

Product # 620A

CERTIFICATE OF ANALYSIS RECOMBINANT LIGHT CHAIN from BOTULINUM NEUROTOXIN TYPE B Lot # 62015A1

Contents:

Each vial contains 10 μ g of light chain from botulinum neurotoxin type B. When reconstituted with 0.1 ml of water, the concentration of the buffer is 20 mM HEPES, pH 7.4 + 1.25% lactose. In order to ensure stability during storage of the protein, 0.05% TWEEN-20 or 1 mg/ml BSA must be included in the reconstitution buffer. Handle the product gently; do not vortex.

The protein was recombinantly expressed in *E. coli* and purified using affinity and anion exchange chromatography. The affinity tag has subsequently been cleaved from the protein prior to quantitation and packaging.

Molecular Weight:

The light chain type B fragment contains amino acids 1 - 436 of the full length botulinum neurotoxin type B. It also contains eight residual amino acids from the affinity tag at the N - terminus to give a total length of 444 amino acids. The molecular weight of the protein has been determined to be 51210 Da based on electorspray-MS analysis.

Concentration:

Protein concentration was determined by absorbance at 280 nm using Abs (0.1%) = 0.80. This value is calculated by ProtParam¹ using an algorithm based on the Edelhoch² method with modifications described in Pace et al³.

Gel Electrophoresis:

When examined on a 4-12% SDS-polyacrylamide gel under reducing conditions, this product migrates as a single major band with an apparent molecular weight of approximately 50,000 Da.

Activity:

The protein has been tested for activity in a FRET endopeptidase assay. Significant cleavage of the fluorogenic peptide substrate VAMPtide® (Product #540) was seen with 2.5, 5, and 10 nM enzyme.

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 product at -20°C. Refrain from multiple freeze/thaw cycles.

Toxicity:

The light chains of the botulinum neurotoxins are non-toxic and unable to penetrate cells in the absence of the heavy chain (binding and translocation domains). The expression and purification of light chain from a recombinant setting ensures there is no possible contamination with heavy chain or full length intact toxin.

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. Nitrile gloves are recommended when handling lyophilized material.
- 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. 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.

QA/QC: KPD Date: 01/22/2021

