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GENOME ANNOUNCEMENT

Complete Genome Sequence of *Bacillus anthracis* H9401, an Isolate from a Korean Patient with Anthrax

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***Bacillus anthracis* H9401 (NCCP 12889) is an isolate from a Korean patient with gastrointestinal anthrax. The whole genome of H9401 was sequenced. It is a circular chromosome containing 5,480 open reading frames (ORFs) and two plasmids, pXO1 containing 202 ORFs and pXO2 containing 110 ORFs. H9401 shows high pathogenicity and genome sequence similarity to Ames Ancestor.**

Bacillus anthracis is the causative agent of anthrax, an infectious, often fatal disease of both animals and humans. *B. anthracis* has become notorious as a bioweapon because of its tough, environmentally resistant endospore and its ability to cause lethal inhalational anthrax. During the course of infection, endospores are taken up by alveolar macrophages, where they germinate in the phagolysosomal compartment. A tripartite exotoxin and a poly-γ-D-glutamic acid capsule, expressed as the major virulence determinant, are essential for full pathogenicity and are encoded by two plasmids, pXO1 and pXO2 (2, 12, 15, 17).

Genotyping methods such as multiple-locus variable-number tandem repeat analysis and an additional typing system based on 12 canonical single-nucleotide polymorphisms have been developed to analyze genetic variation and relatedness (5, 6, 8, 14, 18, 19, 23). The whole chromosome sequence of *B. anthracis* Ames Ancestor isolated in the United States was completed in 2003, and the sequence contributes to the identification of genes that contribute to virulence either by encoding functions for bacterial survival or by enhancing immune evasion and causing host damage. *B. anthracis* H9401 is a fully virulent isolate obtained from a Korean patient with gastrointestinal anthrax in 1994 that has been used as a challenge strain in studies including recombinant anthrax vaccine under development in the Republic of Korea (11, 13, 20, 22).

Genome sequencing was performed with 454 GS-FLX (454 Life Sciences Corporation, Basel, Switzerland), and the results were analyzed using GLIMMER3 (4), tRNAscan-SE (16), and RNAmmer (10). The functions of encoding genes were annotated by using the UniRef90 (24), NCBI nr (1), COG (25), and KEGG (7) databases. Genome structure comparisons were performed by using ACT (3) and MUMMER (9).

Analysis of the H9401 genome sequence produced a total of 467,369 reads with an average read length of 375.3 bp. The complete genome sequence had 30-fold coverage of the chromosome, 88.9-fold coverage of pXO1, and 48.3-fold coverage of pXO2. This coverage level suggests a pXO1-pXO2-chromosome molecular ratio of 3:2:1 in H9401, identical to those in Ames Florida and Ames Ancestor (21).

The sequence homology between H9401 and Ames Ancestor was 99.679% (chromosome, 99.686%; pXO1, 99.570%; pXO2, 99.584%). The amino acid sequence homology was 99.870% (percent amino acid homology: chromosome, 99.876%; pXO1, 99.838%; pXO2, 99.824%). The H9401 genome consists of one circular chromosome (5,218,947 bp containing 5,480 predicted ORFs) and two plasmids, pXO1 (181,700 bp containing 202 predicted ORFs) and pXO2 (94,824 bp containing 110 predicted ORFs). The estimated coding density over the chromosome is 85%, and the average G+C content is 35.0%. The genome of Ames Ancestor consists of one similar-size chromosome (5,227,419 bp with 5,309 encoding genes), pXO1 (1,811,677 bp long and containing 177 ORFs), and pXO2 (94,830 bp long and containing 98 ORFs). The H9401 genome encodes 96 tRNAs and 10 copies of 16S-23S-5S rRNA operons, while that of Ames Ancestor encodes 95 tRNAs and 11 copies of rRNA operons. H9401 has a smaller chromosome size than Ames Ancestor, as short as ~8.5 kbp.

The high pathogenicity and genome sequence similarity of *B. anthracis* H9401 to Ames Ancestor has enabled the use of this Korean isolate as a reference for efficacy testing of anthrax vaccine candidates in the Republic of Korea instead of Ames Ancestor, whose international transfer is prohibited.

Nucleotide sequence accession numbers. The sequences of the *B. anthracis* H9401 main chromosome and plasmids pXO1 and pXO2 have been deposited in the NCBI GenBank database under accession numbers CP002091.1 for the chromosome, CP002092.1.1 for pXO1, and CP002093.1 for pXO2.

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