

Certificate of Analysis for NR-52257

Bacillus coagulans, Strain NRS 609

Catalog No. NR-52257

(Derived from ATCC® 7050™)

Product Description:

Bacillus coagulans (B. coagulans), strain NRS 609 was deposited to ATCC[®] in 1961 by Dr. N. R. Smith. The strain was originally isolated by B. W. Hammer in 1915. NR-52257 lot 70033086 was produced by inoculation of ATCC[®] 7050[™] lot 70025814 into Nutrient broth and grown for 1 day at 37°C in an aerobic atmosphere. Broth inoculum was added to Nutrient agar kolles, which were grown for 1 day at 37°C in an aerobic atmosphere to produce this lot. Quality control testing was completed under propagation conditions unless otherwise noted.

Lot: 70033086 Manufacturing Date: 12FEB2020

TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis		
Cellular morphology	Gram-positive rods	Gram-positive rods
Colony morphology	Report results	Circular, low convex, entire, smooth and cream (Figure 1)
Hemolysis 1 day at 37°C in an aerobic atmosphere on Tryptic Soy agar with 5% defibrinated sheep blood	Report results	Non-hemolytic
Motility (wet mount) Biochemical tests	Report results	Motile
Catalase	Report results	Positive
VITEK® MS (MALDI-TOF)	B. coagulans	B. coagulans (99.9%)
Genotypic Analysis		
Sequencing of 16S ribosomal RNA gene (~ 1290 base pairs)	≥ 99% sequence identity to <i>B. coagulans</i> , strain NRS 609 (GenBank: NR_115727.1)	99.9% sequence identity to <i>B. coagulans</i> , strain NRS 609 (GenBank: NR_115727.1)
Digital DNA-DNA hybridization (dDDH) ¹	≥ 70% for species identification	B. coagulans (99.9%) ²
Purity (post-freeze) 7 days at 37°C in an aerobic atmosphere with 5% CO ₂ on Tryptic Soy agar with 5% defibrinated sheep blood	Growth consistent with expected colony morphology	Growth consistent with expected colony morphology
Viability (post-freeze)	Growth	Growth

¹Relatedness between bacterial strains has traditionally been determined using DDH. For additional information, refer to Auch, A. F., et al. "Digital DNA-DNA Hybridization for Microbial Species Delineation by Means of Genome-to-Genome Sequence Comparison." <u>Stand. Genomic Sci.</u> 2 (2010): 117-134. PubMed: 21304684. *B. coagulans*, strain ATCC 7050 (GenBank: ATUM00000000.1) was used for dDDH analysis.

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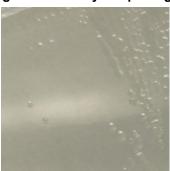
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²The whole genome of *B. coagulans*, strain NRS 609 (3.32 megabase pairs) was sequenced using the Illumina® MiSeq® system. *De novo* contig sequences were generated using Unicycler v0.4.8-beta.



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Figure 1: Colony Morphology



/Heather Couch/ Heather Couch

13 APR 2021

Program Manager or designee, ATCC Federal Solutions

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