

Rhizosphere Engineering: A Novel Strategy to Mitigate Biotic and Abiotic Stress

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Abstract

Rhizosphere engineering presents a promising solution to mitigate the adverse effects of both biotic and abiotic stress on plants. This innovative approach involves deliberate modifications to the rhizosphere, the soil zone influenced by plant roots, with the aim of enhancing plant resilience and productivity. By fostering beneficial microbial interactions, altering root exudation patterns, and optimizing nutrient availability, rhizosphere engineering can effectively suppress pathogenic attacks and improve tolerance to environmental stressors. This article explores recent breakthroughs and potential applications of rhizosphere engineering as an eco-friendly and highly effective tool to safeguard agricultural systems amidst escalating stress challenges.

Introduction

Continuous exposure of biotic and abiotic stress limits the growth and development of plants and check their productivity. Under natural conditions, plants initiate a variety of stress-specific responses to cope with them, modulating various physiological, molecular, and cellular systems in the process. However, plants' natural strategies to deal with stress are not sufficient and they require external assistance. Rhizosphere refers to the narrow region in soil near root zone where complex interactions occur between the roots, soil, and microorganisms. This intimate relationship act as a hotspot for intense microbial activity between plants and their surroundings that plays a crucial role in supporting plant growth, nutrient cycling, and disease suppression. Depending on plant physiology and composition of microbial communities, the physical and chemical environment of rhizosphere results into numerous competing and interacting processes. To harness the full potential of beneficial microorganisms in rhizosphere, it is essential to understand which microorganisms are numerous in the rhizosphere microhabitat and what function they are serving in order to ensure crop productivity and sustainability of agricultural systems. Rhizosphere engineering is the novel strategy that modify rhizosphere microbiome to

understand the processes involved and to help plants to combat the impact of environmental stresses.

What is rhizosphere engineering?

Rhizosphere is the narrow zone of soil next to plant roots that serve as a breeding ground for numerous microorganisms. Apart from microbial secretions, the rhizosphere is also influenced by different rhizo-deposits such as carbohydrates, organic acids, amino acids and secondary metabolites. As a result, rhizosphere harbours wide range of microbial communities which serve a variety of purposes and exert numerous effects on plant growth promotion, nutrient cycling and defend plants from pathogens under biotic and abiotic stress conditions. Environmental stress factors impose major barrier in agricultural systems that dynamically modify the rhizosphere microbial ecology, adversely influencing plant development and growth, and ultimately impeding crop productivity. Plants frequently encounter unfavourable environmental circumstances, which results in low productivity and poor growth. The two main categories of these environmental challenges are biotic and abiotic stresses. Among abiotic stresses, salinity, drought, and extreme temperatures have a negative impact on plant growth and are known to trigger a series of molecular events that alter morphology, physiology and biochemistry of plants. Among biotic stress, phytopathogens are the primary contributors which leads to substantial decrease in crop yields.

Thus, strategic management of rhizosphere microbiome can be essential for mitigating both biotic and abiotic stress. For eco-friendly stress management, there are numerous ways to engineer the rhizosphere, including plant-based, meta organism-based and microbiome-based approaches. Few methods, mechanism and their advantages have been given in table 1, 2 and 3 providing insight to different approaches for shaping the rhizosphere microbiome. The plant-based approach to rhizosphere engineering mainly entails plant breeding techniques and the choice of suitable cultivars, which encourage the production of exudates, either stimulatory or inhibitory in nature that favour the predominance of

certain microbial members in the rhizosphere community. Genetic engineering of plants for enhancing the production of exudates or signalling molecules, creating mutants or transgenics with disease resistance due to the production of antagonistic molecules, transgenics producing quorum sensing signal molecules have direct influence on rhizosphere microbial communities. For in-depth assessment of changes in soil microbial communities, high-throughput sequencing technology can be used to obtain comprehensive information about the composition, diversity and function of soil microbes. Under metaorganism-based approach, opines produced by transgenic plants helps in selecting the host specific microbial community which utilize specific metabolic resources. For many years, research has placed a lot of emphasis on understanding how plants and microbes interact. The physiology of plant-associated microbial community helps plant to combat various stress conditions throughout their life cycle. As a result, the composition and functioning of microbiomes should be prioritised in order to fully utilise their potential.

