

CERTIFICATE OF ANALYSIS
Heavy Chain Binding Domain: Botulinum Neurotoxin Type A
Lot # 6122A1

Contents

Each vial of recombinant Botulinum Neurotoxin Type A Heavy Chain Binding Domain (HccA) contains 50 µg of lyophilized protein. When reconstituted with 100 µl of sterile distilled water, each vial contains 50 µg of HccA in 20 mM HEPES, pH 7.4 + 150mM NaCl + 1.25% lactose. The protein was recombinantly expressed in *E. coli* and purified using affinity and anion exchange chromatography. The affinity tag has subsequently been cleaved off of the protein prior to quantitation and packaging.

Molecular Weight

HccA is 436 amino acids in length. This product contains 10 residual amino acids from the GST tag and amino acids 872-1296 of the Botulinum Neurotoxin Type A Hall Strain. The molecular weight of the protein is approximately 51 kD.

Concentration

Protein concentration was determined by absorbance at 280nm using $Abs(0.1\%) = 1.777$. This value is calculated by ProtParam¹ using an algorithm based on the Edelhoch² method with modifications described in Pace et al³.

Gel Electrophoresis

This product migrates as a single major band on 4-12% SDS polyacrylamide gels with an apparent molecular weight of 50 kD. The protein is approximately 85% pure based on densitometric analysis.

Activity

HccA is reactive to anti-Botulinum Type A antibodies in a Western Blot. HccA was tested in an ELISA assay using GT1b and the receptor domain of SV2c. The binding to GT1b can be detected down to 4 ng of HccA. The binding to SV2c can be detected down to 4 ng of HccA.

Packaging and Storage

This product is supplied as a lyophilized powder which has been stoppered under vacuum. Store lyophilized vials at 2-8°C. Once dissolved, aliquot and store the protein at -20°C. Refrain from multiple freeze/thaw cycles.

Toxicity

HccA is only a fragment of Botulinum toxin and, as such, is a non toxic protein.

(continued)

Handling

Good laboratory technique should be employed in the safe handling of this product. Wear appropriate laboratory attire including a lab coat, gloves and safety glasses. Nitrile gloves are recommended when handling lyophilized material. This product is intended for research purposes by qualified personnel only. It is not intended for use in humans or as a diagnostic agent. List Biological Laboratories, Inc. is not liable for any damages resulting from the misuse or handling of this product.

FOR RESEARCH PURPOSES ONLY. NOT FOR HUMAN USE.

References

1. www.expasy.ch/tools/protparam-doc.html
2. Edelhoch, H. (1967) Biochemistry, 6: 1948-1954.
3. Pace, C.N., Vajdos, F., Fee, L., Grimsley, G., and Gray, T. (1995) Protein Sci., 4:2411-2423.

Production: TC Date: 3/30/12 Management: NS Date: 3/29/12 QA/QC: UP Date: 3/29/12