

**Genomic DNA from *Bacillus anthracis*, Strain Sterne BA721 ( $\Delta$ lef243/ $\Delta$ cya244)**

**Catalog No. NR-9543**

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 BA721 ( $\Delta$ lef243/ $\Delta$ cya244). The designation BA721 refers to the numbering system used in the Stibitz laboratory.

**Lot<sup>1</sup>: 58109382**

**Manufacturing Date: 27FEB2008**

TEST	SPECIFICATIONS	RESULTS
<b>Sequencing of 16S Ribosomal RNA Gene (~ 1380 bp)</b>	Identical to BEI Resources NR-9400 Consistent with <i>B. cereus</i> group <sup>2</sup>	Identical to BEI Resources NR-9400 Consistent with <i>B. cereus</i> group <sup>2</sup>
<b>Presence or Absence of Plasmids Confirmed by PCR Amplification</b> pXO1 pXO2	Positive Negative	Positive Negative
<b>Agarose Gel Electrophoresis</b>	High molecular weight chromosomal DNA	High molecular weight chromosomal DNA (Figure 1)
<b>Content by PicoGreen<sup>®</sup> Measurement</b>	4 to 6 µg in 25 to 100 µL per vial	5.0 µg in 37 µL per vial (136 µg/mL)
<b>PCR Assay of Extracted DNA</b> 16S ribosomal RNA gene Specific chromosomal marker <sup>3</sup> Presence of virulence plasmids <sup>4</sup> pXO1 (two targets) pXO2 (three targets) Verification of $\Delta$ lef/ $\Delta$ cya	~ 1500 bp amplicon; ~ 555 bp amplicon Amplicon present  Amplicons present No amplicons No amplicons	~ 1500 bp amplicon; ~ 555 bp amplicon Amplicon present  Amplicons present No amplicons No amplicons
<b>OD<sub>260</sub>/OD<sub>280</sub> Ratio</b>	1.7 to 1.9	1.8
<b>Bacterial Inactivation</b> 10% of total yield plated on Tryptic Soy Agar with 5% sheep blood <sup>5,6</sup>	No viable bacteria detected	No viable bacteria detected

<sup>1</sup>*B. anthracis*, strain Sterne BA721 ( $\Delta$ lef243/ $\Delta$ cya244) was deposited by Earle Scott Stibitz, Division of Bacterial, Parasitic, and Allergenic Products, Center for Biologics Evaluation and Research, Food and Drug Administration, Bethesda, Maryland. The bacterial preparation used for extraction of genomic DNA was produced by Tryptic Soy Broth culture of the depositor's material. After incubation for 24 hours at 37°C and aerobic atmosphere, genomic DNA was extracted using proprietary technology.

<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>3</sup>This product was verified to a species level using a proprietary (Patent Pending) PCR-based assay to a *Bacillus anthracis*-specific genetic mutation capable of differentiating *B. anthracis* from the remainder of the *B. cereus* group.

<sup>4</sup>Plasmids were verified using a proprietary (Patent Pending) PCR-based assay to a *Bacillus anthracis*-plasmids pXO1 and pXO2.

<sup>5</sup>7 days at 37°C in an aerobic atmosphere

<sup>6</sup>An extraction procedure was used that has been shown to consistently inactivate 100% of Gram-positive and Gram-negative bacteria.

**Date:** 05 SEP 2012

**Signature:**



**Title:**

Technical Manager, BEI Authentication or designee

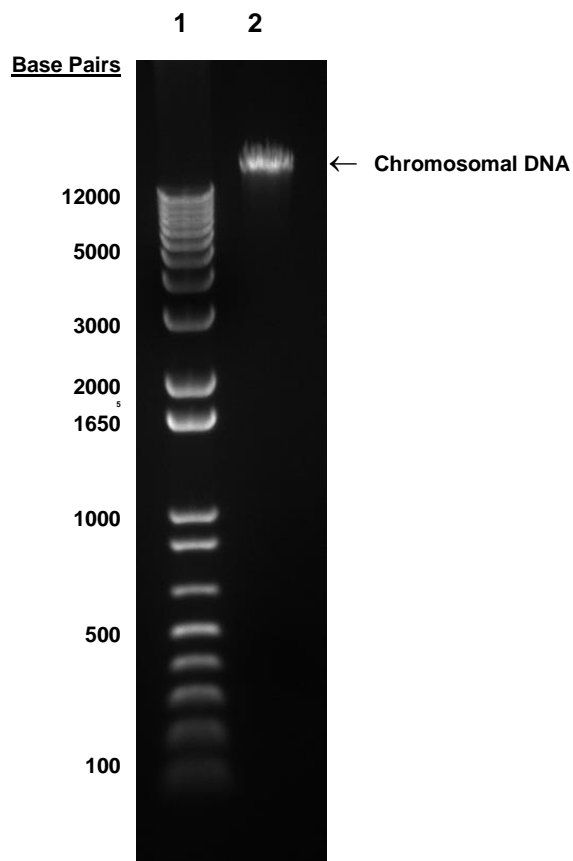
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You are authorized to use this product for research use only. It is not intended for human use.



Figure 1



Lane 1: Invitrogen™ TrackIt™ 1 Kb Plus DNA Ladder  
Lane 2: 200 ng of NR-9543