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SUPPORTING INFECTIOUS DISEASE RESEARCH

Bacillus cereus, Strain VD014

Catalog No. NR-22141

Product Description: Bacillus cereus (B. cereus), strain VD014 was isolated in 2008 from a soil sample collected in Andalusia, Spain. Strain VD014 is reported to contain a pXO1-like plasmid.

Lot¹: 61966651

Manufacturing Date: 21AUG2013

TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis ²		
Cellular morphology	Gram-positive rods	Gram-positive rods
Colony morphology ³	Report results	Irregular, flat, undulate, rough and cream (Figure 1)
Motility ⁴	Motile	Motile
Hemolysis ⁵	Report results	β-hemolytic
Biochemical characterization ⁶⁻⁸		
Production of acid from trehalose	Positive	Positive
Production of acid from salicin	Report results	Positive
Production of acid from glycerol	Report results	Negative
Nitrate reduction	Report results	Positive
Arginine dihydrolase activity	Report results	Positive
PCR Assay of Extracted DNA ⁹		
16S ribosomal RNA gene	~ 560 base pair amplicon	~ 560 base pair amplicon
	~ 1500 base pair amplicon	~ 1500 base pair amplicon
<i>B. anthracis</i> specific chromosomal marker ¹⁰	No amplicon	No amplicon
Presence of virulence plasmid markers ¹¹		
pXO1 (four targets)	Report results	One amplicon present
pXO2 (three targets)	Report results	No amplicons
Genotypic Analysis Sequencing of 16S ribosomal RNA gene (~ 1500 base pairs)	Consistent with <i>B. cereus</i> group	Consistent with <i>B. cereus</i> group ¹²
Viability (post-freeze) ³	Growth	Growth

¹NR-22141 was produced by inoculation of the deposited material into Nutrient broth and grown 24 hours at 37°C in an aerobic atmosphere. After one passage the broth inoculum was added to kolles which were grown 24 hours at 37°C in an aerobic atmosphere to produce this lot. Purity of this lot was assessed for 7 days under propagation conditions.

²Presumptive identification of *B. cereus* was performed using phenotypic tests that eliminate other *B.* cereus group (*B. cereus, B. anthracis, B. thuringiensis* and *B. mycoides*) members (see footnotes 4,7,8).

³24 hours at 37°C in an aerobic atmosphere on Nutrient agar

⁴24 hours at 37°C in an aerobic atmosphere on motility test media with triphenyltetrazolium chloride (TTC). In the *B. cereus* group, *B. cereus* and *B. thuringiensis* are motile, whereas *B. anthracis* and *B. mycoides* are non-motile.

⁵24 hours at 37°C in an aerobic atmosphere on Tryptic Soy agar with 5% sheep blood

⁶Negative tests are observed for >7 days.

⁷B. anthracis is negative for glycerol and salicin.

⁸*B. thuringiensis* is positive for arginine dyhydrolase activity.

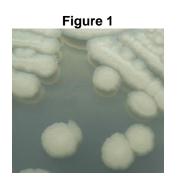
⁹DNA was extracted from a broth culture produced from NR-22141 lot 61966651.

¹⁰A proprietary (Patent Pending) PCR-based assay capable of differentiating *B. anthracis* from the remainder of the *B. cereus* group was used to further eliminate *B. anthracis* as a possible species.

¹¹Presence of markers known to be found on virulence plasmids were verified using a proprietary (Patent Pending) PCR-based assay.

¹²Bacillus cereus group species cannot be classified based on 16S sequence (Spencer, R. C. "Bacillus anthracis." <u>J. Clin. Pathol.</u> 56 (2003): 182-187. PubMed: 12610093). bei resources

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Date: 14 MAR 2014

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