



## Potential Plant Growth-Promoting Microorganism (PGPM) as Biological Control Agents of Paddy in Indonesia

Mahindra Dewi Nur Aisyah<sup>a</sup>, Trisnani Alif<sup>b\*</sup>

<sup>a,b</sup> Jurusan Produksi Pertanian, Politeknik Negeri Jember, Indonesia

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#### *Corresponding Author:*

name author

<sup>b</sup>Jurusan Produksi Pertanian,  
Politeknik Negeri Jember

\*email: [trisnani@polije.ac.id](mailto:trisnani@polije.ac.id)

### ABSTRACT

Today, the main goal of agriculture is increasing crop yields to meet the ever-increasing human population. Climate change has increased the challenges associated with the cultivation of food crops, especially rice. It affects rice production due to the influence of biotic factors (Plant Pest Organisms) and an uncertain environment. To address this phenomenon, Plant Growth Promoting Microbial (PGPM) is considered a better alternative than using chemicals. It has been proven that Plant Growth Promoting Rhizobacteria (PGPR) and Fungi (PGPF) are effective in suppressing plant diseases and controlling pests by producing inhibitory chemicals and inducing immune responses in plants. Furthermore, PGPM increases growth and yields. As biofertilizers and biopesticides, PGPR and PGPF are considered attractive and economically viable approaches to the cultivation of rice in Indonesia. The potential for PGPM utilization is still high considering the diversity of microbes and the fact that these microbes can be found under a variety of environmental conditions. However, it is also a challenge to develop products, especially treatments to maintain the performance of the microbes that will be used

## INTRODUCTION

The global human population is expected to reach 9.8 billion by 2050 and food demand for this growing population will also increase by 50% or even 56% (van Dijk et al., 2021). Based on this phenomenon, it is imperative to improve agricultural production to balance the social and economic challenges caused by population growth. The challenges become even greater when one-third of agricultural land is threatened by desertification and decreased soil fertility due to climate change factors and excessive use of high agricultural inputs (Adl, 2016). For example, decreasing rice production caused by extreme climate change conditions (uncertain rainy and dry seasons) and an increase in the attack and development of plant pest organisms (OPT) is a very serious problem in rice cultivation because it can result in failed harvests (Sudewi et al., 2020).

Plant production and protection systems using a biological approach are currently very developed (Philppot et al., 2013). This approach is based on natural practices and ecological balance to minimize the input of chemicals that have many disadvantages (Singh et al., 2016). Prospects for enriching soil with bioorganic inputs can through changing cultivation practices and manipulating plant rhizosphere using bio-inputs. Bio-inputs are biological products obtained from living organisms such as fungi, bacteria, plants, or their derivatives which can be used as biostimulants, biofertilizers, biocontrollers, biostabilizers, or inoculants for plant protection or to increase soil nutrition and fertility (de Salamone et al. al., 2019).

Microbial inoculants are becoming popularly used to improve plant health and soil fertility, to increase plant resistance to biotic and abiotic agents, and to reduce damage from pests and diseases (Singh et al., 2016). It has been proven to have an impact on plant growth and development, crop production, increasing intrinsic resistance/tolerance of plants to stress (biotic and abiotic), soil remediation. (Kazerooni, et al. 2021). Various studies also provide evidence regarding induced systemic resistance, plant innate immunity, root rhizosphere biology, antagonistic attributes of microbial communities, the impact of inoculated microbes on non-target organisms, and strengthen the facts about the benefits of microbial inoculation in soil and/or plants (Farrar et al., 2014; Reddy & Saravanan, 2013). In particular, Plant Growth Promoting Microorganisms (PGPM), including the Plant Growth Promoting Rhizobacteria (PGPR) and Fungi (PGPF) groups, can also act as an alternative for managing pests and diseases due to the harmful effects of chemical pesticides (Waghunde et al. 2016). Therefore, recently, more attention has been paid to plant growth-promoting rhizobacteria and fungi (PGPR and PGPF, respectively) to replace or supplement agricultural chemicals.

