**TITLE**: Paraburkholderia guartelaensis sp. nov., ISOLATED FROM NATIVE Mimosa gymnas IN A BRAZILIAN ECOTONE

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## ABSTRACT:

The symbioses between plants of the Fabaceae family and diazotrophic bacteria have been studied over the years, due to their ability to fix atmospheric nitrogen in a process denominated as biological nitrogen fixation (BFN). BFN plays a key role in the nitrogen incorporation into the terrestrial environments. This restricted group of bacteria is collectively called rhizobia. Most of the known rhizobia belong to the Alphaproteobacteria class, but also include Betaproteobacteria, with an emphasis on the Paraburkholderia genus. Paraburkholderia have been recognized as the main symbionts of the Mimosa genus in Brazil, the second largest genus of the subfamily Mimosoideae, and the country represents the main radiation center of the genus, many of which are endemic species. Six nitrogen-fixing symbiotic bacteria were isolated from nodules of Mimosa gymnas native and endemic from the Guartelá State Park, PR, an ecotone region between the Brazilian biomes of Atlantic forest and Cerrado. A polyphasic approach was performed in order to determine the taxonomic position of the group. In the analysis of the 16S rRNA, the strains showed highest similarity with *Paraburkholderia nodosa* BR 3437<sup>T</sup> (98.4–98.5 %), *P. mimosarum* PAS44<sup>T</sup> (98.6–99.9 %) and *P. diazotrophica* JPY461<sup>T</sup> (96.5–99.9 %). The Multilocus Sequence Analysis (MLSA) of five (recA, gyrB, trpB, gltB and atpD) concatenated housekeeping genes indicated a distinct position of strain CNPSo 3008<sup>T</sup>, with P. nodosa as the closest species. The nodC phyloheny grouped all CNPSo strains with 100 % of nucleotide identity (NI), revealing a monophyletic character. The CNPSo 3008<sup>T</sup> genome was sequenced and digital DNA-DNA hybridization (dDHH) revealed less than 56 % of relatedness with the closest related species, while average nucleotide identity (ANI) ranged from 81.87 to 93.84 %. All values were below of the suggested threshold for species delimitation, indicating that the CNPSo strains represent a novel Paraburkholderia species. The BOX-PCR showed high genetic diversity between the strains. Phenotypical features were also evaluated. Phylogenetic, genotypic and phenotypical analyses supported the proposal of the novel species Paraburkholderia guartelaensis sp. nov., with CNPSo 3008<sup>T</sup> (=) indicated as the type strain.

Keywords: Paraburkholderia, Mimosoideae, biological nitrogen fixation, rhizobia

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