



# Genome Sequences of *Bacillus thuringiensis* Serovar *kurstaki* Strain BP865 and *B. thuringiensis* Serovar *aizawai* Strain HD-133

Haeyoung Jeong, Soo-Keun Choi, Seung-Hwan Park

Infectious Disease Research Center, Korea Research Institute of Bioscience and Biotechnology (KRIBB), and Biosystems and Bioengineering Program, University of Science and Technology (UST), Daejeon, Republic of Korea

**ABSTRACT** We report the draft genome sequences of two insecticidal strains against lepidopteran pests, *Bacillus thuringiensis* serovar *kurstaki* strain BP865, an isolate from the South Korean phylloplane, and strain HD-133, a reference strain of *B. thuringiensis* serovar *aizawai*.

*Bacillus thuringiensis* is a ubiquitous soil bacterium that produces insecticidal crystal proteins, also known as  $\delta$ -endotoxins (Cry and Cyt toxins), during the sporulation stage (1). Because each type of *B. thuringiensis* toxin is highly active against specific insect larvae, with little or no effect on humans, such toxins have been widely used as environmentally friendly biopesticides for various crops (2).

Strain BP865 (=KCTC 8689P), an isolate from the phylloplane in South Korea, was shown to exhibit both insecticidal and fungicidal activities (J. I. Kim et al., Korean Intellectual Property Office, 28 April 1997). The biochemical and growth characteristics indicate that BP865 is indistinguishable from HD-1, which is the most widely used bioinsecticide and is recognized as the reference standard in the United States (3). Strains belonging to serovar *aizawai*, including HD-133, have elicited special interest owing to their production of a “novel” toxin (CryIc family) that is active against *Spodoptera* spp. (4–7).

Genomic DNA preparation was carried out using a Wizard genomic DNA purification kit (Promega, USA) according to the manufacturer’s instructions. Library construction (average insert size of ~400 bp) and genome sequencing were carried out using an Illumina HiSeq 2000 platform at the National Instrumentation Center for Environmental Management (Seoul, Republic of Korea). Sequence totals of 1.59 Gb and 1.62 Gb ( $2 \times 101$  cycles) were produced from BP865 and HD-133, respectively. A *de novo* genome assembly was applied using the A5-miseq pipeline (8), version 20160825, which yielded 6,408,259 bp in 225 scaffolds with an  $N_{50}$  of 99,607 bp for BP865, and 6,438,280 bp in 214 scaffolds with an  $N_{50}$  of 67,785 bp for HD-133. Genome annotation was carried out using Prokka (9) and NCBI’s Prokaryotic Genome Annotation Pipeline.

Average nucleotide identity analysis using all 78 available *B. thuringiensis* genomes from RefSeq (as of 27 October 2016) indicated that BP865 was closest to serovar *kurstaki* HD-1 (JMHW01; 99.9%), whereas HD-133 was closest to serovar *galleriae* HD-29 (CP010089-99; 99.8%), followed by the two serovar *aizawai* strains Leapio01 (AMXS02; 99.7%) and Hu4-2 (AMXT02; 99.7%). Scaffolds with  $\geq 95\%$  BLAST+ query coverage against complete plasmid sequences for *B. thuringiensis* available from the RefSeq plasmids were placed to putative plasmids. Through this approach, 87 scaffolds from BP865 (910 kb) and 77 scaffolds from HD-133 (687 kb) could be tentatively assigned to the plasmids ([http://genoglobe.kr/kribb/two\\_Bt\\_strains\\_2016](http://genoglobe.kr/kribb/two_Bt_strains_2016)).

**Received** 18 November 2016 **Accepted** 26 November 2016 **Published** 2 February 2017

**Citation** Jeong H, Choi S-K, Park S-H. 2017. Genome sequences of *Bacillus thuringiensis* serovar *kurstaki* strain BP865 and *B. thuringiensis* serovar *aizawai* strain HD-133. *Genome Announc* 5:e01544-16. <https://doi.org/10.1128/genomeA.01544-16>.

