

Mycobacterium tuberculosis*, Strain Indo-Oceanic T17X, Cytosol Fraction*Catalog No. NR-36505**

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Product Description: NR-36505 is a preparation of the cytosol fraction of *Mycobacterium tuberculosis*, strain Indo-Oceanic T17X, 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-alanine-salts 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.

Lot: 62976460**Manufacturing Date: 20FEB2015**

Production and QC testing were performed by Colorado State University (CSU). The CSU documentation for lot 15.IO.02.23.CYT is attached.

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WORK SHEET FOR CYTOSOL

General Information

BEI Catalog Number: NR-36505
Product Lot Number: 15.IO.02.23.CYT
Species: Mycobacterium tuberculosis
Strain: Indo-Oceanic T17X

Sonication and Fractionation Information

Starting material (lot #): 13.IO.11.12.4.WCg.a
Cell amount (wet weight): 103.7g
SOP #: PP008.3 Notebook pages: 51
Date of French Press Sonication: 02/13/15; Buffer used: PBS +1mM EDTA with protease inhibitor
Centrifugation Notes Centrifuged 2x at 100.000xg to separate membrane
Date 10 mM ammonium bicarbonate dialysis started: 02/18/15
date completed: 02/20/15/
Notebook; page(s): PMF/TVTRM #1, 52-53

Quality Control Information

Clarity of product/suspension after dialysis: Clear
BCA: 6.21 $\mu\text{g}/\mu\text{l}$. Notebook and page(s): PMF/TVTRM #1, 54
Total Protein: 656 mg
Silver Stain Date: 02/23/15. Notebook and page(s): PMF/TVTRM #1, 56

QC Gel:

MEM CYT CW



Aliquot Information:

15 x 1mg for BEI
20 x 1 mg for IHO
Bulk 100 ml @ 6.21 $\mu\text{g}/\mu\text{l}$

C. Melchior 02.25.15
(Research Associate) (date)

[Signature] 2/25/2015
(Laboratory Supervisor) (date)