Plant-based approach to engineer rhizosphere

Plant breeding and genetic engineering are two separate methodologies used in plant-based processes to modify desired plant traits. The use of plant breeding techniques to select certain microbial populations is an exciting strategy since the key objective of this technique is to increase crop production by improving plant resilience to various stresses. Thus, the inclusion of microbiome selection in plant breeding techniques focused on highly important taxa and functions. In 1973, Neal *et al.* employed chromosomal substitution to increase the resistance of two wheat lines to the root rot disease and showed that specific alteration of host-plant genotype preserved beneficial bacterial population in the rhizosphere. The gene for mitochondrial citrate synthase (CS) when isolated from *Daucus carota* (DcCS) and introduced into *Arabidopsis thaliana* using *Agrobacterium tumefaciens*-mediated transformation, Koyama *et al.* 2000, compared to the wild type, transgenic plants have a greater capacity for root-based citrate secretions, which helps them to grow more efficiently in phosphate-deficient soil. Few methods, mechanism and their advantages have been

given in table 1 providing insight to plant-based approaches to mitigate biotic and abiotic stress conditions.

Metaorganism-based approach to engineer rhizosphere

Microbial growth in the rhizosphere is generally promoted by carbon and nitrogenous compounds released by the plant, a phenomenon known as rhizodeposition. Since plants and microbes are interrelated, the microbiome, sometimes referred as the secondary genome of the plant which may function as a meta-organism or holobiont. This introduces the "opine concept," which combines orchestrating the host plants to secrete particular root exudates with the inoculation of microbes that are engineered to degrade substrate and often leads to the colonisation of the rhizosphere by a particular type of microbial community. By integrating pulse crops like chickpeas (*Cicer arietinum* L.), field peas (*Pisum sativum* L.) and lentils (*Lens culinaris* Medik.) into the conventional cereal-based cropping systems, crop output in the semiarid region of the Canadian prairies was found to be increased (Gan *et al.* 2015). Berendsen *et al.* 2018 showed that plants can modify their root microbiome in response to pathogen infection and resulted in recruitment of beneficial root-associated microbes that potentially maximizing the chance of plant's survival.

Microbiome-based approach to engineer rhizosphere

The rhizosphere microbial community attached to the root surface is distinct from the microbes in the non-rhizosphere soil, indicating microbial community establishment in the rhizosphere is not random but rather driven by host plant selection. In order to improve the cultivability of the microbes already present in the rhizosphere, several rhizosphere engineering procedures call for the culture of microbes. To understand the functionality and persistence of these microbial isolates focused research is needed to study their positive effects when utilised as an approach to modify the rhizosphere microbiome.

All the three approaches of engineering rhizosphere microbiome help in harnessing natural and beneficial plant-microbe interaction capable of

conferring stress tolerance to host plants. Such rhizobacteria providing resilience against abiotic and biotic stress particularly known as PGPR (Plant growth promoting rhizobacteria). PGPRs are a group of bacteria that colonize the rhizosphere and have an ability to promote plant growth by exerting variety of direct and indirect mechanisms. The mobilisation of nutrients (P, Zn and Fe), nitrogen fixation, and phytohormone synthesis are some of the major direct mechanisms induced by the action of PGPR whereas, indirect mechanisms include the production of antibiotics, siderophores, lytic enzymes, induced systemic resistance etc. There has been a lot of discussion about the potential role of PGPR strains from the genera *Bacillus*, *Pseudomonas*, *Burkholderia*, *Azotobacter*, *Enterobacter*, *Serratia* and *Streptomyces* that play a significant role in the management of plant stresses (Bandyopadhyay *et al.* 2022).

