

Certificate of Analysis for NR-48784

Mycobacterium tuberculosis, Strain H37Rv, Purified Trehalose Monomycolate (TMM)

Catalog No. NR-48784

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

Product Description:

NR-48784 is a preparation of purified trehalose monomycolate (TMM) that was extracted from the lipid fraction obtained from irradiated *Mycobacterium tuberculosis*, strain H37Rv cells. Following purification steps, the TMM was dried under nitrogen gas.

Lot: 70045518 Manufacturing Date: 16SEP2021

Production and QC testing were performed by Colorado State University (CSU). The CSU documentation for lot 21.Rv.9.13.02.TMM is attached.

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WORK SHEET FOR PURIFIED LIPID FRACTION

General Information

BEI Catalog Number: _	NR-48784
CSU Lot Number:	21.Rv.9.13.02.TMM
Fraction Type:	trehalose monomycolate
Species:	M. tuberculosis
Strain:	H37Rv

Purification Information

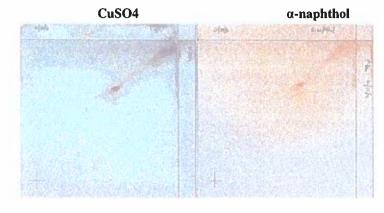
Starting material: 2:1 TL	from H37Rv Starting Material Lot #: 17,Rv.2.11.1.11,WCg.a
Cells Irradiated: Yes	Viability Test Performed: No Viable Organism Detected
Protocol used (SOP #'s):	PP030.1, SP031, SP032, SP033, SP037, SP046
Date started:	5/25/21
Date completed:	9/16/21
Notebook; page(s):	Lipids 10 pp 19-44

Additional notes: 2:1 Rv TL was applied to silica gel flash columns, which were washed with chloroform, then an increasing methanol gradient. The 10% MeOH in CHCl3 fractions were applied to preparatory TLC plates and resolved with 65/25/4 chloroform/methanol/water. Extracted bands were polished on prep plates with 65/25/4.

Quality Control Information:

Total amount of TMM : 4.5 mg	TLC date:	8/27/21
Date dried on N ₂ bath: 9/14/21	Notebook:	Lipids 10 pp 45-48

TLC Analysis:

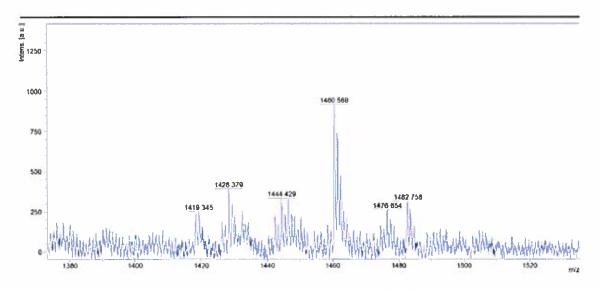


TLC solvent system

1st dimension 65/25/4 chloroform/ methanol/ water, left to right; 2^{nd} dimension 60/30/6, same solvents, bottom to top. Loaded 50 μg .

Aliquot information

 $18 \times 0.25 \text{ mg} = 4.5 \text{ mg}$



Purified $\underline{21.Rv.9.13.02.TMM}$ was loaded at 0.5 μg and 1.0 μg , overlaid with 0.5 DHB matrix, and analyzed with reflector positive mode.

(Research Associate) date (Laboratory Supervisor) date