

***Mycobacterium tuberculosis*, Strain H37Rv, Cell Wall Fraction****Catalog No. NR-14828**

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**Product Description:** NR-14828 is a preparation of the cell wall fraction of *Mycobacterium tuberculosis*, strain H37Rv and contains proteins and non-protein compounds such as mAGP. Culture was grown to late-log phase in glycerol-alanine-salts medium, washed with PBS pH 7.4, and inactivated by gamma irradiation. The bacilli are 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 are removed by low speed (3,000 x g) centrifugation. The cell wall is isolated by centrifugation at 27,000 x g for one hour and washed two times in PBS. The final cell wall pellet is suspended and dialyzed in 0.01 M ammonium bicarbonate, quantified by BCA protein assay for protein content, and frozen.

**Lot: 2045****Manufacturing Date: 18JAN2017**

Production and QC testing were performed by Colorado State University (CSU). The CSU documentation for lot 17.Rv.1.20.CW is attached.

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## WORK SHEET FOR CELL WALL

### General Information

BEI Catalog Number: NR-14828  
CSU Lot Number: 17.Rv.1.20.CW  
Species: *Mycobacterium tuberculosis*  
Strain: H37Rv

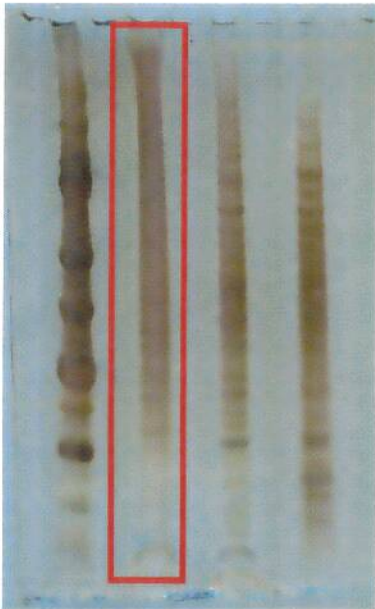
### Sonication and Fractionation Information

Starting material (lot #): 15.Rv.2.2.10.9.WCg.b  
Cells Irradiated: Yes  
Viability Test Performed: No Viable Organism Detected  
Cell amount (wet weight): 75 g  
SOP #: PP008.3 Notebook pages: Kala Early's Notebook #2, pgs. 1-14  
Date of French Press Sonication: 1-9-17; Buffer used: PBS-EDTA with protease inhibitor  
Centrifugation Notes 27,000 x g for 1 hour, 4°C (repeat a second time for wash)  
Date 10 mM ammonium bicarbonate dialysis started: 1/17/17  
date completed: 1/18/17  
Notebook; page(s): First centrifugation at 3,000 x g for 15 minutes, at 4°C to remove unbroken cells = Kala Early's Notebook #2, pg. 1, First and second centrifugations (second to wash) at 27,000 x g = Kala Early's Notebook #2, pg. 1

### Quality Control Information

BCA: 9.249 µg /µl. Notebook and page(s): Kala Early's Notebook #2, pg. 12  
Total Protein: 1094.9 mg  
Silver Stain Date: 1/19/17 Notebook and page(s): Kala Early's Notebook #2, pg. 14

### QC Gel:



Cell Wall in red box

### Aliquot Information:

170 x 1 mg  
1 x 924.4 mg

Kala Early 1/25/17  
(Research Associate) (date)

C. Fleharty 1/25/17  
(Laboratory Supervisor) (date)