Review Article

Use of biostimulants in inducing resistance for phytosanitary management: A review

ABSTRACT

Biostimulants perform functions that improve plant metabolism and can positively interfere with processes such as respiration, photosynthesis, synthesis of nucleic acids and absorption of ions. Furthermore, they improve the roots' ability to absorb nutrients. Thus, bistimulants can help to induce plant resistance by improving the nutritional capacity of plants, making them more tolerant to pathogen attack. Therefore, the use of biostimulants can have beneficial effects, providing an increase in plant productivity, whether through stimulating their physiology or controlling diseases.

Keywords: Physiological processes, increased productivity, plant development.

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1. INTRODUCTION

Biostimulants are any substance or microorganism applied to plants, including commercial products, that stimulate natural processes and improve nutrient absorption and efficient use, providing tolerance to biotic and abiotic stresses and productive quality [1]. The use of biostimulants can increase the growth and development of plants through stimulation of cell division and elongation, in addition to increasing the absorption and use of water and nutrients by plants [2].

Biostimulants have shown a positive effect on several plant metabolisms such as respiration, photosynthesis, nucleic acid synthesis and ion absorption, improving the ability to absorb nutrients by the roots [3]. The components present in biostimulants can change the hormonal status of plants, which are directly responsible for growth and health. These substances generate an increase in the activities of antioxidases in plants, especially when they are under conditions of water stress, severe temperatures, the action of herbicides or pathogen attack [4].

Among plant biostimulant products, the main ones are amino acids and protein hydrolysates, humic substances, microorganisms and algae inoculums and extracts [5]. It is known that the isolated application of these products can result in improvements in crop yield, however, more studies are needed to prove the effectiveness of the combined application of biostimulants, as there may be an antagonistic relationship between these substances [6].

Regarding pathogen attacks, it is known that plants do not have an immune system, however, they have a high degree of resistance to diseases caused by fungi, bacteria, viruses and nematodes. In turn, pathogens can develop different mechanisms to attack host plants and cause diseases [7]. In this sense, the use of bistimulants can help in inducing plant resistance by improving the nutritional capacity of plants, which in many cases makes them more tolerant to pathogen attack [8] and acting on the activation of resistance genes.

responsible for the synthesis of pathogenesis-related proteins (RP), responsible for defenses against pathogen infection [7].

2. USE OF BIOSTIMULANTS IN INDUCING RESISTANCE FOR PHYTOSANITARY MANAGEMENT

Biostimulants are natural substances or microorganisms whose function is to promote improved nutritional efficiency for plants, a positive increase in the capacity for tolerance to abiotic stresses, and causes improvements in the productivity and quality of crops [9]. The use of biostimulants in agriculture allows for greater crop productivity to be achieved, especially in conditions where soil and climate factors are limited [10]. Its use is an interesting alternative to the lack of fertilizers in stimulating root production, especially in soils where there is no water availability and low fertility [11].

Under stress conditions, plants produce high amounts of free radicals or reactive oxygen species, which cause damage to plant cells. Antioxidants produced by plants are substances that minimize the effects of free radicals on plant cells, improving root and aerial part growth, maintaining the ideal water content in the leaves and reducing the attack of diseases, the plants being in ideal conditions of cultivation or under environmental stress. Therefore, the use of biostimulants increases the levels of antioxidants, which will result in an increase in the plants' defense system [3].

It is known that plants do not have an immune system comparable to that of animals, however, they have a high degree of resistance to diseases caused by fungi, bacteria, viruses and nematodes that are present in the environment. Pathogens that are successful in the infection process have developed several mechanisms to attack their host plant and cause disease. Some pathogens can penetrate the plant directly through the cuticle and cell wall, others through the stomatal openings and can also invade the host through injured sites [7].

Basically, there are two ways of inducing resistance. Acquired resistance and induced resistance. Although they are similar, this phenomenon can be differentiated by the hormonal signaling pathways involved. Acquired resistance is mediated by salicylic acid and is generally related to the interaction of plants with virulent, avirulent pathogens or non-pathogenic microorganisms. In this type of resistance, there is an accumulation of proteins related to pathogenesis. Induced resistance is mediated by the hormones jasmonic acid and ethylene, being associated with rhizobacteria or rhizospheric microorganisms [12].

