

**Plasmid pMRLB.47 Containing Gene Rv1886c (Protein Ag85B) from *Mycobacterium tuberculosis***

**Catalog No. NR-13298**

This reagent is the tangible property of the U.S. Government.

**Product Description:** NR-13298 is a recombinant expression vector containing *Mycobacterium tuberculosis* gene Rv1886c, which is a precursor of the 85-B antigen, also referred to as a fibronectin-binding protein (FpbB), that is involved in cell wall mycoloylation. Gene Rv1886c was amplified by PCR and cloned into pET23b for expression in *Escherichia coli*.

**Lot<sup>1</sup>: 58791994**

**Manufacturing Date: 05FEB2010**

TEST	SPECIFICATIONS	RESULTS
<b>PCR Amplification and Sequencing of Insert (835 bp)</b>	Insert identical to <i>M. tuberculosis</i> , strain H37Rv gene Rv1886c	Insert identical to <i>M. tuberculosis</i> , strain H37Rv gene Rv1886c
<b>Agarose Gel Electrophoresis</b> Linearization with <i>EcoRV</i> <sup>2</sup> Linearization with <i>NdeI</i> <sup>3</sup> Linearization with <i>HindIII</i> <sup>4</sup> Digestion with <i>NdeI</i> and <i>HindIII</i> <sup>3,4</sup>	~ 4.5 kb ~ 4.5 kb ~ 4.5 kb ~ 0.9 kb (insert); ~ 3.7 kb (vector)	~ 4.5 kb (Figure 1) ~ 4.5 kb (Figure 1) ~ 4.5 kb (Figure 1) ~ 0.9 kb; ~ 3.7 kb (Figure 1)
<b>Concentration by PicoGreen<sup>®</sup> Measurement</b>	0.7 to 1.5 µg in 25 to 100 µL per vial	1.8 µg in 25 µL per vial (72 µg/mL) <sup>5</sup>
<b>OD<sub>260</sub>/OD<sub>280</sub> Ratio</b>	1.7 to 1.9	1.9
<b>Effective Bacterial Transformation</b>	≥ 100 colonies per 10 ng	≥ 100 colonies per 10 ng

<sup>1</sup>Produced in *Escherichia coli* MAX Efficiency<sup>®</sup> DH5α<sup>™</sup> cells (Invitrogen<sup>™</sup> 18258-012) and extracted using the Qiafilter Plasmid Midi Kit (QIAGEN<sup>®</sup> 12243)

<sup>2</sup>*EcoRV* (New England BioLabs, Inc. R0195M)

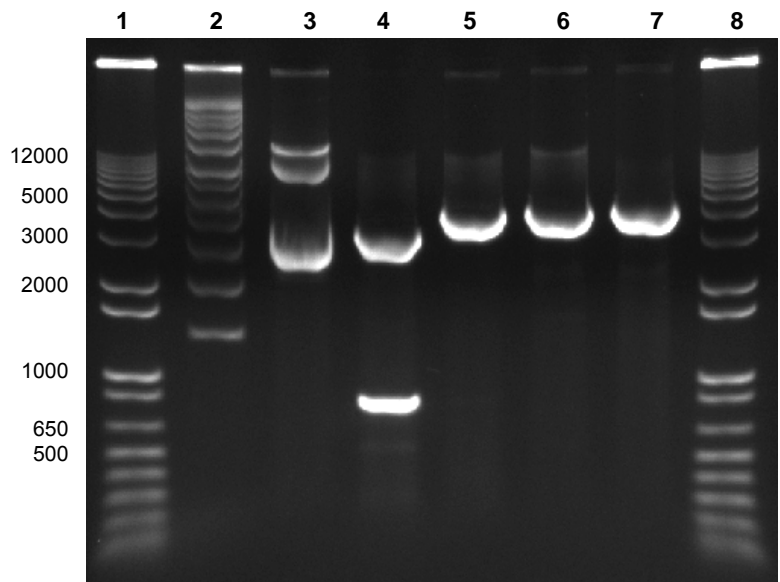
<sup>3</sup>*NdeI* (New England BioLabs, Inc. R0111L)

<sup>4</sup>*HindIII* (New England BioLabs, Inc. R0104M)

<sup>5</sup>The µg of DNA in the vials is greater than required by current specifications

**Figure 1: Agarose Gel of Undigested and Restriction Enzyme Digested NR-13298**

Lane 1: Invitrogen<sup>™</sup> TrackIt 1 Kb Plus DNA Ladder<sup>™</sup>  
 Lane 2: Supercoiled DNA Ladder (Invitrogen<sup>™</sup> 15622-012)  
 Lane 3: Undigested NR-13298  
 Lane 4: NR-13298 digested with *NdeI* and *HindIII*  
 Lane 5: NR-13298 digested with *NdeI*  
 Lane 6: NR-13298 digested with *HindIII*  
 Lane 7: NR-13298 digested with *EcoRV*  
 Lane 8: Invitrogen<sup>™</sup> TrackIt 1 Kb Plus DNA Ladder<sup>™</sup>



**Date:** 21 JUN 2010

**Signature:** Signature on File

**Title:** Technical Manager, BEI Authentication or designee

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