EDITORIAL





Next generation microbe-based bioinoculants for sustainable agriculture and food security

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As the global population continues to surge, the challenge of feeding billions of people has become increasingly urgent. The Green Revolution, a landmark movement in the mid-20th century, significantly boosted agricultural productivity by introducing high-yield seed varieties. However, this revolution came with unintended consequences. While high-yield crops helped meet food demands, they also triggered an overreliance on chemical fertilizers. These synthetic inputs promised copious harvests but came at a cost. The excessive use of fertilizers disrupts natural ecosystems, leading to soil degradation, water pollution, and loss of biodiversity. Local habitats suffered and contamination of water bodies became a pressing concern. Moreover, it posed potential health risks to consumers and threatened the very foundation of sustainable agriculture. As a result, we find ourselves at a crossroad, where the need for increased food production must align with environmental stewardship. After various contemporary and competing thoughts and claims, environmental sustainability issues became a growing concern for governments and stakeholders, gaining prominence as a crucial topic in the Millennium Development Goals (2000). These goals aimed to address pressing issues such as poverty, hunger, disease, and environmental degradation. Subsequently, the United Nations Sustainable Development Goals (SDGs) further elevated these concerns, encompassing critical issues like hunger alleviation, promotion of good health and well-being, responsible consumption and production, as well as the preservation of life below water and on land.

As we grapple with the challenges of sustainable agriculture, microbe based bioinoculants emerge as a beacon of promise. These microbial formulations play a vital role in enhancing nutrient availability, improving soil health, and promoting robust plant growth. By harnessing the power of beneficial microbes, we can reduce our reliance on chemical fertilizers without compromising crop production. Bioinoculants hold the key to a harmonious balance between agricultural yield and environmental preservation. Due to increased concern over the utilization of chemicals in agriculture, the demand for eco-friendly bioinoculants is also rising. It is estimated that the microbial agricultural inoculants market size will grow by USD 243.73 million at a CAGR of 6.78% between 2023 and 2028. Nevertheless, the slow rate of increase in market size is due to farmers' reluctance, specially in Asia and Africa, to shift from the conventional approach of agricultural practices involving the use of chemical fertilizers and pesticides to eco-friendly bioinoculants. The microbial bioinoculants market primarily includes plant growth-promoting microorganisms that help improve soil fertility, micronutrient content, and crop yields.

Bioinoculants act as a method to carry beneficial soil microorganisms (BSM) to plants in the agro-ecosystems. These microorganisms, including bacteria and fungi, play a crucial role in improving soil conditions for plants. One of the primary utility of microbial inoculants is to covert atmospheric nitrogen into available form that plants can easily use. Additionally, the BSM can assist in making essential elements like zinc (Zn), phosphorus (P), and potassium (K) more accessible to plants, promoting nutrient-rich soil and robust plant growth. Bioinoculants also contribute to the regulation of plant hormones, supporting various plant processes. Furthermore, they can act as a defense system for plants, protecting them against phytopathogens. To ensure the effectiveness of these microorganisms, it's essential to choose the right carrier material for them. This carrier material serves as their habitat, providing a safe and suitable

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environment. It needs to be safe for the microorganisms, have good water-absorbing qualities, be easy to handle, and resistant to certain types of radiation. Cost-effectiveness, good adhesion to plants, and non-toxicity are also important considerations. Further, bioinoculants are valuable allies for plants, and selecting the right carrier material ensures their optimal performance, contributing to sustainable and healthy agricultural practices.

Liquid bioinoculants, formulated from broth cultures, mineral oils, organic oils, water and oil mixtures, and polymer-based suspensions, are increasingly being preferred for their user-friendly application on seeds and soil. Despite their convenience, these formulations confront challenges, notably a limited shelf life and sensitivity to environmental stresses, particularly temperature variations. In response, a predominant trend involves producing bioinoculants in solid forms, utilizing diverse carrier materials. Commonly employed soil-originated carriers include peat, coal, clay, lignite, and inorganic soil. Additionally, sustainable alternatives arise from agricultural waste materials such as charcoal, compost, farmyard manure, wheat bran, and soybean meal, offering diverse options for carriers. This strategic use of carrier materials not only enhances the longevity and stability of bioinoculants but also aligns with environmentally conscious practices, contributing to sustainable agricultural solutions.

