, increase plant tolerance to biotic and abiotic stresses enhancing internal and external quality of fruits.

### Introduction

Agricultural intensification was driven by inputs derived from non-renewable energy sources. Although this approach greatly enhanced crop yields, these practices have also resulted in a major decline in ecological heritage as a result of deforestation, soil erosion, industrial pollution, declines in surface and groundwater quality and loss of biodiversity. These negative consequences of food production continue to proceed at an alarming rate and show no signs of reducing. In addition, it is widely acknowledged that an increase in

agricultural activities will further exacerbate the negative impacts of global climate change leading to greater uncertainty in food security. Current unsustainable farming practices therefore need to be reviewed since current models of agricultural intensification are neither socially or environmentally sustainable. The new challenge is to build systems of food production based on alternative intensification strategies which promote nutrient-use efficiency, reduce the need of disease and pest control, increase water-use efficiency and conservation. The term bio-stimulants, often used in the plural form, refers to a group of compounds that act neither as fertilizers nor as pesticides, but have a positive impact on plant performance when applied in small quantities. A plant bio-stimulant is any substance or microorganism applied to plants with the aim to enhance nutrition efficiency, abiotic stress tolerance and crop quality traits, regardless of its nutrients content.

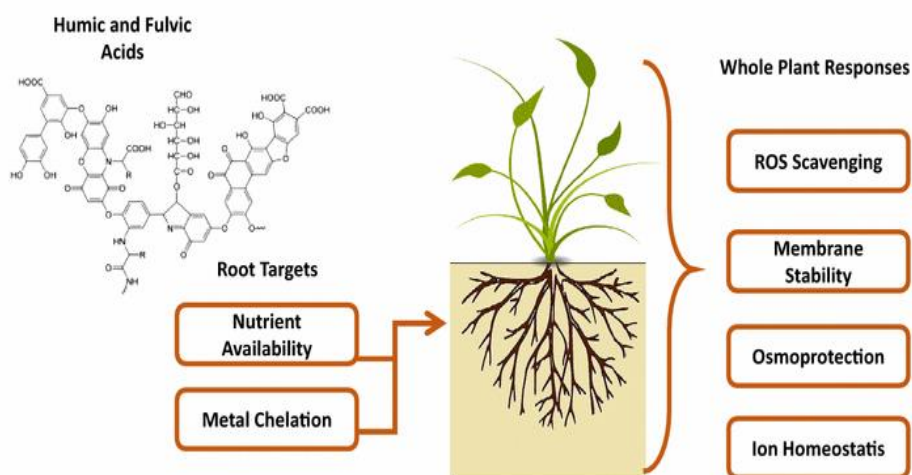
### **Major categories of bio-stimulants**

Major categories of plant bio-stimulants are humic and fulvic acids, protein hydrosylates and other N-containing compounds, seaweed extracts, botanicals, chitosan and other biopolymers, inorganic compounds, beneficial fungi and beneficial bacteria. Major role of plant bio-stimulants is to increase soil fertility, nitrogen assimilation, antioxidant activity, plant nutrient uptake, abiotic stress tolerance, induction of plant defense enzymes, disease resistance and to improve the product quality and shelf life of fruits.

### **Humic and fulvic acid:**

Humic substances (HS) are natural constituents of the soil organic matter, resulting from the decomposition of plant, animal and microbial residues, but also from the metabolic activity of soil microbes using these substrates. HS are collections of heterogeneous compounds, originally categorized according to their molecular weights and solubility into humins, humic acids and fulvic acids. These compounds also show complex dynamics of association and dissociation into supra-molecular colloids, and this is influenced by plant roots via the release of protons and exudates. Humic substances have been recognized for long as essential contributors to soil fertility, acting on physical, physico-chemical, chemical and biological properties of the soil. Most bio-stimulants 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<sup>+</sup>-ATPases, which convert the free energy released by ATP hydrolysis into a transmembrane electrochemical potential used for the import of nitrate and other nutrients (Jindo *et al.*, 2012). HS seem to enhance respiration and invertase activities providing C substrates.



