

Biointensive Management of Bacterial Blight of Rice

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Abstract

Microbiomes of different species have been used to control plant diseases. To date controlling pest and diseases has largely depended on the extensive use of pesticides and farmers are habituated to applying hazardous pesticides in soil and crops. Moreover, most of the pesticides have residual effects and very toxic to non target. Hence, emphasis has been given to bio intensive management of plant diseases. Rice, being a key source of nutrition, offers immediate energy due to its primary carbohydrate content. Throughout its growth stages, rice is highly susceptible to various pathogens, impacting both the quality and quantity of its harvest. Among numerous devastating diseases found in rice, Bacterial blight (BB) caused by *Xanthomonas oryzae* pv. *oryzae* is considered as one of the significant and ancient disease worldwide. The destructive BB disease has also been observed in most of the rice growing states of India including Assam, necessitating development of efficient and environmental friendly approach for management of this endemic disease. Bio-control is an alternative eco friendly, sustainable and cost effective management approaches in BB management and can also be integrated with other management practices to sustain rice yields and provide greater levels of protection.

Keywords Bacterial blight, Biocontrol, Eco-friendly, Management, Rice

1. Introduction

Xanthomonas oryzae pv. *oryzae* incites bacterial blight, which is a good-known and significant disease of rice. Tagami and Mizukami, 1962 revealed that BB was first noticed by farmers in Japan in 1884. Moderate to severe form of the disease incidence lead to a crop loss of 20-50 % of Asian and Southeast Asian countries in highly conducive conditions (Ou 1985). The incidence of bacterial blight has been globally reported in various parts of United States, Asia, Africa and northern Australia. Bihar, Uttar Pradesh, Andhra Pradesh, Haryana, Kerala, Punjab and Odisha are the key rice-growing states of India where BB has been observed widely.

Development of tannish-grey to white lesions along the veins is the systemic condition of BB disease (Mew, 1987). Symptoms become apparent during the tillering stage and the prevalence of the disease increases as the plants grow, reaching its peak during the booting stage (Mew, 1992). Wilting or

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kresek is the most destructive form of the disease, and at this phase the leaves of the entire plant become pale yellow and wilt from the seedling to the early tillering stage, potentially resulting in a partial or complete crop loss. Plants younger than 21 days are most vulnerable and temperatures ranging from 28 to 34 °C promote the development of kresek (Mizukami and Wakimoto, 1969). Symptoms induced by BB as wavy margins on leaf blades with yellow lesions which may extend to leaf sheath. Bacterial ooze from infected tissues has been noticed in warm and humid climates lead to disease's spread. In wilt or kresek phase maximum damage occurs as compared to post-flowering infections which have minimal impact on grain yield. BB thrives in Warm temperatures ranging from 25 to 30 °C, along with high humidity, precipitation and deep water favours the BB infections. The disease is more common in wetland areas where these conditions are frequent. The severity of the disease is more pronounced by the availability of virulent bacterial isolates. BB is particularly severe in susceptible rice varieties when high nitrogen fertilization is applied (Anon, 2004; 2010). The bacterium can spread through irrigation water, splashing or wind-driven rain, plant-to-plant contact, trimming tools used in transplanting and handling plants during transplanting (Mew, 1992).

2. Management of Bacterial Blight of Rice

Efforts to manage BB using chemical treatments such as copper-soap mixtures, copper-mercury fungicides and Bordeaux mixture, with or without sugar, were reported by Sulaiman and Ahmed (1965). It was suggested that spraying a solution of copper oxychloride and streptomycin at short intervals would help in combating this disease (Seki and Mizukami, 1956). Application of stable bleaching powder along with irrigation water could effectively reduce the disease incidence (Chand *et al.*, 1979). Additional synthetic organic bactericides including nickel dimethyl dithiocarbamate, dithianone, phenazine and phenazine N-oxide were likewise advised (Fukunaga, 1966). Using techlothalam for spraying was found to be more effective than applying it to the soil, as it moved easily within the plants and inhibited bacterial growth in rice (Takahi, 1985). Earlier studies suggested treating seeds with hot water at 57°C for 10 minutes or disinfecting with mercury-based compounds to eliminate seed-borne pathogens (Tagami *et al.*, 1963). Srivastava and Rao (1963) found that soaking rice seeds in a 0.07% Agrimycin solution for 12 hours followed by a water bath at 54 °C for half an hour achieved 95 to 100% eradication of the bacterial blight. Rajagopalan (1961) indicated that pre-soaking seeds in cold water, using streptomycin sulfate (27 ppm) or ceresan (1000 ppm) for 8 hours and then applying hot water at 54-55 °C for 20-30 minutes was the most effective method for controlling seed infections. Hori (1973) first time introduced the chemical fungicide dithionone for the control of disease both in vitro and field conditions. Besides, nickel dimethyl dithiocarbamate also significantly reduced the BB disease severity in many countries including India (Lee, 1975).