Soil aggregates, recognized for their role as nutrient reservoirs and organic carbon storage, emerge as effective carrier materials in agricultural applications. Particularly in the context of nodule-forming rhizobia, soil aggregates are favoured due to their cost-effectiveness and natural compatibility. Peat, distinguished as a carrier material for rhizobial, ectomycorrhizal, and arbuscular mycorrhizal fungi (AMF) based bioinoculants, stands out for its versatility with a wide range of microbes. However, peat has a notable drawback with its variable quality and composition arising from diverse sources, prompting the exploration of alternative carriers. In contrast to peat, carriers based on charcoal, such as biochar, are gaining prominence in agricultural practices. Biochar's significance lies in its toxin free elements, substantial surface area, eco-friendly nature, and capacity to enhance the survival rate of microbes. A significant advantage is that biochar can be stored for extended periods without sterilization due to its inherently low water content. This shift towards alternative carriers underscores the ongoing quest for sustainable and effective solutions in the realm of bioinoculant applications, aligning with the broader goals of environmentally conscious agriculture.

To enhance the quality of bioinoculants, recent technological advancements have introduced innovative methods that promise superior shelf life and heightened efficiency. The evolution of bioinoculant preparation techniques has led to the development of robust microorganism entrapment and immobilization processes. These methods encompass flocculation, adsorption, covalent binding, cross-linking, and encapsulation in polymer gels, providing a shield against physical and environmental stresses for microbial cells. Among these techniques, encapsulation stands out as particularly promising, creating a protective shell around microbial cells that gradually degrades in the soil during seed germination. This breakthrough allows for the development of bioinoculants capable of carrying multiple strains simultaneously. Such versatile bioinoculants have the potential to foster the creation of consortia products, combining P or Zn solubilizers with AMF, rhizobia, and various other beneficial microorganisms. These advancements mark a significant stride towards optimizing the effectiveness and versatility of bioinoculants in modern agricultural practices.

In the encapsulation process, it is necessary to select the proper material or polymer for the entrapment of desired microbes and the size of the capsule. The polymers used for capsule preparation can be from natural or synthetic sources. Natural polymers include polysaccharides such as cellulose, chitosan, starch, and protein materials such as keratin, collagen, etc., while synthetic materials include polyacrylamide and polyurethane (Jurić et al. 2021). However, biopolymers have more importance in comparison to synthetic materials as they are more resilient to environmental stresses and also increase the viability of cells. Polyacrylamide and alginate are common polymers used for the encapsulation process, however, polyacrylamide needs efficient handling as it can create toxicity. Some common techniques used for encapsulation include extrusion encapsulation, spray drying, and emulsification. Bioencapsulation offers several advantages for cell-based applications in agriculture. These include stress-free cell protection, reduced contamination risk, and ease of handling. Micro-encapsulated cells can be uniformly distributed on seeds, enhancing efficacy and minimizing off-site drift. Additionally, they exhibit increased P solubilization capacity and plant growth promotion compared to free cells. However, limitations include high production costs and potential physiological changes within encapsulated cells.

Alginate, a natural and biodegradable polymer, is commonly used for cell encapsulation. Unlike polyacrylamide, which is toxic and requires specific handling precautions, when alginate is mixed with multivalent cations such as Ca^{2+} it may form a 3D porous gel. Microbial cells create beads by spreading in the polymer matrix, which is then poured into a cationic solution. Further, nutrients can be added for prolonged shelf life and inoculation efficacy. Technologies like spray drying and thermal gelation control bead size and shape. Smaller beads (10–100 µm) allow direct contact with seeds, while larger macro-encapsulation beads require cells to move through soil towards plants (Deshmukh et al. 2016). Alginate is well documented for entrapping bacteria, either spore-forming or non-spore-forming, AMF, filamentous fungi such as *Aspergillus*, and actinomycetes in their matrix.

