b|e|i RESOURCES

SUPPORTING INFECTIOUS DISEASE RESEARCH

Certificate of Analysis for NR-44101

Mycobacterium tuberculosis, Strain H37Rv, Mycobactin

Catalog No. NR-44101

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

Product Description:

NR-44101 is a preparation of mycobactin derived from irradiated *Mycobacterium tuberculosis*, strain H37Rv.

Lot: 70037073 **Manufacturing Date: 28AUG2020**

Production and QC testing were performed by Colorado State University (CSU). The CSU documentation for lot 20.Rv.08.26.02.MBT 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 MYCOBACTIN

General Information

BEI Catalog Number: NR-44101
CSU Lot Number: 20.Rv.08.26.02.MBT
Fraction Type: mycobactin
Species: *M. tuberculosis*
Strain: H37Rv

Purification Information

Starting material: 2:1 total lipid 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): PP018.1, PP032.2, SP004, SP005, SP031b, SP032, SP033
Date started: 6/16/2020
Date completed: 8/28/2020
Notebook; page(s): Mycobactin Notebook 3 pp 57-80

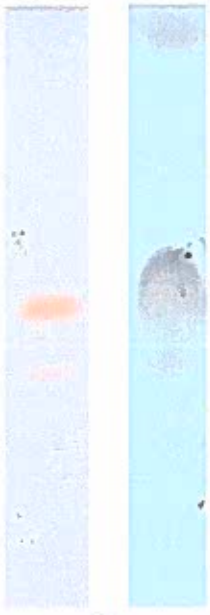
Additional notes (if applicable): 2:1 total lipid was enriched for mycobactin on 7 silica gel columns by eluting with 2% MeOH in CHCl3. These fractions were developed on preparative TLC plates with 95/5 chloroform/ methanol, and the visible mycobactin-bearing silica scraped. Subsequent clean-up was performed by passaging through C18 SepPak columns.

Quality Control Information:

Total amount of mycobactin: 5.6 mg Date dried on N_2 bath: 8/28/20
TLC date: 8/17/2020 Notebook and page(s): Mycobactin 3 pp 81-84
TLC Solvent System: 95/5 chloroform/methanol

QC TLC:

unstained $CuSO_4$

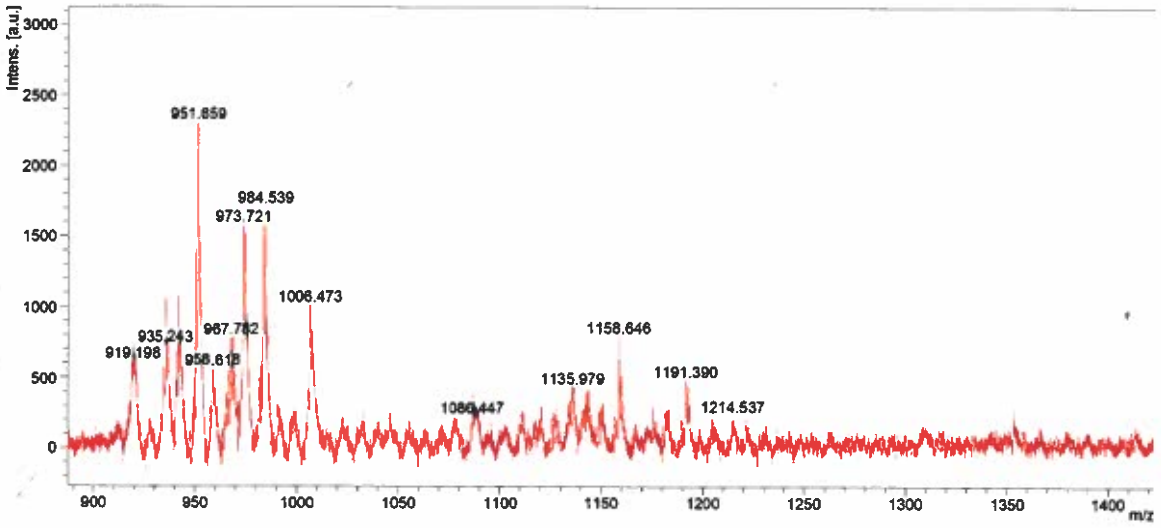


Aliquot Information:

15 x 0.1 mg = 1.5 mg
1 x 4.1 mg = 4.1 mg
5.6 mg

Loaded 100 µg, developed in solvent system described above, and stained one with $CuSO_4$ and charring.

MALDI-TOF:



Applied 1 µg 20.Rv.08.26.02.MBT at 1 µg/µl and fresh DHB matrix was overlaid at ARC-BIO, then analyzed in negative mode.

Daniel Carlson 8/28/20

(Research Associate) date

C. Mehaffy 8/31/20

(Laboratory Supervisor) date