



Bioformulation: A New Frontier in Horticulture for Eco-Friendly Crop Management

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Abstract

Bioformulation represents a groundbreaking approach in horticulture, heralding a new era of eco-friendly crop management. By harnessing the power of naturally occurring organisms and compounds, bioformulation offers sustainable alternatives to conventional agricultural practices. This innovative technique involves the formulation of beneficial microbes, biocontrol agents, and organic substances into products that enhance plant growth, protect against diseases, and improve soil health. Through meticulous research and development, bioformulation has emerged as a potent tool for reducing the reliance on synthetic chemicals and minimizing environmental impact. This abstract delves into the transformative potential of bioformulation in revolutionizing horticulture, emphasizing its role in promoting healthier plants, enhancing agricultural sustainability, and contributing to a greener future for generations to come.

Keywords: bioformulation, diseases, environmental, Conventional

Introduction

There is a growing need for environmentally friendly technical instruments in agricultural production, and global food security is being threatened by climate change. Biostimulants can be used to enhance the effects of chemical inputs. such as beneficial rhizosphere microbiomes including plant growth-promoting rhizobacteria and favorable fungi [1]. Microbial biostimulants can enhance physiological and biochemical processes that improve the absorption of nutrients, increase nutrient utilization, enhance the quality of crops, and boost plant output. When administered to plants by seed, foliar, or rhizosphere treatment, they might be categorized as formulations of microorganisms or microbial consortia [2]. Plant growth-promoting rhizobacteria (PGPR) are a diverse collection of bacteria that live inside plants and can enhance plant growth and yield by producing phytohormones, antioxidants, osmolytes, volatile compounds, exopolysaccharides, and 1-aminocyclopropane-1-carboxylate deaminase. Arbuscular mycorrhizal fungi (AMF) are bio-factors that enhance plant development, enrich nutrients, and aid in phytoremediation [3]. They also protect plants from diseases and increase their resilience to salt, drought, and heavy metal toxicity. The profitability of AMF treatment has been demonstrated in numerous horticultural species, including apple, pepper, citrus, peach, lettuce, strawberry, onions, pineapple, and melon.

The utilization of a combination of PGPRs (Plant Development-Promoting Rhizobacteria) and AMFs (Arbuscular Mycorrhizal Fungi) is a very promising technique for enhancing plant development. This approach capitalizes on the advantages offered by both types of microorganisms and harnesses their combined beneficial effects through synergy [4]. The combined application of plant growth-promoting bacteria (PGPB) and arbuscular mycorrhizal fungus (AMF) was found to have a greater positive impact on both the production and quality of horticultural crops compared to using either PGPB or AMF alone [5].

Nevertheless, the majority of farmers have yet to investigate the potential of microbial biostimulants. Greater endeavor is required to propose and implement them as an ecologically viable method to enhance crop yield and well-being, making a significant contribution to establishing the 21st century as the era of biotechnology. Microbial biostimulants can also enhance the sustainability of medicinal and aromatic plant culture, namely in basil production, especially in situations where growth is limited [6].



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Mechanisms involved in formulations.

Microbial plant growth promoters exert their effects through both direct and indirect routes. Direct mechanisms involve microbes producing substances that enhance the absorption of nutrients, while indirect mechanisms include the solubilization of zinc, the production of siderophores, the biosynthesis of indole acetic acid, the solubilization of phosphorus, the production of ammonia and hydrogen cyanide, the production of antioxidant enzymes, the production of phytohormones, and the biological fixation of nitrogen. Microbial biostimulants, such as fungi and bacteria, can alleviate the adverse effects of environmental pressures by generating hormone-like stimulants that have beneficial effects on plant development [7]. Microbial biostimulants can protect plants by controlling the molecular processes that occur when plants interact with microbes. Additionally, they enhance the production of secondary metabolites in plants. The creation of these protective chemicals occurs via the shikimate pathway, which utilizes the enzyme Phenylalanine Ammonia Lyase (PAL) to create phenylpropanoids in response to microbial elicitation. Plants employ induced systemic resistance (ISR) as a mechanism to deal with external stressors [8]. Plant growth-promoting rhizobacteria (PGPR) stimulates the production of biosurfactants, chelating factors, avermectins, secondary metabolites, fluorescent insecticidal toxins, betaglucanases, and chitinases to enhance disease resistance in plants. In addition, they can enhance antioxidant activity and stimulate the production of phytochemicals, regulate metabolism, and enhance the quality of crops [9]. Additional mechanisms of action encompass the production of cytokinins, ABA, ethylene, auxins, gibberlins, exopolysaccharides, organic acids, siderophores, overexpression of stress-responsive genes, expression of antioxidant enzyme activity, and activation of genes that promote growth.