## **DISCUSSIONS**

### **1. Plant Growth Promoting Microorganism (PGPM) Mechanism as Biological Control Agents**

PGPM which is currently being developed consists of PGPR and PGPF. PGPR consists of a heterogeneous group of nonpathogenic bacteria colonizing roots and improving plant growth, generally found in the rhizosphere of plants. PGPRs are categorized based on their habitat into extracellular (e-PGPR-symbiotics) and intracellular (iPGPR-free-living) PGPRs (Gray and Smith 2005). ePGPR resides in the interspaces in root cortex, rhizosphere, and rhizoplane cells; whereas, iPGPR resides in the nodular structures of root cells (Figueiredo et al. 2010). ePGPR includes different bacterial genera such as *Erwinia*, *Flavobacterium*, *Arthrobacter*, *Agrobacterium*, *Azotobacter*, *Azospirillum*, *Burkholderia*, *Bacillus*, *Caulobacter*, *Chromobacterium*, *Micrococcus*, *Pseudomonas*, and *Serratia* (Ahemad and Kibret 2014). iPGPR includes members of the *Rhizobiaceae* family (such as *Rhizobium*, *Bradyrhizobium*, *Allorhizobium*, *Mesorhizobium*), *Frankia* species, and endophytes, and many other types of bacteria (Bhattacharyya and Jha 2012). Meanwhile, PGPF is a nonpathogenic saprophyte that has beneficial effects on plants. Currently, research on the interaction of PGPF and plants is

also starting to be developed although it is not as much as PGPR. Well-known nonpathogenic fungal genera includes *Aspergillus*, *Piriformospora*, *Fusarium*, *Penicillium*, *Phoma*, *Rhizoctonia*, and *Trichoderma* and stimulates various beneficial plant traits for higher yields (Jaber and Enkerli 2017; Lopez and Sword 2015). Some examples of PGPFs with BCA activity include endophytes, ectomycorrhizae (EcM), arbuscular mycorrhizae (AMF), yeast, *Trichoderma* sp., and certain avirulent phytopathogenic strains such as *Fusarium oxysporum*, *Cryphonectria parasitica*, and *Muscodor albus* (Waghunde et al. 2017).

PGPM produces certain compounds that are useful as biocontrol or Biological Control Agents (BCAs) consisting of direct antagonist, indirect antagonist, and mixed mechanisms (Table 1), specifically in rice plants (Figure 1). PGP Bacteria and fungi through their respective mechanisms are able to encourage healthy plants so that several reports provide a trend of success in the production of cultivated plants. The interactions formed and the resulting benefits have been studied extensively in recent decades because these microorganisms can reduce the use of pesticides. PGPR and PGPF also protect plants from phytopathogenic microorganisms and pests (Etesami and Maheshwari 2018).

**Table 1** Antagonism exhibited by biological control agents

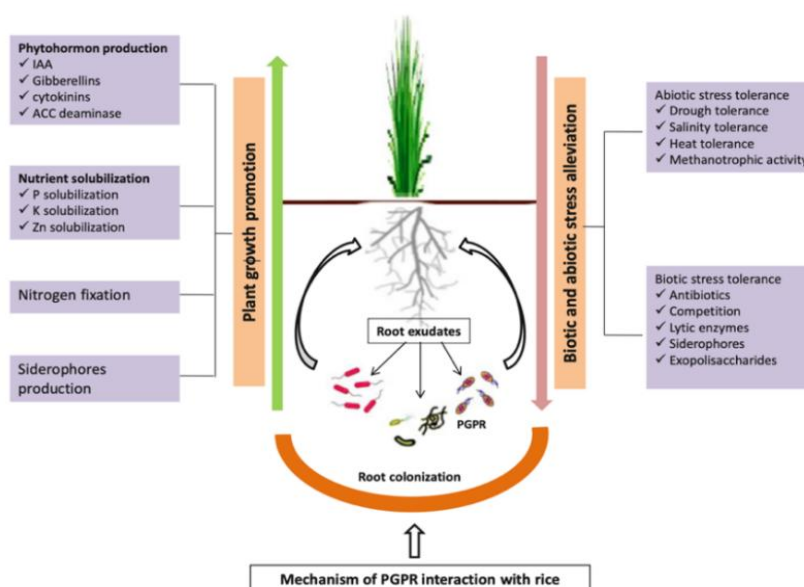
No	Type	Mechanism
1	Direct antagonism	Parasitism (Symbiotic interaction between two phylogenetically unrelated organisms) Hyperparasitism (Parasites using other parasites as their host) Commensalism (one partner benefits while other is neither benefited nor harmed)
2	Indirect antagonism	Competition (interaction harmful to both the partners)  SAR (systemic acquired resistance) ISR (Induced systemic resistance)
3	Mixed path antagonism	Antibiosis, lytic enzymes production, siderophore production, organic and anorganic volatile substances

