Title: PHYLOGENETIC ANALYSIS OF 16S-23S rRNA INTERGENIC SPACER REGIONS FROM *Mucuna aterrima* ROOT NODULES.

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Abstract:

Biological Nitrogen Fixation (BNF), performed by soil microorganisms associated with legumes, is biotechnological techniques, which has a significant role in commercial and environmental activities. The Rhizobium-legume symbioses provide improvement in soil quality and it increases cropping systems productivity, which leads to shrinking with fertilizers, and increasing of soil nutrient supply. Diversity studies have been performed with the purpose of determine factors that regulate bacterial structures over time and space, in order to select most effective strains in the BFN process. The aim of this study was to genetically characterize through Intergenic Spacer Region (ISR), 22 bacteria isolated from mucuna roots, and 10 strains was used as rhizobia control. In the present study, polymerase chain reaction (PCR) amplified fragments between the 16S-23S rRNA region of the 32 isolates tested. The data were evaluated by similarity analysis based on Jaccard's index of similarity and by using UPGMA Algorithm. The strains were separated into groups according to their genetic similarity traits, and represented by dendrogram. The results showed that 11 groups presented 55% of similarity which indicate genetic diversity among the isolates. All bacteria presented at least 10% of similarity between the isolates tested and the strains used as controls. They presented from 1 to 5 fragments, but most of them, 14, presented 3 fragments. The range of size was from 610 to 8160 pb, of which 910-1060 pb were presented in higher amount in 21 isolates. It was possible to observe great variability among the strains, which can be inferred as mutation in the bacterial genome due to DNA recombination and/or natural and evolutionary processes of addition and subtraction bases in the intergenic region. In addition to characterize and show biodiversity, the genotypic study and description can also be used to verify ecological adaptability of different isolates to the prevailing environmental conditions in the Cerrado ecosystem. The characterization can also explain the ecological and evolutionary relationship of soil microorganisms that contributes for selecting and handling effective groups in the process of infection and biological nitrogen fixation in the legume (mucuna), in order to support sustainable agriculture and the improvement of soil quality.

Keywords: rhizobia, BNF, phylogeny

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