b|e|i RESOURCES

SUPPORTING INFECTIOUS DISEASE RESEARCH

Certificate of Analysis for NR-19045

Mycobacterium tuberculosis, Strain HN4699

Catalog No. NR-19045

Product Description: *Mycobacterium tuberculosis* (*M. tuberculosis*), strain HN4699 was isolated in 2007 from human pulmonary tissue in Texas, USA. Strain HN4699 was deposited as a multi-drug resistant (MDR) strain of tuberculosis with resistance to rifabutin, isoniazid and rifampicin.

Lot[1]: 63383545 **Manufacturing Date: 21MAY2015**

TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis[2]		
Cellular morphology	Gram-positive rods	Gram-positive rods
Colony morphology[3]	Report results	Irregular, slight peaked, undulate, rough and cream (Figure 1)
Growth rate	≥ 7 days	20 days
Growth at 26°C	Negative	Negative
Growth at 37°C	Positive	Positive
Acid-fast stain	Positive (red colonies)	Positive (red colonies)
Pigmentation in the dark (Scotochromogen)	Negative (no pigment)	Negative (no pigment)
Photoinduction for 1 hour (Photochromogen)	Negative (no pigment)	Negative (no pigment)
Nonchromogen (no pigment)	Positive (no pigment)	Positive (no pigment)
Biochemical tests		
Niacin production[4]	Positive	Positive
Nitrate reduction	Positive	Positive
Pyrazinamidase	Positive	Positive
Genotypic Analysis Sequencing of Heat Shock Protein 65 gene (~ 440 base pairs)	≥ 99% sequence identity to *M. tuberculosis* type strain (GenBank: AL123456)	100% sequence identity to *M. tuberculosis* type strain (GenBank: AL123456)[5]
Purity (post-freeze)[6]	Growth consistent with expected colony morphology	Growth consistent with expected colony morphology
Viability (post-freeze)[3]	Growth	Growth

[1]NR-19045 was produced by inoculation of the deposited material into Middlebrook 7H9 broth with ADC enrichment and grown for 36 days at 37°C in an aerobic atmosphere with 5% CO_2. Broth inoculum was added to Middlebrook 7H10 agar with OADC enrichment kolles, which were grown for 27 days at 37°C in an aerobic atmosphere with 5% CO_2 to produce this lot.

[2]Information on Mycobacterium testing is available from Ribón, W. "Biochemical Isolation and Identification of Mycobacteria" Biochemical Testing. (2012) Jose C. Jimenez-Lopez (Ed.), InTech, http://www.intechopen.com/books/biochemical-testing/biochemical-isolation-and-identification-of-mycobacteria and Lévy-Frébault, V. V. and F. Portaels. "Proposed Minimal Standards for the Genus *Mycobacterium* and for Description of New Slowly Growing *Mycobacterium* Species." Int. J. Syst. Bacteriol. 42 (1992): 315-323. PubMed: 1581193.

[3]21 days at 37°C in an aerobic atmosphere with 5% CO_2 on Middlebrook 7H10 agar with OADC enrichment

[4]All mycobacteria produce niacin but only *M. tuberculosis* accumulates it, resulting in a positive test for *M. tuberculosis*.

[5]Also consistent with *M. africanum*, *M. bovis*, *M. canettii* and *M. microti*

[6]Purity of this lot was assessed for 21 days at 37°C in an aerobic atmosphere with 5% CO_2 on Middlebrook 7H10 agar with OADC enrichment and 20 days at 37°C in an aerobic atmosphere with 5% CO_2 on Tryptic Soy agar plates.

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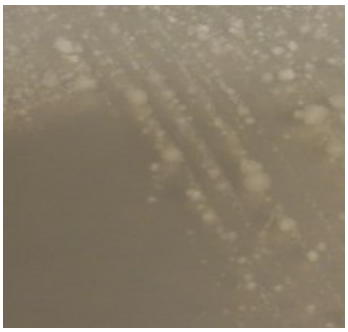
Figure 1: Colony Morphology



Date: 16 AUG 2016 **Signature:**

BEI Resources Authentication

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