

SUPPORTING INFECTIOUS DISEASE RESEARCH

Product Information Sheet for NR-36037

Guinea Pig Expression Clone TNF-α, Recombinant in *Escherichia coli*

Catalog No. NR-36037

For research use only. Not for human use.

Contributor:

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Manufacturer:

BEI Resources

Product Description:

The guinea pig is an animal model for testing novel tuberculosis vaccine candidates because it mimics human tuberculosis. The host response to vaccination and infection can be further investigated utilizing recombinant guinea pig proteins. It has been suggested that tumor necrosis factoralpha, TNF-α, plays multiple roles in immune and pathologic responses in tuberculosis.¹

NR-36037 is an expression clone containing the mature peptide region of TNF- α (GenBank: NM 001173025) from Cavia porcellus (guinea pig). The TNF- α gene was cloned into vector pQE-30 via BamHI and HindIII insertion sites and transformed into Escherichia coli (E. coli), strain M15, competent cells. The M15 strain of E. coli contains a low-copy plasmid, pREP4, which confers kanamycin resistance and constitutively expresses the lac repressor, which is utilized for tight regulation of recombinant protein expression. The pQE-30 vector contains a T5 promoter, a β -lactamase gene for ampicillin resistance, an N-terminal His-tag for purification, a thrombin cleavage site between the His-tag and the protein, and the lacl gene which is used for enhanced protein expression via IPTG induction.² Refer to Table 1 for protein sequence for NR-36037.

Material Provided:

Each vial contains approximately 0.5 mL of *E. coli*, strain M15 in Luria Bertani (LB) broth containing 100 μg/mL ampicillin supplemented with 10% glycerol.

<u>Note</u>: If homogeneity is required for your intended use, please purify prior to initiating work.

Packaging/Storage:

NR-36037 was packaged aseptically in plastic cryovials. The product is provided frozen and should be stored at -60°C or colder immediately upon arrival. For long-term storage, the vapor phase of a liquid nitrogen freezer is recommended. Freeze-thaw cycles should be avoided.

Growth Conditions:

Media:

LB Broth or Agar containing 100 μg/mL ampicillin and 25 μg/mL kanamycin

Incubation:

Temperature: 37°C Atmosphere: Aerobic

Propagation:

- Scrape the top of the frozen vial with a sterile loop or pipette tip and streak onto a selective agar plate and/or inoculate a tube of selective broth. Return the vial to storage at -60°C or colder.
- 2. Incubate the plate and/or tube at 37°C for 18 to 24 hours.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: Guinea Pig Expression Clone TNF- α , Recombinant in Escherichia coli, NR-36037."

Biosafety Level: 1

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. Biosafety in Microbiological and Biomedical Laboratories. 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

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References:

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1. Lasco, T. M., et al. "Evaluating the Role of Tumor

- Necrosis Factor-Alpha in Experimental Pulmonary Tuberculosis in the Guinea Pig." <u>Tuberculosis</u> 85 (2005): 245-258. PubMed: 15958260.
- 2. David N. McMurray, personal communication.

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Table 1. Amino acid sequence of expressed TNF- α

MRGSHHHHHH GALVPRGXYY XRSASQNDND KPVAHVVANQ QAEEELQWLS KRANALLANG MGLSDNQLVV PSDGLYLIYS QVLFKGQGCP SYLLLTHTVS RLAVSYPEKV NLLSAIKSPC QKETPEGAER KPWYEPIYLG GVFQLQKGDR LSAEVNLPQY LDFADSGQIY FGVIAL

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