Source: Hussain et al. (2022)

Several reports have explained that the direct antagonism was carried out by PGPM in controlling pests and diseases of cultivated plants. Parasitism is a type of interaction between two organisms that are phylogenetically unrelated. For example, *Trichoderma* spp., has parasitic activity against various phytopathogens such as *Botrytis cinerea*, *Rhizoctonia solani*, *Pythium* spp., *Sclerotium rolfsii*, *Sclerotinia sclerotiorum*, and *Fusarium* spp. (Waghunde et al. 2016). As it is generally known, *Rhizoctonia solani* causes several plant diseases such as rice blight and black scab disease in potatoes, so *Trichoderma* spp. can potentially be used as a BCA for all these diseases (Jia et al. 2013; Rahman et al. 2014). Some other examples related to hyperparasites and mycoparasites are powdery mildew pathogens parasitized by several hyperparasites such as *Ampelomyces quisqualis*, *Acrodontium kawahiforme*, *A. alternatum*, *Cladosporium oxysporum*, and *Gliocladium virens*, and viruses causing hypovirulence in

*Cryphonectria parasitica*, an ascomycete causing chestnut blight. (Heydari and Pessarakli 2010; Tjamos et al. 2010).

Dominantly, BCA has a more competent nutrient uptake system than phytopathogens. One is controlling *Fusarium* wilt as the result of carbon competition between pathogenic and nonpathogenic *F. oxysporum* strains (Alabouvette et al. 2009). Fire blight, a disease transmitted by *Erwinia amylovora*, can be suppressed by a closely related saprophytic species, *E. herbicola*, which competes for nutrients on the leaf surface. PGPM producing chemical stimuli can induce persistent variations in plants thereby increasing their capacity to tolerate pathogen infection and induce systemic defense against various pathogens, known as induced resistance, which consists of 2 forms, SAR and ISR. SAR is the innate resistance capacity of plants activated upon exposure to chemical elicitors from nonpathogenic, virulent, or avirulent microbes or artificial chemical stimuli (Gozzo and Faoro 2013). Induction of SAR mediated by the accumulation of chemical stimuli such as salicylic acid (SA) is commonly released after pathogen attack. SA is the first chemical signal inducing the production of pathogenesis-related (PR) proteins, for example chitinase,  $\beta$ -1,3 glucanase. The PR gene encodes chitinase and  $\beta$ -1,3-glucanase which play an important role in reducing or preventing pathogen colonization (Sudisha et al. 2012). SAR has been proven to be able to fight several pathogens and pests. Promising results on tomato plants using several strains of *B. firmus* against the root-knot nematode *Meloidogyne incognita* (Kofold and White) (Chitwood) (Tylenchida: Heteroderidae). A serine protease (Sep1) with nematicidal activity was found to be produced by *B. firmus* strains (geng et al. 2016). On the other hand, experimental application of *B. subtilis* decreased intestinal enzyme activity and caused changes in peritrophic membranes and epithelial cells in *Spodoptera litura* (Fabricius) (Lepidoptera: Noctuidae) (Chandrasekaran et al., 2014)



