

## **Certificate of Analysis for NR-856**

## Epsilon Protoxin, from Clostridium perfringens, Strain 34 (Type B)

Catalog No. NR-856

This reagent is the property of the U.S. Government.

**Product Description:** Epsilon protoxin was purified from culture supernatants of *Clostridium* perfringens (*C. perfringens*), strain 34 (type B) (ATCC<sup>®</sup> 3626<sup>™</sup>). The protein is suitable for western blots or can be treated with trypsin to generate the active toxin for cytotoxicity assays.

Lot<sup>1</sup>: 63732546 Manufacturing Date: 28SEP2015

TEST	SPECIFICATIONS	RESULTS
Appearance	Clear and colorless	Clear and colorless
SDS-PAGE (SYPRO Orange densitometer scan) (Figure 1)	Dominant band ~ 32 kDa > 95% pure	Dominant band ~ 32 kDa > 98% pure
SELDI-TOF Mass Spectrometry	Measured mass within 5% of expected mass (32940 Da)	Measured mass (32896 Da) within 0.1% of expected mass
SELDI-TOF Mass Spectrometry of Protease Digest Glutamic C endopeptidase	> 50% of total residues accounted for in peptides of expected mass	78% of total residues accounted for in peptides of expected mass
Concentration by Bicinchoninic Acid Protein Assay	Report results	0.5 mg/mL
Functional Activity Western Blot (Figure 2) <sup>2</sup> Carbonic anhydrase Epsilon protoxin (NR-856)	Non-reactive Reactive	Non-reactive Reactive
Cytotoxicity in MDCK cells (Figure 3) Epsilon protoxin (NR-856)	Report results	CD <sub>50</sub> epsilon protoxin is ~100-fold less than the activated toxin
Sterility	0.22 µm filter-sterilized	0.22 µm filter-sterilized
Endotoxin Content	1 to 10000 EU/mg	< 125 EU/mg
Absorbance Ratio (OD <sub>280</sub> /OD <sub>260</sub> )	Report results	1.6

<sup>&</sup>lt;sup>1</sup>This item was manufactured and subjected to quality control testing by Uniformed Services University of the Health Sciences, Bethesda, Maryland, USA

**Date:** 10 MAY 2016

Signature:

**BEI Resources Authentication** 

ATCC<sup>®</sup>, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected by the contractor to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC<sup>®</sup>'s knowledge.

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<sup>&</sup>lt;sup>2</sup>Completed with primary antibody BEI Resources NR-865 and secondary antibody donkey anti-rabbit IgG F(ab')<sub>2</sub>.

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Figure 1: SDS-PAGE

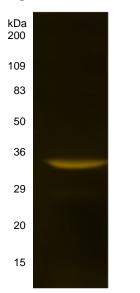
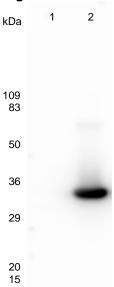
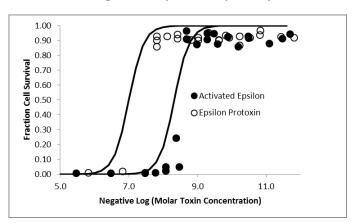


Figure 2: Western Blot



Lane 1: Carbonic Anhydrase (negative control) Lane 2: *C. perfringens* Epsilon protoxin (NR-856)





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