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CERTIFICATE OF ANALYSIS
RECOMBINANT LIGHT CHAIN OF BOTULINUM NEUROTOXIN TYPE C
Lot #6256A1

Contents

Each vial of Light Chain from Botulinum Neurotoxin Type C (BoNT/C) contains 10 µg of lyophilized protein. When reconstituted with 100 µl of water, each vial contains 10 µg of light chain in 20 mM HEPES, pH 7.4 + 1.25% lactose. In order to ensure stability during storage and recovery of the protein, 0.05% TWEEN-20 or 1 mg/ml BSA must be included in the reconstitution buffer.

The protein was recombinantly expressed in *E. coli*. The affinity tag has been cleaved from the protein prior to quantitation and packaging.

Molecular Weight

The light chain C fragment contains amino acids 1 - 436 of the full length Botulinum Neurotoxin Type C. It also contains eight residual amino acids from the affinity tag at the N - terminus to give a total length of 444 amino acids. ESI - MS analysis shows the molecular weight to be 50,874 Da.

Concentration

Protein concentration was determined by absorbance at 280nm using $Abs (0.1\%) = 0.949$. 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 4-12% SDS-polyacrylamide gels, this product migrates as a single major band with an apparent molecular weight of approximately 50,000 Da. The protein is > 95% pure based on densitometric analysis.

Activity

The BoNT/C light chain has been tested for activity in a FRET based assay. Digestion of the FRET substrate was detected within two hours when using ten and five nanomolar BoNT/C Light Chain with 10µM SYNTAXtide substrate (Product #560).

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 chain of Botulinum Neurotoxin is non-toxic and unable to penetrate cells in the absence of the heavy chain. 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.

(continued)

Handling

This product is not known to be hazardous. 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 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.

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