

Draft Whole Genome Sequence of *Bacillus pumilus* Strain 3-19, a Chemical Mutant Overproducing Extracellular Ribonuclease

Vera Ulyanova, Raihan Shah Mahmud, Elena Dudkina, Valentina Vershinina, Olga Ilinskaya

Institute of Fundamental Medicine and Biology, Kazan (Volga region) Federal University, Kazan, Russia

Here, we present a draft genome sequence of *Bacillus pumilus* strain 3-19. It was derived from soil-isolated *B. pumilus* 7P using chemical mutagenesis and is characterized by elevated production of extracellular ribonuclease which is known to possess different biological activities with potential of applications in experimental research, medicine, and biotechnology.

Received 28 June 2014 Accepted 3 July 2014 Published 24 July 2014

Citation Ulyanova V, Shah Mahmud R, Dudkina E, Vershinina V, Ilinskaya O. 2014. Draft whole genome sequence of *Bacillus pumilus* strain 3-19, a chemical mutant overproducing extracellular ribonuclease. *Genome Announc.* 2(4):e00724-14. doi:10.1128/genomeA.00724-14.

Copyright © 2014 Ulyanova et al. This is an open-access article distributed under the terms of the [Creative Commons Attribution 3.0 Unported license](http://creativecommons.org/licenses/by/3.0/).

Address correspondence to Vera Ulyanova, ulyanova.vera@gmail.com.

Bacillus pumilus strain 3-19 was derived from wild-type strain *Bacillus pumilus* 7P by screening on streptomycin-containing media followed by three-step selection of colonies which were resistant to increasing concentrations of antibiotic (10, 250, and 500 $\mu\text{g/ml}$) and were characterized by overproduction of extracellular ribonuclease. The obtained mutant possessed an almost 10-fold higher level of ribonuclease activity in comparison to the parent strain. Both features make the 3-19 strain suitable for industrial production of ribonuclease which can be used as a potential antitumor and antiviral agent (1, 2) and as a RNA-degrading tool in molecular biology. The 3-19 strain was patented in Russia in 2010 (3) and deposited in the Russian National Collection of Industrial Microorganisms (VKPM) under accession number B3833.

Whole-genome shotgun sequencing of *B. pumilus* 3-19 was performed on a 454 GS Junior system (Roche, Switzerland) with approximately 23-fold overall genome coverage. The sequencing 171,451 reads were assembled with GS De Novo Assembler (version May 2014) resulting in 39 contigs (>200 bp). A calculated genome size was 3,573,949 bp with a G+C content of 41.9 mol%. The genome was annotated using the NCBI Prokaryotic Genome Annotation Pipeline (PGAAP) version 2.6 (<http://www.ncbi.nlm.nih.gov/genomes/static/Pipeline.html>). A total of 3,563 genes and 3,363 coding sequences (CDS) were predicted and 77 RNAs were identified, including 72 tRNAs, 4 rRNAs, and 1 non-coding RNA (ncRNA). The total size of the assembly, G+C content, as well as the numbers of coding sequences and ribosomal, transport, and non-coding RNAs were in good agreement with respective figures for the parent strain of *B. pumilus* 7P (3,582,806 bp; 42 mol%; 3,460 CDS; 77 RNAs, respectively), whose draft genome sequence was reported recently (4).

A brief comparison of two genomes made with RAST version 4.0 (5) revealed that 3,522 coding sequences (95% of total CDSs in the 3-19 strain) are identical in both strains while 179 CDSs (5%)

have certain differences. Regulatory and coding regions of a gene encoding the extracellular ribonuclease have no modifications in the 3-19 strain compared with the 7P strain. A more-detailed comparative analysis of the parent and mutant genomes will provide further insight into the mechanism of acquired streptomycin resistance and the nature of ribonuclease overproduction.

Nucleotide sequence accession number. This whole-genome shotgun project has been deposited in DDBJ/EMBL/GenBank under the accession no. [JOJX00000000](http://dx.doi.org/10.1101/000000). The version described in this paper is the first version.

ACKNOWLEDGMENTS

This work was performed within the Russian Government Program of Competitive Growth of Kazan Federal University and supported by Russian Research Foundation grant 14-14-00522.

We are grateful to the staff of the Genome Center of Kazan Federal University for technical support with 454 sequencing.

REFERENCES

1. Ulyanova V, Vershinina V, Ilinskaya O. 2011. Barnase and binase: twins with distinct fates. *FEBS J.* 278:3633–3643. <http://dx.doi.org/10.1111/j.1742-4658.2011.08294.x>.
2. Shah Mahmud R, Ilinskaya ON. 2013. Antiviral activity of binase against the pandemic influenza A (H1N1) virus. *Acta Naturae.* 5:44–51.
3. Ilinskaya ON, Balaban NP, Vershinina VI, Sharipova MR, Kolpakov AI, Zelenikhin PV. March 2010. Streptomycin-resistant strain of *Bacillus* sp. VKPM B-9862, a producer of extracellular alkaline ribonuclease. RU patent 2384619.
4. Shagimardanova EI, Toymentseva AA, Balaban NP, Mardanov AM, Danilova YV, Gusev OA, Kostryukova E, Karpova I, Manolov A, Alexeev D, Sharipova MR. 2014. Draft genome sequence of *Bacillus pumilus* 7P, isolated from the soil of the Tatarstan Republic, Russia. *Genome Announc.* 2(3):e00599-14. <http://dx.doi.org/10.1128/genomeA.00599-14>.
5. Overbeek R, Olson R, Pusch GD, Olsen GJ, Davis JJ, Disz T, Edwards RA, Gerdes S, Parrello B, Shukla M, Vonstein V, Wattam AR, Xia F, Stevens R. 2014. The SEED and the Rapid Annotation of microbial genomes using Subsystems Technology (RAST). *Nucleic Acids Res.* 42:D206–D214. <http://dx.doi.org/10.1093/nar/gkt1226>.