Plant resistance to diseases is mainly related to genetics, however, plants that suffer some type of stress are more susceptible. According to Karnok[13], when plants are in favorable conditions, they develop well, with the effect of biostimulants being difficult to identify, however, when under stress conditions the use of biostimulants results in better development, since their defense becomes more effective. Much of this resistance is associated withimprovements in the nutritional capacity of plants. In general, plants that have unsatisfactory nutritional content are more susceptible to less specific pathogens [8].

Another mechanism for inducing resistance against pathogens associated with biostimulants is the activation of resistance genes responsible for the synthesis of pathogenesis-related proteins (RP). The induction agent (biostimulants) activates resistance genes in plants that, after the transcription process, will synthesize proteins related to pathogenesis (RP), these proteins will accumulate in the intercellular spaces and/or in the vacuole of the cells, when

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under attack of pathogens, RP proteins will act directly on them, preventing the progression of the disease and providing greater resistance to plants [7].

Most of the positive effects associated with biostimulants are their influence on the hormonal activity of plants, which are responsible for interfering with the development of plants in relation to the environment in which they are located. The four main groups of biostimulant substances available are: amino acids and protein hydrolysates, humic substances, microorganisms and inoculums and algae extracts, all of which are presented on the Brazilian market in the form of commercial products [5].

Humic substances are constituents of organic matter in soils resulting from the decomposition process of plant and animal residues that are used as alternative inputs for different cultivars. Its use can generate beneficial effects on soil properties, in addition to improvements in plant metabolism. These substances cause an increase in the movement and absorption of ions, improve respiration and Krebs cycle reactions, increase ATP synthesis in root cells, increase chlorophyll levels and nucleic acid synthesis and decrease nitrogen losses. by volatilization and leaching [9].

Humic substances are made up of humic acids, fulvic acids and humins, these compounds have the function of stimulating the synthesis of natural plant hormones such as auxin, cytokinins and gibberellins that can positively influence physiological mechanisms of plant growth and development [14]. In turn, humic acids act as promoters of increased synthesis of membrane H+ATPase, favoring the activation of H+ pumps, this mechanism being responsible for the rhizogenesis of lateral roots [15]. Thus, humic substances promote the optimization of the absorption of water and nutrients present in the soil [16].

Humic substances stimulate the modification of root architecture, mainly in the emergence of lateral roots. Nitric oxide produced after the application of humic acids stimulates H+ATPase activity and auxin synthesis. Acidification of the apoplast causes the cell wall to loosen, in addition to favoring cell division and expansion through the rearrangement of the cell wall, allowing the growth of lateral roots. These roots increase root volume and contact surface. With the lateral roots being thinner, the plants' absorption capacity is improved, which is an important factor for productive gains [9].

Amino acids have been used in agriculture for many decades in the most varied cultivars and their use is linked to different effects such as greater nitrogen assimilation, increased hormonal activity, chelating and antioxidant effects, as well as indirect effects such as nutrition, growth and development. of plants, through increasing biomass, activity of microorganisms, fertility and soil respiration [1]. These substances are composed of a central carbon, almost always asymmetric, linked to a carboxyl group (COOH), an amino group (NH_2) and a hydrogen atom. Furthermore, they have a radical generically called “R”, which differentiates each of the amino acids. In plants, there are several hypotheses attributed to the functions of amino acids, being that they act in protein synthesis, are precursor compounds of endogenous plant hormones, promote greater resistance to water stress and high temperatures and greater resistance to attack by pests and diseases [9].

In relation to substances containing microorganisms and inocula, they comprise an important category of biostimulants composed of a fundamental group of soil biota that positively affects agricultural production and increases the sustainability of the agroecosystem [17]. The advantages that plants acquire from the symbiotic relationship with mycorrhizae, for example, have beneficial effects on nutrient absorption, as well as tolerance and resistance to biotic environmental stresses such as pests and pathogens and abiotic ones such as drought and salinity [18]. The symbiotic relationship between plants and microorganisms

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allows hyphae to grow in the roots, in this way, the photoassimilates produced by plants are used by fungi, which in turn, supply the plants' root system with soil nutrients, such as phosphorus, nitrogen, copper and zinc [19]. Therefore, the use of microorganisms as plant biostimulants in agricultural practices becomes an ecologically correct alternative to conventional crop management strategies, which are generally dependent on chemical and agrochemical fertilizers [6].