Through the production and release of organic acids and protons, several genera of bacteria help in solubilizing non available form of nutrients to their available forms in the soil. Naqqash *et al.* 2020, demonstrated that TN37 species of bacterial genus *Brevundimonas* showed maximum phosphate solubilization potential and improved growth parameters of potato cultivar. Under salinity, rice plants inoculated with zinc solubilizing bacteria viz. *Bacillus pumilus* and *Pseudomonas pseudoalcaligenes* alleviated osmotic stress by accumulating soluble carbohydrates in leaves (Jha 2019). Under salt stress conditions, inoculation of greenhouse tomato plants with a microbial consortium comprising *Achromobacter*, *Bacillus*, *Delftia* and *Enterobacter* species improved mineral uptake (Na⁺ and K⁺) and alleviated salt stress in comparison to application of a single strain (Kapadia *et al.*, 2021). PGPR combined with *Rhizobium* strains enhanced growth, nodulation, nitrogen fixation and macronutrient contents in white clover under low phosphorus conditions (Matse *et al.* 2020). PGPR also produces a number of phytohormones, including indole-3-acetic acid (IAA), cytokinins, gibberellins (GA), abscisic acid (ABA), 1-aminocyclopropane-1-carboxylate (ACC) deaminase and jasmonates. These hormones play a significant role in regulating plant growth or the response to various stress conditions (Liu *et al.* 2022). Antibacterial compound (C₁₅surfactin A) secreted by

strain HN-2 of *Bacillus velezensis* effectively inhibited *Xanthomonas oryzae* pv. *Oryzae* causing bacterial leaf blight in rice (Jin *et al.* 2020).

Conclusions

Rhizosphere engineering represents a promising strategy to address the challenges of both biotic and abiotic stress in agriculture. The engineering of rhizosphere entails purposeful alteration of the soil environment surrounding plant roots to harness the potential of microbial communities to optimize nutrient availability, to not only improve stress tolerance, but also promote sustainable agriculture practices. This can be accomplished through a variety of strategies, such as plant-based, metaorganism-based, and microbiome-based strategies. While rhizosphere engineering is gaining popularity, it is crucial to highlight that it is still a developing area with continuing study to refine and improve the methodologies and strategies used.

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Table1. Methods, mechanism and advantages of plant-based approach used in shaping the rhizosphere microbiome

| Plant-based approach | | |
|--|---|--|
| Methods | Mechanism | Advantages |
| Plant breeding and choice of suitable cultivars | Production of root exudates | Influences the microbial diversity by enhancing the growth of some selected microbes present in the rhizosphere |
| Genetic modification; change in the amount of signalling molecules, organic exudates, and residues that enters into the soil | Alteration of plant resistance to disease and environmental stress | Improve tolerance to resist adverse environmental conditions (edaphic, biological and climatic) |
| | Plants engineered to secrete exudates that directs specific microbial diversity for providing beneficial services | Plant induces microbiome for beneficial functional traits like production of antibiotics/ siderophores that act as a biocontrol agent |
| | Plants engineered to produce exudates that modify properties of the soil like pH, efflux of anion from the roots | Plant growth is enhanced at acidic or low pH, resistance towards salinity, alkalinity and water stress. Enhance resistance of plant towards Al^{3+} and also solubilize phosphorus |
| | Plants engineered to produce an enzyme that causes degradation of the quorum sensing signals | Prevention of bacterial infection |

Table 2. Methods, mechanism and advantages of metaorganism-based approach used in shaping the rhizosphere microbiome

| Metaorganism-based approach | | |
|--|---|---|
| Methods | Mechanism | Advantages |
| Managing and choosing plants and complementing microbiomes | Crop rotation | Managing soil diversity by introducing suppressive soils |
| Plants engineered to produce compounds and inoculated bacteria are engineered to degrade these compounds | Plants synthesizing opines are co-inoculated with bacteria that are able to utilizing opine | Increasing organic carbon content in soil |
| Agricultural inputs | Utilising mineral fertilizers viz. ammonium nitrate, urea etc | Building a direct connection between two partners of interaction |
| | Using organic fertilizers viz. manures, composts etc | Enhancing soil organic matter through crop residue incorporation which indirectly improve biological activity of soil |

Table 3. Methods, mechanism and advantages of microbiome-based approach used in shaping the rhizosphere microbiome

| Microbiome-based approach | | |
|---|---|---|
| Methods | Mechanism | Advantages |
| Use of microbial formulations viz. biofertilizers | Application of plant growth promoting rhizobacteria (PGPRs), arbuscular mycorrhizae fungi (AMF) etc | Biocontrol against diseases |
| | | Increased production of phytohormones |
| | | Helps in nitrogen fixation and nodulation |
| | | Improve fertility of the soil and enhance performance of the plants |

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