Anti-Cleaved+pro Caspase-3 Antibody [SU38-04] ET1608-64

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IHC-P, IP
Molecular Wt:	Predicted band size: 32/17 kDa
Clone number:	SU38-04

Description: Caspase-3 is a caspase protein that interacts with caspase-8 and caspase-9. It is encoded by the CASP3 gene. CASP3 orthologs have been identified in numerous mammals for which complete genome data are available. Unique orthologs are also present in birds, lizards, lissamphibians, and teleosts. Caspase-3 shares many of the typical characteristics common to all currently-known caspases. For example, its active site contains a cysteine residue (Cys-163) and histidine residue (His-121) that stabilize the peptide bond cleavage of a protein sequence to the carboxy-terminal side of an aspartic acid when it is part of a particular 4-amino acid sequence. This specificity allows caspases to be incredibly selective, with a 20,000-fold preference for aspartic acid over glutamic acid. A key feature of caspases in the cell is that they are present as zymogens, termed procaspases, which are inactive until a biochemical change causes their activation. Each procaspase has an N-terminal large subunit of about 20 kDa followed by a smaller subunit of about 10 kDa, called p20 and p10, respectively.

Immunogen: Recombinant protein within Human Caspase-3 aa 9-175 / 277.

Positive control: HeLa cell lysate, HeLa treated with 1μM staurosporine for 4 hours cell lysate, HeLa treated with 3μM staurosporine for 4 hours cell lysate, C6 cell lysate, C6 treated with 1μM staurosporine for 4 hours cell lysate, C6 treated with 3μM staurosporine for 4 hours cell lysate, U-87 MG cell lysate, MDA-MB-231 cell lysate, NIH/3T3 cell lysate, Jurkat cell lysate, Jurkat treated with 1μM staurosporine for 3 hours cell lysate, human lung carcinoma tissue, human spleen tissue, human liver tissue, human kidney tissue.

Subcellular location: Cytoplasm.

Database links: SwissProt: P42574 Human | P70677 Mouse | P55213 Rat

Recommended Dilutions:

WB	1:1,000-1:2,000
IHC-P	1:50-1:1,000
IP	Use at an assay dependent concentration.

Storage Buffer: 1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20℃ long term.

Purity: Protein A affinity purified.

Images

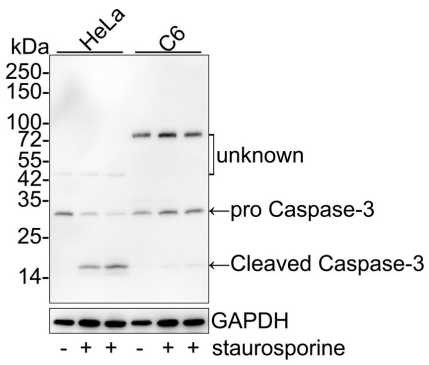

Fig1: Western blot analysis of Cleaved+pro Caspase-3 on different lysates with Rabbit anti-Cleaved+pro Caspase-3 antibody (ET1608-64) at 1/1,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: HeLa treated with 1μM staurosporine for 4 hours cell lysate
Lane 3: HeLa treated with 3μM staurosporine for 4 hours cell lysate
Lane 4: C6 cell lysate
Lane 5: C6 treated with 1μM staurosporine for 4 hours cell lysate
Lane 6: C6 treated with 3μM staurosporine for 4 hours cell lysate

Lysates/proteins at 20 μg/Lane.

Predicted band size: 32/17 kDa
Observed band size: 32/17 kDa

Exposure time: 3 seconds; ECL: K1801;

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 (ET1608-64) at 1/1,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

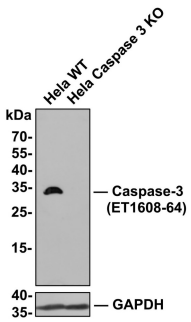

Fig2: Western blot analysis of Caspase-3 with anti-Caspase-3 antibody (ET1608-64) at 1:2,000 dilution.
Lane 1: Wild-type Hela whole cell lysate (10 μg).
Lane 2: Caspase-3 knockout Hela whole cell lysate (10 μg).

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM in TBST for 1 hour at room temperature. The primary antibody (ET1608-64, 1/2,000) was used in 5% BSA at room temperature for 2 hours. Goat Anti-Rabbit IgG-HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

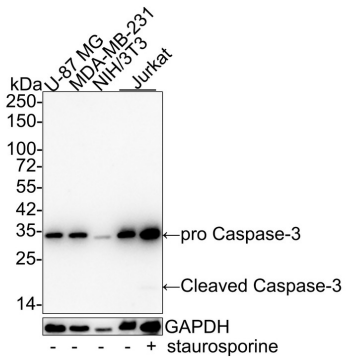

Fig3: Western blot analysis of Cleaved+pro Caspase-3 on different lysates with Rabbit anti-Cleaved+pro Caspase-3 antibody (ET1608-64) at 1/5,000 dilution.

Lane 1: U-87 MG cell lysate
Lane 2: MDA-MB-231 cell lysate
Lane 3: NIH/3T3 cell lysate
Lane 4: Jurkat cell lysate
Lane 5: Jurkat treated with 1μM staurosporine for 3 hours cell lysate

Lysates/proteins at 15 μg/Lane.

Predicted band size: 32/17 kDa
Observed band size: 32/17 kDa

Exposure time: 1 minute 2 seconds;

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 (ET1608-64) at 1/5,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:50,000 dilution was used for 1 hour at room temperature.

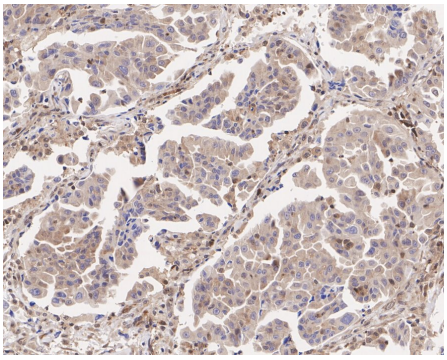

Fig4: Immunohistochemical analysis of paraffin-embedded human lung cancer tissue with Rabbit anti-Cleaved+pro Caspase-3 antibody (ET1608-64) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1608-64) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

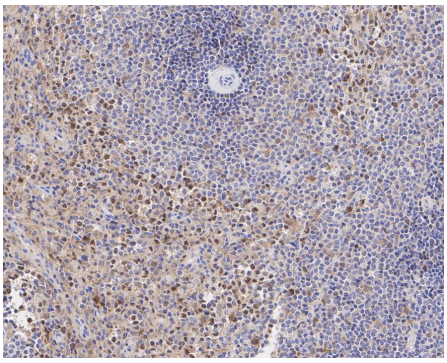

Fig5: Immunohistochemical analysis of paraffin-embedded human spleen tissue with Rabbit anti-Cleaved+pro Caspase-3 antibody (ET1608-64) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1608-64) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

Background References

1. Dong L et al. Echinacoside Induces Apoptosis in Human SW480 Colorectal Cancer Cells by Induction of Oxidative DNA Damages. Int J Mol Sci 16:14655-68 (2015).
2. Nilsonne G et al. Phenotype-dependent apoptosis signalling in mesothelioma cells after selenite exposure. J Exp Clin Cancer Res 28:92 (2009).