In rice, the genetic basis for resistance against various pathogens has been extensively studied. The resistance of rice plants to Xoo changes at different growth stages and depends on the host genotype, exhibiting seedling resistance during the seedling stage and adult plant resistance in the adult stage, although they may be susceptible during the seedling stage (Mew, 1987). Genetic modification of rice presents numerous valuable opportunities for enhancing existing elite strains and creating new varieties. One key benefit of genetic engineering is its ability to quickly produce new varieties through the incorporation of cloned genes into commercial stocks. Research by Zhang *et al.* (1998) demonstrated the successful regeneration of transgenic fertile plants from four elite indica varieties, namely IR64, IR72, Minghui63 and BG90-2, which possessed the bacterial blight resistance gene Xa21. This modification led to a significantly enhanced resistance to the BB pathogen, with this trait shown to be consistently passed down in future generations. Wang *et al.* (1996) introduced the cloned Xa21 gene into the japonica rice variety T309, demonstrating a resistance profile akin to that of the donor line IRBB21. Tu *et al.* (1998) found that the incorporation of the Xa21 gene into the elite indica rice variety IR72 grants resistance to the BB pathogen and this resistance was proven to be consistently passed down through generations.

3. Biological Control of Bacterial Blight in Rice

Various disease management strategies had been employed to reduce the yield losses and to avoid epidemics. Use of chemical substances has been a dominant management strategy for an extended time period but it has several barriers. Improvement of resistant pathogenic races, toxic residues, environmental pollutants and ill impact on beneficial biodiversity are some of the main obstacles of chemo centric control practices (Tjamos *et al.*, 1992). Non availability of effective chemical measures and inconsistent performance of resistant rice varieties have forced the plant pathologists to look for new tactics like exploitation of biocontrol agents for ecofriendly and sustainable disease control, which is a key element of integrated disease management.

20-40% yield reduction has been seen when the bacterial pathogen attack the crop at tillering stage,ⁱTo reduce the incidence of BB various management approaches like, host plant resistance, cropping system alterations, chemical control, and biological control have been employed. Though the use of chemicals and resistance varieties, were the most common management practices of farmers but they have certain limitations. Chemical pesticides are harmful to the environment and host-plant resistance, which is based on a single gene, may not be durable in the field leading to frequent resistance breakdowns. Therefore, it is most important to develop a sustainable, eco friendly management strategies. Biological control is an ecology-conscious, cost-effective and sustainable alternative method in BB management. This approach can also be integrated with other management practices to afford greater levels of protection and sustain rice yields. Antagonistic bacteria are

easy to handle, grow rapidly and colonize the rhizosphere aggressively (Weller, 1988). Certain strains of *Bacillus* sp. and *Pseudomonas* sp., have been used as biocontrol agents to suppress rice BB (Vasudevan *et al.*, 2002). A novel plant growth-promoting strain of *Delftia tsuruhatensis* HR4 has been shown to be promising biocontrol agent against BB (Han *et al.*, 2005).

