

# Complete Genome Sequence of *Burkholderia phenoliruptrix* BR3459a (CLA1), a Heat-Tolerant, Nitrogen-Fixing Symbiont of *Mimosa flocculosa*

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**The genus *Burkholderia* represents a challenge to the fields of taxonomy and phylogeny and, especially, to the understanding of the contrasting roles as either opportunistic pathogens or bacteria with biotechnological potential. Few genomes of nonpathogenic strains, especially of diazotrophic symbiotic bacteria, have been sequenced to improve understanding of the genus. Here, we contribute with the complete genome sequence of *Burkholderia phenoliruptrix* strain BR3459a (CLA1), an effective diazotrophic symbiont of the leguminous tree *Mimosa flocculosa* Burkart, which is endemic to South America.**

Species definition within the genus *Burkholderia* is still challenging and requires polyphasic approaches (2). The genus includes species with contrasting roles, from highly opportunistic pathogens to bacteria with biotechnological potential. Recently a new genus was suggested, encompassing plant-associated beneficial bacteria (4, 6); however, taxonomic problems within the genus continue and will probably be solved only when the genomes of several strains have been fully sequenced. Very few nonpathogenic strains, especially of diazotrophic symbiotic bacteria, have been sequenced. Here, we contribute with the complete genome sequence of *Burkholderia phenoliruptrix* strain BR3459a (CLA1), a mucoid colony variant of strain BR3459 (BR3462), a rhizobial strain isolated from a root nodule of *Mimosa flocculosa*, a small tree endemic to southern South America. This strain is highly specific and effective in fixing nitrogen with the host species and is unusual in its capacity to grow *in vitro* at 40°C without losing its symbiotic properties (3).

The genome sequence of *B. phenoliruptrix* BR3459a was obtained via Roche 454 pyrosequencing with a total of 1,336,569 reads (35-fold genome coverage). The genome was assembled using both the Newbler (version 2.6; 454 Life Sciences, Roche Diagnostics Corporation, Branford, CT) and Celera (WGS; version 7.0) assemblers (5). Annotation and analysis of the sequences were carried out using the System for Automated Bacterial Integrated Annotation (SABIA) (1). An automatic functional annotation was performed using the KEGG database, and a manual annotation by comparison with the UniProt/Swiss-Prot, KEGG, NCBI-NR, and InterPro databases was performed on the remaining open reading frames (ORFs).

The genome of *B. phenoliruptrix* BR3459a consists of two circular chromosomes of 4,152,217 and 2,713,495 bp and one plasmid of 785,419 bp with a total genome size of approximately 7.65 Mbp. The large chromosome (chromosome 1) and the small chromosome (chromosome 2) encode 3,561 and 2,313 predicted ORFs, respectively, and the plasmid contains 731 predicted ORFs. The GC content for the genome is 63.15%.

In chromosome 1, we found 2,975 known protein ORFs, 585 hypothetical ORFs, 55 tRNAs, 3 rRNA operons, and 1 pseudogene. In chromosome 2, we found 1,804 known protein ORFs, 509 hypothetical ORFs, 8 tRNAs, and 3 rRNA operons. In the plasmid, we found 454 known protein ORFs and 277 hypothetical ORFs. The largest number of orthologs was identified with *Burkholderia* sp. strain CCGE1001 (74.47%), isolated from a root nodule of *Mimosa affinis* in Mexico but lacking plasmids and symbiotic genes in its sequenced genome (NCBI BioProject PRJNA42975).

Plasmid pSYMBR3459a shows high sequence identity and considerable gene synteny with the symbiotic plasmid pBPHY02 of *Burkholderia phymatum* strain STM815<sup>T</sup>, including genes for nodulation, nitrogen fixation, auxin synthesis, hydrogenase components, and ACC deaminase activity. The complete genome revealed the presence of an apparatus for synthesis and assembly of flagellar components, several putative ABC transporters, as well as type II, III, IV, and VI secretion systems.

**Nucleotide sequence accession numbers.** The complete genome sequence of *B. phenoliruptrix* BR3459a has been deposited at NCBI GenBank under accession numbers [CP003863](#), [CP003864](#), and [CP003865](#).

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