

## ABSTRACT

An acetylene reduction technique was developed for estimating *in situ* the nitrogenase activity in the rhizosphere of lowland rice. The technique involves the use of only cheap and lightweight equipment, and is quick to set up. A comparison of this technique with the excised root assay, revealed little correlation of the results. This was attributed to the artifacts introduced by the long pre-incubation in the latter technique, including a 40 fold increase in the numbers of nitrogen-fixing bacteria during this period.

Comparisons of the nitrogenase activity in the rhizosphere of different rice varieties at the same growth stage on different soils revealed very little differences between varieties or soils, especially when activity was expressed per unit weight of plant. Nitrogenase activity per plant was maximal at the flowering and early grain filling stages, but when the data was expressed per unit weight of plant, young plants had the highest activity. Increased temperature caused increased nitrogenase activity, but at constant temperature there was little diurnal fluctuation in activity caused by the presence or absence of light. Estimates of the

quantity of nitrogen fixed during the entire cropping season based on data from the *in situ* acetylene reduction assay indicated that the input did not exceed  $6 \text{ kg N. ha}^{-1}$ . It is possible however, that the *in situ* technique used does not detect much of the nitrogenase activity in the soil some distance from the root (the outer rhizosphere).

Nitrogenase activity in the rhizosphere was shown to be due almost exclusively to microaerophilic organisms. Bacteria of the genera *Beijerinckia* and *Azospirillum* were isolated from rice roots and it was found that nitrite reductase negative ( $\text{nir}^-$ ) types of *Azospirillum brasilense* predominated over other *Azospirillum* types isolations were made from surface sterilised roots. Attempts at inoculation of *Beijerinckia* spp or *Azospirillum* spp onto sterile rice roots grown in solid agar, did not establish an effective nitrogen-fixing association.

It was concluded that there was little immediate prospect of selecting plant cultivars and/or bacterial strains to increase the nitrogen input to the plant from biological nitrogen fixation in the rhizosphere. Furthermore, it is probable that considerable modification of the diazotroph/ rice plant system will be required if the nitrogen input from this source is to be significantly increased.