

Certificate of Analysis for NR-860

Shiga Toxin Type 1 Subunit B, Recombinant from Escherichia coli

Catalog No. NR-860

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

Product Description: NR-860 is a recombinant form of the B subunit of Shiga toxin type 1 (Stx1). The amino acid sequence includes a C-terminal hexa-histidine tag and amino acid residues 21 to 89 of the Stx1 subunit B protein (GenPept: AAA98348). The recombinant protein was expressed in *Escherichia coli* and purified by nickel affinity chromatography.

Lot: 57680170 Manufacturing Date: 01NOV2007

TEST	SPECIFICATIONS	RESULTS
Appearance	Clear and colorless, no particulate matter	Clear and colorless, no particulate matter
SDS-PAGE (SYPRO Orange densitometer scan)	Dominant band of expected size (8.513 kDa) > 95% pure	Dominant band of ~ 8 kDa 98% pure (Figure 1)
Mass Spectrometry	Measured mass within 5% of expected mass based on amino acid sequence: 8,513 daltons	Measured mass within 0.1% of expected mass based on amino acid sequence: 8,522 daltons
SELDI-TOF Mass Spectrometry of Lys-C Digest	> 50% of total residues accounted for in peptides of expected mass	77% of total residues accounted for in peptides of expected mass
Concentration by Bicinchoninic Acid Protein Assay	Report results	1.05 mg/mL
Functional Activity by Western Blot Analysis Mouse monoclonal antibody to Stx1 subunit B (BEI Resources NR-844) Carbonic anhydrase	Reactive Non-reactive	Reactive (Figure 2) Non-reactive
Cytotoxicity in Vero Cells NR-860 NR-857 (Stx1 recombinant holotoxin)	Report results Report results	Non-cytotoxic at 1x10 ⁻⁷ M CD ₅₀ ~ 1x10 ⁻¹⁶ M
Sterility	0.22 µm filter sterilized	0.22 µm filter sterilized
Endotoxin Content (Limulus Amoebocyte Lysate assay)	Report results	125 EU/mg
Absorbance Ratio (OD ₂₈₀ /OD ₂₆₀)	Report results	1.57

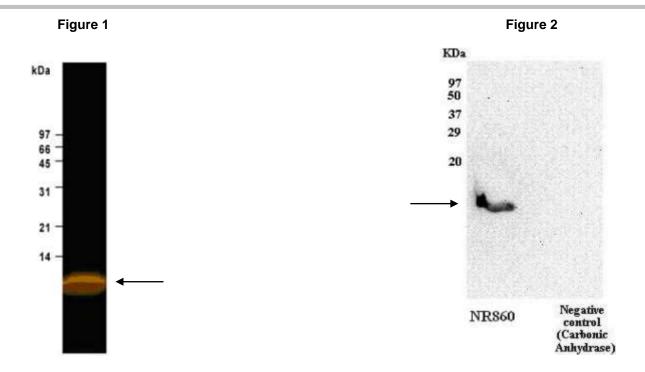
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Date: 04 FEB 2008 **Signature:** Signature on File

Title: Technical Manager, BEI Authentication or designee

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