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Research Article

Dipping Seedling's Rice Root with Indigenous Microorganisms from *Mimosa invisa* to Control Blast Disease and Increased Rice Production in Purwoasri Village, Jember

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ABSTRACT

Rice blast disease has become endemic in Purwoasri Village, Gumukmas District, Jember Regency since 2018; this was due to the use of the Ciherang variety planted continuously. The study aimed to use the indigenous microorganisms (IM) suspension made by farmers from *Pos Pelayanan Agens Hayati* (PPAH, Biological Agent Service Post) in Ambulu District as the control agent for blast pathogens and increased rice production. The roots of rice seeds were dipped in IM suspension at a concentration of 50 mL/L for 2 hours to penetrate to roots. Seedlings with IM were planted in a block of 250 m^2, and control was also planted in the same block size. In the IM-treated block, the incidence and severity of blast disease decreased compared to the control. The root treated by IM dipping was also increased the plant growth, including root architecture, root length, root volume, and the number of tillers/plant; furthermore, it improved the rice production. It was found that IM suspension contained pseudomonad fluorescent and *Bacillus* sp. that belong to Plant Growth Promoting Rhizobacteria (PGPR) group.

Keywords: blast disease; IM; rice seedling root

INTRODUCTION

Rice plants in Purwoasri Village, Gumukmas District, Jember Regency have been infected with blast disease caused by *Pyricularia oryzae*. The disease has been endemic since 2018, and this was due to the continuous cultivation of Ciherang varieties that are known susceptible to blast disease (Suganda *et al.*, 2016). One of the farmers in Purwoasri said that rice yields declined up to 20% in the second planting season in 2020. This fact was supported by Suganda *et al.* (2016), in endemic areas in Sukabumi, losses of Ciherang production due to blast disease reached 3.65 tons/ha.

In 2012–2015, another rice disease, bacterial leaf blight also found in Ambulu, Southern Jember. PPAH farmers in Ambulu have made the suspension of indigenous microorganisms (IM) isolated from *Mimosa invisa* roots to control this disease. The purpose of using IM was to increase rice production and control bacterial leaf blight previously infecting this area. According to Yuliani and Rahayu (2016); Agustina and Syamsiah (2018), *M. invisa* roots are occupied by many bacteria can interact with plants to help nutrient absorption. Plant roots also release exudates into the rhizosphere, which attract beneficial bacteria to colonize the rhizosphere (Vives-Peris *et al.*, 2020). Based on this, the study aimed to determine the effect of the roots dipping of rice seedling in IM suspension (from PPAH Ambulu farmers) on the reduction of rice blast severity and increased rice production in Purwoasri.

MATERIALS AND METHODS

Identification of Bacteria Contained in Indigenous Microorganisms

The isolated bacteria in IM were first grown on NA media, then tested for Gram with 3% KOH solution to determine whether the bacteria were Gram-positive or negative. A hypersensitivity test on tobacco leaves was carried out to determine whether the bacteria belonged to the pathogenic or non-pathogenic. Then, bacteria contained in IM were identified by selective media, Hicrome Bacillus Agar (Alippi, 2019), the growth media for the *Bacillus* group, and King's B media, the growth media for the pseudomonad fluorescent group.

Nursery Preparation

This research area was 500 m^2 which was divided into two blocks. Block P1: dipped root plants with IM and P0 as control. Each block was separated with a dike field. Nine sample plots in each block were taken by diagonal random sampling. One plot was 5 m^2 in size, and 10 rice tillers were sampled randomly from each plot. A total of 90 tillers were collected from each block. Data were analyzed by T-test at a 95% confidence level. IM was obtained from farmers who were made traditionally in Ambulu District. The roots of 14 days old rice seedlings were dipped for 2 hours in IM suspension with a concentration of 50 mL/L. *P. oryzae* was inoculated naturally.

The Incidence and Severity Disease

The incidence of disease was calculated with the formula:

$$I = \frac{n}{N} \times 100\ \%$$

I = Incidence of disease, n = number of infected plants, and N = total plants observed. Based on the disease incidence, the criteria of resistance level was very resistant if 1% plants sick, resistant: 1.1%–10% plants sick, moderate: 10.1%–20% plants sick, susceptible: 20.1–50 % plants sick, and very susceptible > 50.1% plants sick (*Kementerian Pertanian* [Kementan], 2018).

