

## Staphylococcal Enterotoxin B Toxoid, Recombinant from *Escherichia coli*

### Catalog No. NR-10049

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#### Contributor and Manufacturer:

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#### Product Description:

NR-10049 is a recombinant form of Staphylococcal Enterotoxin B (SEB) with genetic mutations which render the protein non-toxic.<sup>1</sup> That is, NR-10049 is a genetically engineered toxoid. The recombinant protein was expressed in *Escherichia coli* and purified by nickel affinity chromatography. NR-10049 contains a hexa-histidine tag near the N-terminus, and has a theoretical molecular weight of approximately 30 kilodaltons. The predicted amino acid sequence of NR-10049 is shown below in Table 1.

SEB is one of several exotoxins produced by *S. aureus* subsp. *aureus*. *S. aureus* subsp. *aureus* is a ubiquitous, nonmotile, Gram-positive coccus found on the skin and mucous membranes of humans and animals. The staphylococcal exotoxins are characterized as enterotoxins, because they exert their effect on the intestinal tract when ingested. SEB has a broad spectrum of biological activity, and depending on the portal of entry (e.g., gastrointestinal, respiratory, or mucosal), the toxin will elicit a different clinical syndrome. The amino acid sequence of SEB from *S. aureus* subsp. *aureus*, strain COL has been determined (GenPept: AAW37877).<sup>2</sup> The crystal structure of SEB has been solved to 1.48 Å (PDB: 3SEB).<sup>3</sup>

Staphylococcal enterotoxins are broadly classified as superantigens because they have the ability to directly bind both class II major histocompatibility complex (MHC) molecules on antigen presenting cells and the variable region of the T cell receptor  $\beta$  chains. This bridging of antigen presenting cells and T cells bypasses the normal antigen processing and presentation system, leading to stimulation of large populations of both CD4 and CD8 T cells. The massive T cell activation in turn, results in rapid overproduction of proinflammatory cytokines and chemokines.<sup>4</sup> Superantigen-induced inflammation is responsible for many of the effects of the staphylococcal enterotoxins. Syndromes associated with SEB exposure range from classic food poisoning to acute toxic shock.

The specific amino acid substitutions in NR-10049 completely

abrogate its ability to bind to class II MHC molecules, thus destroying the superantigenic activity of the toxin.<sup>1</sup>

#### Material Provided:

Each vial of NR-10049 contains approximately 1 mg of SEB toxoid in phosphate buffered saline (pH 7.4). The concentration is shown on the Certificate of Analysis.

#### Packaging/Storage:

NR-10049 was packaged aseptically in plastic cryovials. The product is provided frozen on dry ice and should be stored at -70°C or colder immediately upon arrival. Note: The label for NR-10049, lot 58152420, incorrectly indicates that it should be stored at -20°C. Repeated freeze-thaw cycles should be avoided.

#### Functional Activity:

NR-10049 reacts with rabbit polyclonal antibody to SEB using western blot analysis.

#### Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: Staphylococcal Enterotoxin B Toxoid, Recombinant from *Escherichia coli*, NR-10049."

#### Biosafety Level: 1

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](http://www.cdc.gov/biosafety/publications/bmb15/index.htm).

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**References:**

- Ulrich, R. G., M. A. Olson and S. Bavari. "Development of Engineered Vaccines Effective against Structurally Related Bacterial Superantigens." *Vaccine* 16 (1998): 1857-1864. PubMed: 9795392.
- Gill, S. R., et al. "Insights on Evolution of Virulence and Resistance from the Complete Genome Analysis of an Early Methicillin-Resistant *Staphylococcus aureus* Strain

- and a Biofilm-Producing Methicillin-Resistant *Staphylococcus epidermidis* Strain." *J. Bacteriol.* 187 (2005): 2426-2438. PubMed: 15774886. GenPept: AAW37877.
- Papageorgiou, A. C., H. S. Tanter and K. R. Acharya. "Crystal Structure of Microbial Superantigen Staphylococcal Enterotoxin B at 1.5 Å Resolution: Implications for Superantigen Recognition by MHC Class II Molecules and T-cell Receptors." *J. Mol. Biol.* 277 (1998): 61-79. PubMed: 9514739. PDB: 3SEB.
- Pinchuk, I. V., E. J. Beswick and V. E. Reyes. "Staphylococcal Enterotoxins." *Toxins* 2 (2010): 2177-2197. PubMed: 22069679.
- Nema, V., et al. "Isolation and Characterization of Heat Resistant Enterotoxigenic *Staphylococcus aureus* from a Food Poisoning Outbreak in Indian Subcontinent." *Int. J. Food Microbiol.* 117 (2007): 29-35. PubMed: 17477998.
- Casman, E. P., M. S. Bergdoll and J. Robinson. "Designation of Staphylococcal Enterotoxins." *J. Bacteriol.* 85 (1963): 715-716. PubMed: 14042955.

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| Table 1 – Predicted Protein Sequence for SEB Toxoid |                   |                   |            |            |                     |
|-----------------------------------------------------|-------------------|-------------------|------------|------------|---------------------|
| 1                                                   | <u>MRGSHHHHHH</u> | <u>GIRQPDPKPD</u> | ELHKSSKFTG | LMENMKVLYD | DNHVSAINVK          |
| 51                                                  | SIDQFR*YFDL       | IYSIKDTKLG        | NYDNVRVEFK | NKDLADKYKD | KYVDVFGANA*         |
| 101                                                 | YYQCA*FSKKT       | NDINSHQTDK        | RKTCMYGGVT | EHNGNQLDKY | RSITVRVFD           |
| 151                                                 | GKNLLSFDVQ        | TNKKKVTAQE        | LDYLTRHYLV | KNKKLYEFNN | SPYETGYIKF          |
| 201                                                 | IENENSFWDY        | MMPAPGDKFD        | QSKYLMMYND | NKMVDSKDVK | IEVYLTTL <u>Q</u> P |
| 251                                                 | <u>SLIS</u>       |                   |            |            |                     |

\*Mutagenized residues L56R, Y100A, Y105A based on the recombinant sequence (L45R, Y89A, Y94A in the native sequence). Non-SEB toxin residues are underlined. The recombinant protein does not contain signal peptide residues.