Biological management of plant diseases using antagonistic microorganisms is recognized as an economical, effective and environmentally friendly approach to controlling crop infections (Cook and Baker, 1983). Nowadays, the adoption of biological control agents as substitutes for chemical fungicides is rapidly growing, primarily due to the harmful effects of synthetic pesticides. The genera *Pseudomonas* and *Trichoderma* have been acknowledged for their ability to mitigate plant diseases caused by both fungal and bacterial pathogens (Pant and Mukhopadhyay, 2001). Bacterial antagonists offer the dual benefits of rapid reproduction and greater rhizosphere competence, contributing to organic crop production. Bioagents promote plant growth, leading to improved yields (Mishra and Sinha, 2000). Several biocontrol agents have been registered and can be found in the market as commercial products, including those from the bacterial genera *Pseudomonas*, *Streptomyces* and *Bacillus*, as well as fungal genera such as *Trichoderma* and *Gliocladium*. Biological control has surfaced as a viable and promising approach for managing plant pathogens. Bioagents encourage plant growth even in the absence of disease, leading to improved yields. Numerous researchers have reported the effectiveness of fungal and bacterial bioagents against Xoo, which causes bacterial blight (BB) disease in rice. It has been noted that the severity of BB can be reduced by 40-60% and grain yield improved with the use of *P. fluorescens* as a seed inoculant (Gnanamanickam *et al.*, 1999). The application of *P. fluorescens* as a seed inoculant not only lessens BB severity by 40-60% but also boosts grain yield. Ahmed and Thind (1992) found that using antagonistic sprays of *B. subtilis* and *P. fluorescens* (24 and 36 hours before inoculation) in both greenhouse and field settings yields the best disease control, ranging from 35-48%, while also increasing grain yield by 9%. Nonetheless, the simultaneous application of *Penicillium oxalicum*, *T. harzianum* and *C. cladosporioides* with the pathogen was more effective than their application before or after inoculation, both in greenhouse and field settings. Thirty strains of *B. subtilis* isolated from the rice rhizosphere were found to inhibit the growth of Xoo by 58.1% under in vitro conditions (Chen *et al.*, 1997). According to Sindhan *et al.* (1997), applying *Penicillium acidovorans*, *Aspergillus ochraceus*, *Fusarium chlamydosporum*, *F. pallidoroseum* and *Micrococcus* to rice plants seven days prior to inoculation with Xoo significantly reduced lesion length. Islam and Bora (1998) found that the use of *Azospirillum brasilense* and *Bacillus polymyxa* led to a significant decrease in bacterial blight (BB) of rice, while simultaneously improving growth and yield. Vidhyasekaran *et al.* (2001) discovered that inoculating seeds and seedlings with a talcum powder-based *P. fluorescens* formulation was effective in managing the disease. They found that the integrated use of this product through seed treatment, seedling root

dipping and foliar spray could effectively reduce the incidence of Xoo upto 70% in field conditions. Similarly Manmeet and Thind (2002) conformed that the use of *B. subtilis*, *P. fluorescens* and *T. harzianum* as seed treatment, seedling dip treatment and foliar sprays, significantly lowered disease severity in the field. Nayak *et al.* (2002) observed that as growth of *Bdellovibrio bacteriovorus* in the inoculum increased then esion lengths, kressek incidence and foliage blight in inoculated seedlings get diminished.. Previously, Chetia (1995) investigated the antagonistic properties of *T. harzianum* and *T. viride* by applying their conidial suspensions against Xoo in the field, achieving a 62.03 and 58.69% reduction in disease, respectively. Microbial agents used for biocontrol play a vital role in enhancing soil fertility status through recycling nutrients. Typically, plant growth-promoting rhizobacteria (PGPR) support plant development either by aiding in the acquisition of resources like nitrogen, phosphorus and essential minerals or by influencing plant hormone levels, or they can do so indirectly by reducing the harmful effects of pathogens on plant growth. The availability of nutrients impacts plant growth and alters the microclimate, which significantly affects pathogen infection and sporulation. Additionally, nutrient levels can indirectly influence disease resistance, as plants lacking in nutrients not only show weakened defense responses but are also more susceptible to pathogen attacks (Munees and Mulugeta, 2013). The rhizosphere is the primary zone of microbial activity compared to the bulk soil and other plant areas, due to more favorable conditions for microbial growth. This area can contain as many as 10^{11} microbial cells per gram of root and hosts over 30,000 prokaryotic species that can affect plant productivity (Mendes *et al.*, 2013). Research has shown that soil with a diverse microbiota tends to mitigate the impact of numerous soilborne pathogens. Numerous antagonistic microorganisms are inherently found in soil and provide a certain level of biological control over the activities of plant pathogens. Microorganisms in the rhizosphere are crucial for mitigating soil-borne plant diseases and enhancing plant growth. Research has shown that a diverse microbiota in nutrient-rich soil tends to lessen the impact of various soil-borne plant pathogens. While many antagonistic microorganisms exist naturally in soil and contribute to biological control of plant pathogens, this natural defense often falls short of ensuring consistently disease-free crops. The ability of biocontrol agents to thrive in the rhizosphere is vital, as the specific plant species, soil characteristics and types of pathogens influence the composition of the microbial community present (Schreiter *et al.*, 2014). Antagonistic microorganisms residing in the rhizosphere serve as ideal biocontrol agents, since this area acts as the primary defense for roots against pathogen invasion. The ever-changing nature of microorganisms in the rhizosphere offers a promising avenue for research in disease management. It has been observed that several strains of bacteria isolated from the rhizosphere displayed significant antibacterial properties against the rice bacterial blight (BB) pathogen *Xanthomonas oryzae* pv. *oryzae* (Velusamy *et al.*, 2006). Yasmin *et al.* (2017) discovered that five strains of antagonistic bacteria,

