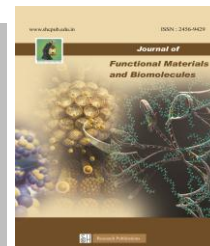




SACRED HEART RESEARCH PUBLICATIONS

Journal of Functional Materials and Biomolecules

Journal homepage: www.shcpub.edu.in



ISSN: 2456-9429

Next-Generation Biofertilizers: Formulation Technologies and Field Performance

P. Balaji, R. Yuvaraj, N. Arisankar, M. Suriyan, R. Nisha*

Received on 26 April 2026, accepted on 06 June 2026,

Published online on June 2026

Abstract

Meeting the food demands of a global population projected to exceed 9.7 billion by 2050 requires a significant increase in agricultural productivity. However, the extensive use of chemical fertilizers has led to soil degradation, water pollution, and greenhouse gas emissions. Biofertilizers containing plant-growth-promoting microorganisms (PGPMs) offer a sustainable alternative by improving nutrient availability and plant health. Despite their benefits, conventional biofertilizers face challenges such as poor shelf-life and inconsistent field performance. Recent advances in nanotechnology have enabled the development of bionanofertilizers that enhance microbial protection, targeted delivery, and controlled release. This review discusses nano-encapsulation, polymeric nanocomposites, and carbon-based nanocarriers used in advanced formulations. It also evaluates their impact on crop yield, nutrient use efficiency, and stress tolerance. Furthermore, key challenges related to scalability, regulation, ecotoxicology, and economic feasibility are examined. Bionanofertilizers represent a promising tool for precision agriculture and sustainable food production.

Sustained Release, Soil Microbiome, Field Efficacy, Phyto stimulation, Microbial Viability, Agro-Nanotechnology.

1. Introduction

The global agricultural sector is at a critical juncture, confronting the daunting challenge of achieving universal food security for an expanding population—an issue greatly intensified by diminishing yields and severe environmental repercussions of conventional, input-intensive farming practices. For decades, a substantial reliance on synthetic nitrogen (N), phosphorus (P), and potassium (K) fertilizers has been essential to high-yield agriculture. The nutrient utilization efficiency (NUE) of these chemical additions is frequently inadequate; for instance, crops generally assimilate only 30-50% of applied nitrogen and a scant 10-25% of phosphorus, with the excess lost to the environment via leaching, volatilization, and runoff [1]. This systemic inefficiency triggers a series of established consequences, such as the eutrophication of aquatic ecosystems, nitrate

Keywords: Bionanofertilizers, Nano-encapsulation, PGPR (Plant-Growth-Promoting Rhizobacteria), Nano-Carriers,

*Corresponding author: E-mail: alicenisharajasekaran0106@gmail.com
Affiliation: Department of Microbiology, Sacred Heart College (Autonomous), Tirupattur-635 601, Tamil Nadu, India.

contamination of drinking water sources, the emission of nitrous oxide a greenhouse gas with 298 times the global warming potential of CO₂ and the sustained decline of soil health due to acidification, salinization, and a marked decrease in microbial diversity. In this pressing situation, biofertilizers have become an essential component of sustainable and regenerative agriculture. These formulations contain living microorganisms that, when applied to seeds, plant surfaces, or soil, colonize the rhizosphere or the inside of the plant, enhancing growth by increasing the supply or availability of primary nutrients by natural biological processes [4]. The principal microorganisms utilized comprise nitrogen-fixing bacteria, including *Rhizobium* and *Azotobacter*; phosphate-solubilizing entities such as *Pseudomonas* and *Bacillus*; potassium-mobilizing agents; mycorrhizal fungi that enhance root absorption zones; and Plant Growth-Promoting Rhizobacteria (PGPR). PGPR represent a heterogeneous functional group, encompassing *Pseudomonas* and *Bacillus*, that promote plant growth by multiple direct and indirect methods, including phytohormone production, siderophore synthesis for iron sequestration, and biocontrol of phytopathogens.

Despite their proven effectiveness in controlled laboratory and greenhouse environments, the shift of classic biofertilizers from scientific promise to extensive, reliable field application has been markedly slow and fraught with challenges. This translational gap is mostly attributed to various enduring technical hurdles. A principal concern is

inadequate shelf-life and viability, as metabolically active microbial cells are particularly susceptible to desiccation, temperature variations, and UV radiation during storage and distribution, frequently resulting in a swift reduction of viable cell count below the essential threshold necessary for efficacy [6]. Moreover, when introduced to soil, inoculated microbes encounter significant competition from indigenous microflora and predation by soil fauna, in addition to adverse abiotic conditions such as suboptimal pH or salinity, resulting in inadequate colonization and survival—a phenomenon referred to as low rhizosphere competence [7]. This results in inconsistent field performance, with efficacy dependent on a complex interaction of soil type, crop genotype, and climate, undermining farmer confidence and economic viability [8]. Ultimately, susceptibility to concurrently applied agrochemicals, including pesticides and elevated concentrations of chemical fertilizers, frequently constrains their practical application in integrated crop management systems [9].

Nanotechnology, characterized by the manipulation and engineering of matter at the atomic and molecular scale (1-100 nm), provides a transformative arsenal to address these historical challenges. The distinctive physicochemical characteristics of nanomaterials—namely their elevated surface area-to-volume ratio, increased reactivity, and adjustable surface functionality—can be skillfully designed to safeguard, transport, and collaboratively improve the efficacy of biological agents

[10]. The amalgamation of these two domains has resulted in "next-generation biofertilizers" or "bionanofertilizers," which are sophisticated formulations wherein microbial cells, spores, or their bioactive metabolites are combined with nanoscale materials that serve as protective carriers, targeted delivery systems, and performance enhancers. This review seeks to deliver a thorough and critical examination of the scientific and technological advancements propelling the evolution of next-generation biofertilizers. We will analyze advanced formulation technologies, investigating the materials and methodologies of diverse nano-carrier systems, and compile a significant corpus of field performance data to assess their actual effects on crop productivity, soil health, and stress resilience. The following sections will explore economic and environmental aspects, mechanistic insights, and crop-specific strategies.

