

# Anti-Stefin B CSTB Antibody Picoband™ (monoclonal, 2B6)

Catalog Number: M02794

#### **About CSTB**

Cystatin B (CSTB), also called STFB, is a small protein that is a member of the superfamily of cysteine protease inhibitors. It has been isolated from human spleen and liver and its amino acid sequence has been fully determined. The cystatin B gene is located on 21q22.3. It is widely distributed and is localized mostly intracellularly, but has been found extracellularly. The protein is able to form a dimer stabilized by noncovalent forces, inhibiting papain and cathepsins I, h and b. Its role is thought to be as a protector against the proteinases leaking from lysosomes. A cystatin B multiprotein complex might have a specific cerebellar function, and that the loss of this function might contribute to the etiopathogenesis of EPM1. Upon differentiation to myotubes, CSTB becomes excluded from the nucleus and lysosomes, suggesting that the subcellular distribution of CSTB is dependent on the differentiation status of the cell.

### Overview

Product Name	Anti-Stefin B CSTB Antibody Picoband™ (monoclonal, 2B6)
Reactive Species	Human
Description	Boster Bio Anti-Stefin B CSTB Antibody Picoband™ (monoclonal, 2B6) catalog # M02794. Tested in Flow Cytometry, IHC, IHC-F, ICC, WB applications. This antibody reacts with Human.
Application	Flow Cytometry, IHC, IHC-F, ICC, WB
Clonality	Monoclonal 2B6
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.05mg NaN3.
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	Mouse
Uniprot ID	P04080

#### **Technical Details**

Immunogen	E. coli-derived human Stefin B recombinant protein (Position: M1-F98). Human Stefin B shares 78.6 % amino acid (aa) sequence identity with both mouse and rat Stefin B.
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Mouse IgG (EK1001) for Western blot, and HRP Conjugated anti-Mouse IgG Super Vision Assay Kit (SV0001-1) for IHC(P), IHC(F) and ICC.
Cross Reactivity	No cross-reactivity with other proteins.
Isotype	Mouse IgG1
Form	Lyophilized





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Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Purification	Immunogen affinity purified.
Suggested Dilutions	Dilute the sample so that the expected range of concentrations fall within the detection range of this kit.  If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples.  Some PubMed article(s) citing the expression level of this target are as follows:  Boster Bio's internal QC testing used:  Western blot, 0.1-0.5ug/ml  Immunohistochemistry (Paraffin-embedded Section), 0.5-1ug/ml  Immunohistochemistry (Frozen Section), 0.5-1ug/ml  Immunocytochemistry, 0.5-1ug/ml  Flow Cytometry, 1-3ug/1x10 <sup>6</sup> cells



## Anti-Stefin B CSTB Antibody Picoband™ (monoclonal, 2B6) (M02794) Images



Figure 1. Western blot analysis of Stefin B using anti-Stefin B antibody (M02794).

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 50ug of sample under reducing conditions.

- Lane 1: human U-87MG whole cell lysates,
- Lane 2: human SW620 whole cell lysates,
- Lane 3: human U-937 whole cell lysates,
- Lane 4: human Caco-2 whole cell lysates.

After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with mouse anti-Stefin B antigen affinity purified monoclonal antibody (Catalog # M02794) 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-mouse IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1001) with Tanon 5200 system.

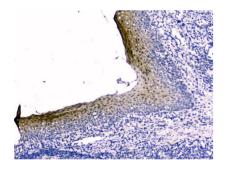


Figure 2. IHC analysis of Stefin B using anti-Stefin B antibody (M02794).

Stefin B was detected in paraffin-embedded section of human tonsil 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 mouse anti-Stefin B Antibody (M02794) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC)(Catalog # SA1021) with DAB as the chromogen.

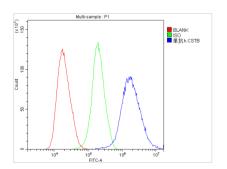


Figure 3. Flow Cytometry analysis of PC-3 cells using anti-Stefin B antibody (M02794).

Overlay histogram showing PC-3 cells stained with M02794 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-Stefin B Antibody (M02794,  $1ug/1x10^6$  cells) for 30 min at 20°C. DyLight® 488 conjugated goat anti-mouse IgG (BA1126, 5- $10ug/1x10^6$  cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG ( $1ug/1x10^6$ ) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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