

SUPPORTING INFECTIOUS DISEASE RESEARCH

Certificate of Analysis for NR-10308

Genomic DNA from Bacillus anthracis, Strain Sterne △GBAA1975-76

Catalog No. NR-10308

This reagent is the tangible property of the U.S. Government.

Product Description: Genomic DNA was isolated from a preparation of *Bacillus anthracis* (B. *anthracis*), strain Sterne Δ GBAA1975-76, a markerless, nonpolar, double deletion mutant of the response regulator/histidine kinase genes from the toxigenic acapsulate original Sterne strain (34F2).

Lot¹: 58253183 Manufacturing Date: 22DEC2008

TEST	SPECIFICATIONS	RESULTS
Sequencing of 16S Ribosomal RNA Gene (~1440 base pairs)	Identical to BEI Resources NR-10004 Consistent with <i>B. cereus</i> group	Identical to BEI Resources NR-10004 Consistent with <i>B. cereus</i> group ²
Presence or Absence of Plasmids Confirmed by PCR Amplification pXO1 (aat) pXO2 (at, capA, capB, capC)	Positive Negative	Positive Negative
Agarose Gel Electrophoresis	High molecular weight chromosomal DNA	High molecular weight chromosomal DNA (Figure 1)
Content by PicoGreen® Measurement	4 to 6 μg in 25 to 100 μL per vial	5.6 μg in 58 μL per vial (97 μg/mL)
Functional Activity by PCR Amplification 16S ribosomal RNA gene Virulence markers on plasmid pXO1 (aat)	~ 1500 bp amplicon ~ 125 bp amplicon	~ 1500 bp amplicon ~ 125 bp amplicon
OD ₂₆₀ /OD ₂₈₀ Ratio	1.7 to 1.9	1.8
Bacterial Inactivation 10% of total yield plated on Tryptic Soy Agar with 5% sheep blood ^{3,4}	No viable bacteria detected	No viable bacteria detected

¹B. anthracis, strain ΔGBAA1975-76 was deposited by Philip Hanna, Associate Professor, Department of Microbiology and Immunology, University of Michigan Medical School, Ann Arbor, Michigan. The bacterial preparation used for extraction of genomic DNA was produced by Tryptic Soy Broth culture of the deposited material. After incubation for 24 hours at 37°C and aerobic atmosphere, genomic DNA was extracted using proprietary technology.

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Date: 25 AUG 2009 **Signature:** Signature on File

Title: Technical Manager, BEI Authentication or designee

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC®'s knowledge.

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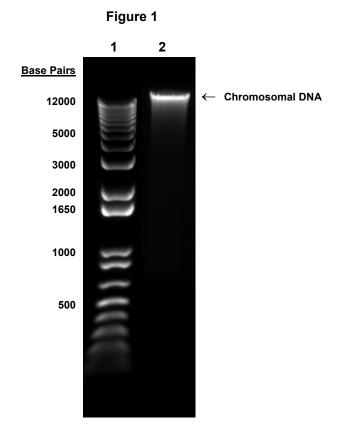
²Bacillus cereus group species (*B. cereus, B. thuringiensis, B. mycoides*, and *B. anthracis*) cannot be classified based on 16S sequence [Spencer, R. C. "Bacillus anthracis." J. Clin. Pathol. 56 (2003): 182-187. PubMed: 12610093].

³7 days at 37°C in an aerobic atmosphere

⁴An extraction procedure was used that has been shown to consistently inactivate 100% of Gram-negative bacteria.

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Lane 1: Invitrogen™ TrackIt™ 1 Kb Plus DNA Ladder

Lane 2: 200 ng of NR-10308

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