The severity of the disease was calculated with the formula:

$$DS = \frac{\sum (n_i \times v_i)}{Z \times N} \times 100\%$$

DS = disease severity, n_i = number of plants per tillers with damage scale value of i, v_i = damage scale value of i-sample, N = number of plants per sample plot observed, Z = highest damage scale value. The disease severity score according to Kementan (2018) was, 0 = no infection, score 1 = needle point spot but not elliptical yet, score 3 = elliptical spots, necrotic size 2–20 mm and 2% leaves infected, score 5 = >2%–10% leaves infected, score 7 = >10%–50% leaves infected and score 9 = > 50%–100% leaves infected or *puso* plants.

Plant Height, Number of Tillers, Root Length, Root Architecture, and Root Volume

Plant height was measured with a meter cloth every week from 14 days after planting (dap) until the vegetative phase ended. The number of tillers was observed at 14 to 49 dap. Three samples of rice roots (49 daps) were taken randomly from each plot, then washed and photographed for their root architecture. Then the root length was measured with a ruler. Root volume = final water volume – initial water volume. The measurement was taken in the *Laboratorium Pengamatan Hama Penyakit Tanaman Pangan dan Hortikultura* (PHPTPH, Laboratory Observation of Pests and Diseases of Food Crops and Horticulture), Tanggul, Jember.

Weight of 1000 Grains (g) and Productivity (tons/ha)

The weight of 1000 grains was calculated per plot from plant samples taken randomly. The productivity was calculated by weighing the yield with IM and control, then converted into ton/hectares.

RESULTS AND DISCUSSION

Identification Bacteria in Indigenous Microorganisms

Indigenous microorganisms (IM) isolated from roots of *M. invisa* were identified as pseudomonad fluorescent group and *Bacillus* sp. When the suspension was grown on NA media, it found several different bacterial colonies; two dominant colonies were Gram-negative and non-pathogenic. After the selection, the two colonies were then planted on different media. When observed under UV light, bacteria grown in King's B media had fine colonies and yellowish-green fluorescent (Figure 1 A). It was suspected that this bacteria belongs to the *pseudomonad fluorescent* group (Kirihio *et al.*, 2017). Bacteria grown on Hicrome Bacillus Agar media have a blue color, round shape, and smooth surfaces. According to Alippi *et al.* (2019), those characteristic bacteria belong to the *Bacillus* sp. group (Figure 1B).

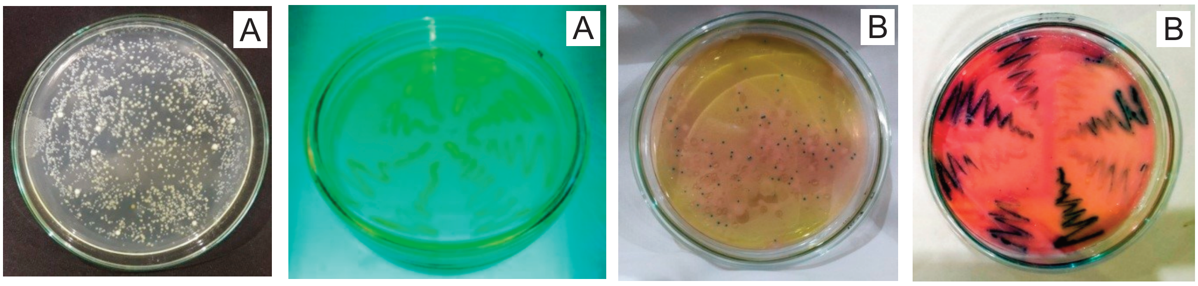

Figure 1. Bacterial isolates in King's B and Hicrome Bacillus Agar Media; (A) Bacteria *pseudomonas fluorescent*, (B) were suspected *Bacillus* sp.

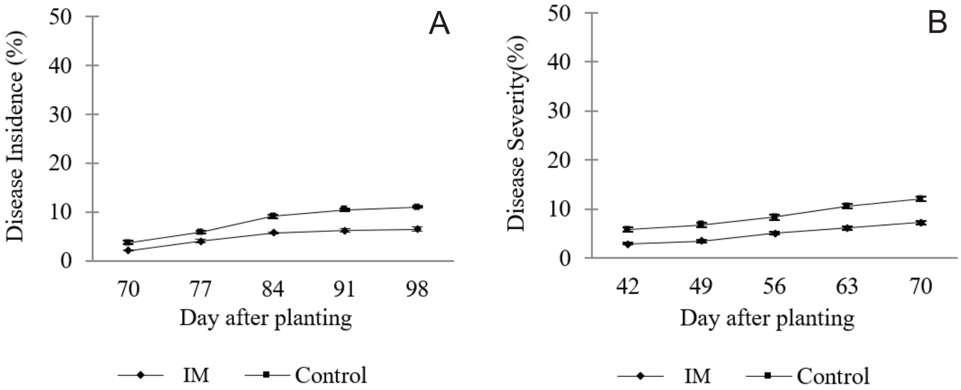

Figure 2. Effect of indigenous microorganisms (IM) on incidence and severity of blast disease; (A) incidence of disease, (B) disease severity

Symptoms of Blast

Infection leaves naturally produced small necrotic spots symptoms on 42 daps samples, and the characteristics were similar to *P. oryzae* infection. Symptoms of neck blast appeared when the plant was 70 dap; at the base of panicles and the rice bran were easily broken, causing rice grains to fall off. These symptoms of neck blast were minor at 42–56 days old, making them difficult to observe and determine disease severity.

