# Whole-Genome Sequence of Bacillus sp. SDLI1, Isolated from the Social Bee Scaptotrigona depilis

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# Whole-Genome Sequence of *Bacillus* sp. SDLI1, Isolated from the Social Bee *Scaptotrigona depilis*

Camila R. Paludo,[a,b,c] Antonio C. Ruzzini,[b] Eduardo A. Silva-Junior,[a,b] Gleb Pishchany,[c] Cameron R. Currie,[d] Fábio S. Nascimento,[e] Roberto G. Kolter,[c] Jon Clardy,[b] Mônica T. Pupo[a]

School of Pharmaceutical Sciences of Ribeirão Preto, University of São Paulo, Ribeirão Preto, São Paulo, Brazil[a]; Department of Biological Chemistry and Molecular Pharmacology, Harvard Medical School, Boston, Massachusetts, USA[b]; Department of Microbiology and Immunobiology, Harvard Medical School, Boston, Massachusetts, USA[c]; Department of Bacteriology, University of Wisconsin, Madison, Wisconsin, USA[d]; Department of Biology, FFCLRP, University of São Paulo, Ribeirão Preto, São Paulo, Brazil[e]

**We announce the complete genome sequence of *Bacillus* sp. strain SDLI1, isolated from larval gut of the stingless bee *Scaptotrigona depilis*. The 4.13-Mb circular chromosome harbors biosynthetic gene clusters for the production of antimicrobial compounds.**

Address correspondence to Jon Clardy, jon_clardy@hms.harvard.edu, or Mônica T. Pupo, mtpupo@fcfrp.usp.br.

A number of *Bacillus* spp. engage in symbiotic relationships with solitary and social bees (1–4). These bacteria can improve insect nutrition (4) and in some instances provide protection against entomopathogens through small-molecule chemical defenses (5, 6). A recent report of a mutualistic relationship between the social bee *Scaptotrigona depilis* and a fungus (7) highlighted the importance of studying bee-associated microbiota. Here, we report a high-quality genome sequence for *Bacillus* sp. SDLI1 from the larval gut of the Brazilian stingless bee *S. depilis*.

The bacterium named *Bacillus* sp. SDLI1 was isolated from *S. depilis* larvae that were subjected to surface sterilization (8). Briefly, the insects were submerged in 70% ethanol for 2.5 min, transferred to 5% sodium hypochlorite for 5 min, then transferred to 70% ethanol for 1 min, and washed with sterile water. The lacerated larvae were inoculated in International *Streptomyces* Project medium number 2 agar plates, then incubated at 30°C.

Genomic DNA was extracted from an overnight monoculture of *Bacillus* sp. SDLI1 in lysogeny broth (Lennox, Sigma-Aldrich) using the Wizard genomic DNA purification kit (Promega). A long insert (15- to 20-kb) DNA library was prepared and sequenced at the Duke Center for Genomic and Computational Biology Sequencing and Genomic Technologies Shared Resource core facility. Two single-molecule real-time (SMRT) cells (9) were run on a PacBio RSII instrument using P6-C4 chemistry. The Hierarchical Genome Assembly Process (10) generated a single 4,143,589-bp polished contig with a mean coverage of 338× and mean quality value of 48.5. Manual inspection of the contig revealed that the termini were degenerate, and one end was removed to produce the final circular assembly of 4,134,042 bp with a G-C content of 45.8%.

A cursory inspection of four *Bacillus* sp. SDLI1 housekeeping genes (*dnaJ*, *rpoB*, *gyrA*, and *recA*) revealed that it is a member of the *B. subtilis* group. A comparison of the complete nucleotide sequences of chromosomes within this group suggests that *Bacillus* sp. SDLI1 is most closely related to *Bacillus siamensis*

KCTC 13613[T] (82.7%, GenBank accession no. AJVF00000000.1) and *B. siamensis* XY18 (82.6%, GenBank accession no. LAGT00000000.1), based on *in silico* genome-to-genome distance calculation and reporting the DNA-DNA hybridization value from formula 2 as a metric (11). Genome annotation using RAST (12) predicted 4,298 coding sequences, including 19 rRNA genes and 86 tRNA genes. Finally, antiSMASH (13) analyses indicated that *Bacillus* sp. SDLI1 genome encodes biosynthetic gene clusters with high sequence identity to those that produce fengycin, bacillomycin, bacillaene, macrolactin, surfactin, bacilysin, bacillibactin, and difficidin.

**Nucleotide sequence accession number.** The complete genome sequence has been deposited to the NCBI under the accession number CP013950.

