JOURNAL OF BACTERIOLOGY, May 2011, p. 2379–2380 Vol. 193, No. 9
0021-9193/11/$12.00 doi:10.1128/JB.00267-11


# Complete Genome Sequence of *Bacillus thuringiensis* Serovar finitimus Strain YBT-020▽

Yiguang Zhu, Hui Shang, Qian Zhu, Fang Ji, Pengxia Wang, Jingjing Fu, Yun Deng, Chengchen Xu, Weixing Ye, Jinshui Zheng, Lei Zhu, Lifang Ruan, Donghai Peng, and Ming Sun*

*State Key Laboratory of Agricultural Microbiology, College of Life Science and Technology, Huazhong Agricultural University, Wuhan 430070, People's Republic of China*

Received 24 February 2011/Accepted 28 February 2011

***Bacillus thuringiensis*** **is a Gram-positive, spore-forming bacterium that forms parasporal crystals at the onset of the sporulation phase of its growth. Here, we report the complete genome sequence of *B. thuringiensis* serovar finitimus strain YBT-020, whose parasporal crystals consist of Cry26Aa and Cry28Aa crystal proteins and are located between the exosporium and the spore coat and remain adhering to the spore after sporulation.**

Typically, the parasporal crystals of *Bacillus thuringiensis* are deposited against the forespore, develop outside the exosporium, and are separated from spores after sporulation (12). However, in a few cases, such as in *B. thringiensis* serovar finitimus strains (2, 7, 13) and serovar oyamesis strain LBIT-113 (9), the parasporal crystals are located between the exosporium and the spore coat and remain adhering to the spore after sporulation. We have previously described this phenotype as spore-crystal association (SCA) and found that the crystal protein genes themselves (*cry26Aa* and *cry28Aa*) and their promoters cannot determine crystal formation inside the exosporium (7). Using a complementation test, the genes determining the SCA phenotype have been isolated from strain YBT-020 (unpublished).

The genome sequence of *B. thuringiensis* strain YBT-020 was determined at the Beijing Genomics Institute (BGI; Shenzhen, China) with a strategy of Solexa paried-end sequencing technology. Filtered paired-end reads (698.65 Mb in total) were obtained to reach a depth of 122-fold coverage of the genome with an Illumina Solexa GA IIx instrument, and about 96.53% of the reads were assembled into 105 contigs (defined as >500 bp) by using the SOAPdenovo alignment tool (http://soap.genomics.org.cn/index.html#intro2). Gaps between contigs were closed by custom primer walks, long-distance PCR amplification, or inverse PCR amplification followed by DNA sequencing with an ABI 3730 sequencer. tRNA and rRNA genes were identified by tRNAscan-SE (10) and RNAmmer (8), respectively.

The complete genome of *B. thuringiensis* strain YBT-020 contains three replicons: a circular chromosome (5,355,490 bp) containing 5,477 open reading frames (ORFs) and two circular plasmids, which were named pBMB26 (187,880 bp) and pBMB28 (139,013 bp), carrying 200 and 149 predicted ORFs, respectively. The G+C content of the chromosome is 35.3%, while those of plasmids pBMB26 and pBMB28 are 33.1% and 33.9%, respectively. This genome carries 107 tRNAs and 14 rRNA operons. Plasmid pBMB26 harbors *cry26Aa* (YBT020_27999) and *cry28Aa* (YBT020_28569) crystal protein genes, while pBMB28 harbors another *cry28Aa* (YBT020_29291) gene. The two *cry28Aa* genes share 99% sequence identity. There are also four truncated crystal protein genes, YBT020_27744, YBT020_27749, YBT020_27819, and YBT020_27994, which are homologous to *cry7Aa1*, *cry8Ka2*, *cry28Aa1*, and *cry26Aa1*, respectively. All the putative crystal genes are located on plasmid pBMB26.

Based on the phylogenetic trees from the chromosome that were inferred by PTreeRec (3), strain *B. thuringiensis* YBT-020 was found to be closest to *B. thuringiensis* Al Hakam (1), *B. thuringiensis* serovar konkukian 97-27 (4), and *B. thuringiensis* BMB171 (5), in order of closeness. The chromosome of strain YBT-020 was phylogenetically more closely related to that of *B. anthracis* Ames (11) than that of *B. cereus* ATCC 14579 (6).