specifically from *Pseudomonas* sp. and *Serratia* sp., exhibited antagonistic activity (with inhibition zones ranging from 1 to 19 mm) against Xoo. All these antagonists also demonstrated plant growth promoting effects, including siderophore production, inorganic phosphate solubilization (82-116 $\mu\text{g mL}^{-1}$) and indole acetic acid production (0.48-1.85 mg L^{-1}). The rice plant serves as a habitat for a variety of microorganisms that inhabit different areas: the aerial parts (phyllosphere), the root surface (rhizoplane), the region surrounding the roots (rhizosphere) and within plant tissues (endophytes). Research indicates that the population of endophytes decreases as one moves from roots to upper parts of the plant, with roots hosting a greater number of endophytes than above-ground tissues (Knief *et al.*, 2012). Changes in the composition of endophytic communities can be linked to factors such as the age of the plant, the source of the plant, the type of tissue, the timing of sampling and environmental conditions. An increasing body of research supports the idea that endophytes (both fungi and bacteria) confer protection to host plants against pathogens and pests, starting from seed germination and continuing throughout the plant's life (Hardoim *et al.*, 2015). Endophytic bacteria may enhance plant growth and combat plant diseases likely through mechanisms similar to those employed by other plant growth-promoting microorganisms (PGPM). Numerous bacterial and fungal endophytes have been documented to support growth and bolster plant health, thereby being considered a vital source of biocontrol agents. The success of endophytes as biological control agents (BCAs) relies on various factors. These factors encompass host specificity, population dynamics, colonization patterns and the ability to move within the host. Among the multiple strategies that endophytes utilize to enhance plant health, one key method is by inhibiting pathogen growth and viability. This suppression involves several mechanisms, including direct competition with pathogens for resources like space and nutrients, production of antimicrobial compounds and the induction of systemic resistance, which boosts the host's defenses against pathogens by enhancing the expression of defense-related genes (Irizarry and White, 2017). Bacterial endophytes contribute to plant growth and increase the ability of hosts to endure pathogen challenges through competition, antibiosis and the induction of induced systemic resistance (ISR). Endophytic bacteria belonging to the genus *Pseudomonas* produce a range of antifungal substances, such as phenazine-1-carboxylic acid, 2,4-diacetylphloroglucinol, pyrrolnitrin, pyoleutin and volatile compounds like hydrogen cyanide, which significantly hinder the growth of fungal and bacterial pathogens (Ongena *et al.*, 2008). Diby (2005) indicated that *Pseudomonas fluorescens* isolated from black pepper can produce siderophores and volatile substances like HCN, which assist in preventing the growth of pathogens. According to Zhang *et al.* (2016), *Bacillus* species have the ability to synthesize volatile compounds and various lipopeptides that may serve as inhibitors of phytopathogenic fungi. Hastuti *et al.* (2012) investigated the effectiveness of ten endophytic *Streptomyces* species in suppressing Xoo disease both *in vitro* and *in planta*. Their research also revealed that *Streptomyces* sp. AB131-

1 produced a significantly higher number of tillers and yields. It has been noted that root infections with endophytic fungi lead to increased phenolic production and stimulate stronger plant defense mechanisms. Fungal endophytes can modify interactions with pests and pathogens, positioning them as viable biocontrol agents. These endophytes release antifungal and antibacterial compounds in minimal concentrations, thereby inhibiting competitors, both endophytic and pathogenic and preserving a balance of antagonism with these competitors (Suryanarayanan *et al.*, 2016).

