STUDY OF MICROBE PLANT INTERACTION IN RELATION TO RICE AND *Rhizobium leguminosarum bv. trifolii* SN10

By

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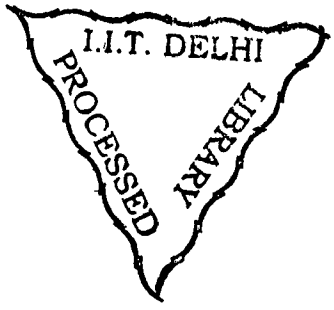
legumes – rice
Harvesting - Non legumes
Rice - microbe plant interaction



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To my parents

In eternal gratitude

For the nest, the compass, and the wings

CERTIFICATE

This is to certify that the thesis entitled **"STUDY OF MICROBE PLANT INTERACTION IN RELATION TO RICE AND *Rhizobium leguminosarum bv. trifolii*SN10"** being submitted by **Ms. RUMPA BISWAS** to the Indian Institute of Technology, Delhi, for the award of the degree of **'DOCTOR OF PHILOSOPHY'**, is a record of the bonafide research work carried out by her, which has been prepared under our supervision in conformity with the rules and regulations of the "Indian Institute of Technology, Delhi". The research reports and the results presented in this thesis have not been submitted for any degree or diploma in any other University or Institute.

Prof.S.N.Mukhopadhyay

Dr. Aqbal Singh

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R Biswas
Rumpa

ABSTRACT

Nitrogen is an essential nutrient which often limits plant growth and productivity. The extension of symbiotic nitrogen fixation to non-legumes, mostly cereal crops has been a long outstanding goal. Presently many workers have found that the nitrogen fixing bacteria are capable of associating endophytically with nonlegumes like rice, wheat and maize which in turn promotes growth and development of the plant. One such growth promoting endophyte is *Rhizobium leguminosarum bv. trifolii* isolated from the rhizosphere and roots of rice in Egypt.

In our study we have used a strain of *R. leguminosarum bv. trifolii* (SN10) isolated from the nodules of its host legume Berseem clover (*Trifolium alexandrium*) from Hissar, India. Its association with a local rice variety *Oryza sativa japonica* (Kdulam), strongly promoted both plant growth and development: shoot and root biomasses were increased up to 61% and 48%, respectively, when compared to the uninoculated rice. In addition, root branching was enhanced from 6 to 12 roots per plant while total nitrogen plant content was augmented to 21%. The bacteria successfully colonized root surfaces of the rice along the grooves, emerging lateral roots and also between epidermal cell junctions. Bacteria were even observed within roots. Moreover, seed and root exudates from this rice variety were capable of inducing an *in vitro* bacterial chemotactic response in *R. trifolii*. However, *O. sativa* does not release any phenolic signaling molecule which could play a role in chemotaxis or effectively induce *nod* genes of the bacteria. On the contrary, *O. sativa japonica* seed and root exudates were found to contain a coumarin, umbelliferone, which constituted almost 37% of the total phenolic content of the

exudates. Umbelliferone, which is known to posses antifungal and antibacterial properties, however had no effects on the bacterial colonization but inhibited *R. trifolii* SN10 *nod* gene expression. This was confirmed using a construct *R. trifolii* SN12 where the inducible *nodA* promoter of *R. trifolii* SN10 was amplified, sequenced and cloned upstream of *E.coli lacZ* in pCM132.By using a nodA-lacZ fusion, we showed that both umbelliferone and rice seed exudates inhibit the nod gene induction confirming that bacterial nod factors are not implied in rice growth. However, umbelliferone had no effect on *R. trifolii* SN10 rice root colonization. Then, the bacterium was found to secrete plant growth promoting compound, indole acetic acid (IAA). We conclude that the growth promotion of *O. sativa japonica* by *R. trifolii* SN10 are due plant growth promoting rhizobacterial (PGPR) interactions.

CONTENTS