

Certificate of Analysis for NR-51333

Bacillus subtilis, Strain 1S28

Catalog No. NR-51333

Product Description: *Bacillus subtilis (B. subtilis)*, strain 1S28 is a sporulation mutant of *B. subtilis*, strain Marburg 168.

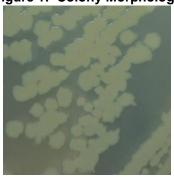
Lot¹: 70017408 Manufacturing Date: 20JUL2018

TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis ²		
Cellular morphology	Gram-positive rods	Gram-positive rods
Colony morphology	Report results	Circular, low convex, undulate, smooth and cream (Figure 1)
Motility	Report results	Motile ³
Hemolysis ⁴	Report results	Non-hemolytic
VITEK® MS (MALDI-TOF)	B. subtilis	B. subtilis (50%) ⁵ and
		B. amyloliquefaciens (50%) ⁵
Genotypic Analysis		
Sequencing of 16S ribosomal RNA gene	≥ 99% sequence identity to	99.9% sequence identity to
(~ 1480 base pairs)	B. subtilis type strain	B. subtilis type strain
	(GenBank: AL009126)	(GenBank: AL009126) ⁶
Purity (post-freeze) ⁷	Growth consistent with expected colony morphology	Growth consistent with expected colony morphology
Viability (post-freeze) ²	Growth	Growth

¹NR-51333 was produced by inoculation of BEI Resources HMC-616 lot 60339796 into Tryptic Soy broth and grown for 1 day at 37°C in an aerobic atmosphere. Broth inoculum was added to Tryptic Soy agar kolles, which were grown for 1 day at 37°C in an aerobic atmosphere to produce this lot.

⁷Purity of this lot was assessed for 7 days at 37°C in an aerobic atmosphere with 5% CO₂ on Tryptic Soy agar.





BEI Resources

www.beiresources.org

E-mail: contact@beiresources.org

Tel: 800-359-7370 Fax: 703-365-2898

²1 day at 37°C in an aerobic atmosphere on Tryptic Soy agar

³Motility test performed in Remel™ Motility Test Medium w/TTC Indicator for 1 day at 37°C in an aerobic atmosphere.

⁴1 day at 37°C in an aerobic atmosphere on Tryptic Soy agar with 5% defibrinated sheep blood

⁵MALDI-TOF MS is inadequate in distinguishing *B. subtilis* from *B. amyloliquefaciens*. *B. amyloliquefaciens* is a member of the *Bacillus subtilis* group comprised of closely related species that cannot be easily distinguished based on phenotypic differences or sequencing of the 16S ribosomal RNA gene. For additional information, please refer to Shu, L. J. and Y. L. Yang. "*Bacillus* Classification Based on Matrix-Assisted Laser Desorption Ionization Time-of-Flight Mass Spectrometry-Effects of Culture Conditions." <u>Sci. Rep.</u> 7 (2017): 15546. PubMed: 29138467.

⁶Also consistent with other *Bacillus* species



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/Heather Couch/ Heather Couch

20 DEC 2018

Program Manager or designee, ATCC Federal Solutions

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