**Fig. 1:** Key mechanisms targeted by humic and fulvic acid based bio-stimulants.

### Protein hydrolysates and other N-containing compounds:

Amino-acids and peptides mixtures are obtained by chemical and enzymatic protein hydrolysis from agro industrial by-products, from both plant sources (crop residues) and animal wastes (e.g., collagen, epithelial tissues). Other nitrogenous molecules include betaines, polyamines and 'non-protein amino acids', which are diversified in higher plants but poorly characterized with regard to their physiological and ecological roles (Halpern *et al.*, 2015). Glycine betaine is a special case of amino acid derivative with well-known anti-stress properties. These compounds have been shown to play multiple roles as bio-stimulants of plant growth. Direct effects on plants include modulation of N uptake and assimilation. Chelating effects are reported for some amino acids (like proline) which may protect plants against heavy metals but also contribute to micronutrients mobility and acquisition. Antioxidant activity is conferred by the scavenging of free radicals by some of the nitrogenous compounds, which contributes to the mitigation of environmental stress. Indirect effects on plant nutrition and growth are also important in the agricultural practice when protein hydrolysates are applied to plants and soils. Protein hydrolysates are known to increase microbial biomass and activity, soil respiration and, overall, soil fertility. Significant

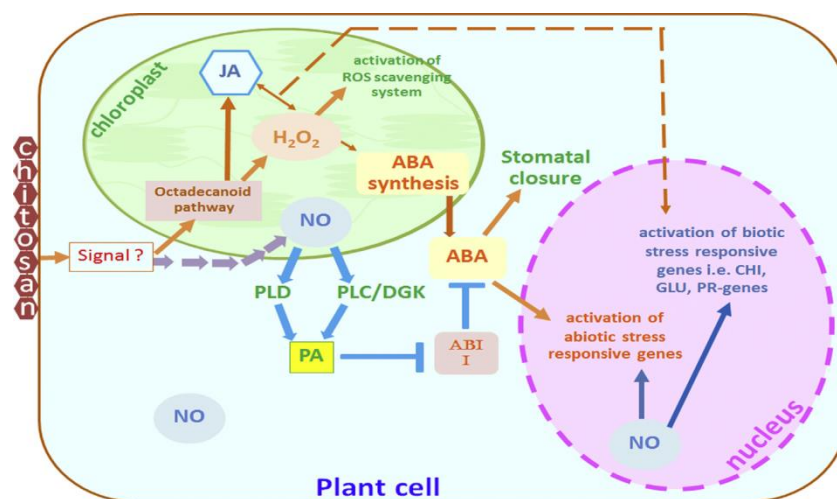
improvements in yield and quality traits have been reported in agricultural and horticultural crops.

#### **Seaweed extracts and botanicals:**

Fresh seaweeds can be used as source of organic matter and as fertilisers. The commercial use of seaweed extracts and of purified compounds, which include the polysaccharides laminarin, alginates and carrageenans and their breakdown products. Other constituents contributing to the plant growth promotion include micro and macronutrients, sterols, N containing compounds like betaines, and hormones. Carrageenans originated from red seaweeds. They can be applied on soils, in hydroponic solutions or as foliar treatments. In soils, their polysaccharides contribute to gel formation, water retention and soil aeration. In plants, nutritional effects via the provision of micro- and macronutrients indicate that they act as fertilisers, beside their other roles.

#### **Chitosan and other biopolymers**

Chitosan is a deacetylated form of the biopolymer chitin, produced naturally and industrially. The physiological effects of chitosan oligomers in plants are the results of the capacity of this polycationic compound to bind a wide range of cellular components, including DNA, plasma membrane and cell wall constituents, but also to bind specific receptors involved in defense gene activation, in a similar way as plant defense elicitors. Chitin and chitosan apparently use distinct receptors and signalling pathways. Horticulture applications of chitosan on plant helps in protection against fungal pathogens, but uses bear on tolerance to abiotic stress (drought, salinity, cold stress) and on quality traits related to primary and secondary metabolisms. Stomatal closure induced by chitosan via an ABA-dependent mechanism participates to the environmental stress protection.



