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Anti-Calpain 2 CAPN2 Antibody Picoband™ (monoclonal, 8I6)

Catalog # ABO14821

Specification

Anti-Calpain 2 CAPN2 Antibody Picoband™ (monoclonal, 8I6) - Product Information

Application	**WB, IHC, IF, ICC, FC**
Primary Accession	P17655
Host	**Mouse**
Isotype	**Mouse IgG2a**
Reactivity	**Human, Mouse, Monkey**
Clonality	**Monoclonal**
Format	**Lyophilized**

Description
Anti-Calpain 2 CAPN2 Antibody Picoband™ (monoclonal, 8I6) . Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Monkey, Mouse.

Anti-Calpain 2 CAPN2 Antibody Picoband™ (monoclonal, 8I6) - Additional Information

Gene ID 824

Other Names
Calpain-2 catalytic subunit, 3.4.22.53, Calcium-activated neutral proteinase 2, CANP 2, Calpain M-type, Calpain large polypeptide L2, Calpain-2 large subunit, Millimolar-calpain, M-calpain, CAPN2, CANPL2

Calculated MW
80 kDa KDa

Application Details
Western blot, 0.1-0.5 µg/ml
 Immunohistochemistry (Paraffin-embedded Section), 0.5-1 µg/ml
 Immunocytochemistry/Immunofluorescence, 2 µg/ml
 Flow Cytometry, 1-3 µg/1x10^6 cells

Subcellular Localization
Cytoplasm. Cell membrane. Translocates to the plasma membrane upon Ca (2+) binding.

Tissue Specificity
Ubiquitous.

Contents
Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.05mg NaN3.

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.

Cross Reactivity
No cross-reactivity with other proteins.

Storage	**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.**

Anti-Calpain 2 CAPN2 Antibody Picoband™ (monoclonal, 8I6) - Protein Information

Name CAPN2

Synonyms CANPL2

Function
Calcium-regulated non-lysosomal thiol-protease which catalyzes limited proteolysis of substrates involved in cytoskeletal remodeling and signal transduction. Proteolytically cleaves MYOC at 'Arg-226' (PubMed:17650508). Proteolytically cleaves CPEB3 following neuronal stimulation which abolishes CPEB3 translational repressor activity, leading to translation of CPEB3 target mRNAs (By similarity).

Cellular Location
Cytoplasm. Cell membrane. Note=Translocates to the plasma membrane upon Ca(2+) binding

Tissue Location
Ubiquitous.

Anti-Calpain 2 CAPN2 Antibody Picoband™ (monoclonal, 8I6) - Protocols

Provided below are standard protocols that you may find useful for product applications.

- Western Blot
- Blocking Peptides
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- Cell Culture

Anti-Calpain 2 CAPN2 Antibody Picoband™ (monoclonal, 8I6) - Images

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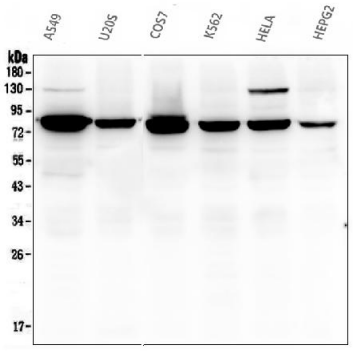

Figure 1. Western blot analysis of Calpain 2 using anti-Calpain 2 antibody (M03492).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.
Lane 1: A549 whole 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 µg/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.

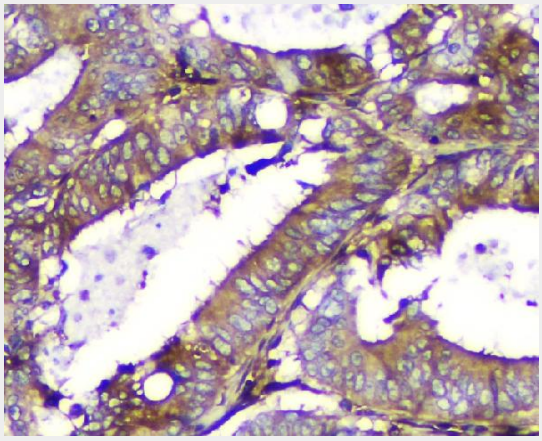

Figure 2. IHC analysis of Calpain 2 using anti-Calpain 2 antibody (M03492).
Calpain 2 was detected in paraffin-embedded section of human intestinal cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10%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

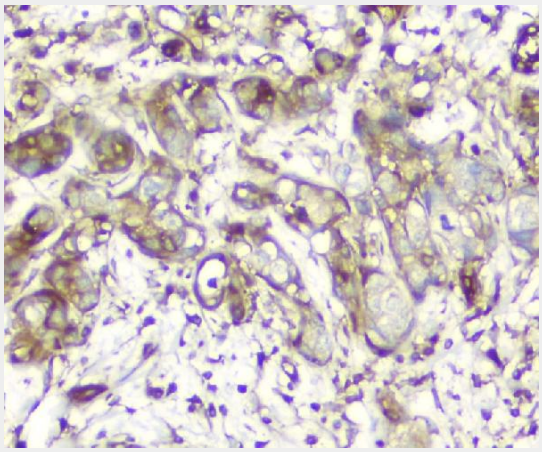

Figure 3. IHC analysis of Calpain 2 using anti-Calpain 2 antibody (M03492).
Calpain 2 was detected in paraffin-embedded section of human mammary cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/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.

