

Certificate of Analysis for NR-13297

Plasmid pMRLB.46 Containing Gene Rv3874 (Protein Cfp10) from *Mycobacterium tuberculosis*

Catalog No. NR-13297

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

Product Description: NR-13297 is a recombinant expression vector containing *Mycobacterium tuberculosis* gene Rv3874, which encodes a 10 kDa culture filtrate protein, Cfp10. Gene Rv3874 was amplified by PCR and cloned into pET23b for expression in *Escherichia coli*.

Lot¹: 58791993 Manufacturing Date: 03SEP2009

TEST	SPECIFICATIONS	RESULTS
PCR Amplification and Sequencing of Insert (300 bp)	Insert identical to <i>M. tuberculosi</i> s, strain H37Rv gene Rv3874	Insert identical to <i>M. tuberculosis</i> , strain H37Rv gene Rv3874
Agarose Gel Electrophoresis Linearization with Ndel ² Linearization with HindIII ³ Linearization with SacII ⁴ Digestion with Ndel and HindIII	~ 3.9 kb ~ 3.9 kb ~ 3.9 kb ~ 0.302 kb (insert); ~ 3.6 kb (vector)	~ 3.9 kb (Figure 1) ~ 3.9 kb (Figure 1) ~ 3.9 kb (Figure 1) ~ 0.302 kb; ~ 3.6 kb (Figure 1)
Concentration by PicoGreen® Measurement	0.7 to 1.5 μg in 25 to 100 μL per vial	1.4 μg in 25 μL per vial (57 μg/mL)
Absorbance Ratio (OD ₂₆₀ /OD ₂₈₀)	Report results	1.9
Effective Bacterial Transformation	≥ 100 colonies per 10 ng	≥ 100 colonies per 10 ng

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

³SacIII (New England BioLabs, Inc. R0157L)

Figure 1: Agarose Gel of Undigested and
Restriction Enzyme Digested NR-13297.

Lane 1: Invitrogen™ TrackIt 1 Kb Plus DNA Ladder™

Lane 2: Undigested NR-13297

Lane 3: NR-13297 digested with Ndel and HindIII

Lane 4: NR-13297 digested with *Ndel* Lane 5: NR-13297 digested with *HindIII* Lane 6: NR-13297 digested with *SacII* 1 2 3 4 5 6 6000 3000 2000 1650 1000 500

Date: 28 OCT 2010 Signature:

Title: Technical Manager, BEI Authentication or designee

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²Ndel (New England BioLabs, Inc. R0111L)

³HindIII (New England BioLabs, Inc. R0104M)