

Plasmid pMRLB73 Containing Gene ML0050 (Protein CFP-10) from *Mycobacterium leprae***Catalog No. NR-19338**

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Product Description: NR-19338 is a recombinant expression vector containing *Mycobacterium leprae* gene ML0050, which encodes culture filtrate protein 10 (CFP-10) [also known as ESAT-6-like protein (EsxB)].^{1,2} Gene ML0050 was amplified by PCR and cloned into pET28b for expression in *Escherichia coli*, strain TOP10. The expressed protein is histidine-tagged and has an observed molecular weight of 10 kDa.

Lot: 61001985**Manufacturing Date: 12MAY2011**

Production and QC testing were performed by Colorado State University (CSU). The CSU documentation for the pre-vial lot pMRLB73 03.25.2011.2A is attached. This lot was received as dry pellets from CSU and rehydrated in TE buffer (10 mM Tris-HCl, 1 mM EDTA buffer, pH 8) at ATCC[®]. Approximately 50 µL of a 0.02 µg/µL solution was dispensed per vial yielding a final amount of 1 µg per vial. A plasmid map of pMRLB73 is attached.

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Recombinant Plasmid Quality Control Record

Plasmid designation pMRLB73 (pET28-CFP-10 ML0050)
 Lot Number 03.25.2011.2A

Notebook/Pgs JNKBook#1/pp142, 145
JNKBook#2/pp15

Notebook detail Plasmid Prep pp.142
A260 and stock dilution pp. 145
QC gel pp.15

Media used LB Broth 50 µg/mL Kanamycin
 Culture size 1 x 1L

Growth conditions: Temp 37°C Time 16 hr. Shaker speed 130 rpm

Plasmid prep type (mini/maxi, kit name or protocol) Qiagen Maxi Prep protocol
 Plasmid prep detail: Maxi prep lysate and tip washing conditions
Purification (Wash & elution)- Qia500 tips

Strain used to produce plasmid TOP-10

E. coli ori? Y/N Y
 Contains Mycobacterial ori? Y/N N

Final concentration Dried, 20 µg/tube
 Number of batches 3 x 20 µg

Buffer TE Buffer

Method used for quantifying Nanodrop

QC gel

Restriction enzymes used in QC analysis NdeI, HindIII & NcoI

Expected size of restriction fragments
 Vector 5.369kb pET28b + NdeI + HindIII
 Insert 0.239 kb CFP-10 + NdeI + HindIII
 Other 5.608kb linearized plasmid- NdeI, HindIII, & NcoI single digests

Gel results All predicted bands visible.

Gel description file number, % agarose, buffer ED CFP-10 2011-05-12, 1.5% 1 X TAE
 Sequence file: sequence CFP-10.ppt
 Plasmid Map: Attached

Generated by Jade Kunch Date 5/12/11

Supervisor John Spencer Date 05/12/11

pET-CFP10 (ML0050)

