Seed treatment using biofertilizer to improve plant growth and yield performances of upland rice cultivars under various planting densities

A Saryoko¹, S Kusumawati¹ and A Pohan²

¹Banten Assessment Institute for Agricultural Technology, Banten, Indonesia
²Directorate General of Food Crops, Ministry of Agriculture, Jakarta, Indonesia

E-mail: andysaryoko@pertanian.go.id

Abstract. Producing upland rice through the use of high yielding cultivars and better agronomical practices is a promising way to improve rice production in Indonesia. The objectives of the study were to evaluate the effect of seed treatment on growth and yield of two upland rice cultivars under two planting densities, and to identify its related traits. A field experiment was conducted in Banten Province, Indonesia. Two rice cultivars, Inpago 8 and Inpago 9 were treated with biofertilizer and were arranged into double-row system with the density of 21 plants m⁻² or 33 plants m⁻². Research results showed that seed treatment significantly improved plant growth and seed yield for both cultivars due to greater panicle number and seed number per panicle. Biofertilizer contributed to a greater amount of total biomass as well as harvest index (HI). Increasing planting density from 21 to 33 plants per m⁻² improved panicle number that affected seed yield increase. The Inpago 9 performed superior in seed yield as compared to Inpago 8 due to better seed number per panicle and HI. High yielding upland rice cultivars in combination with biofertilizer and appropriate planting density would be an approach to improve upland rice production.

1. Introduction

Rice is one of the top leading staple food crops. In general, there are two types of rice cultivation, lowland and upland rice system [1, 2]. Rice is mostly produced from the lowland rice system. Recently, function transformation from agricultural to non-agricultural purpose of lowland becomes a major concern on rice production. On the other side, the dryland area is highly potential to be explored in terms of rice production improvement [3].

The use of high-yielding upland rice cultivars with other agronomical practices, such as planting density arrangement and biofertilizer application as seed treatments, is expected to improve rice production in upland rice system. Indonesian Rice Research Centre – IAARD, has released new high-yielding upland rice cultivars that can produce more than seven tons of rice per ha with an average of four to five t ha⁻¹ [4]. Saryoko et al. [3] reported that Inpago 9 is one of high yielding adaptive cultivars in Banten Province, Indonesia which potentially reach 8.2 t ha⁻¹. One promising candidate for rice seed treatments is the use of a biofertilizer which consists of several micro-organism: Methylobacterium sp., Bacillus cereus, and Azotobacter sp. This type of biofertilizer is claimed to give a positive contribution on plant growth and yield for some crops such as soybean, low land rice and chili.
The objectives of the study were to evaluate the effect of seed treatment using biofertilizer on growth and yield performances of two high yielding upland rice cultivars under two different planting densities, and to identify its related traits.

2. Materials and methods

2.1. Cultivar and agronomical practices
A field experiment with a randomized complete block design with two replications was conducted at Panggarangan Sub-district, Lebak District, Banten Province, Indonesia (Lat. 6.8°S, Long. 106.2°E). On 20 September 2018 seeds of two upland rice cultivars, Inpago 8 (V1) and Inpago 9 (V2) were directly sown immediately after treated without biofertilizer (P1) and with biofertilizer with the doze of 16 g per kg seed (P2). The plants were arranged into double-row system with 25 x 12.5 x 50 cm with the density of 21 plants m$^{-2}$ as normal planting density (J1) and 20 x 10 x 40 cm with the density of 33 plants m$^{-2}$ as dense planting density (J2). The treatments were placed into a 4 x 5 m$^2$ plots. The soil was fertilized with 11.5 g m$^{-2}$, 5.4 g m$^{-2}$ and 4.5 g m$^{-2}$ of N, P$_2$O$_5$ and K$_2$O, respectively. The N fertilizer in the form of urea was applied three times at 10, 25 and 35 days after sowing (DAS) as much as 50, 25 and 25 % respectively. The P$_2$O$_5$ and K$_2$O in the form of SP36 and KCl, respectively, were applied at 10 DAS. Regional recommended management programs were employed for irrigation, weed and pest control to optimize growth condition [3].

