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

**Catalog No. NR-4676**
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**Contributor and Manufacturer:**
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**Product Description:**
NR-4676 is a recombinant toxoid of Shiga toxin type 2 (Stx2) with genetic mutations in the catalytic A subunit which render the protein non-toxic. The recombinant B subunit includes a C-terminal hexa-histidine tag. The recombinant toxoid was expressed in *Escherichia coli* (*E. coli*) and purified by nickel affinity chromatography. NR-4676 has a theoretical molecular weight of approximately 33080 daltons for subunit A and 8640 daltons for subunit B. The predicted amino acid sequence of NR-4676 is shown below in Table 1 (Subunit A) and Table 2 (Subunit B).

The Shiga toxin (Stx) family refers to two types of related toxins: Shiga toxin type 1 (Stx1, Shiga-like toxin 1, or verotoxin 1) and Shiga toxin type 2 (Stx2, Shiga-like toxin 2, or verotoxin 2). Stx1 is almost identical to Shiga toxin produced by *Shigella dysenteriae* (*S. dysenteriae*) at the nucleotide sequence level, while Stx2 shares approximately 55% overall nucleotide sequence homology with Stx1 and Shiga toxin. Shiga toxins are multimeric molecules that are comprised of two polypeptide subunits, A and B. The B subunit is a pentamer that binds the toxin to glycolipids on host cell membranes and the entire toxin molecule can then enter the cell via endocytosis. Once inside the cell, the A subunit undergoes proteolytic cleavage and the reduction of an internal disulfide bond to generate Stx A1 and Stx A2. Stx A1 is an N-glycosidase that catalytically inactivates the 28S ribosomal RNA subunit to inhibit protein synthesis.

The sequences of the structural genes for Shiga toxin from *S. dysenteriae* and Shiga toxin type 2 from *E. coli* have been determined. The crystal structure of Shiga toxin from *S. dysenteriae* and Shiga toxin type 2 from *E. coli* have been solved (PDB: 1DM0 and 1R4P, respectively).

**Material Provided:**
Each vial of NR-4676 contains approximately 50 µg of recombinant Stx2 toxoid suspended in phosphate buffered saline. The concentration, expressed as mg per mL, is shown on the Certificate of Analysis.

**Packaging/Storage:**
NR-4676 was packaged aseptically in plastic cryovials. The product is provided frozen on dry ice and should be stored at -20°C or colder immediately upon arrival. Repeated freeze-thaw cycles should be avoided.

**Functional Activity:**
NR-4676 reacts with rabbit polyclonal antibody to Stx2 and is not cytotoxic in Vero cells.

**Citation:**
Acknowledgment for publications should read “The following reagent was obtained through BEI Resources, NIAID, NIH: Shiga Toxin Type 2 Toxoid, Recombinant from *Escherichia coli*, NR-4676.”

**Biosafety Level:**
1

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References:

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### Table 1 – Predicted Protein Sequence for Stx2 Subunit A

| 1  | REFTIDFSTQ QSYVSSLNSI RTEISTPLEH ISQGTTSVSV INHTPFGSYF |
| 51 | AVDIRGLDVY QARFDHLRLI IEQNNLS*VAG FVTATNTFY RFSDFTHISV |
|101 | PGVTTVSMTT DSSYTLQRV AALERSGMIQI SRHSLVSSYL ALMEFSGNTM |
|151 | TRDARAVLVR FVTVAQ*ALL* FRQIQREQFR ALSETAPVYT MTPGDVDLTL |
|201 | NWGRISNVLIP EYRGEDGVVR GRISFNISAI LGTAVILN CHHQGARSVR |
|251 | AVNESSQPEC QITGDRPVIK INNLWESNT AAAFLNRKSF FLYTGGK |

*Mutagenized catalytic residues Y77S, E167Q and R170L. The recombinant protein does not contain signal peptide residues.

### Table 2 – Predicted Protein Sequence for Stx2 Subunit B

| 1  | ADCAKKGKIEF SKYNEDDTFT VKVDGKEYWT SRWNLQPLLQ SAQLTGMTVT |
| 51 | IKSCTCESGS GFAEVQFNND HHHHHH |

Non-shiga toxin residues are underlined. The recombinant protein does not contain signal peptide residues.