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Product #690B

CERTIFICATE OF ANALYSIS  
GST-SV2c, RECEPTOR FOR BOTULINUM NEUROTOXIN TYPE A  
Lot #6904A1

**Contents**

Each vial, when reconstituted with 100  $\mu$ l of water contains 100  $\mu$ g of GST-SV2c (U.S Patent #8,476,024), in 20 mM HEPES, pH 7.4 + 1.25% lactose. **Handle the product gently. Do not vortex.**

The protein was recombinantly expressed in *E. coli* and purified using affinity chromatography. The GST affinity tag has been retained.

**Molecular Weight**

GST-SV2c contains amino acids 454-579 of the full length SV2c protein. This region of the protein is known as the luminal domain loop, between transmembrane domains 7 and 8, and has been shown to be the location of botulinum neurotoxin type A (BoNT/A) binding.<sup>1,2</sup> The 25 kDa GST affinity tag is on the N-terminal. The total length of the fusion protein is 330 amino acids. The molecular weight of the protein is 41,733 Da based on analysis of the nucleic acid sequence.

**Concentration**

Protein concentration was determined by absorbance at 280nm using Abs (0.1%) = 1.253. This value is calculated by ProtParam<sup>3</sup> using an algorithm based on the Edelhoch<sup>4</sup> method with modifications described in Pace et al.<sup>5</sup>

**Purity**

When examined on 4-12% SDS-polyacrylamide gels under reducing conditions, this product migrates as a single major band with an apparent molecular weight of approximately 40,000 Da. The protein purity is approximately 90% based on densitometric analysis.

**Functionality**

The following posters demonstrate the functionality of the GST-SV2c fusion protein as a receptor for BoNT/A. Recent data from our laboratory indicates improved specificity and reliable detection of as little as 1.25 pg of BoNT/A when captured by the SV2c receptor.

1. K. Suryadi, T. Christian, and N. Shine. Sensitive and Specific Bifunctional Assay for Botulinum Neurotoxin Type A. The 49th Annual Interagency Botulinum Research Coordinating Committee Meeting, September, 2012 in Baltimore, MD.
2. T. Christian and N. Shine. Sensitive, *In Vitro*, Bifunctional Potency Assay for Botulinum Neurotoxin Type A. The 46<sup>th</sup> Annual Interagency Botulinum Research Coordinating Committee Meeting, October, 2009 in Alexandria, VA.
3. T. Christian, A. Rummel, and N. Shine. Functional Assay for Botulinum Neurotoxin Type A Utilizing the Neuronal Receptor Protein SV2c. The 45<sup>th</sup> Annual Interagency Botulinum Research Coordinating Committee Meeting, September 2008 in Philadelphia, PA.

(continued)

4. T. Christian and N. Shine. Capture Assay for Botulinum Neurotoxin Type A Utilizing the Neuronal Receptor Protein SV2c. The 6<sup>th</sup> International Conference on Basic and Therapeutic Aspects of Botulinum and Tetanus Toxins, June, 2008 in Baveno, Italy.

#### **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**

GST-SV2c is non-toxic.

#### **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. Mahrhold, S., Rummel, A., Bigalke, H., Davletov, B., and Binz, T. (2006) FEBS Lett. **580**:2011-2014.
2. Dong, M., Yeh, F., Tepp, W.H., Dean, C., Johnson, E.A., Janz, R., and Chapman, E.R. (2006) Science **312**:592-596.
3. [www.expasy.ch/tools/protparam-doc.html](http://www.expasy.ch/tools/protparam-doc.html)
4. Edelhoch, H. (1967) Biochemistry, **6**:1948-1954.
5. Pace, C.N., Vajdos, F., Fee, L., Grimsley, G., and Gray, T. (1995) Protein Sci., **4**:2411-2423.

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