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Advances in Plant-Microbiome Research : Application to Rice Disease Management

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ABSTRACT

The plant as a holobiont, an ecological entity, interacts and lives with number of microbes to reproduce and survive. As a plant rice is not exceptional. Rice exploits its microbial associates or "hitchhikers" as part of an evolutionary strategy to utilize these beneficial microbial communities in ensuring sustainability with pest and disease control, and improved plant growth and yield. The plant excretes a root exudate into the rhizosphere that acts as a magnet for beneficial microbes such as *Bacillus* and *Pseudomonas* spp. These beneficial microbes act as a biocontrol agent and employ its multi-dimensional activities to help the plant as a defense mechanism against pathogens. They do so by competing for iron, a vital nutrient for most pathogens with a series of mechanisms such as siderophore production, antibiosis mediated by antimicrobial compounds such as lipopeptides and the action of lytic enzymes. These beneficial microbes also act to promote plant growth with the production of plant growth promoting substances and the activity of an enzyme called 1-aminocyclopropane-1-carboxylate deaminase, that mitigates ethylene levels initiated by stress through communicating to the host as a plant growth promoting microbe, allowing the plant to survive and grow under stressful conditions. The beneficial microorganisms also protect the plant through induction of Systemic Acquired Resistance (SAR) and Induced Systemic Resistance (ISR) that has a long-term broad spectrum, durable protection from the most damaging biotic stressors like many important plant diseases. Microbiome-engineering and the use of synthetic microbial communities as a new innovative, environmentally sustainable alternative to synthetic agrochemicals to improve or increase rice productivity that can make a contribution to feed the ever growing global population.

Keywords: ACC-deaminase, *Bacillus*, Disease management, Plant-Microbiome, *Pseudomonas*, Rice, SynCom

Introduction

Microbiome refers to the collection of genomes from all the living microorganisms in the environment, which consists of not only the community of the microorganisms but also the metabolites and microbial structural elements. Many microorganisms have close interactions with both living and non-living components of their

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surrounding environment. Depending on the particular environment, different microbiome exists. For instance, our metabolism is greatly influenced by the complex system of bacteria, fungi, viruses, protists, and archaea that make up the human gut microbiome. Similarly, a microbial ecosystem is formed by the co-existence of the plant microbiome within and surrounding plants. The same is true for the rhizosphere, or the area of soil that surrounds the plant roots. This area is a home for soil microbiome that protects the plants from pathogens and diseases, supplies vital nutrients, and aids in their ability to adapt the changing environmental conditions. As much as antibiotics can actively change the human gut microbiome and its function, similarly, the use of herbicides, fungicides and pesticides in food production shows adverse effects on the plant microbiome in the soil and on the fruits and vegetables that we eat. The journey of biocontrol-bacteria begins in the rhizosphere as it acts as a lively environment for hosting a range of microbial communities, including beneficial as well as pathogenic microorganisms and also microbes that don't show any positive or negative impact (Berendsen *et al.*, 2012). Understanding the role of the rhizosphere in biocontrol-bacteria colonization is pivotal to uncover the mechanisms underlying the successful management of many plant diseases. Biocontrol-bacteria play a pivotal role in sustainable agriculture by helping to manage weeds, pests and diseases. These instrumental microorganisms are primarily setup in the rhizosphere, and some of these bacteria also setup within the roots of crops, where they live inside plant tissues as endophytes. They have plethora of methods by which they can handle plant diseases and pests. Apart from symbiotic associations, these bacteria can also function non-symbiotically, i.e., independently. Symbiotic biocontrol bacteria develop intimate, often lifelong relationships with their host plants, these interactions are typically mutualistic, hence both the plant and the bacteria derive benefits. Usually, they infect particular parts of the plant, e.g., roots or leaves, and may form structures like nodules or biofilms. They can elevate the plant's defense mechanisms, synthesizing antimicrobial agents, and by trembling for nutrients and space, they can perform pathogen exclusion. Also, as they do nitrogen fixation or provide other necessary nutrients, they become one step ahead in the biocontrol by promoting plant growth. On the other hand, non-symbiotic biocontrol-bacteria cannot form unitary, long-lived relationships with plants. One of the most significant characteristics of these organisms is that they may operate separately and thus interact with a wide variety of flora and environmental elements. They manage pathogens through the release of antibiotics, siderophores, enzymes, and volatile organic compounds. In addition, they have the potential to induce systemic resistance in plants, hence, the plant becomes less vulnerable to different types of pathogens. Moreover, in many cases, they also compete with pathogens for nutrients and living space in the rhizosphere or phyllosphere.