Applying PGPR bacteria can enhance the soil with bacterial

inoculums that enhance nutrient availability, boost resistance against non-living stressors, and accumulate antioxidant chemicals to alleviate stress by neutralizing oxidative radicals [10].

PGPR biostimulants are essential in regulating phytohormone signaling, antioxidant defense mechanisms, and photosynthetic processes in abiotic stress conditions such as drought, salt, heavy metals, heat, and cold stress. Research has demonstrated that these biostimulants effectively improve the growth, productivity, and nutrient absorption of plants [11]. Examples of bacteria such as Azospirillum brasilense, Gluconacetobacter diazotrophicus, Burkholderia ambifaria, and Herbaspirillum seropedicae stimulate the synthesis of plant hormones that play a beneficial function in the process of making nutrients soluble and facilitating their absorption in onion plants [12]. Utilizing natural microbial biostimulants derived from soil micro-organisms is a suitable method for mitigating the impact of biotic stresses on plants. Bacillus cereus, Serratia sp. XY21, and Bacillus subtilis SM21 have been discovered to enhance plant resistance against root-knot nematodes in tomato plants. Similarly, Pseudomonas aeruginosa LV has been found to enhance resistance to bacterial stem rot in tomato plants by accumulating extracellular bioactive compounds [13]. Arbuscular mycorrhizal fungi (AMF) have been discovered to enhance crop biomass following their application, perhaps by influencing the complex interaction network of phytohormones and potentially improving nitrogen utilization efficiency through the Glutamine Oxoglutarate Aminotransferase (GS-GOGAT) pathway. AMF inoculation has demonstrated the ability to safeguard Ocimum basilicum plants against the negative effects of salt stress [14]. This is achieved by enhancing the plant's water usage efficiency, promoting chlorophyll synthesis and mineral absorption, and boosting photosynthetic metrics such as net photosynthesis and stomatal conductance [15].



Fig 2 Adopted from [50] and copyright permission from Elsevier.

The mechanisms by which AMFs exert their effects include enhanced antioxidant activity, buildup of osmolytes, upregulation of proline biosynthesis, and higher levels of Mg, Ca, and K. These processes contribute to the promotion of chlorophyll production and enzyme activity. In addition, AMF inoculation has been discovered to limit the accumulation and absorption of sodium (Na) via regulating the expression of AKT2, SOS1, and SKOR genes in the roots. This adjustment enables the roots to maintain a balance of potassium (K+) and sodium (Na+), thereby preserving homeostasis [16]. Recent developments in omics science have uncovered that the use of microbial biostimulants leads to substantial modifications in primary and secondary metabolites, including amino acids, lipids, phenolic acids, and intermediates of the tricarboxylic acid (TCA) cycle. Additionally, it induces changes in protective mechanisms against stress. These functions encompass maintaining a balance in redox levels, regulating osmotic pressure, stabilizing cell membranes, generating energy by breaking down amino acids, and upregulating stressrelated genes [17].

Microbial biostimulants exert indirect effects on plants, such as enhancing water and nutrient absorption, altering root shape, and promoting the growth of new roots. Hormonal activities, such as indole-3-acetic acid, govern several changes in the plant. These changes include cell elongation and division, the growth of new roots, and the creation of hairy roots. Plant growthpromoting bacteria engage with plants through many mechanisms, including Rhizospheric, Endophytic, Symbiotic, and Phyllospheric interactions [18].

Certain microbial biostimulants, such as Paraburkholderia phytofirmans for grapevine and Pseudomonas fluorescens A506 and Pseudomonas chlororaphis for pear and apple trees, can safeguard plants from freezing and cold stress. In addition, they provide protection for crops against heat stress caused by certain bacteria such as Pseudomonas sp. AKMP6 and Pseudomonas putida AKMP7, as well as from Glomus sp. in the case of tomato plants. Pseudomonas fluorescence A506 competes with ice nucleating activity in apples and pears to protect against cold and frost for the crops [19]. Regarding water stress, the introduction of Bacillus licheniformis strain K11 to pepper plants resulted in a higher tolerance to water shortage stress compared to plants that were not introduced to the strain. In greenhouse conditions, when lettuce is subjected to water stress, several strains of arbuscular mycorrhizal fungi (AMF) and Trichoderma koningii have been found to enhance the levels of mineral components and phenolic acids [20]. However, the effects of biostimulants are mostly focused on regulating the production of secondary chemicals rather than enhancing nutrient absorption. Introducing microorganisms into the soil can enhance the ability of roots or fungal hyphae to explore the soil, resulting in a substantial improvement in root conductivity. In addition, the introduction of microorganisms can result in elevated hormone synthesis, namely the formation of Indole-3-acetic acid, in tomato, cucumber, orange, and soybean plants [21].

