Endophytic Bacterial Diversity of Brazilian Sugarcane cultivar RB-72454 MAGNANI, G.S<sup>1</sup>.; CRUZ, L.M.<sup>1</sup>; WEBER, H.<sup>2</sup>; MONTEIRO, R.A.<sup>1</sup>, FAORO, H.<sup>1</sup>; PEDROSA, F. O<sup>1</sup>; YATES, M.G. <sup>1</sup>;; SOUZA, E.M<sup>1</sup>.

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Endophytic bacteria are found in inner tissues of most plants and can promote many benefits. In this work the diversity of sugarcane cultivar RB-72454 endophytes was analyzed by cultivation independent techniques. The stem of sugarcane from a commercial plantation in the Northwest of Parana state (Brazil) was externally sterilized, frozen in liquid nitrogen, and used for DNA extraction. This DNA was used as template for amplification of a 16S rDNA gene fragment using the primers 27f and 805r. Amplicons of 800 pb were cloned into pCR 2.1. Six hundred clones were collected, and the inserts of 107 clones sequenced. Statistical analysis showed that the sequences represented approximately 90% of the operational taxonomic units (OTU) present in the sample. The majority (70 %) of the sequences were highly similar (more than 98% of identity) to Proteobacteria 16S rRNA, among these 40.2% to Pseudomonas sp, 22.4% to Enterobacter sp, 6.5% to Pantoea sp and 0.7% to Klebsiella sp. A high proportion of the sequences (28.9%) were similar to 16S rRNA of uncultured bacteria. Surprisingly, sequences similar to diazotrophic bacteria found associated endophytically to sugarcane such as Herbaspirillum seropedicae, Azospirillum brasilense and Gluconacetobacter diazotrophicus were not identified, suggesting that the use of nitrogen fertilizer in commercial plantation of sugar cane may reduce the endophytic population of nitrogen fixing bacteria. The results also reveal a rich diversity of bacteria and a remarkable number of uncultivable bacteria present in the stem of healthy sugarcane.

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