LIST BIOLOGICAL LABORATORIES, INC.

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Product # 612A

CERTIFICATE OF ANALYSIS
Botulinum Type A Heavy Chain Binding Domain
Lot # 6121A1

Contents:

Each vial of recombinant Botulinum Neurotoxin Type A Heavy Chain Binding Domain (HccA) contains 50 μg of lyophilized protein. When reconstituted with 290 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 A Strain. The molecular weight of the protein is approximately 51 kD.

Concentration:

Protein concentration was determined by a modification of the Bradford[1] method using bovine serum albumin as a standard.

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 greater than 90% pure based on densitometry.

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 with as little as 1 ng of HccA. The binding to SV2c can be detected with as little as 4 ng of HccA.

Packaging and Storage:

This product is supplied as a lyophilized powder which has been stoppered under vacuum. Reconstitution of the powder should be done with syringe through the rubber stopper to avoid any loss of material. Store lyophilized vials at 4°C. Once dissolved, aliquot and store at -20°C. Refrain from multiple freeze/thaw cycles.

(continued)

Toxicity:

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

Handling:

Good laboratory technique should be employed in the safe handling of this product. This requires observing the following practices:

1. **Wear appropriate laboratory attire including a lab coat, gloves and safety glasses.**

2. **Do not mouth pipette, inhale, ingest or allow to come into contact with open wounds. Wash thoroughly any area of the body which comes into contact with the product.**

3. **Avoid accidental autoinoculation by exercising extreme care when handling in conjunction with any injection device.**

4. **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. Bradford, M.M. (1976) *Anal. Biochem.* 72, 248-254.

Approved: TC Date: 2/1/10 Approved: NS Date: 2/1/18