Disease Incidence and Disease Severity

Figure 2 showed that the application of root dipping in IM suspension reduced the incidence and severity of disease every week. At 98 days after dipping in IM, the average incidence rate was 6.51%, while the control was 11.04% (T-calculated > T-table, Figure 2A). The blast severity on IM-treated plants at 70 dap was 7.25%, and the control was 12.07% (T-calculated > T-table, Figure 2B). The bacteria in IM were also categorized as biological agents from the Plant Growth Promoting Rhizobacteria (PGPR) group (Syarifah, 2020) that improve plant growth and controlled pathogens (Agustina & Syamsiah, 2018). Dipping roots in IM suspension reduced the incidence and severity of blast disease compared to the control. This IM inhibited the development of disease pathogens in Ciherang varieties (Ukhra *et al.,* 2016). The bacteria in IM suspension acted as antagonist agents that increased plant defense systems against infection of plant pathogens (Syarifah, 2020).

Plant Height (cm) and Number of Tillers

The average plant height of IM-treated plants was not different from the control. At 63 days, the plant height of IM-treated samples was 111.60 cm, while the control was 99.24 cm (T-calculated = T-table, Figure 3A). The average number of tillers per block of plants treated with IM was also higher than the control. The number of tillers at 50 daps was 30, while the control was 22 (T-calculated > T-table, Figure 3B). The use of IM caused rice roots to grow better and longer, so more beneficial microorganisms prefer to grow toward these roots. As a result, the nutrient absorption improved and increased the growth and number of tillers.

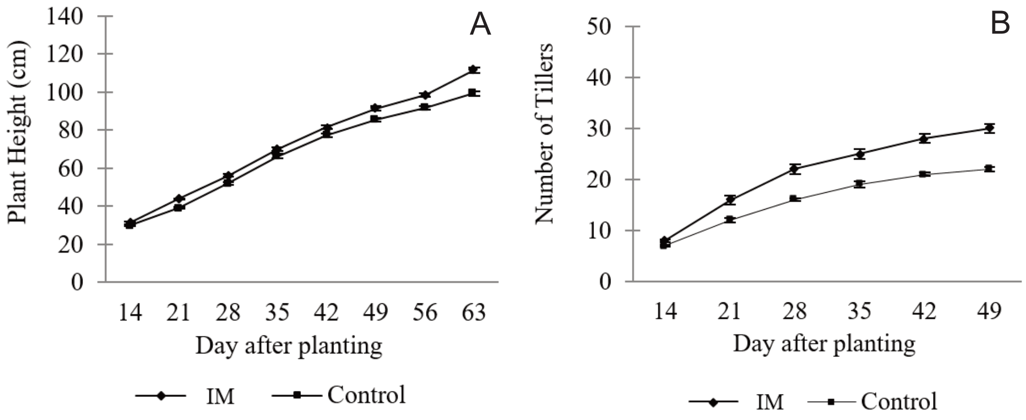

Figure 3. Effect of indigenous microorganisms (IM) on plant growth; (A) plant height (cm), (B) number of rice tillers

