

Certificate of Analysis for NR-49086

Mycobacterium iranicum, Strain M05T

Catalog No. NR-49086

Product Description: Mycobacterium iranicum (M. iranicum), strain M05T was isolated in 2008 from the bronchoalveolar lavage of a 60-year-old female patient with chronic pulmonary disease in Iran.

Lot¹: 64362399 Manufacturing Date: 11JUL2016

TEST	SPECIFICATIONS	RESULTS
		1.255215
Phenotypic Analysis ^{2,3}		
Cellular morphology	Report results	Rods
Colony morphology ⁴	Report results	Circular, convex, entire, smooth and orange (Figure 1)
Growth on MacConkey agar (without crystal violet)	Negative	Negative
Growth rate	≤ 7 days	4 days
Growth at 45°C	Negative	Negative
Growth at 55°C	Report results	Negative
Acid-fast stain	Positive (red colonies)	Positive (red colonies)
Biochemical tests		
Nitrate reduction	Negative	Negative
Aryl sulfate (3 days)	Positive	Negative ⁵
Aryl sulfate (14 days)	Report results	Positive
Iron uptake	Positive	Positive
Growth in the presence of 5% sodium chloride	Positive	Positive
Growth in the presence of thiophene-2-carboxylic	Positive	Positive
acid hydrazide (TCH)		
Genotypic Analysis		
Sequencing of 16S ribosomal RNA gene	≥ 99% sequence identity to	100% sequence identity to
(1450 base pairs)	M. iranicum type strain	M. iranicum type strain
((GenBank: HQ009482.1)	(GenBank: HQ009482.1)
Digital DNA-DNA hybridization (dDDH) ⁶	≥ 70% for species identification	M. iranicum (98.5%) ⁷
Purity (post-freeze)		
Middlebrook 7H10 agar with OADC enrichment ⁸	Growth consistent with expected	Growth consistent with expected
	colony morphology	colony morphology
Tryptic Soy agar ⁸	Report results	Growth consistent with expected
31		colony morphology
Viability (post-freeze) ⁴	Growth	Growth

NR-49086 was produced by inoculation of the deposited material in Middlebrook 7H9 broth with ADC enrichment for 5 days at 37°C in an aerobic atmosphere with 5% CO2. Broth inoculum was added to Middlebrook 7H10 agar with OADC enrichment kolles, which were grown for 5 days at 37°C in an aerobic atmosphere with 5% CO₂ to produce this lot.

BEI Resources

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²Information on Mycobacterium testing is available from Ribón, W. "Biochemical Isolation and Identification of Mycobacteria." <u>Biochemical Testing</u>. (2012) Jose C. Jimenez-Lopez (Ed.), InTech, http://www.intechopen.com/books/biochemical-testing/biochemical-isolation-and-identification-ofmycobacteria 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.

³Phenotypic characterization of *M. iranicum* was performed following: Shojaei, H., et al. "Mycobacterium iranicum sp. nov., a Rapidly Growing Scotochromogenic Species Isolated from Clinical Specimens on Three Different Continents." Int. J. Syst. Evol. Microbiol. 63 (2013): 1383-1389.

 $^{^4}$ 4 days at 37° C in an aerobic atmosphere with 5% CO $_2$ on Middlebrook 7H10 agar with OADC enrichment

⁵NR-49086 was deposited as *M. iranicum* and reported to be positive for 3-day aryl sulfate activity. Testing performed in duplicate by BEI Resources indicates a negative result.

⁶Relatedness between bacterial strains has traditionally been determined using dDDH. 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." Stand Genomic Sci. 2 (2010): 117-134. PubMed: 21304684.

⁷The whole genome of *M. iranicum*, strain M05T (Contig Total Length ~ 6.4 megabase pairs) was sequenced using the Illumina® MiSeq® system and was assembled and analyzed with CLC Genomics Workbench Version 7.0.2.



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⁸Purity of this lot was assessed for 8 days at 37°C in an aerobic atmosphere with 5% CO₂.

Figure 1: Colony Morphology



/Heather Couch/ **Heather Couch**

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Program Manager or designee, ATCC Federal Solutions

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