Plants as Holobionts

It has been used in reference to the fact that plants coexist with different microbes both on and within their tissues. Rice microbiome shows the direct connection between the presence of specific taxa and resistance to diseases. Plants are able to decoy certain microbial taxa for their protection against pathogens and such an action may be altered by airborne signals (volatiles) that are given off by diseased plants. Plants constantly face pressure to protect themselves from different biotic and abiotic stresses, leading them to adopt multiple strategies for resilience. Alongside in internal immune responses, plants are known to use external methods that attract beneficial microorganisms from above as well as below ground (Figure 1). Plants which are continuously stressed by soil borne pathogens are able to attract more antagonistic microorganisms, that result in the formation of a disease-suppressive soil which limits the effects of soil borne diseases. The "cry for help" model offers a mechanistic explanation for the formation of disease-suppressive soils, suggesting that plants call for help from the microbiota by releasing stress-related exudates to cope with the different stresses they undergo. Despite the large population of beneficial microbes in agricultural environment, the presence of pathogens in rice is the major issue that may lead to serious loss in yield. However, harmless microbes have the potential to offer the biocontrol benefits which can increase the disease resistance of the plants by either competing with the harmful microbes or directly suppressing them. One such microbe is from the genus *Bacillus*, like *B. subtilis and B. amyloliquefaciens*. The former is specifically known for producing various antibiotics and secondary metabolites that suppresses the growth of various phytopathogens. These includes lipopeptides such as surfactin, iturin, and fengycin, which disrupt the cell membrane, resulting in cell death. Johnson *et al.* (2025) reported that *B. subtilis* strain UD1022 has notably demonstrated a multifaceted mechanism that leads to the progress in suppressing *Magnaporthe oryzae*, a pathogen responsible for the pandemic blast disease of rice and also activates rice plant defense system involving direct antagonism, volatile organic compound (VOC)-mediated inhibition, and triggering systemic resistance in the host plant, resulting a synergistic approach that replace toxic chemicals in rice production. Furthermore, technological advances in *B. subtilis* application have contributed, in management of many bacterial and fungal diseases complications that includes bacterial blight disease management in rice also. Besides these, a facultative anaerobe, *Pseudomonas fluorescens*, is another important member of the microbial consortia having biocontrol capability. Genus *Pseudomonas* have been studied for a long time as model organisms for biological control and have shown that they can be used to counter various soil-borne plant diseases effectively. This bacterium is a potent weapon, against the *Rhizoctonia solani*. They secrete antimicrobial compounds such as phenazines and pyrrolnitrin that leads to increase in chitinase and peroxidase activity in rice which alters the hypha of *Rhizoctonia*, resulted in flattened, swollen, knotted, ruptured, crumpled, and shriveled, along with leaking of cytoplasm. That results in the destruction or death of the pathogen. The success of these biocontrol agents is a clear indication that these are viable and eco-friendly way of disease management in crops that also decreases the dependency on chemical.

Application of Microbiome Engineering in Rice Crop Management

Application of microbiome engineering is a revolutionary and green method to fight against the diseases. It can be done in two ways: by external supply of beneficial which leads to enhanced plant immunity and pathogen suppression or by reintroduction of locally enriched microbial community that have been provisionally isolated and become a disease-suppressive microbiome reservoir for restoration and maintenance. Synthetic Microbial Communities (SynComs) are designed through an engineering approach like consortia formed between beneficial fungi and bacteria or bacteria and bacteria. These consortia not only enhance protective capabilities but they also address the individual strain's ecological challenges to perform much-enhanced stability and efficacy of biochemicals across diverse field conditions. The use of microbiome engineering as a viable strategy to control diseases in rice is very promising and would eventually lead to the reduction of synthetic agrochemicals. Among fungal biocontrol agents *Trichoderma* species are largely employed as against rice pathogens which involves different strategies such as competition and rhizosphere dominance; mycoparasitism and the production of antibiotics; antibiosis; detoxification of toxins released by pathogens; production of enzymes that degrade the cell wall; inhibition of pathogenic enzymatic processes; and activation of defense responses in host plants. Certain species of *Trichoderma* were not only able to limit the pathogen growth but also to lower the disease severity through the inactivation of the pathogens. They may implant their presence in the rice plants' roots, thus, preventing the pathogens' ingress and increasing the plant's defense potential. They can also damage the pathogens by feeding on them and thus, *Trichoderma* can additionally be regarded as a guardian of a lesser evil in the rice plant. Transcriptomic analyses reveal that *T. asperellum* SL2 inoculation significantly enhances the expression of at least 18 defense-related genes in rice plants. Moreover, *T. asperellum* SL2 induces Systemic Acquired Resistance (SAR) in rice plants by changing molecular signaling in plant cells.

