

# **Diazotrophic Microbes in Rice: A Boon to Save Nitrogen Fertilizers**

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#### Abstract

The potential contribution of diazotrophic (nitrogen fixing) microbes (DM) to the sustainable cultivation of rice crop to exert their most significant effect on crop growth is by enhanced nitrogen uptake as a biological nitrogen fixer. DMs represent a less significant threat to the environment than the use of inorganic N. Development of superior or novel DM strains by improving above traits can be possible using genetic manipulations. These DM-biotechnologies can be exploited as a low-input, sustainable and environment-friendly technology for the management of plant stresses.

Keywords: Diazotrophic Microbes; Rice; Nitrogen Fertilizers

## Introduction

In recent years, the elementary idea of diazotrophic (nitrogen fixing) microbes (DM) has given us a greater deal to fulfill our demands of high production and minimizes the use of agrochemicals [1,3,7]. Root region of plants are rich with organic matters due to exude of root. The rich organic matter in the rhizosphere region not only facilitates but also harbors a number of bacteria, some of them having novel properties of promoting plant growth, popularly known as plant growth promoting rhizobacteria (PGPR) [2,4,6,8]. Diazotrophic PGPR tends to bacteria and archaea that fix atmospheric nitrogen gas into a more usable form such as ammonia [5].

# Significance of diazotrophic microbes in rice crop

Rice is grown in both wetland and upland cultures, with about 85% of the planet's total area of rice being in flooded wetlands. In upland culture with soils drained by gravity, the roots of rice are more aerobic and  $N_2$  may be assimilated as  $NO_3^-$  as a result of fertilizer addition or mineralization and nitrification of N in organic matter. In addition, aerobic diazotrophs may fix atmospheric  $N_2$ . In wetland culture involving flooded paddies, N is mainly available to rice plants from soil as  $NH_4^+$ , requiring less energy to assimilate into amino acids than  $NO_3^-$ . With growing environment-related concerns, various alternatives are being harnessed to reduce the dependence on N fertilizer for plant nutrients. It is, in this context that the use of the nitrogen-fixing (NF) bacteria in agricultural practices is gaining importance. A NF bacterium can exist freely or in symbiosis and in either case entraps atmospheric nitrogen and converts the unreactive  $N_2$  molecule to  $NH_3$ , a form that is readily utilized by plants. This process is termed as biological nitrogen fixation (BNF) and is catalyzed by the oxygensensitive enzyme nitrogenase, present within the bacteria, by the following reaction:

 $N_2 + 8H^+ + 8e^- + 16ATP$  Nitrogenase  $2NH_3 + H_2 + 16ADP + 16Pi$ 

Diazotrophic plant growth-promoting rhizobacteria (DPGPR) may hold the key to achieve BNF as an evolutionary advantage because of their competitive advantages in a situation of adequate C substrates, but of N-deficiency, allowing their selective enrichment in the rhizosphere. Regarding both cereals and other crops, the need to supply extra N by industrial nitrogen fixation or biological N<sub>2</sub> fixation (BNF) to supplement N released to the available pool by mineralization is expected to depend on the following:

- The amount of soil-N removed in the harvested produce
- Leaching of inorganic-N (e.g. NO<sub>3</sub>) to groundwater
- The magnitude of denitrification of soil-N as N<sub>2</sub>O or N<sub>2</sub>
- The extent and duration of immobilisation of N and its rate of remobilisation in the soil biomass

## Association of diazotrophic microbes in rice

Alcaligenes feacalis (strain A15), was isolated from rice roots and was found to be wide spread in paddy soil. Some legume symbionts may also occur as endophyte in the roots of cereals such as rice (*Oryza sativa, Oryza brevigulata*), wheat and maize. These findings have stimulated research on rice growth promotion by rhizospheric and endophytic bacteria. The isolation of plant growth promoting rhizobia capable of enhancing rice yields under greenhouse/ field condition was a remarkable finding, since rice crop is the most important food crop produced in the world. It is now established that in addition to symbiotic association with legumes, rhizobia may occur as an endophyte in non-legume such as rice and can promote its growth and productivity. It has long been known that rice can form natural association with various  $N_2$  fixing bacteria, both phototrophs and heterotrophs. All or some of these may be responsible for supplying the plants with fixed N. Moreover, in addition to the cultivable diazotrophs associated with rice, a substantial molecular diversity of  $N_2$ -fixing bacteria has been detected in field-grown rice based on retrieval of *nifH* or *nifD* gene fragments from root DNA. However, the contribution of the bacteria externally associated with rice is insufficient to sustain a high yield. It has been suggested that bacteria colonizing the plant interior might interact more closely with the host, with less competition for carbon sources and a more protected environment for  $N_2$  fixation, such as that occurring in the relatively efficient  $N_2$ -fixing symbioses between rhizobia and legumes. In view of the above, a global frontier project which aims to transfer the  $N_2$  fixation capability to rice has begun. One of the approaches toward this goal is the use of natural  $N_2$ -fixing endophytic bacteria associated with rice. Some of the diazotrophs associated with various parts of rice crop and their significant effect in terms of plant growth promotion (PGP) is represented in Table 1.

Diazotroph inoculants	Colonization	Condition of cultivation and % increase
Azoarcus	Roots	Gnotobiotic, 16% (total dry weight)
Burkholderia Photosynthetic Bradyrhizobium Gluconacetobacter diazotrophicus	Roots Rhizosphere Stem/Roots	Greenhouse, 68% (shoot biomass) Gnotobiotic, 20% (total plant biomass)Gnotobiotic, 30% (total dry weight)
Herbaspirillum seropedicae	Roots	Gnotobiotic, 38 – 54% (root biomass), 22 – 50% (shoot bio- mass), 37.6% (plant dry weight),Gnotobiotic , 52 – 112%, 71% (fresh and dry weight)
Serratia Marcescens	Roots/Stem	Gnotobiotic, 23% (total dry weight) Greenhouse, increase
Consortia	Roots	Green house, 17% and 8% (Uptake of P and N)

Table 1: Diazotrophic microbes associated with rice plant and their effects as PGP.

## Conclusion

Overall, it concludes that diazotrophic microbes (DM) may hold the key to achieve biological nitrogen fixation (BNF) in rice crop but the problem lies with effective inoculants technology. In recent years, inoculants technology has developed significantly, in terms of scale and quality, particularly for legumes but not for cereal crops. Moreover, genetically engineered strains are possible but remain an expensive and potentially more controversial approach to the technology. The effective utilization of diazotrophic microbes in the future will demand that there is a much more rational approach to the choice and delivery of the particular bacterium into the rice field.

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