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## Complete genome of *Pseudomonas psychrophila* strain L10.10, a psychrotolerant biofertilizer that promote plant growth

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## Abstract

*Pseudomonas psychrophila* strain L10.10 is a psychrotolerent bacteria which was isolated in Lagoon Island, Antarctica. The complete genome sequence revealed genomic information of its role as a plant-growth promoting bacterium through its nitrogen-fixing ability and indole acetic acid (IAA)-producing trait with additional plant disease prevention attribute *via* hydrogen cyanide (HCN) production.

## Highlights

- *Pseudomonas psychrophila* strain L10.10 is a psychrotolerant bacterium.
- This is the first complete genome of *Pseudomonas psychrophila*.
- Various genes for plant promoting properties were identified
- Functional annotations also revealed indole acetic acid (IAA)-producing attribute

**Keyword:** Plant disease control, nitrogen fixing, indole acetic acid, hydrogen cyanide, plant growth-promoting rhizobacteria

Plant growth-promoting rhizobacteria (PGPR) are of significant agricultural importance and biotechnological value due to their plant growth-promoting activity under stressful and nutrient limiting conditions (Nabti et al., 2010). *Pseudomonads* are well known PGPRs which are associated with direct plant growth promotion by production of indole acetic acid (IAA) and involvement in nitrogen fixation activity (Zhao, 2010; Santi et al., 2013). In addition, *Pseudomonads* also confer indirect promotion of plant growth through production of hydrogen cyanide (HCN) which aids in prevention of plant diseases caused by phytopathogens (Schippers et al., 1990). Previously, we isolated a psychrotolerant bacterium, *Pseudomonas psychrophila* strain L10.10 from Antarctica. Psychrotolerent PGPRs are important in improving cold stress tolerance of important agricultural crops such as grapevine plantlets (Ait Barka et al., 2006). Hence, we are interested to investigate the genome feature of *Pseudomonas psychrophila* strain L10.10 and determine if this psychrotolerant strain have the relevant PGP genotype.

Genomic DNA of *Pseudomonas psychrophila* strain L10.10 was isolated from an overnight cell suspension culture using MasterPure<sup>TM</sup> Gram positive DNA purification kit (Epicentre Technologies). Subsequently, the gDNA was constructed into 20-kb SMRTbell template library. Pacific Biosciences (PacBio) RSII sequencing platform was used to perform the whole genome sequencing using C4 chemistry in two single molecule real time (SMRT) cells. A total of 16,782 reads with a mean read length of 11,629 bp were generated. The reads were *de novo* assembled using hierarchical genome assembly process (HGAP) algorithm version 3 into a circular contig with an average genome coverage of 34.96 folds.

Subsequently, genome annotation was performed using NCBI Prokaryotic Genome Annotation Pipeline (PGAP) version 2.10 and Rapid Annotation using Subsystem Technology (RAST) version 3.0 (Aziz et al., 2008, Overbeek et al., 2014 and Brettin et al., 2015, Xia et al., 2015). The genome of *Pseudomonas* psychrophila strain L10.10 consists of a 5,172,488 bp circular chromosome with GC content of 58.2%. A total of 3514 protein coding genes were predicted along with 25 rRNA and 68 tRNA genes (Table 1).

Functional annotation of the genome revealed presence of genes responsible for the two major pathways of bacterial ammonia assimilation namely, glutamate dehydrogenase (GDH) pathway (glutamate dehydrogenase [AOC04\_00325]) and glutamine synthetase (GS)-glutamate synthase (GOGAT) pathway (glutamine synthetase [AOC04\_04085] and glutamate synthase [AOC04\_01625]). In addition, a complete hydrogen cyanide synthase gene cluster [AOC04\_10125, AOC04\_10135, AOC04\_10140] which catalyze synthesis of hydrogen cyanide were identified. Furthermore, we also detected presence of various genes which contribute to IAA biosynthesis namely Indole-3-glycerol phosphate synthase [AOC04\_22470], *N*-(5'-phosphoribosyl) anthranilate isomerase [AOC04\_10045] and anthranilate phosphoribosyltransferase [AOC04\_22465] within the genome of this strain.

Our genome analysis revealed presence of various PGPR traits in strain L10.10 namely nitrogen fixation, hydrogen cyanide production, and phytohormone IAA biosynthesis which reveal its potential as a promising PGPR. These beneficial traits also indicated the potential application of strain L10.10 in development of eco-friendly biofertilizer which can assist in promoting soil fertility and crops yield.

#### Nucleotide sequence accession number

The complete chromosome sequence has been deposited in GenBank under the accession number CP012676

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#### References

Nabti, E., Sahnoune, M., Ghoul, M., Fischer, D., Hofmann, A., Rothballer, M. *et al.* 2010. Restoration of growth of durum wheat (*Triticum durum* var. waha) under Saline conditions due to inoculation with the Rhizosphere Bacterium *Azospirillum brasilense*; NH and extracts of the marine alga; *Ulva lactuca. J Plant Growth Regul* 29, 6–22. Zhao, Y. 2010. Auxin biosynthesis and its role in plant development. Annu. Rev. Plant Biol. 61, 49-64.

Santi, C., Bogusz, D., Franche, C. 2013. Biological nitrogen fixation in non-legume plants. Ann. Bot., 111,743–767

Schippers, B., Bakker, A., van Peer, R. 1990.Beneficial and deleterious effects of HCNproducing pseudomonads on rhizosphereinteractions. Plant Soil. 129,75–83

Ait Barka, E., Nowak, J., Clement, C. 2006. Enhancement of chilling resistance of inoculated grapevine plantlets with a plant growth-promoting rhizobacterium, *Burkholderia phytofirmans* strain PsJN. *Appl. Environ. Microbiol.* 72, 7246–7252

Aziz, R.K., Bartels, D., Best, A.A., DeJongh, M., Disz, T., Edwards, R.A., Formsma, K., Gerdes, S., Glass, E.M., Kubal, M., Meyer, F., Olsen, G.J., Olson, R., Osterman, A.L., Overbeek, R.A., McNeil, L.K., Paarmann, D., Paczian, T., Parrello, B., Pusch, G.D., Reich, C., Stevens, R., Vassieva, O., Vonstein, V., Wilke, A., Zagnitko, O., 2008. The RAST Server: rapid annotations using subsystems technology. BMC Genomics 9, 75.

Overbeek, R., Olson, R., Pusch, G.D., Olsen, G.J., Davis, J.J., Disz, T., Edwards, R.A., Gerdes, S., Parrello, B., Shukla, M., Vonstein, V., Wattam, A.R., Xia, F., Stevens, R., 2014. The SEED and the Rapid Annotation of microbial genomes using Subsystems Technology (RAST). Nucleic Acids Res. 42, D206–D214

Brettin, T., Davis, J.J., Disz, T., Edwards, R.A., Gerdes, S., Olsen, G.J., Olson, R., Overbeek, R., Parrello, B., Pusch, G.D., Shukla, M., Thomason, J.A., Stevens, R., Vonstein, V., Wattam, A.R.,

Xia, F., 2015. RASTtk: A modular and extensible implementation of the RAST algorithm for building custom annotation pipelines and annotating batches of genomes. Sci. Rep. 5, 8365.

#### Table 1

Genome features of *Pseudomonas psychrophila* strain L10.10.

Attributes	Value
Genome size (bp)	5,172,488

Pseudogene	120
GC content (%)	58.2
Total number of genes	4,639
Protein coding genes	4,425
Contigs	1
rRNA genes	25
tRNA genes	68