b|e|i RESOURCES
SUPPORTING INFECTIOUS DISEASE RESEARCH

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: 63050381 **Manufacturing Date: 18MAY2015**

Production and QC testing were performed by Colorado State University (CSU). The CSU documentation for lot 15.Rv.3.11.01.TMM is attached.

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected by the contractor to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC®'s knowledge.

Support Provided by NIAID

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WORK SHEET FOR PURIFIED TMM

General Information

Lot Number: 15.Rv.3.11.01.TMM
Species: *M. tuberculosis*
Strain: H37Rv

Purification Information

Starting material: 10:10:3 Rv
Lot number: 14.Rv.2.6.3.4.WCg.c IHO
Protocol used (SOP #'s): PP030.1, SP031, SP032, SP033, SP037, SP046
Date started: 1/16/15
Date completed: 5/18/15
Notebook; page(s): TMM I pp 58-74

Additional notes: 2:1-soluble 10:10:3 Rv TL was applied to preparatory TLC plates and resolved with 80/20/2 chloroform/methanol/water. Extracted bands were polished on prep plates with 65/25/4, same solvents.

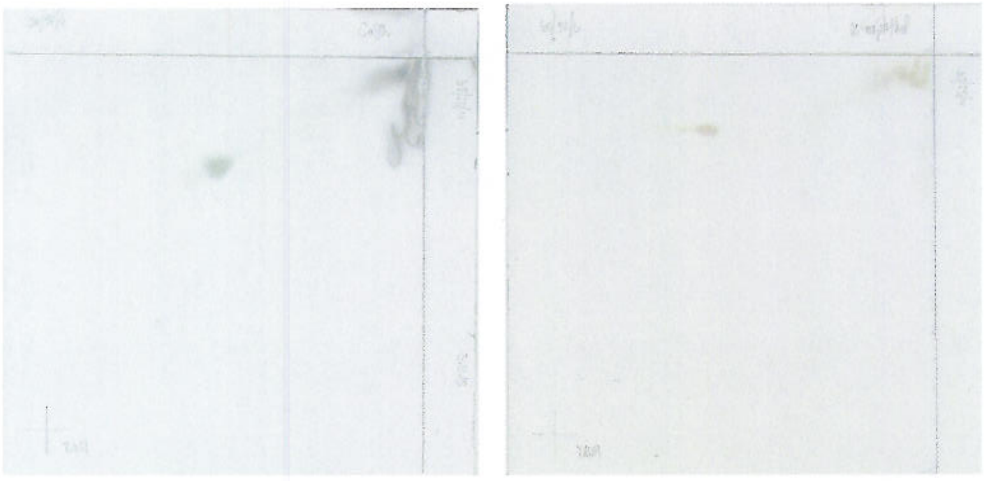
Quality Control Information:

Total amount of TMM: 15.6 mg
TLC date: 3/11/15
Notebook and page(s): TMM 1 pp 75-83

MALDI-TOF completed: 5/18/15
Date dried on N_2 bath: 3/12/15

TLC Analysis:

CuSO4 **α-naphthol**



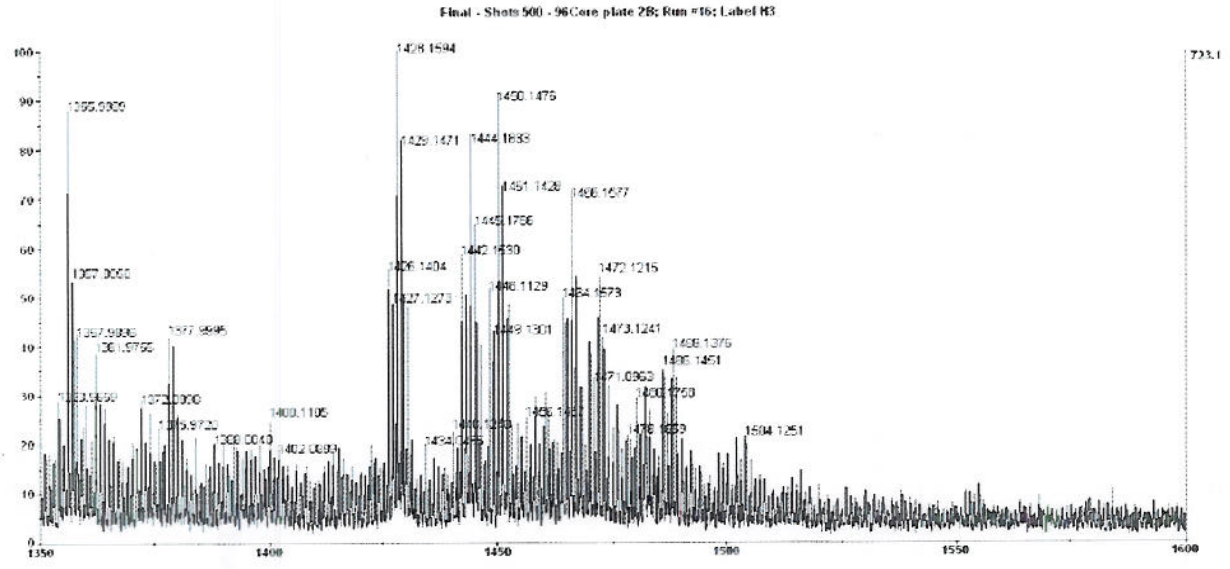
TLC solvent system

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

Aliquot information

15 x 0.25 mg = 3.75 mg
4 x 2.50 mg = 10.00
1 x 1.85 mg = 1.85
15.60 mg

MALDI Analysis



Purified sample (1 µl at 1 µg/µl) was mixed 1:1 with DHB matrix and analyzed in positive electrospray mode.

(Research Associate)	date	(Laboratory Supervisor)	date