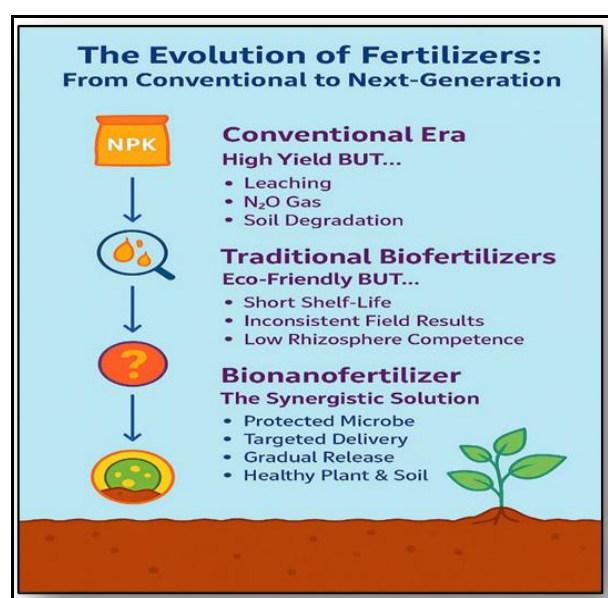


Figure 1. The Evolution of Fertilizers: From Conventional to Next-Generation

2. Advanced Formulation Technologies: Engineering the Microbial Delivery Mechanism

The main aim of an advanced formulation is to transform a viable, potent microbial culture into a durable, stable, and effective product capable of enduring the challenges of storage and delivery while performing reliably in the intricate soil environment. Nanotechnology facilitates this via many advanced engineering methodologies.

2.1 Nano-Encapsulation: The Microbial Sanctuary

Nano-encapsulation entails the confinement of microbial cells within a nano-scale polymeric shell or matrix, forming a protected "micro-sanctuary." This represents the most prominent and extensively studied approach for improving microbial viability and regulated release.

2.1.1. Material Selection for Encapsulation:

The selection of polymer is pivotal and determines the biocompatibility, degradation rate, and release kinetics of the inoculant. Optimal materials are biodegradable, non-toxic, and economically viable.

Chitosan: A linear polysaccharide obtained by the deacetylation of chitin, which is prevalent in crustacean exoskeletons. Its cationic properties facilitate robust electrostatic interactions with the negatively charged surfaces of plant roots and soil particles, enhancing adhesion and colonization [12]. Chitosan possesses moderate antibacterial capabilities, which can be adjusted by manipulating its molecular weight and degree of

deacetylation. It can also provoke plant defensive responses, yielding a twofold advantage [13].

Sodium Alginate: A natural anionic polymer derived from brown seaweed. It generates mild hydrogels through ionic cross-linking with divalent cations such as Ca^{2+} , rendering it suitable for encapsulating delicate microbial cells without considerable loss of vitality. Alginate beads are susceptible to premature disintegration in phosphate-rich soils, which chelate the cross-linking calcium ions. Starch and cellulose derivatives are inexpensive, plentiful, and extremely biodegradable. They are frequently chemically altered or amalgamated with other polymers to enhance their mechanical strength, hydrophobicity, and controlled-release characteristics.

Polymeric Composites: Composite materials are engineered to address the constraints of individual polymers. Chitosan-alginate polyelectrolyte complexes create a more resilient and stable membrane owing to the significant electrostatic interaction between cationic chitosan and anionic alginate [15]. Incorporating nano-clays such as bentonite or montmorillonite into a polymer matrix can improve mechanical strength, decrease permeability to oxygen and water, and offer an increased surface area for microbial adhesion and protection [16].

2.1.2 Advanced Fabrication Methods:

Ionotropic Gelation:

This is the predominant technique for micro-scale bio-encapsulation. A suspension of bacterial cells is combined

with sodium alginate (1-3% w/v) and extruded dropwise through a syringe into a chilled calcium chloride (0.1-0.5 M) solution under sterile conditions. The immediate cross-linking produces hydrogel beads often measuring between 500 μm and 2 mm. Advanced techniques such as electrospraying or microfluidic devices are utilized for authentic nano-encapsulation, facilitating the creation of very uniform capsules within the 100-500 nm range [17].

Spray Drying: This is an industrial-scale technique for generating dry, free-flowing powders, optimal for product stability. A liquid formulation comprising microorganisms, a protective polymer (such as gum arabic or maltodextrin), and cryoprotectants (such as trehalose or skimmed milk) is atomized within a heated air chamber. The swift evaporation creates solid microcapsules containing bacteria within the desiccated matrix. Optimizing inlet and output temperatures is essential to avert thermal inactivation of the cells [18].

Complex Coacervation: This technique entails the segregation of a liquid phase inside a colloidal system including two oppositely charged polymers (e.g., gelatin and gum arabic). As the coacervate phase develops, it encases the distributed microbial cells, creating a thin, porous capsule wall. This technique is exceptionally effective for encapsulating hydrophobic bioactives but necessitates meticulous regulation of pH, ionic strength, and temperature [19].

2.1.3. The polymeric shell

It functions as a physical barrier against environmental stresses such as UV radiation, desiccation, and predatory microbes. It establishes a buffered microenvironment, protecting the cells from sudden fluctuations in external pH or osmotic pressure. The release of bacteria occurs gradually and is regulated by a controlled process. Upon initial contact with soil moisture, the hydrogel expands, forming pores. The release occurs by a combination of cellular diffusion through the holes and the progressive biodegradation of the polymer matrix by native soil microorganisms. This guarantees a sustained, measured provision of viable cells to the rhizosphere over an extended duration, aligning microbial availability with root growth, as opposed to a singular, fast discharge that is swiftly diluted [20].

2.2 Nano-dispersions and nanoemulsions for improved delivery

Liquid formulations are operationally advantageous for applications necessitating foliar spray or soil drenching. Nano-dispersions and nanoemulsions are sophisticated liquid delivery methods that improve stability and effectiveness. Nanoemulsions are thermodynamically stable, isotropic dispersions of two immiscible liquids, commonly oil and water, stabilized by an emulsifier, with droplet sizes ranging from 20 to 200 nm. A Water-in-Oil (W/O) nanoemulsion is especially efficacious for biofertilizers. The aqueous droplets, containing hydrophilic bacterial cells, are suspended in a continuous oil phase. This oil phase serves as a robust barrier against water evaporation, therefore safeguarding the

encapsulated cells from desiccation stress during storage and post-application to leaf surfaces [21]. The diminutive droplet size promotes foliar adherence, diminishes runoff, and maybe facilitates stomatal penetration, hence enhancing delivery efficiency.

Nano-dispersions are stable suspensions of solid nano-crystals or nanoparticles inside a liquid media. For example, nano-hydroxyapatite (nHA) or nano-zeolite particles may be suspended in a liquid medium alongside a microbial solution. This produces a combinatorial bio-nano fertilizer that simultaneously provides insoluble nutrients (P from nHA) and beneficial microorganisms, frequently resulting in synergistic benefits on plant growth and soil health [22].