Bioformulation in horticultural crops

Microbial biostimulants are essential in contemporary agriculture, especially given the challenges posed by climate change and the growing world population. These cutting-edge instruments enhance plant nutrition by absorbing mineral elements beyond the areas where the plant roots are actively depleting them. This leads to alterations in secondary metabolism and a rise in the amount of beneficial chemicals in the plants. An example of a biostimulant, which is based on microorganisms, consists of two strains of arbuscular mycorrhizal fungi (AMFs) and Trichoderma koningii. This biostimulant enhanced the quality of plants, independent of the amount of water available [22].

Utilizing AMFs and PGPRs can enhance the absorption of nutrients from the soil, hence enhancing plant growth, improving fruit quality, and increasing overall output. They can also be utilized in circumstances of abiotic stress, where crop growth is hindered or meets substantial constraints. As an illustration, the fungus Aspergillus flavipes can synthesize indole-3-acetic acid (IAA) by utilizing soybean bran as a growth substrate [23].

The concurrent use of biostimulants, such as plant growthpromoting bacteria and freshwater algae, had a substantial impact on the plant weight of romaine and leaf lettuce over the summer and spring seasons. The greatest enhancement in the weight of romaine lettuce (18.9%) was attained during the spring harvest, whereas the use of a biostimulant therapy resulted in a 22.7% increase in weight for the leaf lettuce during the summer harvest [24].

Microbial biostimulants can provide defense against salt stress, drought stress, and other environmental adversities. Some commercially available plant biostimulants that are based on PGPR and beneficial fungi are FZB24®fl, Rhizovital 42®, Inomix® Biostimulant, Inomix® phosphore, and Inomix® Biofertilisant[25].

Plant Growth-Promoting Rhizobacteria (PGPRs) are essential for sustainable horticultural crop production. They enhance germination, stimulate growth, and improve the look, nutritional content, and texture of vegetables, even in challenging situations. Rhizobia, the predominant genus in this classification, consists of 13 species that form a symbiotic relationship with legumes and induce the formation of nodules, as well as five species that cause tumor formation. Rhizobia's ability to tolerate soil moisture shortage can have many advantages for agronomic production, particularly when applied to seeds in dry soil conditions [26].

Azospirillum is a well-studied kind of bacteria that promotes the development of plants in the roots. It primarily functions by fixing nitrogen and producing phytohormones. Multiple reports have emphasized the beneficial impacts of applying Azospirillum bacteria. These include mitigating the harmful effects of salt stress on chickpea growth and performance, enhancing product quality, improving chlorophyll content, prolonging storage life, and promoting higher germination and vegetative growth compared to control treatments [27]. Azotobacter promotes plant development through many actions, including the generation of growth hormones, siderophores, nitrogen fixation, and the ability to remove oil contamination, tolerate heavy metals, and degrade pesticides. Azotobacter salinestris exhibited a high tolerance towards metal-oxide nanoparticles (NPs). Furthermore, the introduction of these bacteria into tomato plants resulted in enhanced photosynthesis, greater flower development, higher fruit yield, and elevated lycopene levels [28].

Application of Azotobacter chroococcum and AMF species greatly improved the survival rate of saplings exposed to salt stress. It also increased all growth parameters and microbial count in the rhizosphere of mulberry plants. Furthermore, it had a positive impact on the desirable growth parameters of saplings, which is beneficial for the early grafting of apple trees cultivated under solarized black plastic mulching [29]. Two rhizobacteria with plant growth-promoting properties were isolated from the rhizosphere of Prunus domestica. These bacteria were identified as Pseudomonas stutzeri and Bacillus toyonensis. They were found to enhance the growth of tomato plants under salt-stress conditions. Additionally, they improved the establishment of Vitis vinifera and peach rootstock GF305 when these plants were moved from in vitro conditions to the greenhouse [30]. The introduction of Cd- and Pb-resistant PGPR (Plant Growth Promoting Rhizobacteria) strains Bacillus sp. QX8 and QX13, isolated from soil polluted with heavy metals, resulted in enhanced growth of Solanum nigrum and increased extraction of Pb and Cd from the soil through plants [31].