The most studied and used microorganisms in Brazil and Latin America is the genus *Trichoderma*, which is composed of saprophic and mycoparasitic fungi found mainly in soil and decomposing plant remains. The potential of this genus as a plant disease control agent lies in its ability to compete for energy sources, produce antibiotics and other metabolites that inhibit pathogen activity, predatory effects or mycoparasitism. Furthermore, the genus *Trichoderma* has a direct effect on plants, inducing resistance through the production of jasmonic acid, salicylic acid and ethylene [20].

Algae extracts are anti-stress agents, affecting the oxidative system of plants, thus increasing tolerance to unfavorable environmental conditions and improving their recovery capacity after experiencing a stress condition, which enhances gains or, at least, maintains productivity, even in non-optimal conditions [9]. These substances are sustainable alternatives to improve the productive yields of cultivars without adverse impacts on the environment. Furthermore, algae extracts are cheap and easy to prepare for use and are efficient in small doses, and in their composition these substances contain macronutrients, micronutrients, amino acids, vitamins, carbohydrates and plant growth hormones, all of which substances affect cellular metabolism, increasing plant growth and yield [21]. Algae extracts are believed to act on plant performance by regulating root biomass, which can increase nutrient absorption and translocation, resulting in increased carbohydrates, proteins, phenolic compounds, stress tolerance and disease resistance [6].

For the assay of defense responses in tobacco leaves (*Nicotiana tabacum*) to tobacco mosaic virus (TMV) Klarzynski et al. [22] used sulfated fucan oligosaccharides, which are structural compounds extracted from seaweed applied to tobacco leaves at concentrations of 0 to 200µg, compared to the control solution containing only water. The authors found that there was a releaseherethe significant with explosionsThethat of espItisactive oxygen ciesItisninium (H_2O_2) in the presencewto 200µg mL^{-1} (black square) of oligofucans,eatswl walk approximately 3 min after addition of oligosaccharides(Figure 1). These espItisactive oxygen ciesItisThey cannot act directly by destroying pathogen cells and preventing their proliferation in the plant. Furthermore, according to the authors, tobacco leaves treated with oligofucans. This way, they showed fewer symptoms of the disease on their leaves (Figure 2).

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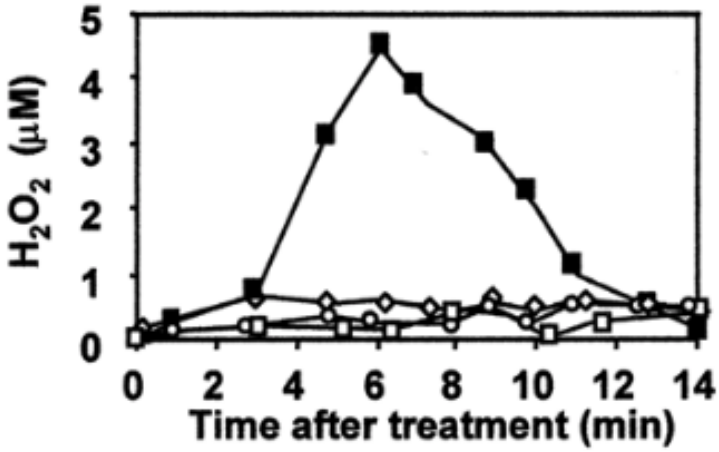

Figure 1.Oxidative explosion induced by oligofucans in cells tobacco leaves. The cells tobaccowere treated with 200µg of oligofucans ml^{-1} (black square) or with the water (open square).

Figure 2.Representation of tobacco leaves (*Nicotiana tabacum*) inoculated with tobacco mosaic virus (TMV). A) Leaves treated with control solution containing only water. B) Leaves treated with seaweed extract.

In studies carried out by Gonzalez et al. [6], biostimulant compounds based on growth-promoting mycorrhizal fungi (AMF) and seaweed extract (SE) were used, which regulate or improve the physiological processes of plants in physiological characteristics related to growth and biochemical effect on protein contents, lipids, carbohydrates, nitrogen and phosphorus from tomato plants (*Solanum lycopersicum* L. cv"Rio Fuego). Thus, the treatments were tested: only the nutrient solution (NS); AMF; AMF+NS; SE; SE+NS; AMF+SE+NS.

In general, for all characteristics evaluated, whether growth (Table 1) (Figure 3) or biochemical content (Table 2), biostimulants based on mycorrhizal fungi and seaweed extract presented higher averages when compared to treatments in which used only nutrient solution. When used individually, mycorrhizal fungi and seaweed extract stimulated the growth of tomato plants in different ways. While mycorrhizal fungi promoted greater leaf growth, seaweed extract was more favorable to root growth. However, the association of biostimulates with each other and with the nutrient solution must be emphasized, where there was an additive effect from the combination of these substances, positively affecting the quality and performance of the plants (Figure 4).