**Fig. 2:** Model of Chitosan responses at the cellular level in plants

### Inorganic compounds

Chemical elements that promote plant growth and may be essential to particular taxa but are not required by all plants are called beneficial elements. The five main beneficial elements are Al, Co, Na, Se and Si, present in soils and in plants as different inorganic salts and as insoluble forms like amorphous silica ( $\text{SiO}_2 \cdot n\text{H}_2\text{O}$ ). These beneficial functions can be constitutive, like the strengthening of cell walls by silica deposits, or expressed in defined environmental conditions, like pathogen attack for selenium and osmotic stress for sodium which promote plant growth, the quality of plant products and tolerance to abiotic stress. This includes cell wall rigidification, osmoregulation, reduced transpiration by crystal deposits, thermal regulation via radiation reflection, enzyme activity by co-factors, plant nutrition via interactions with other elements during uptake and mobility, antioxidant protection, interactions with symbionts, pathogen and herbivore response, protection against heavy metals toxicity.

### Beneficial fungi:

Fungi interact with plant roots in different ways, from mutualistic symbioses to parasitism. Mycorrhizal fungi are a heterogeneous group of taxa which establish symbioses with over 90% of all plant species. Among the different forms of physical interactions and taxa involved, the Arbuscule-Forming Mycorrhiza (AMF) are a widespread type of endomycorrhiza associated with crop and horticultural plants, where fungal hyphae of Glomeromycota species penetrate root cortical cells and form branched structures called arbuscules. There is an increasing interest for the use of mycorrhiza to promote sustainable



agriculture, considering the widely accepted benefits of the symbioses to nutrition efficiency (for both macronutrients, especially P, and micronutrients), water balance, biotic and abiotic stress protection. The fungal endophytes, like *Trichoderma* and Sebaciniales, distinct from the mycorrhizal species, are able to live at least part of their life cycle away from the plant, to colonize roots and, as shown recently, to transfer nutrients to their hosts, using poorly understood mechanisms.

### **Beneficial bacteria**

Bacteria interact with plants in all possible ways (i) as for fungi there is a continuum between mutualism and parasitism; (ii) bacterial niches extend from the soil to the interior of cells, with intermediate locations called the rhizosphere and the rhizoplane; (iii) associations may be transient or permanent, some bacteria being even vertically transmitted via the seed; (iv) functions influencing plant life cover participation to the biogeochemical cycles, supply of nutrients, increase in nutrient use efficiency, induction of disease resistance, enhancement of abiotic stress tolerance, modulation of morphogenesis by plant growth regulators. With regard to the agricultural uses of bio-stimulants, two main types should be considered within this taxonomic, functional and ecological diversity: (i) mutualistic endosymbionts of the type *Rhizobium* and (ii) mutualistic, rhizospheric PGPRs ('plant growth-promoting rhizobacteria'). *Rhizobium* and related taxa are commercialised as biofertilizers, PGPRs are multifunctional and influence all aspects of plant life: nutrition and growth, morphogenesis and development, response to biotic and abiotic stress, interactions with other organisms in the agroecosystem.

### **Conclusion:**

Bio-stimulants, is a group of compounds that act neither as fertilizers nor as pesticides, but have a positive impact on plant performance when applied in small quantities. The application of bio-stimulants in fruit crops cultivation helps in many aspects. It increases the levels of sustainability by reduction in use of fertilizers and environmental contamination and, at the same time, increases plant tolerance to abiotic and biotic stresses which leads to enhancement in internal and external quality. The characterization of a bio-stimulant should be performed on the basis of the plant responses, indicating the physiological targets and metabolic network involved.

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