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SUPPORTING INFECTIOUS DISEASE RESEARCH

Mycobacterium canettii Cytosol Fraction

Catalog No. NR-36507

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Contributor and Manufacturer:

Karen Dobos, PhD., Colorado State University, Fort Collins, Colorado, USA

Product Description:

NR-36507 is a preparation of the cytosol fraction of *Mycobacterium canettii* and contains cytosolic proteins and soluble material released from the cell wall during disruption of the bacilli.

The culture was grown to late log phase in glycerol-alaninesalts medium, washed with PBS and inactivated by gamma irradiation. The bacilli were suspended at a concentration of 2 g/mL in PBS containing 8 mM EDTA, DNase, RNase, and a proteinase inhibitor tablet, and broken in a French Press pressure cell at 4°C. Unbroken cells were removed by low speed (3,000 x g) centrifugation. The cell wall was isolated by centrifugation at 27,000 x g. The supernatant was subjected to a 100,000 x g centrifugation for four hours, then collected and dialyzed against 10 mM ammonium bicarbonate. The protein content was determined using the BCA protein assay.

Material Provided:

Each vial contains approximately 1 mg of protein in 10 mM ammonium bicarbonate provided as a frozen pellet.

<u>Note</u>: NR-36507 is soluble in 100 mM to 500 mM aqueous buffered salt solutions, such as phosphate buffered saline. A 10 mM ammonium bicarbonate solution can also be used. Cytosol fractions may not fully solubilize, but a uniform suspension can be achieved with bath sonification.

Packaging/Storage:

NR-36507 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 canettii* Cytosol Fraction, NR-36507."

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>. 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

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References:

- Lee, B. Y., S. A. Hefta and P. J. Brennan. "Characterization of the Major Membrane Protein of Virulent *Mycobacterium tuberculosis*." <u>Infect. Immun.</u> 60 (1992): 2066-2074. PubMed: 1563797.
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- Van Soolingen, D., et al. "A Novel Pathogenic Taxon of the Mycobacterium tuberculosis Complex, Canetti: Characterization of an Exceptional Isolate from Africa." <u>Int. J. Syst. Bacteriol.</u> 47 (1997): 1236-1245. PMID: 9336935.

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