Bradyrhizobium japonicum and Bradyrhizobium elkanii isolates recovered from soils cultured with soybean in a zero tillage management that differ in th

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Rhizobia nodulating soybeans were isolated from soils cultivated under zero tillage management for approximately 10 years. The isolates (250) were found to be siblings of strains of B.japonicum and B. elkanii used in commercial inoculants. A subset of 12 of them were selected based upon either their high or low nitrogen fixing activity, compared to control strains E109, Semia 5080, Semia 5079, Semia 587 and Semia 5019, which are the strains used in commercial inoculants in Argentina, Brasil and Uruguay. The similarity of the isolates was assessed by means of BOX, REP-PCR and RSα sequences. Our hypothesis is that differences in the strains ability to fix nitrogen might be due to structural alterations that occurred in the two genome fragments containing most of the nitrogen fixing genes (Kaneko et al., 2002) or to differential expression of nitrogen fixation genes. We are currently analyzing the structural features of the DNA fragments containing the nitrogen fixation genes. Our preliminary assays consisted in the amplification of a 1169 bp fragment containing nifD that was restricted with EcoRV. We found three different restriction patterns. Also we amplified a 3.5 kb fragment containing RSα, RSβ and nifD that is sequences where mutations might occur at different rates. We are currently setting up amplification reactions aimed at amplifying in four different reactions the nitrogen fixation genes in order to evaluate if the strains lack similarity at this regions.