

Certificate of Analysis for NR-140

Anthrax Protective Antigen (PA), Recombinant from Bacillus anthracis

Catalog No. NR-140

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

Product Description: Recombinant anthrax protective antigen (PA) was produced in a non-sporulating avirulent strain of *Bacillus anthracis* lacking both of the wild type plasmids, pX01 and pX02. Recombinant PA was purified using conventional chromatographic techniques. The resulting purified protein lacks all other anthrax virulence factors.

Lot: 1715AA Manufacturing Date: 10OCT2002

TEST	SPECIFICATIONS	RESULTS
Visual Appearance (after reconstitution)	Clear and colorless	Clear and colorless
SDS-PAGE (Coomassie Blue densitometer scan)	~80–90 kDa band is > 90% of total density	~80–90 kDa band is 96% of total density
Electrospray Mass Spectrometry (MW)	Report results (expected MW is 82,674 Da)	Interference by 17,688 Da species
MALDI-TOF Mass Spectrometry of Trypsin Digest	> 50% of total residues accounted for in peptides of expected mass	75% of total residues accounted for in peptides of expected mass
Functional Activity ELISA ¹	Report results	Within 5.3% of reference PA
Cytotoxicity assay [determination of effective concentration (EC ₅₀) by titration of PA in J774A.1 macrophage cell line]	PA with 1 μg/mL LF: $EC_{50} \le 500$ pM 1 μg/mL PA alone: $EC_{50} \ge 10,000$ pM 1 μg/mL LF alone: $EC_{50} \ge 10,000$ pM	$EC_{50} = 122 \text{ pM}$ $EC_{50} \ge 10,000 \text{ pM}$ $EC_{50} \ge 10,000 \text{ pM}$
Microbial Content	No detectable colony-forming units in 0.2 mL final product	No detectable colony-forming units in 0.2 mL final product
Endotoxin Content (Limulus Amoebocyte Lysate assay) ²	< 0.5 EU endotoxin per μg protein	0.012 EU endotoxin per μg protein
Contaminating Protease Detection 3 hour incubation of 1 mg/mL solution at 37°C	Major band is > 85% initial density	Major band is 98% initial density
Absorbance Ratio (OD ₂₈₀ /OD ₂₆₀)	≥ 1.7	1.73
Absorbance Ratio (OD ₂₈₀ /OD ₃₁₀)	≥ 10	14.6

Quinn, C. P., et al. "Specific, Sensitive and Quantitative Enzyme-Linked Immunosorbent Assay for Human Immunoglobulin G Antibodies to Anthrax Protective Antigen." Emerg. Infect. Dis. 8 (2002): 1103–1110. PubMed: 12396924.

²Performed by LAL Testing Service, Cambrex BioScience

Date: 24 MAY 2012

Signature: Dorothy C. Young

Title: Technical Manager, BEI Authentication or designee

BEI Resources www.beiresources.org E-mail: contact@beiresources.org

Tel: 800-359-7370 Fax: 703-365-2898



Certificate of Analysis for NR-140

ATCC[®], on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected by the contractor to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC[®]'s knowledge.

ATCC® is a trademark of the American Type Culture Collection. You are authorized to use this product for research use only. It is not intended for human use.

BEI Resources www.beiresources.org E-mail: contact@beiresources.org

Tel: 800-359-7370 Fax: 703-365-2898