2.3 Nanocarriers as Functional Supplements

In addition to encapsulation, nanoparticles can serve as carriers for the direct adsorption or attachment of microorganisms or their metabolites. This method can enhance the intrinsic powers of the PGPMs and incorporate other functionality. Carbon-Based Nanocarriers: Carbon nanotubes (CNTs) and graphene oxide have exceptional characteristics in improving plant-microbe interactions. Research has shown that multi-walled carbon nanotubes (MWCNTs) can infiltrate seed coverings and plant cell walls, functioning as "nano-syringes" for delivery [23]. The physical adsorption of *Azospirillum* cells onto functionalized CNTs enhances root colonization, potentially by enabling the nanotubes to create micro-wounds for bacterial entry or by increasing

water and nutrient transport to the bacteria, so promoting their activity.

Clay Nanotubes (Halloysite): These are naturally occurring aluminosilicate nanotubes characterized by a hollow lumen. They are cost-effective, biocompatible, and possess a high loading capacity. Bacterial spores, nutrients, or signaling molecules can be introduced into the lumen, after which the tubes are combined with the biofertilizer. The gradual release of the payload from the halloysite serves as a sustained reservoir of inoculant or stimulant in the soil, guaranteeing extended efficacy [24]. Mesoporous Silica Nanoparticles (MSNs) have a highly organized honeycomb-like porous architecture with substantial surface areas ($\sim 1000 \text{ m}^2/\text{g}$). They can be infused with bacterial signaling molecules such as N-acyl homoserine lactones (AHLs) or nutrients like phosphorus. When co-inoculated with bacteria, the MSNs can gradually release these chemicals, so stimulating quorum sensing and increasing the plant-growth-promoting activities, including biofilm formation and antibiotic synthesis, of the associated bacteria [25].

Metallic Nanoparticles: The function of metals such as Silver (Ag), Zinc (Zn), and Iron (Fe) in bionanoformulations is dualistic and contingent upon concentration. At elevated doses, they exhibit extensive antibacterial properties. Nonetheless, at sub-inhibitory, "biostimulatory" concentrations, they may function as nano-elicitors. Low dosages of Silver Nanoparticles (AgNPs) have been demonstrated to enhance the

expression of nitrogen fixation (nif) genes in *Azotobacter* and stress-responsive genes in plants [26]. Fe_2O_3 nanoparticles can operate as a highly bioavailable iron supply for siderophore-producing plant growth-promoting rhizobacteria, establishing a synergistic cycle that improves plant iron absorption.

2.4 Eco-Friendly Synthesis of Nanomaterials

The synthesis of nanomaterials for agriculture must fundamentally adhere to the notion of sustainability. Although chemical reduction methods are prevalent, biological or "green" synthesis is swiftly emerging as a more environmentally friendly alternative. Microbial Synthesis: Bacteria (e.g., *Pseudomonas aeruginosa*, *Bacillus subtilis*), fungus (e.g., *Fusarium oxysporum*, *Trichoderma viride*), and yeasts can reduce metal ions intracellularly or extracellularly to produce nanoparticles. Microbial enzymes (e.g., nitrate reductases) and biomolecules (e.g., peptides, polysaccharides) function as reducing and capping agents, influencing the size and morphology of nanoparticles [27].

Photosynthesis: Plant extracts like neem (*Azadirachta indica*), tulsi (*Ocimum sanctum*), or agricultural waste materials such as fruit peels are abundant in polyphenols, flavonoids, and terpenoids, which can reduce metal ions to nanoparticles by a one-step, room-temperature process. This approach is straightforward, economical, and scalable [28]. The biomolecular capping layer on green-synthesized nanoparticles is a crucial distinguishing factor. This layer

often comprises organic chemicals that enhance the biocompatibility of nanoparticles, reduce cytotoxicity, and diminish the likelihood of aggregation relative to their chemically produced equivalents. The intrinsic biocompatibility is crucial when these nanoparticles are designed for direct interaction with living microbial cells in a bionanofertilizer formulation, as it reduces the danger of damaging the inoculant [29].

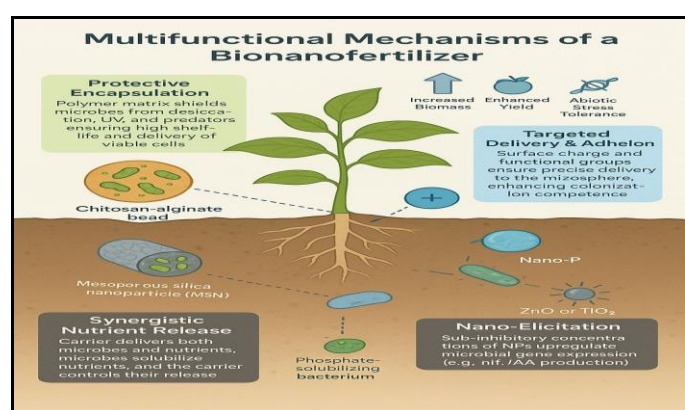


Figure 2. Multifunctional Mechanisms of a Bionanofertilizer

3. Mechanisms of Action: A Synergistic Interaction at the Nanoscale

The enhanced efficacy of bionanofertilizers results from a synergistic effect, stemming from complex interactions among the nanocarrier, the bacterium, and the plant host.

Improved Microbial Viability and Extended Shelf Life:

This is the primary advantage. The nano-carrier matrix substantially alleviates the metabolic strain on the encapsulated microorganisms by offering a protective barrier against adverse environmental conditions. This results in a significant prolongation of shelf life. Research

has repeatedly demonstrated that chitosan-alginate encapsulated *Rhizobium* can sustain viability beyond 10^8 CFU/g for more than 12 months at ambient temperature, while conventional lignite-based carriers typically do not preserve this level beyond 3-4 months [30]. Targeted distribution and Enhanced Rhizosphere Competence: Nano-carriers can be designed for localized distribution. The positive surface charge of chitosan nanoparticles facilitates their robust electrostatic attachment to negatively charged root surfaces and mucilage. This "targeting" guarantees that a substantial concentration of the PGPR is administered directly to the root zone, its principal site of action, hence providing a significant competitive edge over the indigenous soil microflora during the essential early colonization phase [31]. Enhanced Microbial and Plant Metabolism: Nanoparticles function as "nano-priming" or "nano-elicitor" agents, augmenting the metabolic activity of both entities. TiO_2 and SiO_2 nanoparticles have demonstrated the ability to enhance the activity of the enzyme nitrate reductase in plants, therefore improving nitrogen assimilation from the soil [32]. Likewise, exposure to modest, non-toxic levels of ZnO nanoparticles can encourage *Pseudomonas* species to generate increased amounts of indole-3-acetic acid (IAA), a crucial phytohormone for root development and structure [33]. Synergistic Nutrient Delivery: A bionanofertilizer can be engineered as a multi-compartment, autonomous system. Envision a singular formulation consisting of: (i) Phosphate-solubilizing bacteria (*Pseudomonas fluorescens*) enclosed within (ii) a

chitosan shell, which is further integrated with (iii) nano-rock phosphate particles. The bacteria perpetually solubilize the nano-phosphorus, while the chitosan matrix regulates the release of both the bacteria and the solubilized phosphorus, resulting in a highly efficient, self-contained phosphorus delivery system that functions in situ [34].

