

**TITLE:** RALSTONIA sp. AND METAL RESISTANCE GENES RECOVERED FROM THE GUT MICROBIOME METAGENOME OF A HUNTER-GATHERER BRAZILIAN GROUP

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**ABSTRACT:**

Studies focused on human gut microbiome allow the exploration of taxonomic and functional traits that can raise particular features from specific organisms. Here, we previously generated and analyzed shotgun metagenomes from the gut microbiome of a hunter-gatherer Yanomami group from Brazil. This data was compared with shotgun metagenomes from other traditional groups and an urban-industrialized group. In the taxonomic analysis, LefSe identified the genus *Ralstonia* as a biomarker of the Yanomami from Brazil gut microbiome. *Ralstonia* is a bacterial genus known to have a set of metal resistance determinants, and interestingly, the functional analysis performed by SUPER-FOCUS demonstrated that the gut microbiome from Yanomami had a high abundance of gene families related to cobalt, zinc and cadmium resistance. Therefore, we studied more deeply the genus *Ralstonia* and the metal resistance genes in these microbiomes. Seven *Ralstonia* housekeeping genes (*gdhA*, *gyrB*, *rplB*, *leuS*, *adk*, *mutS* and *16S*) were recovered using bowtie2 with customized parameters. Phylogenetic analysis showed a cluster containing three *Ralstonia* from Yanomami separate from other clusters formed by different *Ralstonia* species. This result indicates that they may belong to a species not previously identified. To explore the metal resistance capacity, we searched for *czc* genes in the metagenomic contigs assembled with metaSPADES. The positive contigs, with length ranging from 15 kb to 25 kb, were blasted against the ncbi database confirming the presence of *czc* genes and moreover, that the contigs containing the *czc* genes have high identity (>90%; query cover > 92%) with *Ralstonia* genomes, addressing the metal resistance trait to *Ralstonia*.

**Keywords:** Gut microbiome, Metal resistance genes, MLSA, Phylogenetic analysis, *Ralstonia*

**Development Agency:** CAPES, CNPq