Bacterial antagonists have been assessed with varying levels of effectiveness for controlling rice diseases caused by fungi (Vasudevan *et al.*, 2002). The *P. fluorescens* strain 7-14 has demonstrated a well-established capability to inhibit both rice blast and sheath blight (Chatterjee *et al.*, 1996; Krishnamurthy and Gnanamanickam, 1998). The potential for biocontrol agents to effectively reduce BB has yet to be thoroughly investigated. Nevertheless, the use of antagonistic bacteria such as *Bacillus* sp. for managing BB in rice has been recorded (Vasudevan, 2002). The application of plant growth promoting rhizobacteria (PGPR) has gained significant traction in agriculture, providing an appealing method to enhance crop growth and development while replacing or complementing fertilizers and pesticides. Regarding environmental health, biocontrol agents are viewed as a more eco-friendly alternative to chemical protections. Up to now, several biocontrol agents have been registered and are accessible on the market as commercial products, including strains from the bacterial genera *Pseudomonas*, *Streptomyces* and *Bacillus*, as well as fungal genera like *Trichoderma* and *Gliocladium*. A prominent PGPR from the *Pseudomonas* order, *Pseudomonas fluorescens*, serves as an effective biocontrol agent against soil-borne diseases in a variety of crop plants (Zhang *et al.*, 2000).

The significance of sustainable agriculture has emerged as a critical concern in today's context of agricultural advancement and research globally. In this landscape of enhanced farming practices, pests and diseases continue to significantly hinder crop yields. Over the past few decades, the utilization of plant growth-promoting rhizobacteria (PGPR) for sustainable farming has surged in various regions worldwide. Typically, PGPR can enhance either the quantity or quality of plant growth through three main mechanisms: producing specific compounds for plants, aiding in the absorption of certain nutrients from the soil and mitigating or reducing diseases in plants, generally classified into direct and indirect methods. The direct methods of PGPR involve making phosphorus available for plant uptake, fixing nitrogen for plant utilization, sequestering iron for plant access via siderophores, producing phytohormones such as auxins, cytokinins and gibberellins and lowering levels of ethylene in plants, among others (Glick, 1995 and Glick *et al.*, 1999). Indirect mechanisms involve mitigating the harmful impacts of plant pathogens, achieved through the production of a diverse array of antibacterial and antifungal substances, which supports their extensive application as biocontrol agents (Hayat *et al.*, 2010).

The inconsistency in the effectiveness of these biocontrol agents raises concerns and needs to be addressed to enhance their performance against diseases and establish a dependable substitute for chemical control. Increasing trust in biocontrol agents is essential not just for researchers but also for growers. The variability in effectiveness often stems from various abiotic and biotic influences. Consequently, managing habitats may help minimize inconsistencies by identifying factors that, alongside biocontrol agents, can produce a synergistic effect and enhance their antagonistic actions in the suppression of crop diseases. Biological systems respond to the salt levels present in their environments. The nutrient elements within the nutrient solution, along with specific micronutrients, can be adjusted to improve the management of root diseases or to boost the antagonistic capabilities of biocontrol strains (Zahran, 1997).

4. Conclusion

The positive attributes of application of microbial bioagents can be adopted as an effective and sustainable disease management agent for enhancing crop productivity, nutrient availability, imparting tolerance against pest and diseases and thus eliminating the possibilities of crop failure or poor crop productivity. Thus utilizing microbial bioagents can serve as an effective approach in organic farming to manage various rice diseases that are closely linked to soil nutrient levels, changes in microbial populations, their influence on suppressing pathogens, or the interactions between the host and pathogens.

5. References

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