2.2. Agronomical traits, plant growth and yield components
The growth of plant was observed every one or two weeks within four weeks after planting (WAP) to 9 WAP. Canopy size and development were measured at 2, 3, 6 and 7 WAP using a digital imaging technique [5-8] using ImageJ (National Institute Health, USA) with some modifications, and expressed as a percentage of soil coverage by the rice canopy. For this purpose, plots were maintained as weed-free as possible during the growth stage. Leaf area (LA) was measured at 6 WAP. Two sample plants per plot were taken and leaves from were separated as fast as possible to avoid leaf withered, and the digital image was taken. Digital image technique analysis was performed to calculate leaf area comparing the area with specific leaf dry weight.

At the maturity stage, a 1.2 m$^2$ for J1 or 1.5 m$^2$ for J2 sub plots were created for each plot, and plant samples were taken. Four plants per sub plot were randomly harvested, and yield and yield components were observed. Panicle number per plant was counted, and panicle length and seed number per panicle were observed from each per plant sample. Seed number was counted and weighted, and 1000 seed weight (1,000SW) and seed moisture content was measured. Grain yield per plant was calculated as a multiplication of grain number, 1,000SW and seed moisture content was measured. Grain yield per plant was calculated as a multiplication of grain number, 1,000SW, panicle number per plant at 14% moisture content. The remaining plant organs (leaves and straws) were dried at 80° C for 48 h. Harvest index (HI) was calculated as a ratio of dry grain yield (Y) and total above-ground biomass at maturity (TDW), as follows:

\[ HI = \frac{Y}{TDW} \] (1)

2.3. Data Analysis
The variability and interaction among treatments were evaluated using an analysis of variance (ANOVA) and were followed by Tukey’s test using MINITAB 16.

3. Results and discussion

3.1. Plant growth and canopy development
The plant height of Inpago 8 and Inpago 9 under two different planting densities were shown in figure 1. There was a significant different in plant height caused by planting density. In general, cultivars grown under high planting density (J2) tend to grow taller as compared to them grown under normal density
(J1) specifically at the later stages from 6 to 9 WAP. Under dense condition, plants tend to grow taller as a consequence of light competition [9]. Between cultivars, Inpago 9 was taller under each planting density as compared to Inpago 8. There was no significant different found in plant height caused by biofertilizer application.

![Figure 1](image1.png)

**Figure 1.** Growth of plant height of upland rice cultivar Inpago 8 (V1) and Inpago 9 (V2) without biofertilizer (P1) and with biofertilizer (P2) under normal (J1) and dense planting density (J2).

The variation of LA measured at 6 WAP was provided in figure 2. Across all treatments, the Inpago 9 significantly greater in LA as compared to Inpago 8. Significant different was also found in LA between normal and dense planting density. All cultivars tend to produce greater LA under the dense conditions only when it was treated without biofertilizer (P1). The leaf area tended to be smaller under dense condition when it was treated with the biofertilizer. From this result, a normal planting density (J1) in combination with biofertilizer application (P2) could improve the leaf area of Inpago 8 and Inpago 9.

![Figure 2](image2.png)

**Figure 2.** Leaf area at 6 WAP of upland rice cultivar Inpago 8 (V1) and Inpago 9 (V2) without biofertilizer (P1) and with biofertilizer (P2) under normal (J1) and dense planting density (J2).

The canopy size was expressed as canopy coverage. The growth of canopy size was provided in figure 3. The canopy size between cultivar was significantly different specifically at 3 WAP under dense condition (J2). At 3 WAP, Inpago 9 showed greater canopy size as compared to Inpago 8. The canopy
size of Inpago 9, however, tends to be similar with that of Inpago 8 at earlier or later stages (figure 3). There was no significant different found in canopy size caused by biofertilizer application. Besides, under dense condition plants tend to produce larger canopy size from early to late growth stage as well.

![Figure 3](image)

**Figure 3.** Growth of canopy size of upland rice cultivar Inpago 8 (V1) and Inpago 9 (V2) without biofertilizer (P1) and with biofertilizer (P2) under normal (J1) and dense planting density (J2).