Blast (*Magnaporthe oryzae*), brown spot (*Bipolaris oryzae*), sheath blight (*Rhizoctonia solani*), false smut (*Ustilaginoidea virens*), bacterial blight (*Xanthomonas oryzae* pv. *oryzae*), bacterial leaf streak (*Xanthomonas oryzae* pv. *oryzicola*), the rice tungro disease, rice yellow mottle, and rice hoja blanca are the major damaging diseases of rice (Laha *et. al.*, 2017). The diseases that were once considered insignificant have turned into major problems in different rice-growing areas all over the world. Some diseases that have always been there, like neck blast, brown spot, foot rot, and bakanae (*Fusarium moniliforme*), as well as sheath rot (*Sarocladium oryzae*) and stem rot (*Sclerotium oryzae*), have even arisen as major threats and possibly have a great influence on rice production in some regions. Gbadenya (2024) reported that false smut disease is one of the leading causes in rice production that has been getting out of control in the last few years. In addition to that, new diseases such as red stripe have been found in different rice-growing areas that face the problem of sustainable rice cultivation. Patel *et al.* (2021) reported that *Pseudomonas* emits a volatile organic compound (pyrazine) that can lead to a decrease in the infection of rice by *M. oryzae*. The antifungal components synthesized by *B. subtilis* have marked synergistic inhibitory effect on the hyphal development of both *M. oryzae* and *R. solani*. A proteomic study found that rice treated with this *Bacillus* strain led to higher expression of several disease-resistance proteins such as RGA1, NBS-LRR, serine, threonine protein kinase, chitinase, beta-1–3 glucanase, ascorbate peroxidases, hydroxyl methyl CoA ligase, Phenylalanine Ammonia Lyase (PAL), and iron superoxide dismutase. The elevated levels of these defense proteins in the *Bacillus* spp. treated plants most probably played a crucial role in the protection of the plants from *R. Solani* . Microorganisms can serve as effective biocontrol agents to enhance both the profitability and sustainability of rice crop production, particularly in managing fungal diseases while improving rice yields (Table 1).

Table 1. Microbes and their biocontrol regulation activities against fungal diseases in rice

Microbe	Target Phytopathogen and disease	Key Mechanisms & Observed Effects
Pseudomonas fluorescens	*Magnaporthe oryzae* (Rice Blast)	Triggered ISR in rice, leading to reduced lesion formation and disease incidence.
Bacillus subtilis	*Rhizoctonia solani* (Sheath Blight of Rice)	Reduced disease incidence and severity by producing antifungal lipopeptides (e.g., surfactins, kurstakins).
Trichoderma harzianum	*Rhizoctonia solani* (Sheath Blight of Rice)	Mycoparasitism (coiling around and lysing hyphae) and strong competition for nutrients in the rhizosphere.
Glomus intraradices	*Magnaporthe oryzae* (Rice Blast)	Up-regulated key defense response genes (*OsNPR1*, *OsAP2*), significantly enhancing plant resilience.
Burkholderia oryzicola	*Gibberella fujikuroi*gro wth (Bakane of Rice)	Reduced severity by inhibiting fungal and modulating phytohormone balance.
Streptomyces spp.	*Magnaporthe oryzae* (Rice Blast)	Accelerated defense enzyme activities like catalase, PAL, beta-1,3-glucanase and inhibited pathogen colonization.
Paenibacillus polymyxa	*Rhizoctonia solani* (Sheath Blight of Rice)	Induced systemic resistance by increasing activity of defense enzymes like PAL.
Pantoea agglomerans	*Magnaporthe oryzae* (Rice Blast)	Induced systemic resistance (ISR) via Jasmonic acid (JA) and Ethylene (ET) signaling pathways.

