

## Certificate of Analysis for NR-10446

## Genomic DNA from Bacillus anthracis, Strain Weybridge

## Catalog No. NR-10446

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 Weybridge. Strain Weybridge is an avirulent strain that contains the toxigenic pXO1 plasmid but lacks the pXO2 capsule plasmid.

Lot<sup>1</sup>: 58646093 Manufacturing Date: 09JUN2009

TEST	SPECIFICATIONS	RESULTS
Sequencing of 16S Ribosomal RNA Gene (~ 720 base pairs)	Consistent with B. cereus group	Consistent with <i>B. cereus</i> group <sup>2,3</sup>
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 μg in 41 μL per vial (123 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 <sub>260</sub> /OD <sub>280</sub> Ratio	1.7 to 1.9	1.8
Bacterial Inactivation  10% of total yield plated on Tryptic Soy Agar with  5% sheep blood <sup>4.5</sup>	No viable bacteria detected	No viable bacteria detected

<sup>&</sup>lt;sup>1</sup>NR-10446 was produced by Tryptic Soy Broth culture of NR-10350 (Lot: 58485551). After incubation for 24 hours at 37°C and aerobic atmosphere, genomic DNA was extracted using proprietary technology.

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**Date:** 07 OCT 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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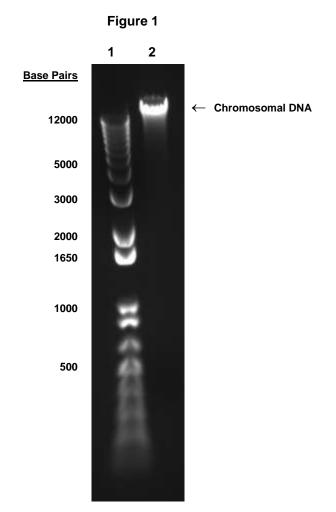
<sup>&</sup>lt;sup>2</sup>Bacillus cereus group species (*B. cereus, B. thuringiensis, B. mycoides*, and *B. anthracis*) cannot be classified based on 16S sequence [Spencer, R. C. "Bacillus anthracis." J. Clin. Pathol. 56 (2003): 182-187. PubMed: 12610093].

<sup>&</sup>lt;sup>3</sup>Also consistent with *B. coagulans* 

<sup>&</sup>lt;sup>4</sup>6 days at 37°C in an aerobic atmosphere

<sup>&</sup>lt;sup>5</sup>An 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-10446

800-359-7370