Seed inoculation with Bacillus species showed a favorable correlation with the growth characteristics and nutritional status of cucumber plants cultivated in conditions of elevated salinity. Utilizing rhizobacteria during periods of water restriction also enhanced the levels of antioxidants and photosynthetic pigments in basil plants [32].

The PGPR consortium exhibited a higher accumulation of AsIII in leaves, while also triggering plant defense systems that mitigated the harmful effects of AsIII in grapevine plants. Inoculating grapevines with PGPR (Bacillus licheniformis, Micrococcus luteus, and Pseudomonas fluorescens) under As(III) stress conditions enhanced antioxidant activity and effectively mitigated the toxic effects of NaAsO2 in vitro grapevine plants. Specifically, the inoculation with M. luteus demonstrated promising potential for bioremediation of As(III) contamination [33].

Co-inoculation with various bacterial strains has been observed to provide positive impacts on the growth, yield, and quality characteristics of crops. For example, the introduction of Bacillus amyloliquefaciens during seed germination led to the greatest improvement in seed germination (84.75%) and seedling vigor (1423.8), as well as an increase in the vegetative development parameters of chili (Capsicum annum L.). Furthermore, the synergistic interaction between Pseudomonas BA-8 and Bacillus OSU-142 significantly influenced the productivity, development, and nutrient levels of sweet cherry plants (Prunus avium L.) [34].

Mycorrhizas are a mutually beneficial relationship between fungus and plant roots, which can take many shapes depending on the classification of the fungi and the dispersion of the host plants. They can greatly enhance the efficiency of mineral absorption and may be classified into two main categories: endotrophic and ectotrophic. Arbuscular mycorrhizal fungi (AMFs) have been extensively researched in vegetable production as part of sustainable agriculture. They have been found to enhance plant nutrient absorption, promote plant growth and yield, and improve the quality of the final product. Additionally, AMFs have shown significant potential in suppressing phytopathogens [35].

The primary categories of arbuscular mycorrhizal fungi (AMF) associated with the sub-phylum Glomeromycotina of the phylum Mucoromycota have been identified within this sub-phylum. Recent research studies have examined the impact of arbuscular mycorrhizal fungi (AMF) on enhancing the development of horticultural plants, including fruit trees, vegetables, flower crops, and ornamental plants. These studies have investigated the effects of AMF on stimulating vegetative and reproductive growth, improving yield quality, enhancing stress physiology, and increasing disease resistance. AMF raised the nutrition and water provision for horticulture plants,

resulting in greater output and improved quality. Additionally, AMF improved the plants' ability to withstand environmental stress and resist infections [36].

There have been several reports documenting the beneficial impact of applying arbuscular mycorrhizal fungi (AMF) on horticultural crops. An instance of this is mycorrhiza Y-037, which exhibits a strong level of infection and significantly enhances the development of Guizhou blueberry plants. The introduction of fungus by inoculations somewhat enhanced the quality of the fruit and the composition of mineral elements, with the extent of improvement varying depending on the specific species of fungi. Piriformospora indica, a fungus with characteristics similar to mycorrhiza, has been found to be a more effective alternative to AMF in its use on citrus trees [37]. The use of AMF (Arbuscular Mycorrhizal Fungi) and controlled fertilization in a soil with low phosphorus content and moderate mycorrhizal potential can enhance the growth and productivity of tomato plants by optimizing biomass yield and output. AMF can enhance the availability of phosphorus in the rhizosphere and greatly improve nitrogen consumption in onion plants that

have been infected [38]. The symbiotic association between Mycorrhiza (Glomus mossea) and growth-promoting bacteria (Azospirillum) has been observed to enhance the productivity of fennel plants by increasing their yields, total carotenoids, and chlorophyll content, particularly when the plants are subjected to water deficiency stress. AMF (Arbuscular Mycorrhizal Fungi) and vermicompost have the ability to enhance the absorption of water in cactus plants and reduce the negative effects of drought, while also reducing the presence of oxidative stress indicators. When tomato plants are subjected to restricted watering, certain strains of arbuscular mycorrhizal fungi (AMF) have the ability to enhance plant development and recover the dry weight of both the shoots and roots [39].

