



Genome Announcement

Complete genome sequence of *Paenibacillus riograndensis* SBR5^T, a Gram-positive diazotrophic rhizobacterium[☆]Luciana Fernandes Brito^{a,b}, Evelise Bach^c, Jörn Kalinowski^b, Christian Rückert^b, Daniel Wibberg^b, Luciane M. Passaglia^c, Volker F. Wendisch^{a,b,*}^a Genetics of Prokaryotes, Faculty of Biology, Bielefeld University, Universitätsstraße 25, 33615 Bielefeld, Germany^b Center for Biotechnology (CeBiTec), Bielefeld University, Universitätsstraße 25, 33615 Bielefeld, Germany^c Departamento de Genética, Instituto de Biociências, Universidade Federal do Rio Grande do Sul. Av. Bento Gonçalves, 9500, Caixa Postal 15.053, 91501-970 Porto Alegre, RS, Brazil

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ABSTRACT

Paenibacillus riograndensis is a Gram-positive rhizobacterium which exhibits plant growth promoting activities. It was isolated from the rhizosphere of wheat grown in the state of Rio Grande do Sul, Brazil. Here we announce the complete genome sequence of *P. riograndensis* strain SBR5^T. The genome of *P. riograndensis* SBR5^T consists of a circular chromosome of 7,893,056 bps. The genome was finished and fully annotated, containing 6705 protein coding genes, 87 tRNAs and 27 rRNAs. The knowledge of the complete genome helped to explain why *P. riograndensis* SBR5^T can grow with the carbon sources arabinose and mannitol, but not *myo*-inositol, and to explain physiological features such as biotin auxotrophy and antibiotic resistances. The genome sequence will be valuable for functional genomics and ecological studies as well as for application of *P. riograndensis* SBR5^T as plant growth-promoting rhizobacterium.

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Plant growth-promoting bacteria may be beneficial for crop production (da Costa et al., 2014). Bacterial communities can be characterized by metagenomics approaches as e.g. applied to monitor changes of root bacterial communities associated to two different development stages of canola (*Brassica napus* L. var *oleifera*; de Campos et al., 2013). Studies focusing on isolation of plant growth-promoting bacteria are also often performed, e.g. for sugar cane (Beneduzi et al., 2013), maize (Arruda et al., 2013), rice (Souza et al., 2013), and wheat (Beneduzi et al., 2008). *Paenibacillus riograndensis* SBR5^T, a diazotrophic bacterium isolated from the rhizosphere of *Triticum aestivum* L. cultivated in Southern Brazil, has been described as a new species of the genus *Paenibacillus* (Beneduzi et al., 2010). The Gram-positive rod-shaped, facultative aerobic, motile, spore-forming *P. riograndensis* SBR5^T has been investigated for its plant growth promotion characteristics and its potential use as wheat inoculant (Beneduzi et al., 2008). The strain is available from the Brazilian type collection LFB-FIOCRUZ

as CCGB1313 and from Spanish type collection CECT as CECT7330. The draft genome sequence has previously revealed the presence of *nif* genes as well as of genes related to the alternative nitrogen fixation system (*anf* genes; Beneduzi et al., 2011). Since there are few studies about *anf* genes in Gram-positive diazotrophs, this species constitutes an interesting model for the study of the regulation of nitrogen fixation in this group of bacteria (Fernandes et al., 2014).

To perform the sequencing of *P. riograndensis* SBR5^T, two shotgun Paired-End and Mate-Pair libraries were generated. The libraries were prepared using Nextera DNA sample preparation kit and Nextera Mate-Pair sample preparation kit, respectively (Illumina, U.S.A.). The sequencing run was carried out using the Illumina MiSeq System. The genome sequencing resulted in 6,781,183 reads, assembled in 4 scaffolds and 198 contigs by the Newbler v.2.8 (Roche, Switzerland), with 198 fold average coverage. The largest scaffold had 7,885,596 bps and the largest contig had 437,460 bps. The average read lengths were 743 ± 249 bps for the Paired-End library and 9692 ± 2423 bps for the Mate-Pair library.