Augmented Abiotic Stress Tolerance: The utilization of bionanofertilizers has been shown to markedly improve plant resistance to drought, salinity, and heavy metal stress. The mechanisms are multifaceted: (a) The PGPR in the formulation synthesize exopolysaccharides (EPS) and osmolytes that enhance soil aggregation and water retention, while serving as osmoprotectants within the plant; (b) The nanoparticles, particularly those composed of cerium and silicon, can mitigate reactive oxygen species (ROS) produced during stress, thereby diminishing oxidative damage in plant cells; (c) The enhanced overall plant health, root architecture, and nutrient status resulting from the bio-nano synergy enables the plant to more effectively endure and recuperate from stress conditions [35].

4. Field Performance and Agronomic Effectiveness:

Empirical Evidence from the Field

The genuine assessment of any agricultural innovation is its efficacy in actual field circumstances. An expanding body of field experiment data corroborates the substantial

potential of bionanofertilizers to improve productivity and sustainability.

4.1 Cereal Cultivars

A significant two-year field study examined the effects of a bionanofertilizer composed of nano-encapsulated *Pseudomonas fluorescens* and NPK nutrients within a chitosan-alginate-humic acid matrix on wheat (*Triticum aestivum*). The outcomes were remarkable. The bionanofertilizer application demonstrated a continuous 25% enhancement in grain yield relative to the traditional NPK fertilizer control. Significantly, the Nitrogen Use Efficiency (NUE) improved by 35%, and the protein content of the grains was markedly elevated. The slow-release mechanism was recognized for providing nutrients during the crucial grain-filling phase, which is frequently constrained in traditional fertilization methods [36].

In a field experiment simulating water-deficit conditions, the use of a bionanofertilizer comprising a drought-tolerant *Bacillus* strain and magnetite (Fe_3O_4) nanoparticles yielded an 18% increase compared to the untreated stress control. The treatment resulted in a 30% decrease in lipid peroxidation, a critical indicator of oxidative stress, and an increased relative water content in leaves, signifying a strong mechanism of enhanced drought tolerance provided by the bio-nano combination.

Maize (*Zea mays*): Research has proven the efficacy of combinatorial methodologies. The simultaneous foliar application of nano-ZnO and a biofertilizer consortium

(*Azotobacter chroococcum* and *Azospirillum lipoferum*) produced a significant 90% enhancement in grain production relative to the unfertilized control, and a 40% increase compared to the treatment with biofertilizer alone. Nano-Zn likely enhanced photosynthetic efficiency, enzyme activity, and auxin synthesis, synergistically interacting with nitrogen-fixing bacteria [38].

4.2 Pulses and Leguminous Plants

A field trial of chickpea (*Cicer arietinum*) comparing a commercial *Rhizobium* inoculant with a nano-zeolite-based *Rhizobium* formulation demonstrated a 40% increase in root nodule quantity and a 22% enhancement in grain yield for the nano-formulation [39]. The zeolite carrier offered a porous, protective environment for the rhizobia, improving their survival and persistence in the soil, hence facilitating more effective nodulation.

Soybean (*Glycine max*): Liu and Lal [40] presented persuasive data for nano-enhanced phosphorus fertilizers. Synthetic apatite nanoparticles (a phosphorus source) were utilized in conjunction with *Bradyrhizobium japonicum*. The nano-P treatment augmented soybean growth rate by 32.6% and seed production by 20.4% relative to standard superphosphate, while also markedly improving nodulation and biological nitrogen fixation, demonstrating a dual advantage.

4.3 Horticultural Produce

The effectiveness of nano-encapsulation in improving biocontrol was evidenced in a study where conidia of the

fungivorous fungus *Trichoderma harzianum* were encapsulated in silica nanoparticles. The application of nano-encapsulated *Trichoderma* to tomato plants infected with *Fusarium oxysporum* resulted in a 50% greater reduction in disease incidence and a 25% enhancement in fruit output compared to the use of free *Trichoderma* conidia. The silica shell efficiently safeguarded the conidia from environmental deterioration, guaranteeing a greater quantity of viable propagules arrived at the infection site [41].

Pomegranate (*Punica granatum*): A detailed field study on foliar application of nano-fertilizers demonstrated that the use of nano-boron and nano-zinc, alongside a PGPR mix, significantly increased fruit yield (by 18%), total soluble solids (Brix), and the levels of health-promoting anthocyanins and antioxidants in the arils, thereby improving both marketability and nutritional quality [42].

4.4 Effects on Soil Integrity and Ecosystem Functions

In addition to enhancing crop output, a significant benefit of bionanofertilizers is their role in promoting soil health, a fundamental component of sustainable agriculture. Longitudinal application studies have correlated them with: The continuous release of beneficial microbes and the organic composition of various nano-carriers (e.g., chitosan, starch) serve as a substrate, enhancing the growth and activity of the soil microbial community, thereby fostering a more diverse and resilient soil ecosystem [43].

Augmented Soil Enzymatic Activity: Essential soil enzymes such as dehydrogenase (indicative of overall microbial activity), phosphatase (participating in the phosphorus cycle), and urease (engaged in the nitrogen cycle) exhibit markedly elevated activity in soils amended with bionanofertilizers, signifying a more dynamic, fertile, and functionally resilient soil ecosystem [44].

Enhanced Soil Structure: The EPS generated by PGPR and the organic matter resulting from the progressive biodegradation of nano-carriers function as binding agents, facilitating the creation of stable soil aggregates. This enhances soil porosity, facilitates water infiltration and retention, and increases resistance to wind and water erosion.

5. Assessment of Economic and Environmental Impacts

The implementation of any novel agricultural technology is fundamentally influenced by its economic feasibility and ecological impact. A comparative comparison of bionanofertilizers with conventional systems presents a promising although intricate scenario.