AMF colonization can enhance drought resistance in citrus leaves by enhancing non-structural carbohydrates, calcium, potassium, and magnesium. Additionally, it can mitigate the adverse impacts of water deficiency stress by increasing the activity of primary and secondary metabolic processes and maintaining a high level of water potential in the stems of olive plants. AMF can mitigate the negative effects of salinity on Ligustrum vicaryi plants by increasing the levels of nitrogen, calcium, zinc, magnesium, and soluble proteins [40]. Vitis vinifera L. plants treated with mycorrhizal fungi exhibit improved physiological and nutritional conditions, as well as greater relative water content (RWC) and photosynthetic rate throughout the hardening process. Introduction of F. mosseae and R. intraradices enhances the synthesis of essential oils in Thymus daenensis and T. vulgaris L., particularly when subjected to water scarcity [41].

The symbiotic relationship with AMF does not affect the growth of corms, but it enhances the creation of new corms in saffron plants and reduces the occurrence of fungal infections. Inoculating Ocimum tenuiflorum with Rhizophagus intraradices enhances production and improves the quality of the final products [42].

The establishment of arbuscular mycorrhizal fungi (AMF) greatly enhances the ability of lettuce to withstand high temperatures. It also decreases the levels of sodium (Na+) and chloride (Cl-) ions in the plant, while increasing the relative water content, total fresh and dry weight, and photosynthetic activity of olive trees. The AMF treatment reduces the uptake of Cd by plants, but the addition of biochar hinders the

accumulation of Cd in plant roots [43].

The mycorrhizal consortium has the ability to suppress Fusarium wilt in cucumber and demonstrates potential as a biological control agent in greenhouse agro ecosystems. The application of AMF has a substantial impact on the polyphenolic compounds and antibacterial activity of Tamarix gallica. Additionally, the presence of Rhizophagus intraradices and Funneliformis mosseae greatly enhances the levels of root proline, total soluble sugars, and total phenolics in both the shoots and roots of valerian plants, as compared to valerian plants that were not treated with mycorrhizal fungi [44].

Factors affecting bioformulation.

The effectiveness of microbial formulation is affected by a range of biotic and abiotic parameters, including as strain choice, carrier materials, storage conditions, shelf life, competition in the environment, application technique, ambient conditions, quality control, and genetic stability [45].

The selection of strains is critical since various strains possess differing capacities to flourish in diverse environmental situations. The selection of carrier materials or additives in the formulation has a direct impact on the safeguarding, transportation, and discharge of the microorganisms. Optimal storage conditions, encompassing factors such as temperature, humidity, and packaging, are crucial for preserving the viability of the bacteria in the formulation [46].

The efficiency of the formulation can be considerably affected by its shelf life. Formulations with shorter shelf lives may need to be applied more frequently, whilst those with longer shelf lives lessen the requirement for frequent reapplication. Environmental factors, such as UV light, chemical exposure, and competition with local microorganisms, might impact the efficacy of the created microbes [47].

The technique of administration, whether by means of spraying, irrigation, or injection, can have an impact on the distribution and efficacy of the formulation in the desired region. The efficacy of microbial formulations can be influenced by environmental factors, such as seasonal fluctuations and climatic variations. Implementing quality control methods throughout the production process is crucial to guarantee the uniformity and dependability of microbial compositions [48]. It is important to take into account the genetic stability of microbial strains in the formulation to ensure that desirable features are maintained throughout time. Maximizing the operating efficiency of microbial formulations requires optimizing these parameters according to specific application and environmental circumstances [49].

Conclusion

In conclusion, bioformulation stands poised as a revolutionary force in horticulture, offering a paradigm shift towards more sustainable and eco-friendly crop management practices. Through the integration of natural organisms and compounds, bioformulation provides effective solutions for enhancing plant health, mitigating pests and diseases, and improving soil quality. The adoption of bioformulation not only reduces reliance on synthetic chemicals but also contributes to the preservation of biodiversity and the protection of environmental ecosystems. As we navigate the challenges of feeding a growing global population while safeguarding our planet's resources, bioformulation emerges as a beacon of hope, offering a path towards a more resilient and harmonious agricultural future. By embracing the principles of bioformulation, we can cultivate healthier crops, promote environmental stewardship, and cultivate a sustainable food system for generations to come.

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