

Shiga Toxin Type 2, Recombinant from *Escherichia coli*

Catalog No. NR-4478

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Contributor and Manufacturer:

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Product Description:

Recombinant Shiga toxin type 2 was expressed in *Escherichia coli* (*E. coli*) and purified by affinity chromatography.

The *E. coli* Shiga toxins (Stx) refer to two types of related toxins: Shiga toxin type 1 (Stx1; also referred to as Shiga-like toxin 1 or verotoxin 1) and Shiga toxin type 2 (Stx2; also referred to as Shiga-like toxin 2 or verotoxin 2).¹ Stx1 is almost identical at the nucleotide level to the Shiga toxin produced by *Shigella dysenteriae*, while Stx2 shares only 55% nucleotide sequence homology with both 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.² 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.³

The sequences of Stx2 for multiple strains of *E. coli* have been determined and the crystal structure of Stx2 from *E. coli* has been resolved (PMB: 1R4P).⁴ The predicted amino acid sequence of NR-4478 is shown below in Table 1 (Stx2A) and 2 (Stx2B).

Material Provided:

Each vial of NR-4478 contains approximately 10 µg of Stx2 suspended in PBS. The concentration, expressed as mg per mL, is shown on the Certificate of Analysis.

Packaging/Storage:

NR-4478 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-4478 (lot 57680181) has been shown to react with a mixture of monoclonal antibody to Stx2A (BEI Resources NR-846) and rabbit polyclonal antibody to Stx2B (BEI Resources NR-9352). NR-4478 demonstrates cytotoxicity in Vero cells.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: Shiga Toxin Type 2, Recombinant from *Escherichia coli*, NR-4478."

Biosafety Level: 2

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. Biosafety in Microbiological and Biomedical Laboratories. 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see www.cdc.gov/biosafety/publications/bmb15/index.htm.

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References:

1. Nakao, H. and T. Takeda. "Escherichia coli Shiga Toxin." *J. Nat. Toxins* 9 (2000): 299–313. PubMed: 10994531.
2. Sandvig, K., et al. "Endocytosis from Coated Pits of Shiga Toxin: A Glycolipid-binding Protein from *Shigella dysenteriae* 1." *J. Cel Biol.* 108 (1989): 1331-1343. PubMed: 2564398.
3. Skinner, L. M., and M. P. Jackson. "Investigation of Ribosome Binding by the Shiga Toxin A1 Subunit, Using Competition and Site-Directed Mutagenesis." *J. Bacteriol.* 179 (1997): 1368-1374. PubMed: 9023224.

4. Fraser, M. E., et al. "Structure of Shiga Toxin Type 2 (Stx2) from *Escherichia coli* O157:H7." *J. Biol. Chem.* 279 (2004): 27511–27517. PubMed: 15075327. PDB: 1R4P.
5. Lee, J. E., et al. "Phylogenetic Analysis of Shiga Toxin 1 and Shiga Toxin 2 Genes Associated with Disease Outbreaks." *BMC Microbiol.* 7 (2007): 109. PubMed: 18053224.

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Table 1

Predicted Protein Sequence, Subunit A					
1	REFTIDFSTQ	QSYVSSLNSI	RTEISTPLEH	ISQGTTSVSV	INHTPPGSYF
51	AVDIRGLDVY	QARFDHLRLI	IEQNNLYVAG	FVNTATNTFY	RFSDFTHISV
101	PGVTTVSMIT	DSSYTTLQRV	AALERSGMQI	SRHSLVSSYL	ALMEFSGNTM
151	TRDASRAVLR	FVTVTAEALR	FRQIQREFRQ	ALSETAPVYT	MTPGDVDLTL
201	NWGRISNVLP	EYRGEDGVRV	GRISFNNISA	ILGTVAVILN	CHHQGARSVR
251	AVNEESQPEC	QITGDRPVIK	INNTLWESNT	AAAFLLNRKSQ	FLYTTGK

Table 2

Predicted Protein Sequence, Subunit B					
1	ADCAK GKIEF	SKYNEDDTFT	VKVDGKEYWT	SRWNLQPLLQ	SAQLTGMTVT
51	IKSSTCESGS	GFAEVQFNND			