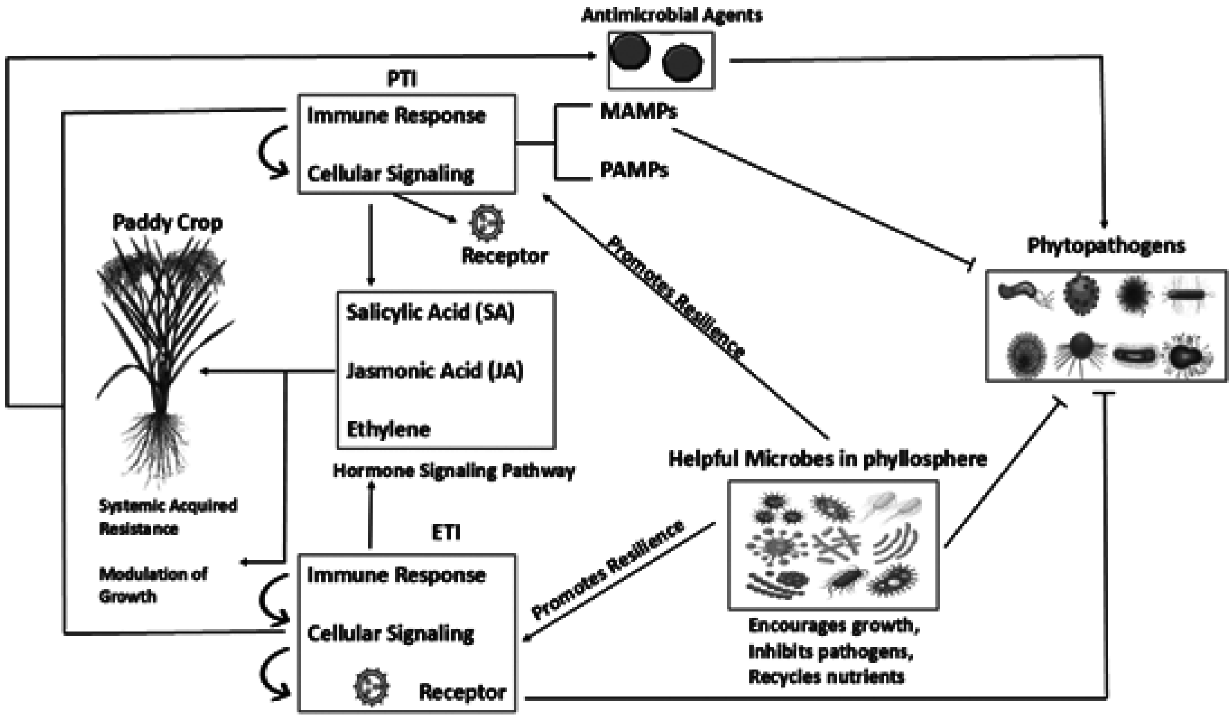

Figure 1. Harnessing Phyllosphere Microbes to Promote Resilience and Disease Resistance in Paddy Crops (Danso *et. al.*, 2023)

Bacterial blight disease out of rice caused by *X. oryzae* pv. *oryzae* and *Pantoea* spp. are some of the significant challenges to rice growers in different countries. In recent years, diverse microbial inoculants have been taken as biocontrol agents of bacterial diseases on which experiments have been conducted. For example, *Bacillus amyloliquefaciens* showed its capability to control *X. oryzae* pv. *oryzae* and *X. oryzae* pv. *oryzicola*, the pathogens that cause bacterial blight and bacterial leaf streak, respectively, in rice. *B. amyloliquefaciens* produces two antibiotic agents (difficidin and bacilysin) that negatively impact the expression of genes related to virulence, cell division, protein, and cell wall synthesis of the *Xanthomonas* pathogen. Individually or jointly, *T. harzianum* and *P. fluorescens* helped to strongly destroy the antagonist of *X. oryzae* pv. *oryzae*. The treatment of plants with a mixture of *T. harzianum* and *P. fluorescens* barely resulted in any symptoms of leaf blight as compared to other plants that did not receive the treatment of *T. harzianum* and *P. fluorescens*. The manifestation of prevention against *X. oryzae* pv. *oryzae* with the help of fungal-bacterial agents is because the cell wall components are being more lignified besides the rising enzymes activities like peroxidase, PAL, and 4-coumarate-CoA ligase in the rice leaves. The use of beneficial microbiomes for the control of bacterial diseases in rice cultivation has been considered as a very promising strategy over the years (Table 2).

Table 2. Examples of biocontrol effects of beneficial microbes in protecting rice plants from bacterial diseases.

