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SUPPORTING INFECTIOUS DISEASE RESEARCH

Mycobacterium tuberculosis, Strain H37Rv, Purified Lipomannan (LM)

Catalog No. NR-14850

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For research use only. Not for use in humans.

Contributor:

BEI Resources or NIH - TB Vaccine Testing and Research Materials Contract

Manufacturer:

Karen Dobos, Ph.D., Colorado State University, Fort Collins, Colorado, USA or NIH - TB Vaccine Testing and Research Materials Contract

Product Description:

NR-14850 is a preparation of the lipomannan (LM) derived from the cell wall of irradiated *Mycobacterium tuberculosis*, strain H37Rv.

Mycobacterium tuberculosis, strain H37Rv was grown to latelog phase in glycerol-alanine-salts, washed with PBS and inactivated by gamma irradiation. The cells were delipidated, suspended in buffer containing 4% Triton X-114 and broken by French Press. Insoluble material was removed from the lysate by repeated centrifugation. The Triton X-114 extract was collected, heated to allow biphasic partitioning and centrifuged. The detergent layer was collected and macromolecules, including LM, were recovered by ethanol precipitation. The ethanol insoluble material was suspended in PBS, and the proteins were digested and dialyzed out. The crude carbohydrate mixture was fractionated by size exclusion chromatography and the pure LM pooled. Buffer contaminants were removed by extensive dialysis. Contaminating LPS was avoided as all buffers and water used were endotoxin-free.

Material Provided:

Each vial contains approximately 100 µg of lyophilized purified LM from *Mycobacterium tuberculosis*, strain H37Rv. Lyophilized products may aggregate as a loose powder near the lid. Please take precautionary measures (such as tapping the bottom of the tube on the lab bench) to reduce escape of the reagent when opening the vial to reconstitute contents.

<u>Note</u>: LM can be reconstituted in water. A 100 mM to 500 mM aqueous buffered salt solution, such as phosphate buffered saline, may also be used.

Packaging/Storage:

NR-14850 was packaged aseptically in cryovials. The product is provided frozen on dry ice and should be stored at -80°C or colder immediately upon arrival. Freeze-thaw cycles should be avoided.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: *Mycobacterium tuberculosis*, Strain H37Rv, Purified

BEI Resources www.beiresources.org Lipomannan (LM), NR-14850."

Biosafety Level: 1

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. <u>Biosafety in Microbiological and Biomedical Laboratories</u>. 6th ed. Washington, DC: U.S. Government Printing Office, 2020; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

Disclaimers:

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References:

 Chatterjee, D., et al. "Lipoarabinomannan of Mycobacterium tuberculosis. Capping with Mannosyl Residues in Some Strains." <u>J. Biol. Chem.</u> 267 (1992): 6234-6239. PubMed: 1556132.

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