

Anti-Calpain 1 Antibody Picoband™ (monoclonal, 2I10)

Catalog # ABO15062

Product Information

Application	WB, IF, ICC, FC
Primary Accession	P07384
Host	Mouse
Isotype	Mouse IgG2a
Reactivity	Human
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-Calpain 1 Antibody Picoband™ (monoclonal, 2I10) . Tested in Flow Cytometry, IF, ICC, WB applications. This antibody reacts with Human.
Reconstitution	Add 0.2ml of distilled water will yield a concentration of 500ug/ml.

Additional Information

Gene ID	823
Other Names	Calpain-1 catalytic subunit, 3.4.22.52, Calcium-activated neutral proteinase 1, CANP 1, Calpain mu-type, Calpain-1 large subunit, Cell proliferation-inducing gene 30 protein {ECO:0000303 Ref.3}, Micromolar-calpain, muCANP, CAPN1 (HGNC:1476), CANPL1
Calculated MW	81890
Application Details	Western blot, 0.25-0.5 µg/ml, Human Immunocytochemistry/Immunofluorescence, 5 µg/ml, Human Flow Cytometry, 1-3 µg/1x10 ⁶ cells, Human
Source	Eukaryota
Contents	Each vial contains 4mg Trehalose, 0.9mg NaCl and 0.2mg Na ₂ HPO ₄ .
Clone Names	Clone: 2I10
Immunogen	E.coli-derived human Calpain 1 recombinant protein (Position: Q396-A555). Human Calpain 1 shares 86% amino acid (aa) sequence identity with both mouse and rat Calpain 1.
Purification	Immunogen affinity purified.
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.

Protein Information

Name	CAPN1 (HGNC:1476)
Synonyms	CANPL1
Function	Calcium-regulated non-lysosomal thiol-protease which catalyzes limited proteolysis of substrates involved in cytoskeletal remodeling and signal transduction (PubMed: 19617626 , PubMed: 21531719 , PubMed: 2400579). Proteolytically cleaves CTBP1 at 'Asn-375', 'Gly-387' and 'His-409' (PubMed: 23707407). Cleaves and activates caspase-7 (CASP7) (PubMed: 19617626).
Cellular Location	Cytoplasm. Cell membrane. Note=Translocates to the plasma membrane upon Ca(2+) binding. In granular keratinocytes and in lower corneocytes, colocalizes with FLG and FLG2 (PubMed:21531719)
Tissue Location	Ubiquitous.

Background

CAPN1 is also known as CANP or muCL. 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 1. Several transcript variants encoding two different isoforms have been found for this gene.

Images

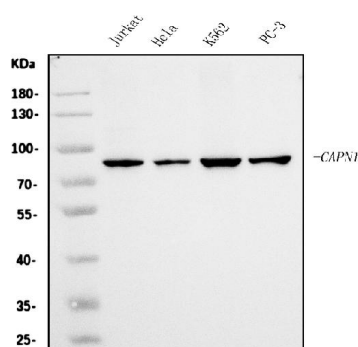


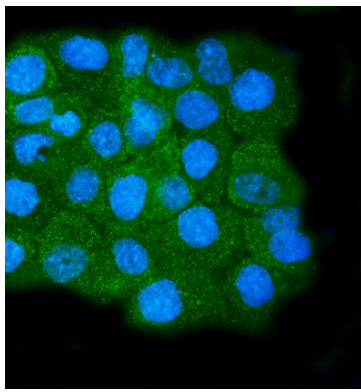
Figure 1. Western blot analysis of Calpain 1 using anti-Calpain 1 antibody (M01943-3).

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 30ug of sample under reducing conditions.

Lane 1: human Jurkat whole cell lysates,
Lane 2: human Hela whole cell lysates,
Lane 3: human K562 whole cell lysates,
Lane 4: human PC-3 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 1 antigen affinity purified monoclonal antibody (Catalog # M01943-3) 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 1:10000 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 1 at approximately 82KD. The expected band size for Calpain 1 is at 82KD.

Figure 2. IF analysis of Calpain 1 using anti-Calpain 1 antibody (M01943-3).



Calpain 1 was detected in immunocytochemical section of A431 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 5 µg/mL mouse anti-Calpain 1 Antibody (M01943-3) 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.

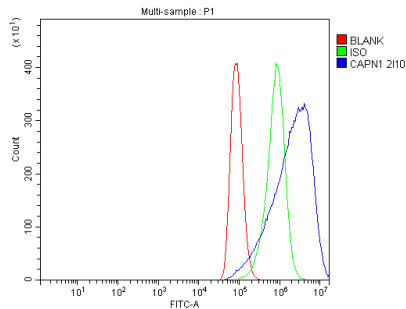


Figure 3. Flow Cytometry analysis of A549 cells using anti-Calpain 1 antibody (M01943-3). Overlay histogram showing A549 cells stained with M01943-3 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-Calpain 1 Antibody (M01943-3, 1 µg/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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