

Anti-SNAP25 Antibody Picoband™

Catalog Number: A01625

About SNAP25

Synaptosome-associated protein of 25,000 daltons, also known as SNAP-25, is a protein which in humans encodes a 25-kD protein of 206 amino acids. It was first investigated as a neuron-specific gene preferentially expressed in mouse hippocampus. The tSNARE (the target-membrane soluble NSF-attachment protein receptor, where NSF is N-ethylmaleimide-sensitive fusion protein) synaptosomal-associated protein of 25 kDa (SNAP-25) is expressed in pancreatic B-cells and its cleavage by botulinum neurotoxin E (BoNT/E) abolishes stimulated secretion of insulin. In the nervous system, two SNAP-25 isoforms (a and b) have been described, which are produced by alternative splicing. It is identified mammalian Snap25a and Snap25b as targets of protein kinase A, a key regulator of neurosecretion that primes slowly releasable pools and readily releasable pools of secretory vesicles. SNAP-25 inhibits P/Q- and L-type voltage-gated calcium channels located presynaptically and interacts with the synaptotagmin C2B domain in Ca²⁺-independent fashion. In glutamatergic synapses SNAP-25 decreases the Ca²⁺ responsiveness, while it is naturally absent in GABAergic synapses.

Overview

Product Name	Anti-SNAP25 Antibody Picoband™
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-SNAP25 Antibody Picoband™ catalog # A01625. Tested in ELISA, Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
Application	ELISA, Flow Cytometry, IF, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na ₂ HPO ₄ , 0.05mg NaN ₃ .
Storage Instructions	Store at -20°C for one year from date of receipt. After reconstitution, at 4°C for one month. It can also be aliquotted and stored frozen at -20°C for six months. Avoid repeated freeze-thaw cycles.
Host	Rabbit
Uniprot ID	P60880

Technical Details

Immunogen	E. coli-derived human SNAP25 recombinant protein (Position:M1-L203).
Predicted Reactive Species	Human
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western blot, and HRP Conjugated anti-Rabbit IgG Super Vision Assay Kit (SV0002-1) for IHC(P) and ICC.
Cross Reactivity	No cross-reactivity with other proteins.
Isotype	Rabbit IgG

Form	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Purification	Immunogen affinity purified.
Suggested Dilutions	<p>Dilute the sample so that the expected range of concentrations fall within the detection range of this kit.</p> <p>If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples.</p> <p>Some PubMed article(s) citing the expression level of this target are as follows:</p> <p>Boster Bio's internal QC testing used:</p> <p>Western blot, 0.1-0.5ug/ml</p> <p>Immunohistochemistry (Paraffin-embedded Section), 0.5-1ug/ml</p> <p>Immunocytochemistry/Immunofluorescence, 5ug/ml</p> <p>Flow Cytometry, 1-3ug/1x10⁶ cells</p> <p>Direct ELISA, 0.1-0.5ug/ml</p>

Anti-SNAP25 Antibody Picoband™ (A01625) Images

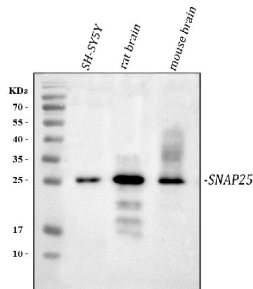


Figure 1. Western blot analysis of SNAP25 using anti-SNAP25 antibody (A01625).

Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.

Lane 1: human SH-SY5Y whole cell lysates,

Lane 2: rat brain tissue lysates,

Lane 3: mouse brain tissue lysates.

After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-SNAP25 antigen affinity purified polyclonal antibody (Catalog # A01625) at 0.5 ug/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for SNAP25 at approximately 25 kDa. The expected band size for SNAP25 is at 23 kDa.

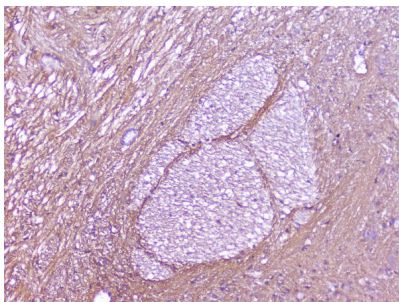


Figure 2. IHC analysis of SNAP25 using anti-SNAP25 antibody (A01625).

SNAP25 was detected in paraffin-embedded section of rat brain tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml rabbit anti-SNAP25 Antibody (A01625) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.