**Figure 1.** Mechanism of PGP interaction with rice (Hussain et al. 2022)

Meanwhile, ISR naturally exists in plants and is generally associated with stimulation by non-pathogenic rhizobacteria originating from plants (Pieterse and Van Wees 2015). ISR does not depend on the SA-mediated pathway and PR proteins are not involved. The mechanism of ISR is plant specific and depends on the plant genotype. Application of nonpathogenic PGP induces ISR facilitated by the production of phytohormones (i.e., jasmonic acid and ethylene). PGP induces ISR in several plant organs used to combat various environmental stressors. The plant defense system produces a large number of enzymes involved in plant defense, such as polyphenol oxidase,  $\beta$ -1, 3-glucanase, chitinase, phenylalanine ammonia lyase, peroxidase, etc. Although ISR does not directly fight specific pathogens, it plays an important role in plant defense. So the mechanism can be used to control various diseases in plants (Kamal et al. 2014). For example, ISR activity induced by the use of three bacteria as *Bacillus halotolerans*, *Massilia alkalitolerans*, and *Bacillus aryabhatai* was shown to be effective reduced the growth of *P. ultimum* and *R. solani* in tomato and maize (Abdelaziz et al, 2023).

This mixed mechanism, sometimes almost similar to PGPR and PGPF, induces resistance to insect pests through the synthesis of phytohormones, increases uptake of phosphorus and nitrogen, and increases solubility of iron and minerals through chelation growth. Systemic Resistance by rhizobacteria resembles systemic acquired resistance induced in pathogens including nematodes, insects, bacteria, fungi and viral pathogens ([Hossain et al., 2016](#); [Rashid and Chung, 2017](#)). PGPR against pest with various mode of action, such as degradation of gut epithelium gut, tissue, antimicrobial activity and toxicity, action on neuro-system, antifeeding, pathogenesis, septicaemia and cellular immunity suppression (Ruiu, 2020). Rhizobacterial inoculation also induces phytohormonal signals or regulators such as, jasmonic acid, ethylene, indole-3-acetic acid (IAA), gibberellic acid, cytokinin and 1-amino-cyclopropane-1-carboxylate (ACC) deaminase (Pieterse et al., 2014; [Vejan et al., 2016](#); [Gouda et al., 2018](#)) that regulates plant secondary metabolite concentrations. Several strains of *Bacillus* and *Pseudomonas* are used as biofertilizer inoculants which have direct and indirect effects on insect pest resistance. And suppresses soil-borne pathogens through the production of siderophores and antimicrobial metabolites (Kennedy et al. 2014; Hussain et al, 2016). Aisyah et al. (2015) stated that plants given PGPR increased phenol content and affected instar development, feeding activity and egg laying locations of *P. xylostella*. Microbial induction in strawberry plants increases polyphenol oxidase, flavonoids and anthocyanins having a negative impact on *T. urticae* reproduction (Mouden et al. 2021).

PGPR also inhibits plant pests through the release of various volatile and diffuse metabolites (e.g., pyoluteorin and pyrrolnitrin) that are toxic to insect pests thereby reducing their populations as observed (Naaem et al. 2018). Direct mechanisms include the production of plant hormones, solubilization of phosphates, and increased uptake of iron. Indirect effects include antibiotics production, nutritional competition, parasitism, pathogen toxin inhibition, and induced resistance (Elnahal et al., 2022). In addition, PGPR helps increase phosphorus and nitrogen uptake and increases the activity of indole acetic acid which helps wheat plants absorb. In addition, PGPR can compete with other bacteria by colonizing quickly and accumulating a greater supply of nutrients, thereby preventing the growth of other organisms such as pathogens

(Salomon et al., 2017; Abd El-Mageed et al., 2020). Bacteriocins, antibacterial proteins, and enzymes are examples of antimicrobial peptides produced by PGPM that can inhibit metabolic processes or growth activities of pathogens, most of which are specific to certain strains, can target ribosomal RNA (rRNA), change membrane structure, and damage pathogen cell walls (Nazari and Smith, 2020). Furthermore, the siderophores produced by PGPM are special iron-chelating agents that inhibit phytopathogens from gaining access to iron and are also able to maintain plant health, especially in iron-deficient environments (Shen et al., 2013; Radzki et al., 2013).