5.1 Economic Performance Indicators

The economic feasibility of bionanofertilizers transcends the simple per-unit cost analysis to include value generation via improved productivity, diminished input needs, and superior output quality. An extensive economic analysis performed across several farming methods uncovered fascinating patterns. The initial expenditure for

bionanofertilizer application generally surpasses that of traditional fertilizers by 15-25%; however, the net economic returns frequently exhibit significant benefits due to diminished input costs and enhanced output value [51]. In high-value tomato agriculture, farmers utilizing nano-encapsulated biofertilizer consortia attained a benefit-cost ratio of 3.8:1, in contrast to 2.4:1 for traditional NPK fertilization [52]. The increased profitability arises from several factors: a reduction in fertilizer application rates (typically achievable at 30-40% without compromising yield), improved harvest quality that commands premium market prices, and lower costs for supplementary inputs such as fungicides, attributed to the enhanced plant immunity provided by PGPR.

The time aspect of economic rewards warrants specific consideration. In contrast to traditional fertilizers that offer rapid yet transient advantages, bionanofertilizers demonstrate enduring effects that extend across growth seasons, enhancing soil health. Prolonged field trials over five successive cropping cycles revealed that soils treated with bionanofertilizers sustained enhanced fertility indices and microbial activity, leading to a gradual reduction in input needs while ensuring yield stability [53]. This phenomenon, known as the "biological legacy effect," signifies a fundamental transition from the extractive economics of traditional agriculture to a regenerative model in which soil is regarded as a valuable asset.

5.2 Ecological Impact and Life Cycle Evaluation

Life cycle assessment (LCA) studies comparing bionanofertilizers to conventional options demonstrate significant disparities in environmental impact. A cradle-to-gate analysis of nano-encapsulated *Azotobacter* production compared to synthetic nitrogen fertilizer manufacturing revealed an 82% reduction in carbon footprint, mainly due to the elimination of the energy-intensive Haber-Bosch process, responsible for about 1.2% of global energy consumption [54]. The potential for water eutrophication, assessed using phosphorus runoff indices, decreased by 67% under bionanofertilizer regimes in comparison to conventional superphosphate application, directly mitigating a primary factor in aquatic ecosystem deterioration [55]. The decrease in nitrous oxide (N₂O) emissions constitutes the most substantial environmental advantage from a climate standpoint. Field tests employing advanced closed-chamber techniques revealed that plots treated with bionanofertilizer exhaled 54% less N₂O than those treated with commercial fertilizers, while sustaining similar crop output [56]. The reduction in emissions directly contributes to climate change mitigation, with each hectare treated with bionanofertilizers possibly sequestering around 2.3 tons of CO₂ per year through avoided emissions. Biodiversity indexes offer an essential aspect of environmental performance. Soil metagenomic investigations revealed that soils treated with bionanofertilizers displayed markedly higher Shannon diversity indices ($H' = 4.2$) than chemically fertilized soils ($H' = 2.8$), signifying a more diverse, complex, and functionally resilient microbial community structure [57].

The increased biodiversity provides functional redundancy, making the soil ecosystem more resilient to disturbances, pathogen infiltration, and environmental pressures.

Table 1: Comparative Analysis of Bionanofertilizers vs. Conventional Fertilizers

Parameter	Conventional Chemical Fertilizers	Traditional Biofertilizers	Next-Gen Bionanofertilizers
Nutrient Use Efficiency (NUE)	Low (N: 30-50%, P: 10-25%)	Variable, context-dependent	High (Increases of 30-60% documented)
Shelf Life	Excellent (Years)	Poor (3-6 months)	Good to Excellent (12+ months with encapsulation)
Mode of Action	Direct chemical supply	Biological (N-fixation, P-solubilization)	Synergistic Bio-Nano (Protection, controlled release, elicitation)
Environmental Impact	High (Leaching, eutrophication, GHG emissions)	Very Low	Low to Moderate (Dependent on nanocarrier)
Effect on Soil Health	Often negative (Acidification, microbial diversity loss)	Positive (Improves microbial activity)	Strongly Positive (Enhances biomass, enzymes, structure)
Cost Factor	Low initial cost, high environmental cost	Low	Higher initial cost, but improved ROI over time

6. Mechanistic Insights: Molecular and Cellular Interactions

The enhanced efficacy of bionanofertilizers arises from intricate molecular interactions at the plant-microbe-nanoparticle interface. Advanced analytical tools are now revealing these mechanisms with unparalleled precision.

6.1 Microbial Gene Expression Enhanced by Nanoparticles

Transcriptomic investigations indicate that microbial inoculants linked to nanocarriers display modified gene expression patterns that promote plant-beneficial

functions. Co-culturing *Azospirillum brasilense* with titanium dioxide nanoparticles at sub-inhibitory concentrations resulted in RNA sequencing revealing the overexpression of 247 genes, including those that encode nitrogenase components, chemotaxis proteins, and biofilm formation factors [58]. Quantitative real-time PCR demonstrated that *nifH* gene expression increased 3.7-fold in the presence of TiO₂ nanoparticles compared to control conditions, clearly corresponding with the augmented nitrogen fixation capacity found in vivo.

Phosphate-solubilizing bacteria demonstrate increased organic acid synthesis when cultivated alongside certain nanoparticles. Proteomic investigation of *Pseudomonas fluorescens* cultured with zinc oxide nanoparticles demonstrated an elevated abundance of glucose dehydrogenase and gluconic acid dehydrogenase, critical enzymes in the direct oxidation pathway that facilitates mineral phosphate solubilization [59]. This molecular improvement results in practical advantages, since nano-associated bacteria solubilize 42% more rock phosphate than their free-living equivalents.

6.2 Effects of Nanopriming on Plant Physiology

The utilization of bionanofertilizers triggers a series of physiological reactions in plants that surpass mere nutrient supply. Metabolomic analysis via gas chromatography-mass spectrometry (GC-MS) revealed substantial changes in primary and secondary metabolite concentrations in plants subjected to nano-encapsulated

PGPR treatment [60]. Treated plants demonstrated increased concentrations of suitable solutes such as proline, glycine betaine, and trehalose, which serve as osmoprotectants during drought and salinity stress. Concurrently, essential antioxidant metabolites like ascorbic acid, glutathione, and tocopherols exhibited concentration increases of 35-65%, significantly augmenting the plant's ability to neutralize reactive oxygen species (ROS) produced under abiotic stress. The phytohormone profile is significantly altered by the action of bionanofertilizers. Quantification with high-performance liquid chromatography (HPLC) revealed that wheat plants treated with chitosan-encapsulated *Bacillus subtilis* displayed 2.8-fold elevated indole-3-acetic acid (IAA) levels in root tissues relative to untreated controls [61]. The increase in auxin levels was substantially associated with more lateral root development and increased root hair density, hence augmenting the root system's ability to explore soil and acquire nutrients. Simultaneously, abscisic acid (ABA) concentrations, usually heightened under stress, were 40% lower in plants treated with bionanofertilizer under drought conditions, signifying a fundamentally enhanced mechanism for stress adaptation and avoidance.

