# Anti-Caspase-1 Antibody [SU40-07]

## ET1608-69



Product Type: Species reactivity: Applications: Molecular Wt: Clone number:	Recombinant Rabbit monoclonal IgG, primary antibodies Human, Mouse, Rat WB, FC, IF-Cell Predicted band size: 45 kDa SU40-07
Description:	Caspase-1, originally designated ICE (for IL-1 converting enzyme), is a member of the group of caspases with large prodomains. Caspase-1 promotes maturation of interleukin IL- 1 $\beta$ and interleukin18 (IL-18) by proteolytic cleavage of precursor forms into biologically active pro-inflamatory cytokines. Active caspase-1, a (p20/p10)2 tetramer, is necessary and sufficient for cleavage of precursor IL-1 as well as for induction of apoptosis in some cell lines. The highly conserved family of caspases mediate many of the morphological and biochemical features of apoptosis, including structural dismantling of cell bodies and nuclei, fragmentation of genomic DNA, destruction of regulatory proteins and propagation of other pro-apoptotic molecules. The human Caspase-1 gene maps to chromosome 2q14 and encodes a cytoplasmic protein expressed in liver, heart, skeletal muscle kidney and testis. Caspase-1 has been implicated in inflammation, septic shock, and other situations such as wound healing and the growth of certain leukemias.
Immunogen:	Recombinant protein within Human Caspase-1 aa 213-404 / 404.
Positive control:	THP-1 cell lysate, HCT 116 cell lysate, HL-60 cell lysate, RAW264.7 cell lysate, Mouse spleen lysate, Rat spleen lysate, U937 cell lysates, PC-12, HL-60, THP-1.
Subcellular location:	Cytoplasm, Cell membrane.
Database links:	SwissProt: P29466 Human   P29452 Mouse   P43527 Rat
Recommended Dilutions: WB FC IF-Cell	1:1,000-1:100,000 1:1,000 1:100
Storage Buffer:	1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Store at +4 $^\circ\!{\rm C}$ after thawing. Aliquot store at -20 $^\circ\!{\rm C}$ or -80 $^\circ\!{\rm C}$ . Avoid repeated freeze / thaw cycles.
Purity:	Protein A affinity purified.

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Orders:0086-571-88062880

Technical:0086-571-89986345

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#### Images



**Fig1:** Western blot analysis of Caspase-1 on different lysates with Rabbit anti-Caspase-1 antibody (ET1608-69) at 1/100,000 dilution.

Lane 1: THP-1 cell lysate Lane 2: HCT 116 cell lysate Lane 3: HL-60 cell lysate Lane 4: RAW264.7 cell lysate Lane 5: Mouse spleen lysate Lane 6: Rat spleen lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 45 kDa Observed band size: 45 kDa

Exposure time: 3 minutes;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1608-69) at 1/100,000 dilution was used in 5% NFDM/TBST at  $4^{\circ}$ C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

**Fig2:** Western blot analysis of Caspase-1 on U937 cell lysates with Rabbit anti-Caspase-1 antibody (ET1608-69) at 1/1,000 dilution.

Lysates/proteins at 10 µg/Lane.

Predicted band size: 45 kDa Observed band size: 45 kDa

Exposure time: 1 minute;

10% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1608-69) at 1/1,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:300,000 dilution was used for 1 hour at room temperature.







**Fig3:** Immunocytochemistry analysis of PC-12 cells labeling Caspase-1 with Rabbit anti-Caspase-1 antibody (ET1608-69) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-Caspase-1 antibody (ET1608-69) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at  $+4^{\circ}$ C. Goat Anti-Mouse IgG H&L (iFluor 1594, HA1126) was used as the secondary antibody at 1/1,000 dilution.



Fig4: Flow cytometric analysis of PC-12 cells labeling Caspase-1.

Cells were fixed and permeabilized. Then stained with the primary antibody (ET1608-69, 1/1,000) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4 °C for an hour, the cells were stained with a iFluor TM 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4 °C. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

**Fig5:** Immunocytochemistry analysis of HL-60 cells labeling Caspase-1 with Rabbit anti-Caspase-1 antibody (ET1608-69) at 1/100 dilution.

Cells were fixed in 80% precooled methanol for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-Caspase-1 antibody (ET1608-69) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at  $+4^{\circ}$ C. Goat Anti-Mouse IgG H&L (iFluor  $^{TM}$  594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

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**Fig6:** Immunocytochemistry analysis of THP-1 cells labeling Caspase-1 with Rabbit anti-Caspase-1 antibody (ET1608-69) at 1/100 dilution.

Cells were fixed in 80% precooled methanol for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-Caspase-1 antibody (ET1608-69) at 1/100 dilution in 1% BSA in PBST overnight at 4  $^{\circ}$ C. Goat Anti-Rabbit IgG H&L (iFluor<sup>TM</sup> 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at  $+4^{\circ}$ C. Goat Anti-Mouse IgG H&L (iFluor <sup>TM</sup> 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

#### **Background References**

- 1. Zhao J et al. Synthetic Oligodeoxynucleotides Containing Multiple Telemeric TTAGGG Motifs Suppress Inflammasome Activity in Macrophages Subjected to Oxygen and Glucose Deprivation and Reduce Ischemic Brain Injury in Stroke-Prone Spontaneously Hypertensive Rats. PLoS One 10:e0140772 (2015).
- 2. Zhang X et al. Porcine Mx1 fused to HIV Tat protein transduction domain (PTD) inhibits classical swine fever virus infection in vitro and in vivo. BMC Vet Res 11:264 (2015).

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