

The complete genome sequence of *Bacillus halotolerans* isolated from *Solanum tuberosum* in Tunisia

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Abstract

In this work, we present the complete genome sequence (CGS) of *Bacillus halotolerans* strain SpS5 isolated from *Solanum tuberosum* from Tunisia, which consists of 4133407 bp including 4136 protein-coding sequences, 88 tRNA, 1 tmRNA and 43.68 % GC content. The CGS of *B. halotolerans* strain sequence will bring important information into the implication of key genes associated with its endophytic behaviour, interactions with its hosts as well as its biocontrol mechanisms.

Introduction

To date, a great number of *Bacillus* species have been isolated and identified from different origins. However, there are few studies on their genome information. (Shen et al. 2021).

Bacillus species are considered important rhizobacteria that help to promote plant growth and development by various mechanisms involving the promotion of alternative nutrient pathways, through the fixation of atmospheric nitrogen, the acquisition of iron by siderophores, the solubilization of phosphate, and the production of growth phytohormones and organic volatile compounds (Petrillo et al. 2021).

Bacillus sp. are highly used as potential bio-control agents against plant pathogens due to the production of different antimicrobial compounds like hydrolytic enzymes and lipopeptides

(Wang et al 2021). Indeed, Ben Slama *et al.* (2019) revealed that *Bacillus halotolerans* could efficiently inhibit *Fusarium oxysporum* on the tomato fruit, and showed in vitro inhibition of four main phytopathogens i.e. *Alternaria. alternata*, *Botrytis. cinerea*, *Rhizoctonia bataticola*, and *Phytophthora infestans*. The complete genome sequence of bacteria is an efficient method enveloped to understand the genetic basis of bacteria that confers tolerance, adaptation and improvement of plant growth and production. For example Zhang et al., (2018) studied the resistance potential to drought and salt stress of *Bacillus halotolerans* through complete genome sequence of this bacteria.

Therefore, this work present the complete genome sequence of *B. halotolerans* SpS5 strain in order to go insight the genetic basis of its mechanisms of biocontrol against the fungal pathogens *Rhizoctonia solani* and *Fusarium solani*.

Material and methods

Bacteria isolation and culture

Bacillus halotolerans strain SpS5 isolated from stems of *Solanum tuberosum* was used in this study. The isolation of the endophytic bacteria was performed by disinfecting stems of *potato* cv. Spunta with 70 % ethanol for 2 min, then with 3.5 % sodium hypochlorite for 10 min and followed by washing them with sterile distilled water five times. Disinfected stems pieces were placed on Luria-Bertani (LB) medium at 28 °C in dark until appearance of bacterial colonies. The last washing water was poured on LB medium to ensure the success of the surface disinfection. The SpS5 strain was purified on LB medium at 28 °C then stored in 25 % glycerol at -20 °C (Azaiez et al., 2018).

Sequencing of bacterial genomic DNA

DNA extraction, Illumina genome sequencing and assembly were carried out following the protocols provided by MicrobesNG (Birmingham, UK, <https://microbesng.com>). Complete genome sequencing of *Bacillus halotolerans* was performed using Illumina HiSeq, reads were trimmed by means of Trimmomatic (version 0.39) and the quality was evaluated using in-house scripts combined with the Samtools version 1.4 ([git://github.com/samtools/samtools.git](https://github.com/samtools/samtools.git)), BedTools version 2.18 (Quinlan and Hall, 2010) and bwa-mem (Li and Durbin, 2009) software. Sequenece reads were assembled into contigs using QUAST software version 5.0.2 (Gurevich et al., 2013) and the genome was annotated with

Prokka 1.14.3 (<https://github.com/tseemann/prokka>). The taxonomic labels identification of the genome sequence was achieved by Kraken.

Results

Genomic proprieties

A summary of the genomic features for SpS5 strain is listed in Table 1. Genome assembly accounted for 4.13 Mb with a mean coverage of 105.241. The number of protein-coding genes of *Bacillus halotolerans* was 4136 and the G + C content was 43.68. The results based on the Average Nucleotide Identity (ANI) test and current taxonomic nomenclature disclosed an identity above 98 % of the submitted genome sequence to *Bacillus* genus.

Nucleotides sequences accession numbers

The *B. halotolerans* SpS5 strain complete genome sequence has been deposited in the NCBI GenBank under accession number as indicated in Table 1.

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Table 1. Summary statistics of *Bacillus halotolerans* SpS5 genome assembled from Illumina reads.

Bacterial strain	Host plant	Bio-sample	# contigs ≥1000 bp	Largest contig	Total length	GC (%)	Mean coverage	N50	CDS	tRNA	tm RNA	GenBank Accession (Assembly)	GenBank Accession (Raw reads)
<i>Bacillus halotolerans</i> SpS5	<i>Solanum tuberosum</i>	SAMN219 71836	15	787391	4133407	43.68	105.241	504381	4136	88	1	JAJERC0000 00000	SRR16149 277