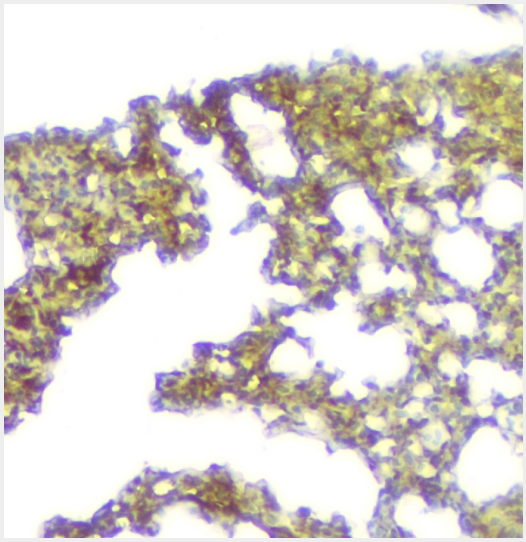

Figure 4. IHC analysis of Calpain 2 using anti-Calpain 2 antibody (M03492).
Calpain 2 was detected in paraffin-embedded section of mouse lung tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/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.

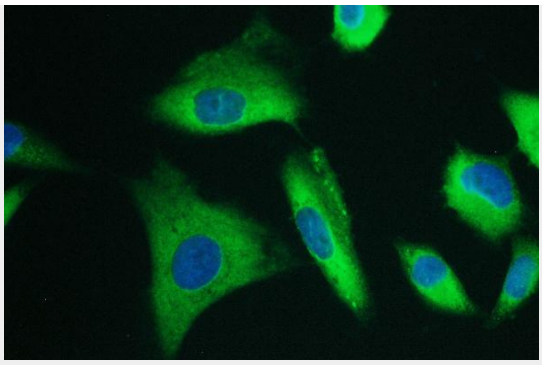

Figure 5. IF analysis of Calpain 2 using anti-Calpain 2 antibody (M03492).
Calpain 2 was detected in immunocytochemical section of A549 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 2 µg/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.

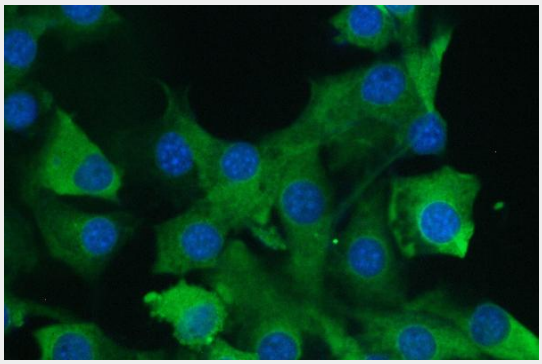

Figure 6. IF analysis of Calpain 2 using anti-Calpain 2 antibody (M03492).
Calpain 2 was detected in immunocytochemical section of U251 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 2 µg/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.

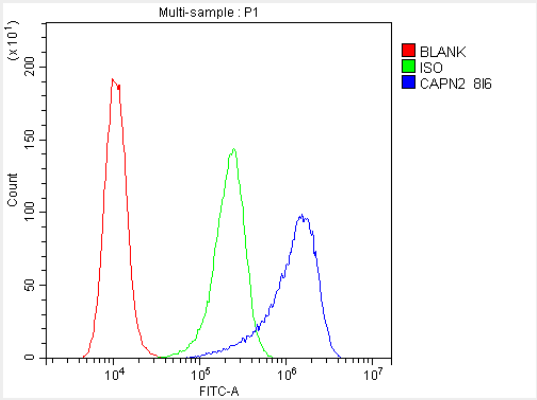

Figure 7. Flow Cytometry analysis of A431 cells using anti-Calpain 2 antibody (M03492).
Overlay histogram showing A431 cells stained with M03492 (Blue line).The cells were blocked with

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10% normal goat serum. And then incubated with mouse anti-Calpain 2 Antibody (M03492,1 µg/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

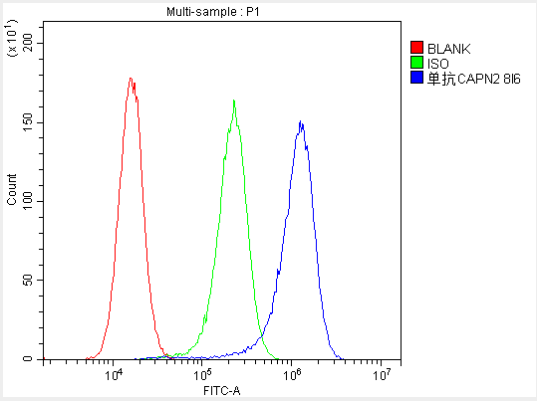

Figure 8. Flow Cytometry analysis of U87 cells using anti-Calpain 2 antibody (M03492). Overlay histogram showing U87 cells stained with M03492 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-Calpain 2 Antibody (M03492,1 µg/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

Anti-Calpain 2 CAPN2 Antibody Picoband™ (monoclonal, 8I6) - Background

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.