

Mycobacterium smegmatis*, Purified Lipoarabinomannan (LAM)*Catalog No. NR-14849**

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

Product Description: NR-14849 is a preparation of lipoarabinomannan (LAM) derived from the cell wall of *Mycobacterium smegmatis*. LAM possesses many biological activities including immunogenicity, induction of TNF and the release of other cytokines, and inhibition of antigen processing. The culture was grown to late-log phase in glycerol-alanine-salts medium, washed with PBS and inactivated by gamma irradiation. The cells were delipidated and suspended in PBS buffer containing 4% Triton X-114 and broken by French Press. The lysate was incubated at 4°C for 18 hours with rocking and insoluble material was removed by centrifugation. The Triton X-114 extract was collected, heated at 37°C to allow biphasic partitioning, and centrifuged. The detergent layer was collected and macromolecules, including LAM, were recovered by ethanol precipitation. The ethanol-insoluble material was suspended in PBS, and the proteins digested and dialyzed out. The crude carbohydrate mixture was fractionated by size exclusion chromatography and the pure LAM pooled. Buffer contaminants were removed by extensive dialysis. Contaminating LPS is avoided as all buffers and water used are endotoxin-free.

Lot: 61699475**Manufacturing Date: 10APR2013**

Production and QC testing were performed by Colorado State University. The Colorado State University documentation for the 13.Smeg.2.2.27.SOS.LAM is attached.

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You are authorized to use this product for research use only. It is not intended for human use.



WORK SHEET FOR LIPOARABINOMANNAN

General Information

Lot Number: 13.Smeg.2.2.27.SOS.LAM
Species: *M. smegmatis*
Strain: N/A

Purification Information

Starting material (lot #): Pooled material from lots 12.Smeg.2.11.30.SOS and 12.Smeg.2.7.17.SOS
Cell amount (wet weight): N/A
Protocol used (SOP #): PP015.1, PP016.3, PP017.2
Date started: 2/27/2013
Date completed: 4/10/2013
Notebook; pages: Shane Orlin Severs, Book #3, SOS-Smeg, pages 1-8
Additional notes (if applicable):

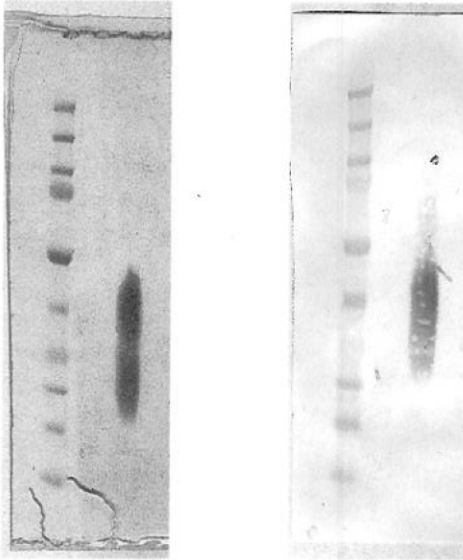
Quality Control Information

Clarity of product: Clear
NMR results: Pass
Endotoxin amount: 0.64 ng ET/mg LAM
GC Results: 5.87 mg/ml
Total volume: 3.096 ml
Silver stain date: 4/9/2013
Western blot date: 4/9/2013

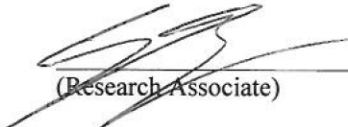
Antibody used: CS35

Notebook and page(s): SOS-Smeg, 4
Notebook and page(s): SOS-Smeg, 7
Notebook and page(s): SOS-Smeg, 5-6
Total amount of LAM: 18.17 mg
Notebook and page(s): SOS-Smeg, 7-8
Notebook and page(s): SOS-Smeg, 7-8

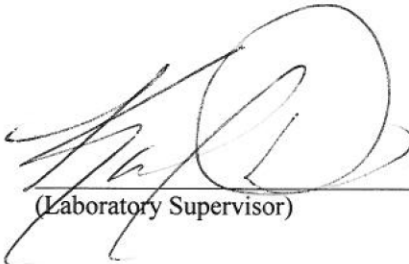
QC Gel and Blot:



Aliquot Information:
36 vials @ 500 ug each
18000 ug total


(Research Associate)

April 16th, 2013
(date)


(Laboratory Supervisor)

4/17/2013
(date)