

***Mycobacterium tuberculosis*, Strain 96-3459**

Catalog No. NR-30746

This reagent is the tangible property of the U.S. Government.

Product Description: *Mycobacterium tuberculosis* (*M. tuberculosis*), strain 96-3459 was isolated between 1995 and 2000 from human sputum from an HIV-negative patient infected with pulmonary tuberculosis in North America.

Lot¹: 64496316

Manufacturing Date: 14SEP2016

| TEST | SPECIFICATIONS | RESULTS |
|---|--|--|
| Phenotypic Analysis² Cellular morphology Colony morphology ³ Growth rate Growth at 26°C Growth at 37°C Acid-fast stain Pigmentation in the dark (Scotochromogen) Photoinduction for 1 hour (Photochromogen) Nonchromogen (no pigment) Biochemical tests Niacin production ⁴ Nitrate reduction Pyrazinamidase | Gram-positive rods Report results ≥ 7 days Negative Positive Positive (red colonies) Negative (no pigment) Negative (no pigment) Positive (no pigment) Positive Positive Positive | Gram-positive rods Irregular, slight peaked, undulate, rough and cream (Figure 1) 21 days Negative Positive Positive (red colonies) Negative (no pigment) Negative (no pigment) Negative (no pigment) Positive (no pigment) Positive Positive Positive |
| Genotypic Analysis Sequencing of Heat Shock Protein 65 gene (~ 330 base pairs) | ≥ 99% sequence identity to <i>M. tuberculosis</i> type strain (GenBank: AL123456) | 100% sequence identity to <i>M. tuberculosis</i> type strain (GenBank: AL123456) ⁵ |
| Purity (post-freeze) Middlebrook 7H10 agar with OADC enrichment ⁶ Tryptic Soy agar ⁷ | Growth consistent with expected colony morphology Report results | Growth consistent with expected colony morphology No growth |
| Viability (post-freeze)³ | Growth | Growth |

¹NR-30746 was produced by inoculation of the deposited material into Middlebrook 7H9 broth with ADC enrichment. Broth inoculum was added to Middlebrook 7H10 agar with OADC enrichment kolles, which were grown for 29 days at 37°C in an aerobic atmosphere with 5% CO₂ to produce this lot.

²Information on Mycobacterium testing is available from Ribón, W. "Biochemical Isolation and Identification of Mycobacteria." [Biochemical Testing](http://www.intechopen.com/books/biochemical-testing/biochemical-isolation-and-identification-of-mycobacteria). (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.

³21 days at 37°C in an aerobic atmosphere with 5% CO₂ on Middlebrook 7H10 agar with OADC enrichment

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

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

⁶Purity of this lot was assessed for 52 days at 37°C in an aerobic atmosphere with 5% CO₂.

⁷Purity of this lot was assessed for 21 days at 37°C in an aerobic atmosphere with 5% CO₂.

Figure 1: Colony Morphology



Date: 30 AUG 2017

Signature:

A handwritten signature in black ink, which appears to read "David C. Cook". The signature is written in a cursive style.

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