Table 1.Growth characteristics of tomato treated with plant biostimulant

Treatment	Length (cm)			Area (cm^2)		Mass (g)	
	aerial	Source	Total	sheet	Source	Fresh	Dry
NS	16.9 b	24.4b	39.1 a	11.3 a	43.4b	1.65 a	0.71 b
AMF	21.1c	18.0 a	39.1 a	23.8b	11.5 a	1.67 a	0.18 a
AMF+NS	21.9c	31.5c	53.6d	104.3 d	143.1d	12.65c	1.30 cd
SE	12.5 a	30.8c	45.4c	31.8c	50.9b	4.13 ab	1.23c
SE+NS	12.4 ab	33.8c	44.7 bc	34.8c	58.6b	4.32b	1.31 cd
AMF+SE+NS	24.8d	32.5c	58.4 e	120.5 e	117.8c	14.46d	1.56 d

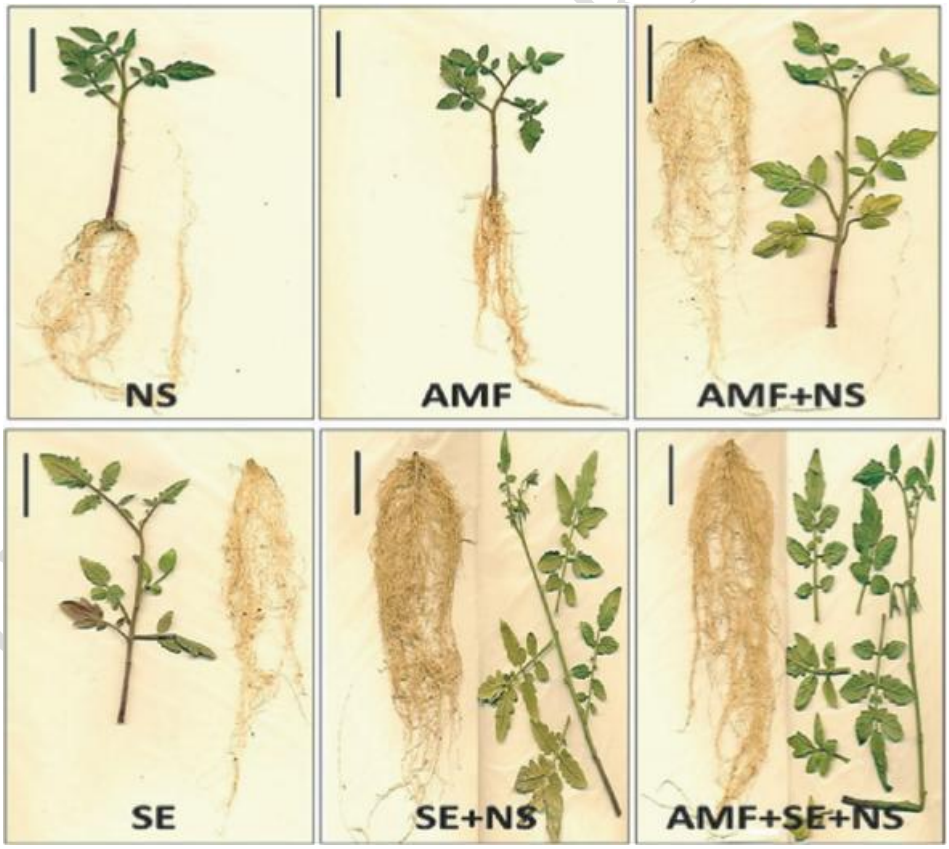

Figure 3. Tomato plant growth after the 96-day experiment.