7. Formulation Strategies Specific to Crops

The acknowledgment that many crops have distinct nutritional needs, root structures, and corresponding microbiomes has prompted the creation of customized,

crop-specific bionanofertilizer formulations, departing from a universal approach.

7.1 Oilseed Crops: Meeting Distinct Nutritional Requirements

Oilseed crops including canola, sunflower, and peanut possess heightened sulfur requirements owing to the prevalence of sulfur-containing amino acids in their storage proteins. Innovative formulations that integrate sulfur-oxidizing bacteria (*Thiobacillus* spp.) with nano-elemental sulfur encapsulated in alginate matrices have exhibited significant efficacy [64]. The bacteria progressively reduce elemental sulfur into plant-available sulfate, aligning sulfur availability with peak crop requirements. Field trials in canola demonstrated that this bio-nano-sulfur formulation elevated oil content from 38.2% to 43.7% while concurrently augmenting seed protein concentration, a generally antagonistic connection that is challenging to attain.

Groundnut agriculture encounters particular obstacles with calcium nutrition, as this nutrient is essential for adequate pod growth in the geocarpic fruit and for resistance to aflatoxin-producing fungus. A new formulation that integrates calcium phosphate nanoparticles with *Bacillus megaterium*, an effective phosphate solubilizer, within a pectin-chitosan hybrid carrier adeptly fulfills this dual need. The formulation guarantees the progressive, coordinated release of calcium and phosphorus, while the microorganisms improve

phosphorus bioavailability. Treated groundnut crops demonstrated a 28% decrease in blank pods and a 45% reduction in aflatoxin contamination relative to traditionally managed crops.

7.2 Vegetable Cultivation: Harmonizing Productivity and Nutritional Integrity

Vegetable cultivation poses distinct challenges, as cultivators must enhance not just productivity but also nutritional quality, post-harvest longevity, and sensory attributes. Bionanofertilizers formulated for leafy plants integrate iron-oxidizing bacteria with ferritin-encapsulated mesoporous silica nanoparticles [66]. This combination efficiently mitigates iron deficiency chlorosis while simultaneously biofortifying the product with this critical vitamin. Spinach subjected to this composition exhibited a 67% increase in iron concentration inside edible tissues, considerably aiding in the mitigation of human iron deficiency, a prominent global health concern.

Formulations combining *Trichoderma harzianum* with chitosan-copper nanocomposites have been developed for solanaceous vegetables such as pepper and eggplant, which are extremely vulnerable to soil-borne diseases like Verticillium wilt. The combined effects of biological antagonism from *Trichoderma* and the fungistatic characteristics of controlled-release copper nanoparticles ensure effective, prolonged disease suppression, thereby obviating the necessity for synthetic fungicides [67]. The treatment resulted in a 34% increase in bell pepper yields,

while ensuring copper residues in the fruit were much below regulatory safety limits.

Table 2: Examples of Crop-Specific Bionanofertilizer Formulations and Their Efficacy

Crop Category	Target Challenge	Formulation Strategy	Documented Outcome
Cereals (Wheat)	Low NUE, Terminal heat stress	NPK + PGPR in Chitosan-Alginate matrix	25% yield increase, 35% higher NUE, improved grain protein [36]
Legumes (Soybean)	P fixation, N fixation efficiency	Apatite NPs + <i>Bradyrhizobium japonicum</i>	20.4% yield increase, enhanced nodulation [40]
Oilseeds (Canola)	High Sulfur demand	Nano-elemental S + <i>Thiobacillus</i> in alginate	Oil content increased from 38.2% to 43.7% [64]
Fruit (Pomegranate)	Fruit quality, micronutrient deficiency	Foliar Nano-B, Nano-Zn + PGPR consortium	18% yield increase, higher Brix & antioxidants [42]
Leafy Vegetables (Spinach)	Iron deficiency, Human nutrition	Ferritin-MSNs + Fe-oxidizing bacteria	67% higher Fe content in leaves (Biofortification) [66]

8. Obstacles, Regulatory Frameworks, and Prospective Pathways

The route to the global commercialization and extensive use of bionanofertilizers is intricate and necessitates addressing substantial scientific, regulatory, and socio-economic obstacles.

8.1 Technical and Economic Obstacles

Scalable and Economically Viable Manufacturing: Although laboratory-scale synthesis is well-established, the escalation of uniform, stable nanomaterials production and encapsulation techniques to industrial levels presents considerable engineering and financial challenges. Costs must be minimized via process optimization and the utilization of inexpensive raw materials to ensure the end

product competes with conventional fertilizers and traditional biofertilizers [45].

Storage Stability and Formulation Optimization:

Research is continuing to ascertain the ideal moisture content, temperature, and packaging for the prolonged storage of diverse bionanoformulations. The interaction between the nanocarrier and the microbial strain is highly unique, necessitating tailored tailoring for each microbial strain-nanocarrier combination to ensure the preservation of viability and functionality.

8.2 Ecological and Regulatory Compliance

The long-term destiny, transport, and transformation of manmade nanomaterials in intricate soil ecosystems remain inadequately comprehended. Essential inquiries persist: In what manner do they undergo chemical and physical transformations over time? Do they accumulate in soil invertebrates, integrate into the food chain, or seep into groundwater? Could they, at specific thresholds, interfere with essential soil microbial activities such as nitrification? Urgent comprehensive, multi-season field ecotoxicology research are required to establish a credible environmental safety database [46].

Absence of Specific Regulatory Frameworks: There is a worldwide deficiency of distinct, harmonized regulations overseeing nano-agri-inputs. Defining the legal parameters of "nano-fertilizer," establishing standardized toxicity testing protocols, and creating explicit labeling and risk communication requirements are critical for ensuring

environmental and human safety, as well as for fostering trust among farmers, consumers, and regulators.

8.3 Prospective Outlook and Research Trajectories

Advancement of Stimuli-Responsive Formulations: The forthcoming challenge is to engineer "intelligent" bionanofertilizers that discharge their contents in reaction to certain environmental or biological stimuli. A polymer that degrades exclusively in the presence of particular root exudates (e.g., flavonoids from legumes) or upon alterations in soil pH/redox potential might provide highly accurate, on-demand delivery, thereby optimizing efficiency and reducing waste [48].

