

***Mycobacterium tuberculosis*, Strain H37Rv, Purified Demannosylated Lipoarabinomannan (DLAM)****Catalog No. NR-56329**

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**Product Description:**

NR-56329 is a preparation of demannosylated lipoarabinomannan (DLAM) derived from the cell wall of irradiated *Mycobacterium tuberculosis* (*M. tuberculosis*), strain H37Rv. LAM possesses many biological activities including immunogenicity, induction of TNF and the release of other cytokines, and inhibition of antigen processing. The nonreducing termini of strain H37Rv LAM are extensively capped with mannose. Mannose-capped LAM (ManLAM) has demonstrated immunomodulatory effects, such as inhibition of T cell activation and proliferation and influences cytokine production. Variability in mannose capping observed in clinical isolates and among different strains of *M. tuberculosis* may contribute to the variation of biological activities *in vitro*. Removal of the mannose caps of LAM from virulent strain H37Rv provides the opportunity to study the biological features attributed to LAM that are not associated with mannose capping.

**Lot: 70058656****Manufacturing Date: 03AUG2023**

Production and QC testing were performed by Colorado State University (CSU). The CSU documentation for lot 23.Rv.8.3.DLAM is attached.

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## WORK SHEET FOR DEMANNOSYLATED LIPOARABINOMANNAN

### General Information

BEI Catalog Number: NR-56329  
CSU Lot Number: 23.Rv.8.3.DLAM  
Species: *Mycobacterium tuberculosis*  
Strain: H37Rv

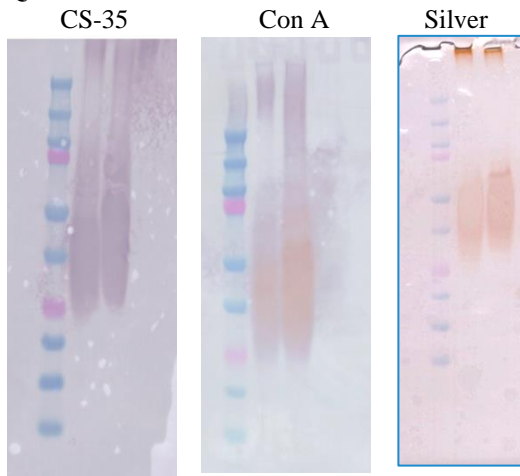
### Purification Information

Cells Irradiated: Yes  
Viability Test Performed: No Viable Organism Detected  
LAM starting material (lot #): 23.Rv.7.13.LAM  
LAM starting material (mg): 11.648 mg  
Protocol used (SOP #): PP059 and SP079  
Date started: 7/13/23  
Date completed: 8/3/23  
Notebook; pages Megan Stookey NB 3 pg: 57, 58, 63, 66, 67 – 71  
Additional notes (if applicable): Protein assay quantification by BCA resulted in quantification of 0.159mg protein per mg of demannosylated LAM; no protein content was detected in subsequent silver stain analyses. Product meets purification criteria.

### Quality Control Information

BCA: 0.159 mg protein/mg LAM \*see note Notebook and page(s): Megan Stookey Notebook 3 pg: 66,71  
Endotoxin assay used: EndoZyme II  
Endotoxin amount: 4.482 ng/mg Notebook and page(s): Megan Stookey Notebook 3 pg: 68,69  
Image J Concentration: 0.2514 mg/ml Notebook and page(s): Megan Stookey Notebook 3 pg: 66, 67, 70  
Total amount of LAM: 3.208 mg  
Silver stain date: 08/2/23 Notebook and page(s): Megan Stookey Notebook 3 pg: 70, 71  
Western blot: 8/2/23 Antibody used: CS-35 Notebook and page(s): Megan Stookey Notebook 3 pg: 70,71  
Western blot: 8/1/23 Antibody used: Con A Notebook and page(s): Megan Stookey Notebook 3 pg: 70  
Amount Loaded on gel/s: 5 µg

### QC Gel and Blots:



#### CS-35 and Con A:

Lane 1= Ladder  
Lane 2= Demannosylated LAM product  
Lane 3= H37Rv LAM  
Lane 4= HSPX (negative control)

#### Silver Stain:

Lane 1=Ladder  
Lane 2= Demannosylated LAM product  
Lane 3=H37Rv LAM

#### Aliquot Information:

\*Aliquot information reflects those made at the time of QC.  
Bulk aliquots will be broken down as needed.

12 vials x 0.25 mg aliquots  
1 vial x 0.208 mg aliquot

Megan Stookey 8/25/23  
Research Associate Date

Rebecca Staudenmaier 8/25/2023  
Laboratory Supervisor Date