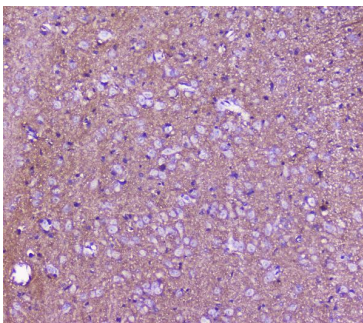
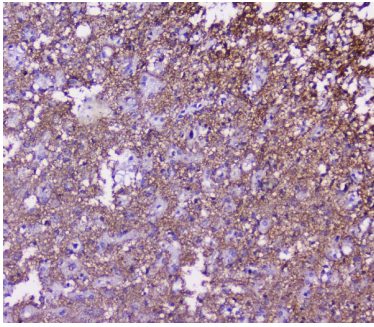


Figure 3. IHC analysis of SNAP25 using anti-SNAP25 antibody (A01625).

SNAP25 was detected in paraffin-embedded section of rat brain tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml rabbit anti-SNAP25 Antibody (A01625) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.

Figure 4. IHC analysis of SNAP25 using anti-SNAP25 antibody



(A01625). SNAP25 was detected in paraffin-embedded section of human glioma tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml rabbit anti-SNAP25 Antibody (A01625) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.

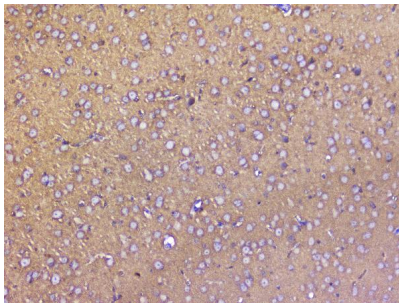


Figure 5. IHC analysis of SNAP25 using anti-SNAP25 antibody (A01625).

SNAP25 was detected in paraffin-embedded section of mouse brain tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml rabbit anti-SNAP25 Antibody (A01625) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.

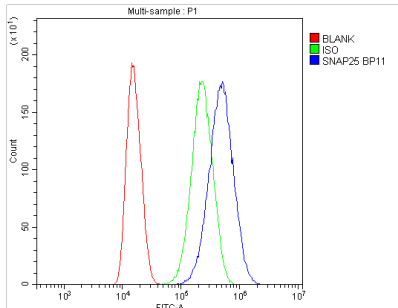


Figure 6. Flow Cytometry analysis of U87 cells using anti-SNAP25 antibody (A01625).

Overlay histogram showing U87 cells stained with A01625 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-SNAP25 Antibody (A01625, 1ug/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1ug/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

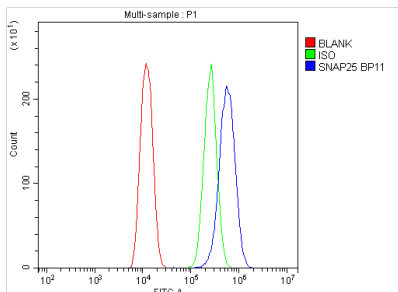
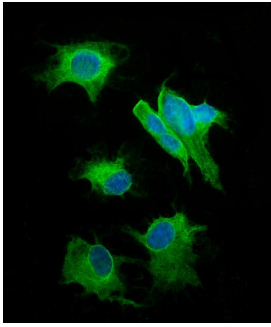


Figure 7. Flow Cytometry analysis of U20S cells using anti-SNAP25 antibody (A01625).

Overlay histogram showing U20S cells stained with A01625 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-SNAP25 Antibody (A01625, 1ug/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1ug/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

Figure 8. IF analysis of SNAP25 using anti-SNAP25 antibody (A01625).

SNAP25 was detected in immunocytochemical section of SH-SY5Y cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins.



The cells were blocked with 10% goat serum. And then incubated with 5ug/mL rabbit anti-SNAP25 Antibody (A01625) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Rabbit IgG (BA1127) was used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

4 Publications Citing This Product

1. PubMed ID: 10.1016/j.tox.2017.03.021, Disruption of glutamate neurotransmitter transmission is modulated by SNAP-25 in benzo[a]pyrene-induced neurotoxic effects
2. PubMed ID: 10.1080/00207454.2017.1387115, Effects of spontaneous recurrent seizures on cognitive function via modulation of SNAREs expression
3. PubMed ID: 32930942, Guan Y,Chen X,Zhao B,Shi Y,Han F.What Happened in the Hippocampal Axon in a Rat Model of Posttraumatic Stress Disorder.Cell Mol Neurobiol.2020 Sep 15.doi:10.1007/s10571-020-00960-w.Epub ahead of print.PMID:32930942.

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