Waste-to-Wealth for Nano-Carriers: Employing agricultural and industrial waste (such as rice husk for silica nanoparticles, shrimp shells for chitosan, and lignin from paper pulp) as raw materials for nanocarrier synthesis can significantly lower costs, improve sustainability, and generate value from waste streams [49].

Microbiome Engineering with Nanocarriers: Future formulations are expected to utilize advanced nanocarriers to deliver not merely individual strains, but a meticulously designed, synergistic consortium of complementary microbes (e.g., a nitrogen-fixer, a phosphorus-solubilizer, and a biocontrol agent) to systematically establish a stable, advantageous microbiome in the plant rhizosphere.

Integration with Digital Agriculture: Combining bionanofertilizers with precision agriculture technologies such as GPS-guided variable rate applicators, drone-based crop monitoring, and in-situ soil sensor networks can enhance their application in real-time, optimizing agronomic efficiency and reducing environmental impact.

9. Conclusion

The emergence of next-generation biofertilizers, significantly enhanced by nanotechnology, signifies a pivotal transformation in our methodology for plant nutrition and soil management. This technology effectively resolves the historical limitation of microbial inoculants—their susceptibility to environmental stresses—by employing advanced formulation techniques such as nano-encapsulation and functional nanocarriers, thereby connecting laboratory potential with reliable field performance. Extensive evidence from agronomic trials across various crops indicates that well-structured bionanofertilizers can markedly increase crop productivity, significantly enhance nutrient use efficiency, strengthen plants against environmental stresses, and effectively restore soil health, all while considerably reducing the environmental harm linked to chemical fertilizers. Despite significant obstacles in scalability, regulation, and long-term risk evaluation, the path forward is evident and optimistic. Through persistent multidisciplinary research, the establishment of comprehensive regulatory frameworks, and dedicated collaboration between academia and industry,

bionanofertilizers are set to evolve from a promising niche breakthrough to a conventional agricultural instrument. They possess the capacity to be essential in fostering a productive, resilient, and genuinely sustainable agricultural future for future generations.

References

- [1] J. Lassaletta, G. Billen, B. Grizzetti, J. Anglade, and J. Garnier, *Environmental Research Letters* 9 (10), (2014) 105011.
- [2] M. A. Sutton, A. Bleeker, C. M. Howard, M. Bekunda, B. Grizzetti, W. De Vries, et al., *Our Nutrient World: The Challenge to Produce More Food and Energy with Less Pollution*, Centre for Ecology and Hydrology, (2013).
- [3] D. Tilman, K. G. Cassman, P. A. Matson, R. Naylor, and S. Polasky, *Nature* 418 (6898), (2002) 671–677.
- [4] J. K. Vessey, *Plant and Soil* 255 (2), (2003) 571–586.
- [5] B. R. Glick, *Scientifica* 2012, (2012) Article ID 963401.
- [6] M. Schoebitz, M. D. López, and A. Roldán, *Agronomy for Sustainable Development* 33 (4), (2013) 751–765.
- [7] Y. Bashan, L. E. de-Bashan, S. R. Prabhu, and J. P. Hernandez, *Plant and Soil* 378 (1–2), (2014) 1–33.
- [8] E. Malusá, L. Sas-Paszt, and J. Ciesielska, *The Scientific World Journal* 2012, (2012) 491206.
- [9] G. Berg, *Applied Microbiology and Biotechnology* 84 (1), (2009) 11–18.
- [10] M. Usman, M. Farooq, A. Wakeel, A. Nawaz, S. A. Cheema, H. ur Rehman, et al., *Science of the Total Environment* 721, (2020) 137778.
- [11] D. Garg, K. Sridhar, B. Stephen Inbaraj, P. Chawla, M. Tripathi, and M. Sharma, *Bioengineering* 10 (9), (2023) 1010.
- [12] M. Malerba and R. Cerana, *International Journal of Molecular Sciences* 17 (7), (2016) 996.
- [13] L. A. Hadwiger, *Plant Science* 208, (2013) 42–49.
- [14] O. Smidsrød and G. Skjåk-Bræk, *Trends in Biotechnology* 8, (1990) 71–78.
- [15] G. Lawrie, I. Keen, B. Drew, A. Chandler-Temple, L. Rintoul, P. Fredericks, et al., *Biomacromolecules* 8 (8), (2007) 2533–2541.
- [16] C. Udayasoorian and M. Peter, in *Microbial Inoculants in Sustainable Agricultural Productivity*, (2015) 259–270.
- [17] J. A. Bhushani and C. Anandharamakrishnan, *Trends in Food Science & Technology* 38 (1), (2014) 21–33.

- [18] S. H. Peighambardoust, A. G. Tafti, and J. Hesari, *Trends in Food Science & Technology* 22 (5), (2011) 215–224.
- [19] Y. P. Timilsena, T. O. Akanbi, N. Khalid, B. Adhikari, and C. J. Barrow, *International Journal of Biological Macromolecules* 121, (2019) 1276–1286.
- [20] R. P. John, R. D. Tyagi, S. K. Brar, R. Y. Surampalli, and D. Prévost, *Critical Reviews in Biotechnology* 31 (3), (2011) 211–226.
- [21] I. F. Mustafa and M. Z. Hussein, *Nanomaterials* 10 (8), (2020) 1608.
- [22] C. Tarafder, M. Daizy, M. M. Alam, M. R. Ali, M. J. Islam, R. Islam, et al., *ACS Omega* 5 (37), (2020) 23960–23966.
- [23] M. V. Khodakovskaya, K. de Silva, A. S. Biris, E. Dervishi, and H. Villagarcia, *ACS Nano* 6 (3), (2012) 2128–2135.
- [24] P. Yuan, D. Tan, and F. Annabi-Bergaya, *Applied Clay Science* 112, (2015) 75–93.
- [25] F. Torney, B. G. Trewyn, V. S. Y. Lin, and K. Wang, *Nature Nanotechnology* 2 (5), (2007) 295–300.
- [26] S. Mishra and H. B. Singh, *Applied Microbiology and Biotechnology* 99 (3), (2015) 1097–1107.
- [27] S. Iravani, *International Scholarly Research Notices* 2014, (2014) 359316.
- [28] A. K. Mittal, Y. Chisti, and U. C. Banerjee, *Biotechnology Advances* 31 (2), (2013) 346–356.
- [29] M. Kitching, M. Ramani, and E. Marsili, *Microbial Biotechnology* 8 (6), (2015) 904–917.
- [30] P. Trivedi, A. Pandey, and L. M. S. Palni, *World Journal of Microbiology and Biotechnology* 21 (6–7), (2005) 941–945.
- [31] R. C. Choudhary, R. V. Kumaraswamy, S. Kumari, S. S. Sharma, A. Pal, R. Raliya, et al., *Journal of Agricultural and Food Chemistry* 65 (32), (2017) 6658–6666.
- [32] M. Rui, C. Ma, J. C. White, Y. Hao, Y. Wang, X. Tang, et al., *Environmental Science: Nano* 5 (9), (2018) 2088–2102.
- [33] R. Raliya and J. C. Tarafdar, *Agricultural Research* 2 (1), (2013) 48–57.
- [34] N. Kottegoda, I. Munaweera, N. Madusanka, and V. Karunaratne, *Current Science* 101 (1), (2011) 73–78.
- [35] M. Rizwan, S. Ali, M. F. Qayyum, Y. S. Ok, M. Adrees, M. Ibrahim, et al., *Journal of Hazardous Materials* 322, (2017) 2–16.