Some other biopolymers are well known for their effective role in encapsulation such as chitosan (polymer of glucosamine and N-acetyl-glucosamine units), which is biodegradable and easily available. Chitosan polymerizes in cross-links and forms a matrix in the presence of anions like alginate. Apart from alginate and chitosan some other natural sources are also used such as complex polysaccharides. Gum Arabic, which is a rich source of micronutrients and obtained from the plant *Acacia senegal* is one such example. Furthermore, starch, maltodextrins, gelatin, and carrageenan (obtained from seaweeds) are also used for preparing polymer matrix.

There are also some other promising technologies that are thought to be futuristic approaches to developing good quality and highly effective bioinoculants. These include the utilization of water and oil mix formulations, which are understudied and could be used as liquid bioinoculants. Oil around the microbial cell could trap water, help in reducing evaporation and protect cells from environmental stresses. Apart from this, biofilms produced by bacteria offer an intriguing innovation as potential carriers (Seneviratne et al. 2011). Beyond bacterial inocula, they can also serve as fungi-bacteria consortia. By applying biofilms, we can enhance microbial delivery systems in agriculture and other fields. For selecting biofilms-based bioinoculants, it is imperative to select beneficial microbes that have proven capacity to produce exopolysaccharides (EPS) (Thakur and Yadav 2024). EPS help in root colonization by the introduced microbe, protection against desication and other stresses and in improving shelf-life of the bioinoculants.

Bio-nanotechnologies offer exciting new avenues for developing carrier-based microbial inocula. These technologies leverage nanoparticles, which can be made from inorganic or organic materials and typically measure 100 nm or less. Integrating whole cells within hybrid nanostructures has far-reaching applications across various fields, including agriculture. For instance, the use of biocompatible titania nanoparticles in bioinoculant aid the performance of the soil bacterium under biotic and abiotic stresses (Timmusk et al. 2018). The physical stability and the high surface area of nanotubes, combined with their cost-effective fabrication hold potential for expanding bioinoculant production. Moreover, nano-formulations can enhance the stability of bioinoculants. By incorporating hydrophobic silica nanoparticles (ranging from 7 to 14 nm) into water-in-oil emulsion formulations, the desiccation of the mycelium can be reduced (Ghormade, 2011). Remarkably, even after 12 weeks of storage at room temperature, the microorganisms remained viable and active. These advancements underscore the transformative role of nanotechnology in bioinoculant preparation for eco-friendly agriculture, offering solutions for sustainable crop production and environmental management. An alternative strategy involves fortifying bioinoculants by the incorporation of metabolites as additives; such as protectants, adjuvants, attractants, stimulants, antimicrobials, or biologically derived precursors. This enrichment serves to enhance shelf life and viability, contributing to the overall effectiveness of the bioinoculants (Arora and Mishra 2016). The additives can also help in the process of quorum sensing thus promoting root colonization or root nodule formulation.

In the realm of sustainable agriculture, the integration of the latest technologies with bioinoculants holds immense promise. By combining synergistic approaches and incorporating cutting-edge innovations, the effectiveness of microbial inoculants can be significantly improved. From precision agriculture and nanotechnology to meta-omics and synthetic biology, researchers are exploring novel avenues. By harnessing these advancements, we can create highquality bioinoculants tailored to specific crops, soils, and environmental conditions. As we cultivate a greener future, this harmonious blend of science and nature will propel us toward resilient, productive, and sustainable agricultural systems. SDG2 sets forth an ambitious vision: eradicating hunger, ensuring food security, and promoting sustainable agriculture by 2030. Achieving this goal demands unwavering commitment, especially in the face of climate change, land degradation, and persistent droughts or floods. While conventional bioinoculants have their place, relying solely on them won't suffice. It's time to think beyond traditions and embrace cutting-edge tools. The hour calls for innovation, resilience, and a holistic approach to make agriculture sustainable and more regenerative.

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