

Certificate of Analysis for HM-318

Clostridium bolteae, Strain WAL-14578

Catalog No. HM-318

Product Description:

Clostridium bolteae (C. bolteae), strain WAL-14578 was isolated from the stool of an autistic male child. HM-318 was produced by inoculation of BEI Resources seed lot 60110260 into Modified Chopped Meat broth and grown for 3 days at 37°C in an anaerobic atmosphere. Broth inoculum was added to Modified Chopped Meat broth, which was grown for 2 days at 37°C in an anaerobic atmosphere to produce this lot. Quality control testing was completed under propagation conditions unless otherwise noted.

<u>Note</u>: Quality control of HMP material is only performed to demonstrate that the material distributed by BEI Resources is identical to the deposited material. It should not be considered a complete characterization of the deposited organism.

Lot: 70036700 Manufacturing Date: 01JUL2020

TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis Cellular morphology ¹ 3 days at 37°C in an anaerobic atmosphere in Tryptic Soy agar with 5% defibrinated sheep blood	Report results	Gram-negative rod
Colony morphology 3 days at 37°C in an anaerobic atmosphere in Tryptic Soy agar with 5% defibrinated sheep blood	Report results	Circular, convex, undulate, smooth and gray (Figure 1)
Motility (wet mount)	Report results	Motile
Genotypic Analysis Sequencing of 16S ribosomal RNA (rRNA) gene (~ 1390 base pairs)	≥ 99% identical to <i>C. bolteae</i> , strain WAL-14578 (GenBank: ADLI01000057.1)	99.9% identical to <i>C. bolteae</i> , strain WAL-14578 (GenBank: ADLI01000057.1)
Purity (post-freeze) Anaerobic 7 days at 37°C in an anaerobic atmosphere in Tryptic Soy agar with 5% defibrinated sheep blood Aerobic 7 days at 37°C in an aerobic atmosphere with 5% CO ₂ in Tryptic Soy agar with 5% defibrinated sheep blood	Growth consistent with expected colony morphology Report results	Growth consistent with expected colony morphology No growth
Viability (post-freeze) 3 days at 37°C in an anaerobic atmosphere in Tryptic Soy agar with 5% defibrinated sheep blood	Growth	Growth

¹In Clostridial cultures, a decrease in peptidoglycan thickness occurs during growth, thus aged cultures may stain Gram-negative or Gram-variable (Beveridge, T. J. "Mechanism of Gram Variability in Select Bacteria." <u>J. Bacteriol.</u> 172 (1990): 1609-1620. PubMed: 1689718.).

Figure 1: Colony Morphology



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Program Manager or designee, ATCC Federal Solutions

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