Certificate of Analysis for NR-4769

Clostridium Botulinum Neurotoxin Type E Heavy Chain, Recombinant from *Pichia pastoris*

It has been determined that the level of production documentation for Lot 0015040P/60-0404 of NR-4769 does not support its use in studies for product licensure. BEI Resources is releasing Lot 0015040P/60-0404 for research use only.

Catalog No. NR-4769

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

Product Description: A recombinant form of a portion of the *Clostridium botulinum* type E heavy chain was expressed in *Pichia pastoris.* NR-4769 is a non-toxic, non-infective protein antigen that has been tested in rodents with no ill effects.

Lot: 0015040P/60-0404 **Manufacturing Date: 25JAN2008**

TEST	SPECIFICATIONS	RESULTS
Appearance	Clear and colorless; may contain visible flocculation	Clear and colorless
SDS-PAGE (Reducing gel densitometer scan)	Major band ~ 52 kDa Conforms to reference molecular weight (MW) > 95% pure	Major band ~ 52 kDa Conforms to reference molecular weight (MW) > 99.6% pure
Western Blot Analysis with Specific Antibody	Major band ~ 52 kDa Conforms to reference MW	Major band ~ 52 kDa Conforms to reference MW
Reverse Phase HPLC	Major peak elution time is within 20% of the elution time of the major peak of the reference Conforms to reference standard	Major peak elution time (3.8 minutes) is within 20% of the elution time of the major peak of the reference (3.72 minutes) Conforms to reference standard
Strong Cation Exchange Chromatography	Report results	100% pure
Concentration by Absorbance at OD_{280}	0.20 ± 0.05 mg/mL[1]	0.14 mg/mL[2]
pH	3.8 – 4.2	3.94
Sterility	0.22 µm filter sterilized	0.22 µm filter sterilized
Bioburden	≤ 1 colony-forming unit per mL	0 colony-forming unit per mL
Endotoxin Content (Limulus Amoebocyte Lysate assay)	< 17.5 EU per mg	< 6.75 EU per mg

[1]Limit is set to gather information and not used for disposition.

[2]The result reported is 0.01 mg/mL below the limits value. This is potentially due to inherent assay variability and/or due to a post-thaw 0.22 µm absolute filtration performed prior to filling and/or due to the additional freeze/thaw cycle which the vialed product underwent prior to product release analysis. The concentration does not negatively affect the product quality or use of the product.

Biodefense and Emerging Infections Research Resources Repository
P.O. Box 4137
Manassas, VA 20108-4137 USA
www.beiresources.org
800-359-7370
Fax: 703-365-2898
E-mail: contact@beiresources.org

Certificate of Analysis for NR-4769

Date: 16 APR 2008

Signature: Signature on File

Title: Technical Manager, BEI Authentication or designee

ATCC®, 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®'s knowledge.

Biodefense and Emerging Infections Research Resources Repository
P.O. Box 4137
Manassas, VA 20108-4137 USA
www.beiresources.org

800-359-7370
Fax: 703-365-2898
E-mail: contact@beiresources.org