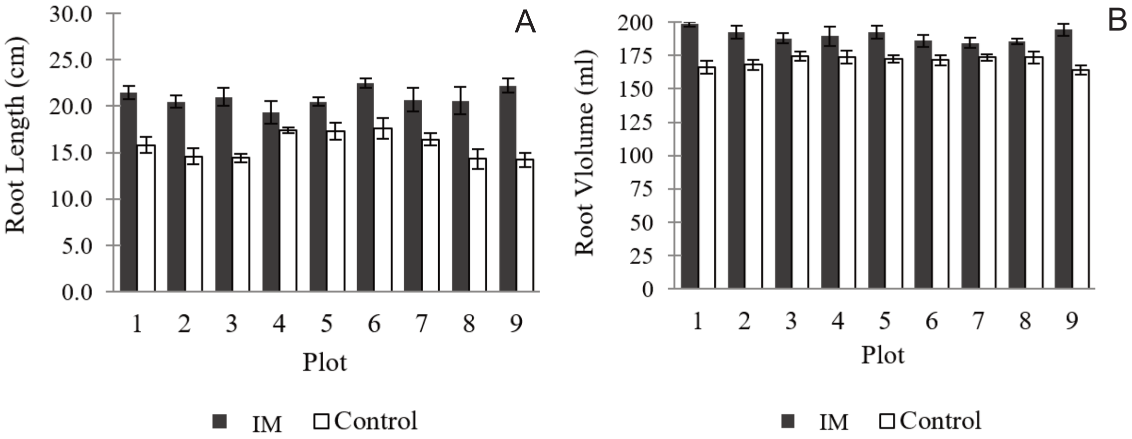

Figure 4. Effect of indigenous microorganisms (IM) on root growth; (A) root tillers length (cm), (B) rice root volume (ml) in each plot at the age of 63 days after planting (dap)

Maybe, this was also because there was an effect of PGPR which acts as a biofertilizer to the plant. This result was in line with Anhar *et al.* (2011) that pseudomonad fluorescent isolates improved the number of tillers.

Root Length (cm), Root Architecture and Root Volume (ml)

Figure 4 showed that IM improved root architecture, roots elongation, and increased root volume. The root architecture of plants treated with IM appeared denser and longer, and the root volume was better than the control (T-calculated > T-table, Figure 4B). As Agustina and Syamsiah (2018) found, using IM from *Mimosa pudica* to soak the seeds also increased root length and the growth of Cianjur rice at 30 dap. IM contains macro and micronutrients (Handayani *et al.,* 2015), so the root growth was better than the control. This result also supposed that bacteria in IM stimulated the formation of growth hormones in the meristematic area of the root tip to increase the root cell elongation (Anhar *et al.*, 2011). As a result, root growth becomes denser and longer, improving nutrient absorption. These bacteria also play a role in dissolving P to be available for plant absorption, as in line with the result of Anhar *et al.'s* (2011) research. The longer and denser roots are correlated with nutrients and water absorption area. Thus, these also affected the number of productive tillers (Kusumawati & Istiqomah, 2020).

Weight of 1000 Rice Grain (g) and Productivity (tons/ha)

The 1000 grains were taken from each plot randomly. The weight of 1000 grains from IM-treated plants was not significantly different from the control (T-calculated=T-table, Figure 5A).

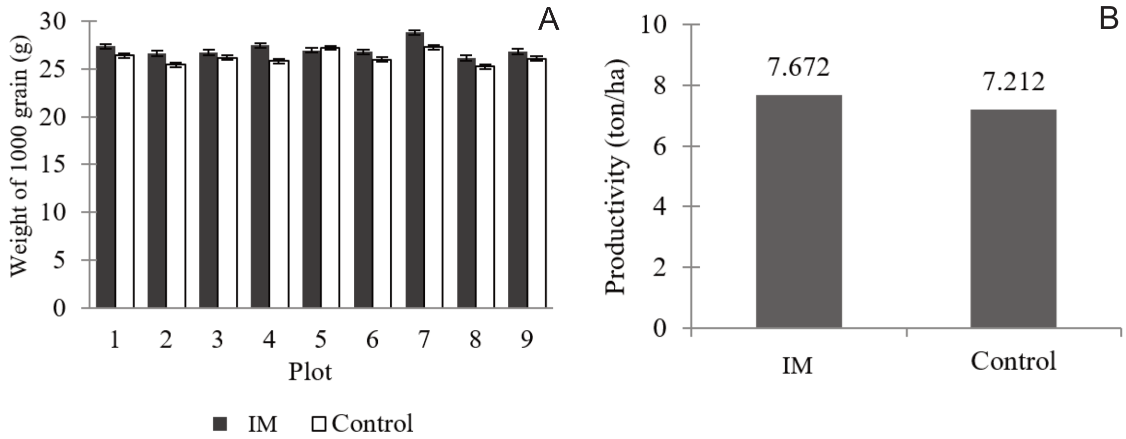

Figure 5. Effect of indigenous microorganisms (IM) on yield; (A) mean weight of 1000 (g) of grain per plot, (B) rice productivity (ton/ha)

Figure 5B showed the productivity of rice yield with IM was high, 7,672 tons/hectare, while the control was 7,212 tons/hectare (Figure 5B). The difference in productivity between the rice yield with IM and the control (0.460 tons/hectare) indicated that IM's application increased rice productivity, as Kusumawati and Istiqomah (2020) stated.

CONCLUSION

The IM suspension contains nutrients and bacteria of pseudomonad fluorescent and Bacillus groups commonly used as PGPR. The roots of rice seedlings dipped in IM suspension for 2 hours effectively reduced the incidence and severity of blast disease. IM suspension also increased the growth of rice plants and their productivity.

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