**Copyright** © 2017 Jeong et al. This is an open-access article distributed under the terms of the [Creative Commons Attribution 4.0 International license](https://creativecommons.org/licenses/by/4.0/).

Address correspondence to Soo-Keun Choi, [sookeun@kribb.re.kr](mailto:sookeun@kribb.re.kr), or Seung-Hwan Park, [shpark@kribb.re.kr](mailto:shpark@kribb.re.kr).

BP865 encodes  $\delta$ -endotoxin genes for Cry1Aa, Cry1Ab, Cry1Ac, Cry1Ia, and Cry2Ab, whereas HD-133 encodes for Cry1Aa, Cry1Ab, Cry1Ca, Cry1Da, Cry1Ia, Cry2Ab, and Cry9Ea, thereby suggesting that the latter has a wider insecticidal spectrum. However, these include partial or apparently fragmented genes, which necessitates further validation of the complete gene structure. We also identified at least two nonribosomal peptide synthetase (NRPS) genes and two NRPS/polyketide synthetase hybrid genes from both genome assemblies, implying that the potential use of *B. thuringiensis* strains can be extended beyond the biological control of insect pests.

**Accession number(s).** These whole-genome shotgun projects have been deposited in DDBJ/ENA/GenBank under the accession numbers [MOXR000000000](#) (BP865) and [MOXQ000000000](#) (HD-133). The versions described in this paper are the first versions, MOXR01000000 and MOXQ01000000.

## REFERENCES

1. Schnepf E, Crickmore N, Van Rie J, Lereclus D, Baum J, Feitelson J, Zeigler DR, Dean DH. 1998. *Bacillus thuringiensis* and its pesticidal crystal proteins. *Microbiol Mol Biol Rev* 62:775–806.
2. Sanahuja G, Banakar R, Twyman RM, Capell T, Christou P. 2011. *Bacillus thuringiensis*: a century of research, development and commercial applications. *Plant Biotechnol J* 9:283–300. <https://doi.org/10.1111/j.1467-7652.2011.00595.x>.
3. Dulmage HT. 1977. *B. thuringiensis* U.S. assay standard. *Bull Entomol Soc Am* 19:200–202.
4. Lecadet MM, Martouret D. 1987. Host specificity of the *Bacillus thuringiensis*  $\delta$ -endotoxin toward lepidopteran species: *Spodoptera littoralis* Bdv. and *Pieris brassicae* L. *J Invertebr Pathol* 49:37–48. [https://doi.org/10.1016/0022-2011\(87\)90123-6](https://doi.org/10.1016/0022-2011(87)90123-6).
5. Chak KF, Ellar DJ. 1987. Cloning and expression in *Escherichia coli* of an insecticidal crystal protein gene from *Bacillus thuringiensis* var. *aizawai* HD-133. *J Gen Microbiol* 133:2921–2931. <https://doi.org/10.1099/00221287-133-10-2921>.
6. Lecadet MM, Sanchis V, Menou G, Rabot P, Lereclus D, Chauvaux J, Martouret D. 1988. Identification of a delta-endotoxin gene product specifically active against *Spodoptera littoralis* Bdv. among proteolysed fractions of the insecticidal crystals of *Bacillus thuringiensis* subsp. *aizawai* 7.29. *Appl Environ Microbiol* 54:2689–2698.
7. Sanchis V, Lereclus D, Menou G, Chauvaux J, Lecadet MM. 1988. Multiplicity of delta-endotoxin genes with different insecticidal specificities in *Bacillus thuringiensis aizawai* 7.29. *Mol Microbiol* 2:393–404. <https://doi.org/10.1111/j.1365-2958.1988.tb00044.x>.
8. Coil D, Jospin G, Darling AE. 2015. A5-miseq: an updated pipeline to assemble microbial genomes from Illumina MiSeq data. *Bioinformatics* 31:587–589. <https://doi.org/10.1093/bioinformatics/btu661>.
9. Seemann T. 2014. Prokka: rapid prokaryotic genome annotation. *Bioinformatics* 30:2068–2069. <https://doi.org/10.1093/bioinformatics/btu153>.