Rice Cultivar Variation in the Growth Response to Inoculation of Free-Living Rhizobacteria

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Our previous studies (Alam et al., 2001) clearly showed that the growth of rice plants was significantly improved by inoculation of mix-cultured rhizobacteria. However, several studies (Garcia and Dobereiner, 1996; Smith et al., 1984) showed that an inappropriate combination of bacteria and crop plant often resulted in a negative effect on the nitrogen accumulation and growth of the host plant. Moreover, a number experiments showed that the extent of the positive effect of the bacteria on nitrogen accumulation and crop growth varied with the species or variety of the host plant (Bouton and Brooks, 1982; Chanway et al., 1988). The main objective of the present study was to investigate the effects of inoculating mix-cultured free-living rhizobacteria onto several rice cultivars including indica and japonica types of Oryza sativa, and Oryza glaberrima, on the nitrogenase activity and growth of the plant.

Materials and Methods

The indica type cultivars of Oryza sativa L. selected were IR 26, Kinungdang Puti (KP) and Boro 8; the japonica type cultivars were Nipponbare, Akihikari and Sasanishiki; and the Oryza glaberrima Steud. cultivars were CG 20, TOG 5810 and CG 14. The plants were grown in pots, each containing 3.5kg soil without addition of any chemical fertilizer in the year of 2000. The experiments were conducted in a completely randomized block design with three replicates for each treatment.

Twenty-nine-day-old seedlings of rice were transplanted to the pots on May 26, 2000. The first inoculation was conducted by immersing the roots of the seedlings into the culture suspension of a mixture of four different bacteria inocula: Azotobacter armeniacus, Azotobacter nigricans, Bacillus sphaericus and B. megaterium for 24h. The density of each bacterium in the mixture of culture suspension was the same (about 0.25 X 10^8 cells mL^-1). The second inoculation was made 30 days after transplanting by injecting 5 mL culture suspension of bacteria into the soil. For the control pots, the same amount of autoclaved suspension was applied to the soil.

Plants were harvested on August 18, 2000, and roots were excised for the measurement of nitrogenase activity. Nitrogenase activity of the excised roots was determined according to the method reported by Alam et al. (2001). Dry matter weight of the aboveground part was determined after drying at 80°C for 72h in an oven with forced air.

Results and Discussion

Inoculation of a mixture of four bacteria species led to the significant increase in nitrogenase activity in most of the cultivars investigated. The average increase in indica and japonica type of Oryza sativa L. was 33.6, 32.4%, respectively, and that in Oryza glaberrima Steud. was 36.4% (Table 1). No significant difference was observed

| Cultivar               | Control | Inoculated |
|-----------------------|---------|------------|
| Oryza sativa (Indica type) |         |            |
| IR 26                 | 34.9    | 50.1*      | (43.6) |
| KP                    | 33.1    | 35.6       | (7.6)  |
| Boro 8                | 31.1    | 46.6*      | (49.8) |
| Average               | 33.0    | 44.1       | (33.6) |
| Oryza sativa (Japonica type) |         |            |
| Nipponbare            | 29.0    | 42.9*      | (44.9) |
| Akihikari             | 27.8    | 38.3*      | (41.4) |
| Sasanishiki           | 24.7    | 26.7       | (8.0)  |
| Average               | 27.2    | 36.0       | (32.4) |
| Oryza glaberrima      |         |            |
| CG 14                 | 36.5    | 56.2*      | (44.0) |
| TOG 5810              | 44.4    | 69.4*      | (56.3) |
| CG 20                 | 51.0    | 54.4       | (6.7)  |
| Average               | 44.0    | 60.0       | (36.4) |

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Table 2. Aboveground dry matter weight in *Oryza sativa* (Indica type), *Oryza sativa* (Japonica type) and *Oryza glaberrima* cultivars inoculated with rhizobacteria. * significantly different at 5% level. Values in parentheses are percent increase in the inoculated plants as compared to the control.

| Cultivar            | Dry matter (g plant⁻¹) | Control | Inoculated | % Increase |
|---------------------|-------------------------|---------|------------|------------|
| *Oryza sativa* (Indica type) |                      |         |            |            |
| IR 26               | 2.8                     | 3.5*    | (25.0)     |            |
| KP                  | 5.1                     | 5.5     | (7.8)      |            |
| Boro 8              | 4.9                     | 5.8*    | (18.4)     |            |
| Average             | 4.3                     | 4.9     | (13.9)     |            |
| *Oryza sativa* (Japonica type) |                   |         |            |            |
| Nipponbare          | 3.3                     | 4.0*    | (21.2)     |            |
| Akihikari           | 5.1                     | 5.8*    | (13.7)     |            |
| Sasanashiki         | 4.0                     | 4.2     | (5.0)      |            |
| Average             | 4.1                     | 4.7     | (14.0)     |            |
| *Oryza glaberrima*  |                        |         |            |            |
| CG 14               | 3.1                     | 4.1*    | (32.3)     |            |
| TOG 5810            | 3.0                     | 4.1*    | (36.7)     |            |
| CG 20               | 3.7                     | 3.9     | (5.4)      |            |
| Average             | 3.3                     | 4.0     | (21.2)     |            |

in the increase of nitrogenase activity due to inoculation between indica type and japonica type cultivars, or between *Oryza sativa* L., and *Oryza glaberrima* Steud.

Dry matter production was influenced by the inoculation with bacteria in all nine cultivars (Table 2). The average increase in dry matter production was 13.9%, and 14.6%, in indica type, and japonica type of *Oryza sativa*, respectively, and that in *Oryza glaberrima* was 21.2%. Thus, the cultivar variation in the relative increase of dry matter weight was well correlated with that in nitrogenase activity. These findings indicate that free-living rhizobacteria inoculation increased the nitrogenase activity, and also the growth activation of the host plant. A similar variation in growth activation was also found in several genotypes of maize (Garcia et al., 1996). Some mechanisms for the growth promotion by inoculating with free-living rhizobacteria have been proposed, including the enhancement of nitrogen fixation, nutrient uptake and exudation of growth regulating substances (Nayak et al., 1986; Barea and Brown, 1976; Lee, 1982), which might be associated with the cultivar variation in plant growth. In the present experiment, two cultivars of *Oryza glaberrima* showed a significantly higher response to the inoculation with mix-cultured bacteria in terms of nitrogenase activity and dry matter production. Therefore, favorable interaction was seen between the free-living rhizobacteria and dry matter production in *Oryza glaberrima* cultivars.

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