### 3.2. Yield and yield components

Across all planting densities and cultivars, the seed yield of upland rice cultivars treated with biofertilizer (P2) was significantly higher than that of non-treated (P1). The seed yield of P2 (615 g m\(^{-2}\)) was 28% higher than that of P1 (480 g m\(^{-2}\)) (table 1). Better seed yield of P2 was due to greater biomass produced as well as longer panicle. Also, seed yield was positively correlated with TDW (R\(^2\) = 0.85) (data not shown). Seed treatment using biofertilizer also improved the panicle number and seed number (figure 4). Seed yield was positively correlated with panicle number and seed number with R\(^2\) of 0.58 and 0.97, respectively.

The increase of plant density from 21 plants m\(^{-2}\) (J1) to 33 plants m\(^{-2}\) (J2) significantly increased seed yield (table 1). The greater seed yield in J2 was due to the greater TDW and longer panicle. Across cultivars and seed treatments, the seed size under J2 (30.8 g 1000seed\(^{-1}\)) however, was slightly smaller than that of J1 (31.8 g 1000seed\(^{-1}\)), but it was offset by a longer panicle of J2 (table 1).

The seed yield of Inpago 9 across seed treatments and planting densities was higher than that of Inpago 8. Even though Inpago 8 produced greater biomass, the harvest index of Inpago 8 was smaller (table 1). Across all seed treatments and planting densities, Inpago 8 was longer in panicle and greater in seed size (table 1). However, an Inpago 9 produced greater number of panicles resulted in better seed number (figure 4A and 4B).
Table 1. Yield and yield components of upland rice cultivar Inpago 8 (V1) and Inpago 9 (V2) without biofertilizer (P1) and with biofertilizer (P2) under normal (J1) and dense planting density (J2).

| Treatment | Seed Yield (g m⁻²) | TDWᵃ (g m⁻²) | HIᵇ | Panicle Length (cm) | 1000SWᶜ (g 1,000 seed⁻¹) |
|-----------|---------------------|--------------|-----|---------------------|--------------------------|
| P1        |                     |              |     |                     |                          |
| J1        | V1 441              | 773          | 0.36| 24.9                | 32.2                     |
|           | V2 575              | 993          | 0.37| 21.7                | 32.1                     |
| J2        | V1 398              | 915          | 0.31| 22.8                | 31.6                     |
|           | V2 509              | 703          | 0.42| 21.6                | 29.4                     |
| P2        |                     |              |     |                     |                          |
| J1        | V1 453              | 1,056        | 0.30| 24.5                | 32.5                     |
|           | V2 461              | 609          | 0.43| 22.0                | 30.4                     |
| J2        | V1 686              | 1,192        | 0.36| 25.2                | 31.6                     |
|           | V2 867              | 1,169        | 0.41| 24.7                | 30.7                     |
| P1        | 480ᵇ                | 846ᵇ         | 0.37| 23.7                | 31.3                     |
| P2        | 617ᵃ                | 1,006ᵃ       | 0.38| 24.1                | 31.3                     |
| J1        | 483ᵇ                | 858          | 0.37| 23.3                | 31.8                     |
| J2        | 615ᵃ                | 995          | 0.38| 24.6                | 30.8                     |
| V1        | 494ᵇ                | 984          | 0.33| 24.3                | 32.0ᵇ                    |
| V2        | 603ᵃ                | 868          | 0.41| 23.5                | 30.6ᵇ                    |

Values followed by the same letters in each column are not significantly different as determined by Tukey’s test at the 5% level.

ᵃ TDW = Total Dry Weight
ᵇ HI = Harvest Index
ᶜ 1,000SW = 1,000 Seed Weight

Figure 4. Panicle number (A) and seed number (B) of upland rice cultivar Inpago 8 (V1) and Inpago 9 (V2) without biofertilizer (P1) and with biofertilizer (P2) under normal (J1) and dense planting density (J2).

The above results indicated that seed treatments using biofertilizer (P2) and increasing plant density from 21 to 33 plants m⁻² positively affected seed yield for both cultivars Inpago 8 and Inpago 9 followed by the increased of panicle number and seed number.
4. Conclusions
Biofertilizer application as a seed treatment on upland rice improved plant performance in growth and yield. Increasing planting density improved panicle number and seed number, then affected seed yield increase. The Inpago 9 performed superior compared to Inpago 8. High yielding of upland rice cultivars in combination with biofertilizer and appropriate planting density would be an approach to improve upland rice production.

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