Microbe	Target Phytopathogen	Key Mechanisms & Observed Effects
Bacillus spp. (e.g., *B. subtilis*, *B. amyloliquefaciens*)	*Xanthomonas oryzae* pv. *oryzae* (Bacterial blight of rice)	Activation of ISR and enhanced activity of defense-related enzymes like peroxidase
Pseudomonas aeruginosa	*Xanthomonas oryzae* pv. *oryzae* (Bacterial blight of rice)	Direct antagonism via antibiotic production and strong induction of host defense-related enzymes and proteins.
Streptomyces spp.	*Burkholderia glumae* (Bacterial panicle blight)	Inhibition of pathogen growth through antifungal compounds which leads to increased overall plant growth and yield.
Bacillus amyloliquefaciens and *Aspergillus pseudoporous* (Consortium)	*Xanthomonas oryzae* pv. *oryzae* (Bacterial blight of rice)	Synergistic effect of up-regulation of defense related genes and acceleration of defense proteins.
Bacillus velezensis	*Xanthomonas oryzae* pv. *oryzae* (Bacterial blight of rice)	Production of lipopeptides (fengycin) that directly inhibit pathogen quorum sensing and induce systemic defense pathways.
Pseudomonas putida	*Acidovorax avenae* subsp. *Avenae* (Bacterial stripe of rice)	Produces siderophores to restrict iron availability results in limiting pathogen growth.

Mechanism of Suppression of Microbial Diseases

Besides resisting diseases, many microorganisms also make a significant contribution to the growth of plants. Microbes such as *Pseudomonas*, *Bacillus*, and *Streptomyces* utilize siderophore-mediated iron competition, antibiosis, and lytic enzyme production to combat diseases in plants. Moreover, a bacterium that generates the enzyme 1-aminocyclopropane-1-carboxylate (ACC) deaminase is capable of enhancing plant growth, especially when subjected to environmental stress (Glick *et al.*, 2012). ACC-deaminase converts ACC, which is an ethylene precursor, into á-ketobutyrate and ammonia, thus decreasing ethylene production in plants and supporting their growth and development even under the occurrence of biotic and abiotic stresses Along with being the producers of indole-

3-acetic acid, gibberellic acid, and ACC deaminase (each one of them being very important for the control of intracellular phytohormone levels, the activation of antioxidants and defense mechanisms, and the facilitation of plant resistance to stresses), *Bacillus* species are also known to be the producers of these substances. Siderophores are the tiny molecules secreted by bacteria and fungi that are of great help in getting iron from the environment. They are the main agents that determine the high-affinity iron uptake by both pathogenic and non-pathogenic bacteria and fungi. On top of that, siderophores are very helpful in the promotion of biocontrol activities of the beneficial microbes through which they make iron a very scarce commodity for the pathogens thus leading to the major cause of increasing crop yields. The importance of siderophores in the field is yet to be fully understood. Apart from that, they could be virulence factors in pathogenic organisms by easing their iron uptake from the host plant. Therefore, the mechanism through which siderophores lead to disease relief involves the microbial production of these molecules that are intended to capture iron from the environment, thus depriving pathogens of the iron they require for virulence (Figure 2). Plants however, did

