

**Shiga Toxin Type 2 Toxoid, Recombinant from *Escherichia coli***

**Catalog No. NR-4676**

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

**Product Description:** NR-4676 is a recombinant toxoid of Shiga toxin type 2 (Stx2) with genetic mutations (Y77S, E167Q and R170L; based on the recombinant sequence) in the catalytic A subunit which render the protein non-toxic. The recombinant protein was expressed in *Escherichia coli* and purified by nickel affinity chromatography.

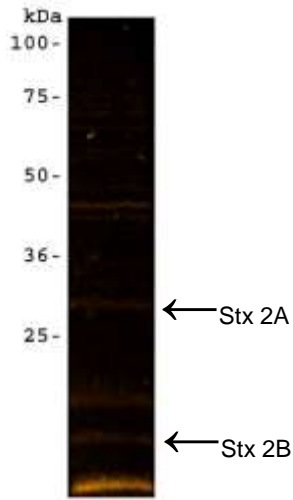
**Lot: 59062961**

**Manufacturing Date: 05JUN2011**

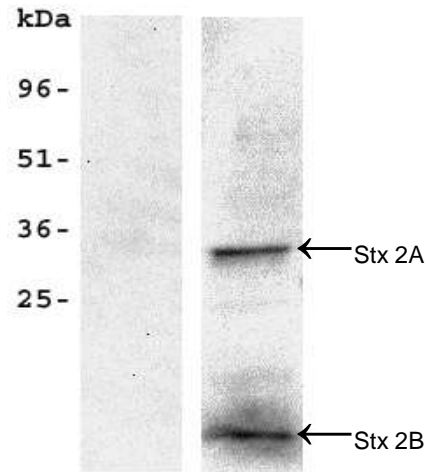
TEST	SPECIFICATIONS	RESULTS
<b>Appearance</b>	Clear and colorless, no particulate matter	Clear and colorless, no particulate matter
<b>SDS-PAGE (SYPRO Orange densitometry)</b>	Dominant bands of expected size: Stx2A: 33.1 kDa Stx2B: 8.6 kDa	Dominant bands of expected size (Figure 1): Stx2A: ~ 33 kDa Stx2B: ~ 8 kDa
<b>SELDI-TOF Mass Spectrometry</b>	Measured mass within 5% of expected mass based on amino acid sequence: Stx2A: 33074 Da Stx2B: 8640 Da	Measured mass within 0.1% of expected mass based on amino acid sequence: Stx2A: 33394 Da Stx2B: 8596 Da
<b>SELDI-TOF Mass Spectrometry of Trypsin Digest</b>	> 50% of total residues accounted for in peptides of expected mass	Stx2A: 56% of total residues accounted for in peptides of expected mass Stx2B: Resistant to trypsin digestion
<b>Concentration by Bicinchoninic Acid Protein Assay</b>	0.05 mg/mL ± 5%	0.05 mg/mL
<b>Functional Activity by Western Blot Analysis<sup>1</sup></b> NR-4676 Carbonic anhydrase	Reactive Non-reactive	Reactive (Figure 2) Non-reactive
<b>Cytotoxicity in Vero Cells (48 hours)</b> NR-4676 Stx2	≥ 10 <sup>4</sup> or more less toxic than holotoxin Report results	Non-cytotoxic at 1 x 10 <sup>-7</sup> M (Figure 3) CD <sub>50</sub> ~ 1 x 10 <sup>-14</sup> M
<b>Sterility</b>	0.22 µm filter-sterilized	0.22 µm filter-sterilized
<b>Endotoxin Content (Limulus Amoebocyte Lysate assay)</b>	Report results	≤ 25 EU per mg protein

<sup>1</sup>Rabbit polyclonal antibody to Stx2

**Figure 1**

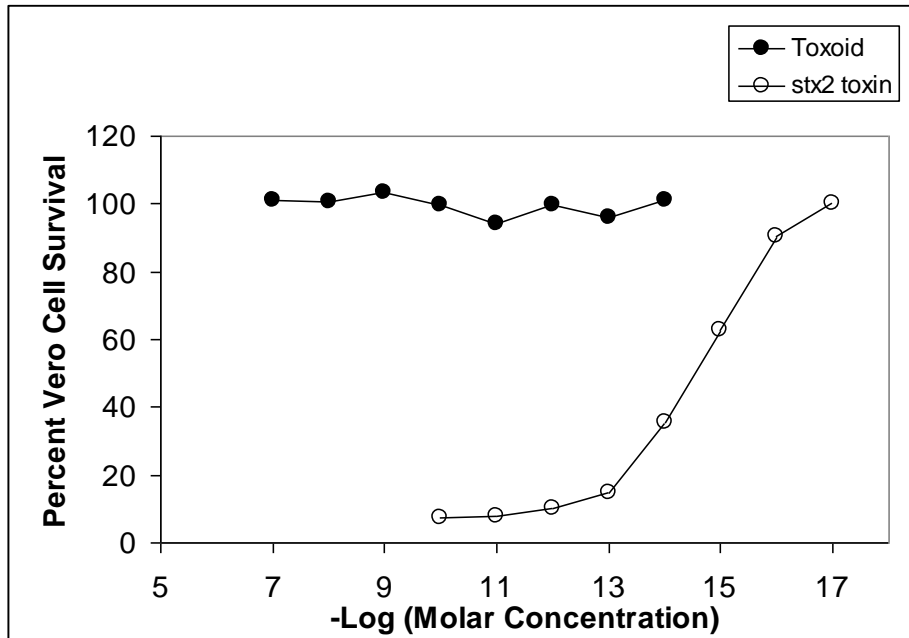


**Figure 2**



Lane 1: Carbonic anhydrase  
(negative control)  
Lane 2: NR-4676

**Figure 3**



**Date:** 17 OCT 2011

**Signature:** *Dorothy C. Young*

**Title:** Technical Manager, BEI Authentication or designee

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