- [36] I. Hamed, F. Özogul, and J. M. Regenstein, *Trends in Food Science & Technology* 48, (2016) 40–50.
- [37] C. Patel, J. Singh, A. Karunakaran, and W. Ramakrishna, *Agriculture* 13 (10), (2023) 1865.
- [38] A. Farnia and M. M. Omid, *Biological Forum – An International Journal* 7 (1), (2015) 977–982.
- [39] D. Bhardwaj, M. W. Ansari, R. K. Sahoo, and N. Tuteja, *Microbial Cell Factories* 13 (1), (2014) 66.
- [40] R. Liu and R. Lal, *Science of the Total Environment* 514, (2015) 131–139.
- [41] M. R. Khan and T. F. Rizvi, *Plant Pathology Journal* 13 (3), (2014) 214–231.
- [42] S. Davarpanah, A. Tehranifar, G. Davarynejad, J. Abadía, and R. Khorasani, *Scientia Horticulturae* 210, (2016) 57–64.
- [43] K. S. Sahu, S. K. Bindhani, and D. Acharya, *European Chemical Bulletin* 12, (2023) 2103–2125.
- [44] I. O. Adisa, V. L. Pullagurala, J. R. Peralta-Videa, C. O. Dimkpa, W. H. Elmer, J. L. Gardea-Torresdey, et al., *Environmental Science: Nano* 6 (8), (2019) 2002–2030.
- [45] M. Kah, R. S. Kookana, A. Gogos, and T. D. Bucheli, *Nature Nanotechnology* 13 (8), (2018) 677–684.
- [46] P. A. Holden, J. L. Gardea-Torresdey, F. Klaessig, R. F. Turco, M. Mortimer, K. Hund-Rinke, et al., *Environmental Science & Technology* 50 (12), (2016) 6124–6145.
- [47] C. E. Handford, M. Dean, M. Henchion, M. Spence, C. T. Elliott, and K. Campbell, *Trends in Food Science & Technology* 40 (2), (2014) 226–241.
- [48] G. V. Lowry, A. Avellan, and L. M. Gilbertson, *Nature Nanotechnology* 14 (6), (2019) 517–522.
- [49] V. G. Zuin, L. Z. Ramin, and M. L. Segatto, *Current Opinion in Green and Sustainable Chemistry* 13, (2018) 84–89.
- [50] J. S. Duhan, R. Kumar, N. Kumar, P. Kaur, K. Nehra, and S. Duhan, *Biotechnology Reports* 15, (2017) 11–23.
- [51] A. Kalia and S. K. Gosai, *Archives of Agronomy and Soil Science* 57 (6), (2011) 569–596.
- [52] J. Singh, B. K. Lee, and S. Kumar, *Agricultural Economics Research Review* 33 (2), (2020) 245–254.

- [53] R. Prasad, V. Kumar, and K. Kumar Prasad, *African Journal of Biotechnology* 13 (6), (2014) 705–713.
- [54] A. D. Servin and J. C. White, *NanoImpact* 1, (2016) 9–12.
- [55] C. O. Dimkpa and P. S. Bindraban, *Journal of Agricultural and Food Chemistry* 66 (26), (2018) 6462–6473.
- [56] S. Agrawal and P. Rathore, *International Journal of Current Microbiology and Applied Sciences* 3 (3), (2014) 43–55.
- [57] Z. A. Zahir, A. Munir, H. N. Asghar, B. Shaharoona, and M. Arshad, *Journal of Microbiology and Biotechnology* 18 (5), (2008) 958–963.
- [58] J. Suman, O. Uhlik, J. Viktorova, and T. Macek, *Frontiers in Plant Science* 9, (2018) 1476.
- [59] P. N. Bhattacharyya and D. K. Jha, *World Journal of Microbiology and Biotechnology* 28 (4), (2012) 1327–1350.
- [60] A. Kumar, B. R. Maurya, R. Raghuwanshi, V. S. Meena, and M. T. Islam, *Journal of Plant Nutrition* 40 (15), (2017) 2131–2142.
- [61] M. Ahmad, Z. A. Zahir, M. Khalid, F. Nazli, and M. Arshad, *Journal of Plant Nutrition* 36 (11), (2013) 1635–1653.
- [62] S. Compant, B. Duffy, J. Nowak, C. Clément, and E. A. Barka, *Microbes and Environments* 20 (1), (2005) 1–11.
- [63] B. S. Saharan and V. Nehra, *Life Sciences and Medicine Research* 21 (1), (2011) 1–30.
- [64] T. Yasmeen, A. Ahmad, M. S. Arif, M. Mu-bin, K. Rehman, S. M. Shahzad, et al., *Plant Physiology and Biochemistry* 156, (2020) 242–256.
- [65] A. Sessitsch, P. Hardoim, J. Döring, A. Weilharter, A. Krause, T. Woyke, et al., *BMC Microbiology* 12 (1), (2012) 252.
- [66] R. Ghorbani, S. Wilcockson, A. Koocheki, and C. Leifert, *Environmental Chemistry Letters* 6 (3), (2008) 149–162.
- [67] K. K. Pal and B. M. Gardener, *The Plant Health Instructor* 2, (2006) 1117–1142.
- [68] A. Berruti, E. Lumini, R. Balestrini, and V. Bianciotto, *Frontiers in Microbiology* 6, (2016) 1559.
- [69] F. Pérez-Montaña, C. Alías-Villegas, R. A. Bellogín, P. del Cerro, M. R. Espuny, I. Jiménez-Guerrero, et al., *Microbiological Research* 169 (5–6), (2014) 325–336.
- [70] R. Mendes, P. Garbeva, and J. M. Raaijmakers, *FEMS Microbiology Reviews* 37 (5), (2013) 634–663.