## REFERENCES

1. **Gilliam M.** 1997. Identification and roles of non-pathogenic microflora associated with honey bees. FEMS Microbiol Lett **155**:1–10. http://dx.doi.org/10.1111/j.1574-6968.1997.tb12678.x.
2. **Keller A, Grimmer G, Steffan-Dewenter I.** 2013. Diverse microbiota identified in whole intact nest chambers of the red mason bee *Osmia bicornis* (Linnaeus 1758). PLoS One **8**:e78296. http://dx.doi.org/10.1371/journal.pone.0078296.
3. **Sinacori M, Francesca N, Alfonzo A, Cruciata M, Sannino C, Settanni L, Moschetti G.** 2014. Cultivable microorganisms associated with honeys

of different geographical and botanical origin. Food Microbiol **38:** 284–294. http://dx.doi.org/10.1016/j.fm.2013.07.013.

4. **Wang M, Zhao W-Z, Xu H, Wang Z-W, He S-Y.** 2015. *Bacillus* in the guts of honey bees (*Apis mellifera*; Hymenoptera: Apidae) mediate changes in amylase values. Eur J Entomol **112:**619–624. http://dx.doi.org/10.14411/eje.2015.095.

5. **Sabaté DC, Carrillo L, Audisio MC.** 2009. Inhibition of *Paenibacillus larvae* and *Ascosphaera apis* by *Bacillus subtilis* isolated from honeybee gut and honey samples. Res Microbiol **160:**193–199. http://dx.doi.org/10.1016/j.resmic.2009.03.002.

6. **Benitez LB, Velho RV, de Souza da Motta AS, Segalin J, Brandelli A.** 2012. Antimicrobial factor from *Bacillus amyloliquefaciens* inhibits *Paenibacillus larvae*, the causative agent of American foulbrood. Arch Microbiol **194:**177–185. http://dx.doi.org/10.1007/s00203-011-0743-4.

7. **Menezes C, Vollet-Neto A, Marsaioli AJ, Zampieri D, Fontoura IC, Luchessi AD, Imperatriz-Fonseca VL.** 2015. A Brazilian social bee must cultivate fungus to survive. Curr Biol **25:**2851–2855. http://dx.doi.org/10.1016/j.cub.2015.09.028.

8. **Bacon CW.** 1990. Isolation, culture, and maintenance of endophytic fungi of grasses, p. 259–282. *In* Labeda DP (ed), Isolation of biotechnological organisms from nature. McGraw-Hill, New York, NY.

9. **Eid J, Fehr A, Gray J, Luong K, Lyle J, Otto G, Peluso P, Rank D, Baybayan P, Bettman B, Bibillo A, Bjornson K, Chaudhuri B, Christians F, Cicero R, Clark S, Dalal R, Dewinter A, Dixon J, Foquet M, Gaertner A, Hardenbol P, Heiner C, Hester K, Holden D, Kearns G, Kong X, Kuse R, Lacroix Y, Lin S, Lundquist P, Ma C, Marks P, Maxham M, Murphy D, Park I, Pham T, Phillips M, Roy J, Sebra R, Shen G, Sorenson J, Tomaney A, Travers K, Trulson M, Vieceli J, Wegener J, Wu D, Yang A, Zaccarin D, Zhao P, Zhong F, Korlach J, Turner S.** 2009. Real-time DNA sequencing from single polymerase molecules. Science **323:**133–138. http://dx.doi.org/10.1126/science.1162986.

10. **Chin C-S, Alexander DH, Marks P, Klammer AA, Drake J, Heiner C, Clum A, Copeland A, Huddleston J, Eichler EE, Turner SW, Korlach J.** 2013. Nonhybrid, finished microbial genome assemblies from long-read SMRT sequencing data. Nat Methods **10:**563–569. http://dx.doi.org/10.1038/nmeth.2474.

11. **Meier-Kolthoff JP, Auch AF, Klenk H-P, Göker M.** 2013. Genome sequence-based species delimitation with confidence intervals and improved distance functions. BMC Bioinformatics **14:**60. http://dx.doi.org/10.1186/1471-2105-14-60.

12. **Aziz RK, Bartels D, Best AA, DeJongh M, Disz T, Edwards RA, Formsma K, Gerdes S, Glass EM, Kubal M, Meyer F, Olsen GJ, Olson R, Osterman AL, Overbeek RA, McNeil LK, Paarmann D, Paczian T, Parrello B, Pusch GD, 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. http://dx.doi.org/10.1186/1471-2164-9-75.

13. **Weber T, Blin K, Duddela S, Krug D, Kim HU, Bruccoleri R, Lee SY, Fischbach MA, Müller R, Wohlleben W, Breitling R, Takano E, Medema MH.** 2015. antiSMASH 3.0—a comprehensive resource for the genome mining of biosynthetic gene clusters. Nucleic Acids Res **43:** W237–W243. http://dx.doi.org/10.1093/nar/gkv437.