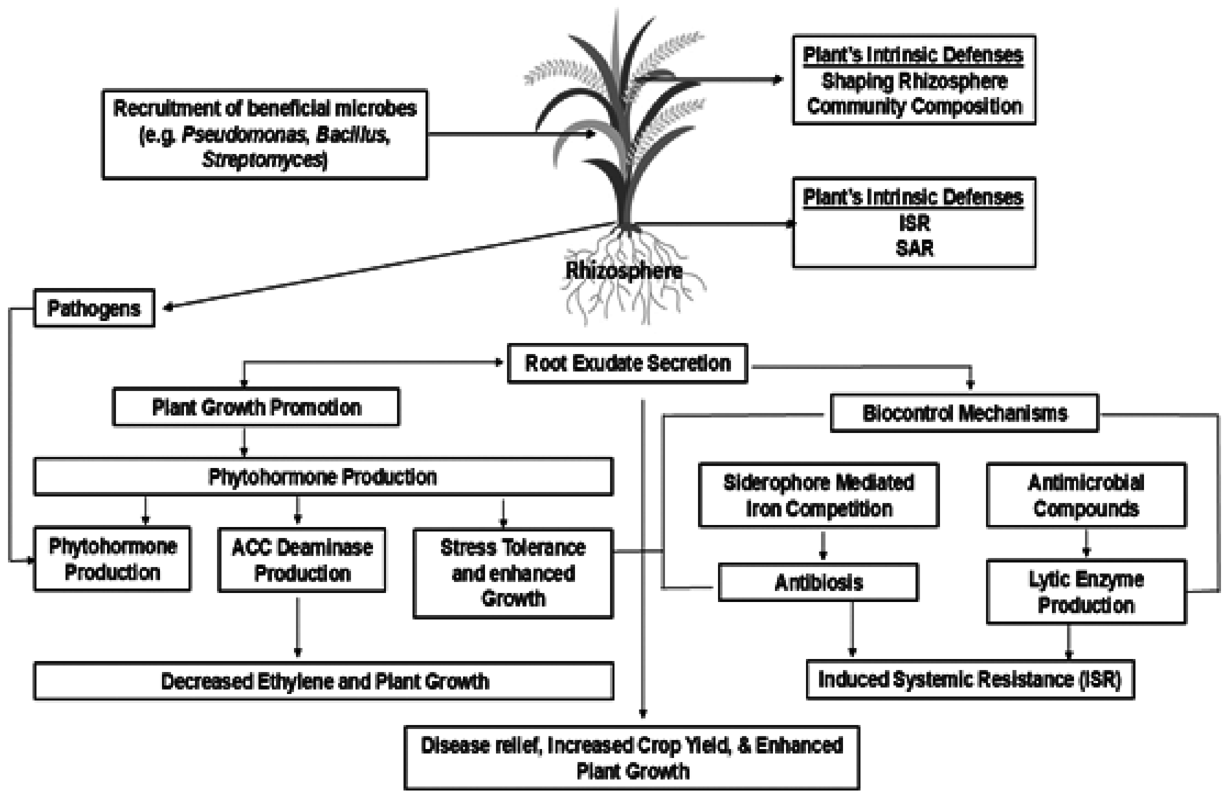

Figure 2. Mechanism of disease resistance and growth promotion mediated by rhizosphere microbes

not wait idly for pathogens to attack but rather sent out a whole arsenal of their own defensive measures. Among others, these are exudate secretion, ISR, and SAR. Both ISR and SAR are the reactions of tissues that are directly attacked by the pathogen, whereas root exudates carry out their function by recruiting microbial helpers to the infection site. The role of root exudates in not only defending the plants through the microbial community in the rhizosphere but also in shaping this community will be our point of discussion. After root exudate release, microbes respond by moving toward the plant rhizosphere. The nature of the exudate determines the microbial kinds that will be drawn to the rhizosphere which in turn means that the character of the exudate is an actor in the composition of the rhizosphere microbial community.

Conclusion

Plant microbiome are a sustainable and efficient alternative to synthetic agro-chemicals in rice production. Neither phyllosphere nor rhizosphere ecosystems are fully passive, these ecosystems are shaped actively by the plant itself, which attracts beneficial microorganisms selectively to protect against bacterial and fungal pathogens. This strengthens the concept of a rice plant as a holobiont. Key species such as *Pseudomonas fluorescens*, *Bacillus subtilis*, and *Trichoderma* spp. can effectively implement biocontrol through various mechanisms. This includes antibiosis by producing diffusible, broad-spectrum secondary metabolites and some lytic enzymes. Direct mycoparasitism, which involves the physical breakdown of pathogen structures and competitive exclusion for essential resources, such as iron-mediated siderophores. Considerably, they systemically induced resistance in the host plant (SAR and ISR), which primes its immune system for a faster and stronger response against pathogen attack. This dependency on natural and multi-dimensional defense mechanisms shows complexity on a level that is difficult for pathogens to overwhelm, unlike the single-target action of many chemical treatments. These successful case studies show that the use of powerful biocontrol agents against the major rice diseases like blast, sheath blight, and bacterial blight shows the immediate practicality of microbial inoculants in significantly lowering the disease rates. Single-strain applications have been effective, but their success has often been hampered by specific environmental conditions. In basic science, future research should seek for a better understanding of complex interactions between microbes and between microbes and plants. This includes decoding the exact chemical signals, like root exudates-plants use to send out a distress signal by attracting protective microbiota. This also includes an analysis of how signals from the nearby diseased plants affect this recruitment. Translation of this deep understanding into practice requires establishing strong, cost-effective, and field-compatible delivery systems. Despite the great progress made in using microbial communities for plant protection, research should focus on improving microbial formulations, understanding microbe-microbe interactions, and developing delivery systems suitable for the field to enhance their effectiveness. Ultimately, employing the natural resilience of the rice microbiome could lead to consistent increases in crop yields, reduced financial losses, an environmentally friendly approach to disease management and a sustainable global rice production system.

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