## GENETIC DIVERSITY OF FAST GROWING RHIZOBIA ISOLATED FROM DIFFERENT GEOGRAPHICAL AREAS.

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Fast growing rhizobia that induce nodules on soybean were isolated from China, Vietnam and Papua New Guinea (Balatti, 1994). These organisms were partially characterized by several authors based on physiological and symbiotic characteristics (Sadowsky et al., 1983; Cleyet-Marel, 1987, Navarro et al., 1996) Several evidences suggest that some of the fast growing isolates might be evolutionary intermediates Therefore, the aim of this project was to fingerprint the genome of fast growing organisms by different PCR methods to analyze the genetic relationship of these organisms.

DNA was extracted from fast growing strains isolated from the soils of China, Vietnam and Papua New Guinea, respectively. We fingerprinted the microorganisms genome with Enterobacter Repetitive Intergenic Consensus (ERIC), Repetitive Extragenic Palindromic elements (REP) and Random Amplified Polymorphic DNA (RAPD) primers. Also, amplified 16SARN and Intergenic Sequence (IGS) *nifH-nifD* DNA fragments were analyzed by restriction analysis with *Alu* I, *Hae* III, *Rsa* I, *Hind* III and *Hinf* I. UPGMA algorithm was used to perform a hierarchical cluster analysis. Pairwise comparisons were calculated using Simple Matching (SM) coefficient. Principal coordinate Analysis (PCA) was also performed.

The analysis of ERIC, REP and RAPD fingerprints revealed considerable diversity between fast growing rhizobia and confirmed the results of Perret et al (1994) that these are closely related organisms. However, these fingerprinting techniques suggest that *Rhizobium* sp. NGR234 is more closely related to *S. fredii* strains, than at least some *S. f* strains are between each other. In addition the analysis of the results suggest that the strains isolated from the soils of Vietnam were the most similar *S. fredii* with a SM coefficient of 90%.

ARDRA analysis of the 16SRNA and IGS *nifD-nifH* suggest that *R*. sp. NGR 234 is, as expected, though closely related to *S. fredii* isolates, the most distantly related organism. Cluster analysis performed either with results from ARDRA or from REP, ERIC or RAPD, grouped the strains based on the place of origin. Though more work has to be done the similarity coefficient found between *S. fredii* strains and *Rhizobium sp.* NGR234 suggest that this strain should most probably be named *Sinorhizobium fredii*.

References

Balatti PA (1994) Thesis. University of Missouri, Columbia, USA. Cleyet-Marel JC (1987) Thesis Universite Claude Bernard, Lyon, France. Peret et al (1994) Nucl. Acids Res. 22, 1335-1341. Rodriguez-Navarro DN et al (1996) Syst. Appl. Microbiol. 19, 240-248. Sadowsky et al (1983) Int. J. System. Bacteriol. 33, 716-722. Videira LB et al (1999) Soil Biol. and Biochem. (Submitted)

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