## **2. Opportunities and Challenges in Utilizing Plant Growth Promoting Microorganism (PGPM) as Biological Control Agents for Paddy in Indonesia**

In recent decades, interest in beneficial microorganisms in rice has increased due to their potential use as plant growth regulators and various pest and disease control measures (Jha and Subramanian, 2012). The results showed that all PGPRs promoted rice growth (grain yield increased by 10.50-51.30% in greenhouse conditions and 4.83-9.16% in paddy fields) and reduced As damage to rice. In particular, S10 reduced As accumulation in brown rice under greenhouse (3.50-26.01%) and paddy field (9.26-10.50%) conditions by significantly reducing the available As concentration in the rhizosphere soil, especially in Dabaoshan-B land (34.00%). In contrast, under greenhouse conditions, strains S6 and S7 increased As concentrations by 6.10-20.10% and 2.14-14.60%, respectively. Our results show that PGPR inoculation can be used to reduce As accumulation and promote rice growth in As-contaminated rice fields. However, because the impact of PGPR varies by strain and depends on environmental factors, careful selection of strains and environmental conditions as well as initial testing will be essential before applying PGPR to As-contaminated rice fields. PGPR in rice increased grain yield by 51.3% and 9.16% in greenhouse and paddy fields respectively, in arsenic-accumulating soils in China (Aw et al., 2020). The highest grain yield in both wheat varieties due to the application of PGPR may be due to better grain per stalk and productive tillers. It is well known that productive tillering and grain per stalk are important traits contributing to the outcomes resulting in better grain yield in this study.

Various PGPM research has been carried out in Indonesia, especially on rice plants (table 2) showing a positive trend. The use of PGPM needs to be massively socialized to farmers, so that farmers slowly switch to environmentally friendly cultivation. Apart from that, farmers need to be given evidence that in the long term the use of PGPM will reduce the chemical inputs currently used. Alawiyah and Cahyono (2018) stated that farmers understand that the use of biological agents will reduce the use of fertilizers and pesticides, but many farmers do not understand in detail how to apply the use of biological agents on land. Demonstration plots and FFD (Farmer Field Day) are methods that can be used to introduce the use of PGPM in plant cultivation. Rachmawatie et al. (2022) carried out sharing, direct practice, and on-farm monitoring of farmer groups that utilize *Trichoderma* sp. Continuous recognition efforts accompanied by assistance and concrete evidence in the field will change farmers' paradigm in sustainable cultivation practices.

Biological control through the use of PGPM is the most promising effort for sustainable agriculture because the agricultural pest control approach is proven to be environmentally friendly. The use of live microorganisms to reduce pest and disease populations in a conservative, reliable, and ecologically friendly manner. In developed countries, biological control is considered a sustainable, cheaper and safer way of managing pest control; thus, it benefits farmers as producers and consumers compared to synthetic (chemical) pest management (El-Saadony et al. 2022). Considering that Indonesia has high microbial biodiversity, of course, this opportunity needs to be optimized to support environmentally friendly cultivation practices, especially paddy.

