

# Anti-Calpain 2 CAPN2 Antibody Picoband™ (monoclonal, 816)

Catalog Number: M03492

### **About CAPN2**

Calpain-2 catalytic subunit is a protein that in humans is encoded by the CAPN2 gene. The calpains, calcium-activated neutral proteases, are nonlysosomal, intracellular cysteine proteases. The mammalian calpains include ubiquitous, stomach-specific, and muscle-specific proteins. The ubiquitous enzymes consist of heterodimers with distinct large, catalytic subunits associated with a common small, regulatory subunit. This gene encodes the large subunit of the ubiquitous enzyme, calpain 2. Multiple heterogeneous transcriptional start sites in the 5' UTR have been reported.

#### Overview

Product Name	Anti-Calpain 2 CAPN2 Antibody Picoband™ (monoclonal, 816)
Reactive Species	Human, Monkey, Mouse
Description	Boster Bio Anti-Calpain 2 CAPN2 Antibody Picoband™ (monoclonal, 816) catalog # M03492. Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Monkey, Mouse.
Application	Flow Cytometry, IF, IHC, ICC, WB
Clonality	Monoclonal 816
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	P17655

### **Technical Details**

Immunogen	E. coli-derived human Calpain 2 recombinant protein (Position: R500-L700). Human Calpain 2 shares 93.5% and 92.5% amino acid (aa) sequence identity with mouse and rat Calpain 2, respectively.
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) and ICC.
Cross Reactivity	No cross-reactivity with other proteins.
Isotype	Mouse IgG2a
Form	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Suggested Dilutions	Dilute the sample so that the expected range of concentrations fall within the detection range of this



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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  Immunocytochemistry/Immunofluorescence, 2ug/ml  Flow Cytometry, 1-3ug/1x10 <sup>6</sup> cells
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## Anti-Calpain 2 CAPN2 Antibody Picoband™ (monoclonal, 816) (M03492) Images

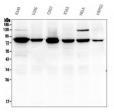


Figure 1. Western blot analysis of Calpain 2 using anti-Calpain 2 antibody (M03492).

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: A549 whole cell lysates,

Lane 2: U20S whole cell lysates.

Lane 3: COS-7 whole cell lysates,

Lane 4: K562 whole cell lysates,

Lane 5: HELA whole cell lysates,

Lane 6: HEPG2 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-Calpain 2 antigen affinity purified monoclonal antibody (Catalog # M03492) 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 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1001) with Tanon 5200 system. A specific band was detected for Calpain 2 at approximately 80KD. The expected band size for Calpain 2 is at 80KD.

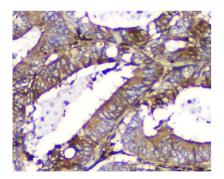


Figure 2. IHC analysis of Calpain 2 using anti-Calpain 2 antibody (M03492).

Calpain 2 was detected in paraffin-embedded section of human intestinal cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml mouse anti-Calpain 2 Antibody (M03492) 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.

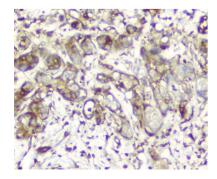


Figure 3. IHC analysis of Calpain 2 using anti-Calpain 2 antibody (M03492).

Calpain 2 was detected in paraffin-embedded section of human mammary cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml mouse anti-Calpain 2 Antibody (M03492) 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





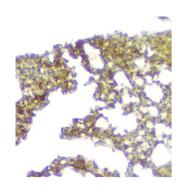


Figure 4. IHC analysis of Calpain 2 using anti-Calpain 2 antibody (M03492).

Calpain 2 was detected in paraffin-embedded section of mouse lung tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml mouse anti-Calpain 2 Antibody (M03492) 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.

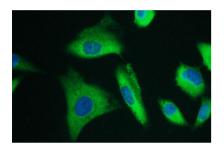


Figure 5. IF analysis of Calpain 2 using anti-Calpain 2 antibody (M03492).

Calpain 2 was detected in immunocytochemical section of A549 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 2ug/mL mouse anti-Calpain 2 Antibody (M03492) overnight at 4°C. DyLight®488 Conjugated Goat Anti-mouse IgG (BA1126) 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.

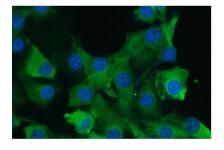


Figure 6. IF analysis of Calpain 2 using anti-Calpain 2 antibody (M03492).

Calpain 2 was detected in immunocytochemical section of U251 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 2ug/mL mouse anti-Calpain 2 Antibody (M03492) overnight at 4°C. DyLight®488 Conjugated Goat Anti-mouse IgG (BA1126) 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.

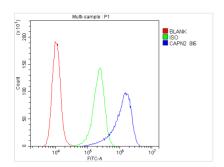


Figure 7. Flow Cytometry analysis of A431 cells using anti-Calpain 2 antibody (M03492).

Overlay histogram showing A431 cells stained with M03492 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-Calpain 2 Antibody (M03492,1ug/1x10 $^6$  cells) for 30 min at 20 $^\circ$ C. DyLight® 488 conjugated goat anti-mouse IgG (BA1126, 5-10ug/1x10 $^6$  cells) was used as secondary antibody for 30 minutes at 20 $^\circ$ 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.



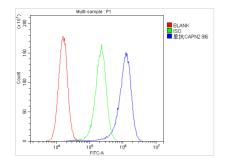


Figure 8. Flow Cytometry analysis of U87 cells using anti-Calpain 2 antibody (M03492).

Overlay histogram showing U87 cells stained with M03492 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-Calpain 2 Antibody (M03492,1ug/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.

# 1 Publications Citing This Product

1. PubMed ID: 25415668, Chen Hx, Tang Sp, Gao Ft, Xu Jl, Jiang Xp, Cao J, Fu Gb, Sun K, Liu Sz, Shi W. Medicine (Baltimore). 2014 Nov;93(23):E138. Doi: 10.1097/Md.00000000000138. Fibrosis, Adipogenesis, And Muscle Atrophy In Congenital Muscular Torticollis.

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