

Certificate of Analysis for NR-857

Shiga Toxin Type 1, Recombinant from Escherichia coli

Catalog No. NR-857

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

Product Description: Recombinant Shiga toxin type 1 was expressed in *Escherichia coli* and purified by affinity chromatography.

Lot: ¹ 6176493 Manufacturing Date: MAR2006

TEST	SPECIFICATIONS	RESULTS
Appearance	Clear and colorless	Clear and colorless
SDS-PAGE (SYPRO Orange densitometer scan)	2 dominant bands ~ 31 kDa; ~ 8 kDa > 95% pure	2 dominant bands ~ 31 kDa, ~ 8 kDa > 98% pure (see Figure 1)
Mass Spectrometry	Measured mass within 5% of expected mass based on amino acid sequence: ~ 32,200 daltons, A subunit ~ 7,700 daltons, B subunit	Measured mass within 3% of expected mass based on amino acid sequence: 32,180 daltons, A subunit ² 7,709 daltons, B subunit
SELDI-TOF Mass Spectrometry of Trypsin Digest	> 50% of total residues accounted for in peptides of expected mass	> 50% of total residues accounted for in peptides of expected mass
Concentration by Bicinchoninic Acid Protein Assay	Report results	0.05 mg/mL
Functional Activity by Western Blot Analysis Rabbit polyclonal antibody ³ to Shiga toxin 1	Reactive	Reactive (see Figure 2)
Cytotoxicity 50% cytotoxic dose in Vero cells	Report results	~ 1 X 10 ⁻¹⁶ M
Sterility	0.22 µm filter sterilized	0.22 µm filter sterilized

¹NR-857 was not tested for endotoxin activity due to the inherent toxicity of the protein itself. The toxicity of Shiga toxin is so much greater than that of any contaminating endotoxin that any contribution by endotoxin to the overall toxicity is minor.

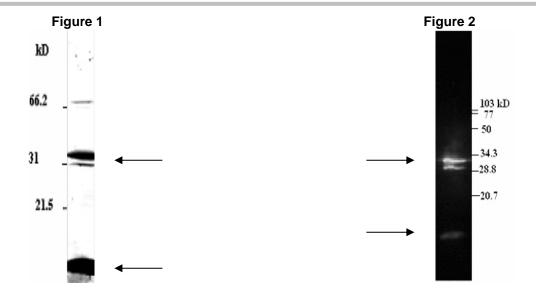
800-359-7370

²As expected, due to post-translational modification.

³O'Brien, A. D., G. D. LaVeck, D. E. Griffin, and M. R. Thompson. "Characterization of *Shigella dysenteriae* 1 (Shiga) Toxin Purified by Anti-Shiga Toxin Affinity Chromatography." <u>Infect. Immun.</u> 30 (1980): 170–179. PubMed: 7002787.



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Date: 01 FEB 2008 **Signature:** Signature on file

Title: Technical Manager, BEI Authentication or designee

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