**Table 2.** PGPM research on paddy in Indonesia

Kind of Paddy	PGPM	Treatment	Reference
<b>Bacteria</b>			
Beras merah	<i>Serratia marcescens</i>	Seed treatment can increase can increase chlorophyll, panicles number, tillers number, weight of 1000 seeds, yield of dry seeds harvested and reduce the intensity of leaf spot disease	Nurmala et al, 2021
Inpari Unsoed 79 Agritan	Bakteri diazotrof	Seed treatment can reduce disease intensity by up to 70%.	Isnaeni et al. 2021
IR 64, Cisantana, Inpari Unsoed 79 Agritan	Rizobakter	Seed treatment was able to induce resistance to <i>Xanthomonas oryzae</i> pv. <i>Oryzae</i> and can increase growth and crop yields	Khaeruni et al. 2014
LokaL Kamba	<i>Bacillus</i> sp	Seed treatment can increase the number of productive tillers, the number of grains per panicle, and the total number of grains.	Sudewi, 2020.
Inpari 42	<i>Bacillus amyloliquefaciens</i>		
Ciherang	Kombinasi <i>Pseudomonas aruginosa</i> dan <i>Bacillus cereus</i>	Spraying plants every afternoon can increase grain weight and resistance to blast disease ( <i>Pyricularia grisea</i> ) attacks	Jannah, 2016
IPB 3S	<i>Bacillus polumixa</i> dan <i>Pseudomonas fluorescens</i>	Soaking seeds for 12 hours can reduce fungal infections and increase rice seed germination	Andini dan Tondok, 2020
<b>Fungi</b>			

Inpari 13	Mikoriza	Applied to planting media, it can increase plant height, number of panicles, and plant carbohydrate levels.	Mustaqimah, et al. 2019
-	<i>Trichoderma</i> sp.	Soaking seeds and spraying can reduce the severity of blast disease in rice, increase plant height, number of tillers and grain weight.	Hidayat, et al. 2014 Yanti, et al. 2021,
Kuriak Kusuik	<i>Trichoderma spp</i>	Soaking seeds can increase the seed vigor index.	Aldo dan Anhar, 2021
Ciherang	<i>Nigrospora</i> sp	Soaking seeds and spraying plants can reduce the severity of leaf blast disease	Hariyanti, et al. 2022
Ciherang	<i>Aspergillus niger</i> , <i>Penicillium</i> sp, dan <i>Trichoderma viridae</i>	Mycelium mixed into the planting medium can increase the growth of rice plants in saline soil	Subowo, 2015
Beras merah	<i>Aspergillus niger</i> dan <i>Aspergillus oruzae</i>	Can increase germination capacity, resist water stress and increase production.	Yustisia, 2020
PAK-TIWI 1	<i>Monosporium</i> sp, <i>Curvularia</i> sp dan <i>Nigrospora</i> sp	Soaking seeds can increase the physiological activity of plants and can increase the production of the hormone IAA.	Rachmawati, 2022
Padi	<i>Bauveria bassiana</i>	Increases mortality of brown planthoppers	Aristyawan et. al. 2020

Apart from that, there are various challenges in applying PGPM as a pest and disease control agent. Successful colonization of root tissue and/or rhizosphere is an important component for PGPM to become an effective BCA. However, the performance of inoculated PGPM can vary, depending on survival rates in the soil, plant compatibility, interactions with other local microbial species, and environmental factors (Vejan et al., 2016). PGPR performance is generally assessed based on geographic region, soil type, species host plants, and various environmental conditions (El-Saadony et al. 2022). In addition, in vitro PGPM testing is important to carry out before trials in the greenhouse and/or field (Bashan et al., 2014). This testing stage will guarantee the suitability of PGPM for increasing production, managing pests, diseases, and resilience to climate change conditions.

Furthermore, PGPM stability is also influenced by various technical factors such as method, formulation, transportation, and storage conditions. To achieve the survival rate of PGPM as



BCA, it is necessary to have the formulation technology used and the shelf life of the microbes measured and optimize the production of the targeted microbial types, and if it has already been in the large-scale production stage, the costs required are not too expensive (Lobo et al., 2019; Carrasco-Espinosa et al., 2015; Zhang et al., 2019) (Kang et al., 2017). Due to the variety of factors influencing PGPM performance, time and academic-industry collaboration is needed to ensure its usefulness.

## CONCLUSIONS

Plant Growth Promoting Microbial (PGPM) is considered capable of increasing rice production amidst uncertain climate conditions and excessive use of pesticides. Plant Growth Promoting Rhizobacteria (PGPR) and Fungi (PGPF) have been proven effective in suppressing plant diseases and controlling pests and pathogens with various mechanisms and inducing immune responses in plants. As biofertilizers and biopesticides, PGPR and PGPF are considered an attractive and economically viable approach to support sustainable agriculture.

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