Table 2. Biochemical content of tomato plants treated with plant biostimulant

Treatment	Protein (%)	Lipids (%)	Carbohydrates ($mg \cdot g^{-1}$)	Polyphenols ($mg\ g^{-1}$)	Nitrogen (%)	Fósforo ($mg \cdot Kg^{-1}$ P)
NS	10.81b	4.51c	4.672e	0.050a	1.2a	0.119b
AMF	10.58b	2.44b	2.134b	0.687d	2.3b	0.014a
AMF+NS	9.29a	2.73b	3.750d	0.352c	2.1b	0.223c
SE	12.20c	2.46b	0.952a	0.108b	1.9a	0.080b
SE+NS	10.44b	5.49d	2.944c	0.057a	2.1b	0.100b
AMF+SE+NS	13.22d	1.42a	5.164f	0.114b	1.8a	0.201c

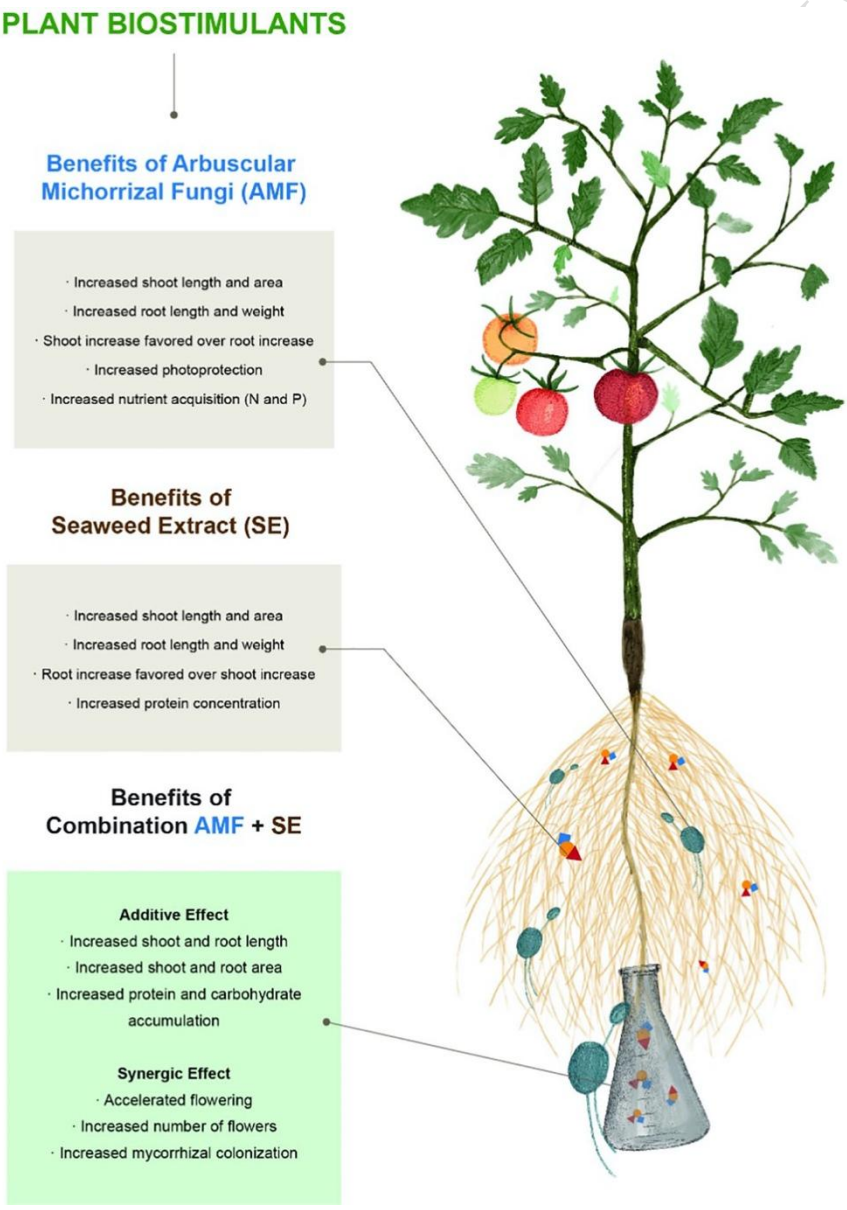

Figure 4.Influence of the individual application of mycorrhizal fungi and seaweed extract and their combination in stimulating the growth of tomato plants

3. CONCLUSION

Given the current market demand for food produced in a more sustainable way and free from products harmful to human health and harmful to environmental quality, there is a need to search for natural products, which provide quality vegetable cultivation and are free from agrochemicals. In this sense, biostimulants can provide increases in plant productivity, whether by stimulating their physiology or controlling diseases. Furthermore, natural products may present greater cost/benefit, as they do not require the synthesis of new molecules, facilitating the availability of these products on the market and favoring their acquisition and use by farmers.

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