This is the first genome sequence of *B. thuringiensis* harboring crystal protein genes, and its availability will facilitate the understanding of evolutionary relationships among *B. cereus* group organisms.

**Nucleotide sequence accession numbers.** The sequences of the *B. thuringiensis* serovar finitimus strain YBT-020 have been deposited in GenBank under accession numbers CP002508 (chromosome), CP002509 (plasmid pBMB26), and CP002510 (plasmid pBMB28).

This study was supported by the National Natural Science Foundation of China (grants 30870066 and 30470026), National High Technology Research and Development Program (863 Program) of China grants 2011AA10A203 and 2006AA02Z174, the National Basic Research Program (973 Program) of China (grant 2009CB118902), Genetically Modified Organisms Breeding Major Projects of China grant 2009ZX08009-032B, and the China 948 Program of the Ministry of Agriculture (grant 2011-G25), the Ministry of Forestry (grant 2006-4-41), and the China National Fundamental Fund of Personnel Training (grant J0730649).

* Corresponding author. Mailing address: State Key Laboratory of Agricultural Microbiology, College of Life Science and Technology, Huazhong Agricultural University, Wuhan 430070, People's Republic of China. Phone: 86-27-87283455. Fax: 86-27-87280670. E-mail: m98sun@mail.hzau.edu.cn.

▽ Published ahead of print on 11 March 2011.

## REFERENCES

1. **Challacombe, J. F., et al.** 2007. The complete genome sequence of *Bacillus thuringiensis* Al Hakam. J. Bacteriol. **189:**3680–3681.
2. **Debro, L., P. C. Fitz-James, and A. Aronson.** 1986. Two different parasporal

inclusions are produced by *Bacillus thuringiensis* subsp. *finitimus.* J. Bacteriol. **165:**258–268.
3. **Deng, R., et al.** 2006. PTreeRec: Phylogenetic Tree Reconstruction based on genome BLAST distance. Comput. Biol. Chem. **30:**300–302.
4. **Han, C. S., et al.** 2006. Pathogenomic sequence analysis of *Bacillus cereus* and *Bacillus thuringiensis* isolates closely related to *Bacillus anthracis*. J. Bacteriol. **188:**3382–3390.
5. **He, J., et al.** 2010. Complete genome sequence of *Bacillus thuringiensis* mutant strain BMB171. J. Bacteriol. **192:**4074–4075.
6. **Ivanova, N., et al.** 2003. Genome sequence of *Bacillus cereus* and comparative analysis with *Bacillus anthracis*. Nature **423:**87–91.
7. **Ji, F., et al.** 2009. Promoters of crystal protein genes do not control crystal formation inside exosporium of *Bacillus thuringiensis ssp. finitimus* strain YBT-020. FEMS Microbiol. Lett. **300:**11–17.
8. **Lagesen, K., et al.** 2007. RNAmmer: consistent and rapid annotation of ribosomal RNA genes. Nucleic Acids Res. **35:**3100–3108.
9. **Lopez-Meza, J. E., and J. E. Ibarra.** 1996. Characterization of a novel strain of *Bacillus thuringiensis*. Appl. Environ. Microbiol. **62:**1306–1310.
10. **Lowe, T. M., and S. R. Eddy.** 1997. tRNAscan-SE: a program for improved detection of transfer RNA genes in genomic sequence. Nucleic Acids Res. **25:**955–964.
11. **Read, T. D., et al.** 2003. The genome sequence of *Bacillus anthracis* Ames and comparison to closely related bacteria. Nature **423:**81–86.
12. **Schnepf, E., et al.** 1998. *Bacillus thuringiensis* and its pesticidal crystal proteins. Microbiol. Mol. Biol. Rev. **62:**775–806.
13. **Wojciechowska, J. A., E. Lewitin, L. P. Revina, I. A. Zalunin, and G. G. Chestukhina.** 1999. Two novel delta-endotoxin gene families *cry26* and *cry28* from *Bacillus thuringiensis* ssp. *finitimus*. FEBS Lett. **453:**46–48.