The genome finishing was performed using the CONSED Software package (Gordon, 2003) to order and join the contigs, close gaps (repetitive sequences, which were confirmed by PCR) and resolve SNPs in repetitive regions. The whole genome of SBR5^T consists of a circular chromosome of 7,893,056 bps, with GC content of 50.97% (Table 1). The 523,056 bps absent from the draft genome sequence consisting of 2276 contigs (Beneduzi et al., 2011) were

[☆] Sequence accession numbers: The complete genome sequence has been deposited in EMBL/GenBank with accession number LN831776.

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Table 1
Genome features of *P. riograndensis* SBR5^T.

Features	Chromosome
Length (bp)	7,893,056
G + C content (%)	50.97%
CDS	6705
rRNA genes (operons)	27 (9)
tRNA genes	87

not clustered, but scattered over the whole genome. The finished sequence was submitted to GenDB Software (Meyer et al., 2003) for automatic identification and annotation of the genes, resulting in 6705 protein coding genes, 87 tRNAs and 27 rRNAs (Table 1). The rRNA genes (named Prio.6706 to Prio.6732) are organized in nine individual operons (*rnnA*, *rnnB*, *rnnC*, *rnnD*, *rnnE*, *rnnF*, *rnnG*, *rnnH* and *rnnI*) located in different regions of the genome. Each operon encodes the 5S, 16S, and 23S rRNAs in varied order except for operon *rnnG* which lacks a 5S rRNA gene while operon *rnnH* contains two 5S rRNA genes.

The genome of SBR5^T contains genes putatively involved in resistance to several antibiotics such as encoding the antibiotic efflux systems belonging to the RND (e.g. Prio.4911), ABC (e.g. Prio.6246), MFS (e.g. Prio.6658), and MATE (e.g. Prio.2495) protein families. Furthermore, genes that possibly confer specific antibiotic resistance including 10 *van* (e.g. Prio.6068) and 18 genes related to the general β -lactamase mediated resistance were found (e.g. Prio.6596). For example, the growth of SBR5^T on LB agar plates containing 200 $\mu\text{g ml}^{-1}$ erythromycin or 600 $\mu\text{g ml}^{-1}$ kanamycin (data not shown) may be explained by the gene encoding a multidrug exporter of the Emr protein family (Prio.3171), which confers erythromycin resistance in *Escherichia coli* (Nishino and Yamaguchi, 2001), and the kanamycin nucleotidyltransferase gene (Prio.3529), respectively.

SBR5^T is not able to grow in minimal medium without biotin and this biotin auxotrophy is reflected by the absence of all biotin biosynthesis genes (*bioWAFDBI*), although the Prio.5347 encoded P450 enzyme shows similarity to Biol of *Bacillus subtilis*.

P. riograndensis SBR5^T is characterized by the ability to grow with the carbon sources arabinose and mannitol, but not *myo*-inositol (Beneduzi et al., 2010). A cluster of three adjacent genes (Prio.4651–4653) encoding uptake system AraE and the AraC-family two-component regulatory system and a cluster of four genes (Prio.6589–6592) encoding enzymes AraB, AraA and AraD as well as repressor AraR may explain uptake, utilization and regulation of arabinose. Mannitol uptake, phosphorylation and conversion to fructose-6-phosphate is commensurate with the presence of four adjacent genes (Prio.1805–1808) coding for mannitol specific PTS and mannitol-1-phosphate 5-dehydrogenase. *P. riograndensis* SBR5^T is unable to utilize *myo*-inositol which is reflected by the lack of the genes *iolB*, *iolD* and *iolJ*, although homologs of *idhA*, *iolE*, *iolC* and *iolA* are present (Prio.3014, Prio.4831, Prio.2204 and Prio.6323). Albeit SBR5^T was negative in a nitrate reduction assay (Beneduzi et al., 2010), its genome encodes putative nitrate reductase NarGHJI (Prio.3572–3574), while there is no evidence for assimilatory nitrate reductase NasACKBDEF. The complete genome sequence will be valuable for future characterization

of the physiology of the diazotroph *P. riograndensis* SBR5^T, functional genomics and its application in agrobiotechnology.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at <http://dx.doi.org/10.1016/j.jbiotec.2015.04.025>

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