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Product Information

Anti-Caspase 1 (ICE)
antibody produced in rabbit, IgG fraction of antiserum

Catalog Number **C4851**

Product Description

Anti-Caspase 1 is produced in rabbit using a synthetic peptide (PEHKTSDSTFLVFMSHGIREG) conjugated to KLH as immunogen. This sequence corresponds to amino acid residues 129-152 of human Caspase 1. The IgG fraction of antiserum is purified using protein A.

Anti-Caspase 1 recognizes the p20 (20 kDa) subunit of reduced human caspase 1 isoforms and the p45 (45 kDa) proenzyme. The antibody reacts with human, mouse and rat caspase 1.

Anti-Caspase 1 may be used for the detection and localization of caspase 1 in immunoblotting and immunocytochemistry.

Caspase 1/Interleukin-1β converting enzyme (ICE) is a cytoplasmic cysteine protease that cleaves the inactive form of interleukin 1-β (IL-1β) to generate the active form of this proinflammatory cytokine.[1,2] Caspase 1/ICE is expressed in many tissues as a proenzyme of 45 kDa, which is activated by proteolytic cleavage.[3] Active ICE is composed of two subunits, known as p10 (10 kDa) and p20 (20 kDa), respectively. Analysis of Caspase 1/ICE protein crystals indicates that these subunits associate as a tetramer with the stoichometry $(p20)_2/(p10)_2$.[4,5] Caspase 1/ICE has an unusual substrate specificity as it preferentially cleaves its substrates after Asp residues,[6] that is shared by a family of cystein proteases called caspases (caspase = cysteine aspartic-specific proteases). This family of cysteine proteases are important in regulating programmed cell death and apoptosis.[7,8] Caspase 1/ICE is functionally homologous to the protein encoded by the *C. elgans* death gene ced-3,[9] and overexpression of Caspase 1/ICE in fibroblasts induced apoptosis.[7,9] In addition, overexpression of Crm A, a poxviral protein that specifically inhibits Caspase 1 /ICE,[10,12] prevented apoptosis in fibroblasts and protected ganglion cells from apoptosis following NGF withdrawl.[9,11]

Reagent

Supplied as a solution in 0.1 M Tris-glycine, pH 7.4, 0.15 M NaCl, and 0.05% sodium azide.

Antibody concentration: ~1 mg/ml.

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Storage/Stability

Store at –20 °C. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use.

Procedure

Immunocytochemistry

1. Plate approximately 200 µL of a cell suspension into each well of a slide. Incubate 24 hours in a 37 °C. CO_2 incubator.
2. Wash the cells 3 X for 5 minutes with PBS. Do not shake cells.
3. Add fixative (95% ethanol, 5% acetic acid) for 1 minute at room temperature.
4. Wash the cells with PBS, 2 X for 15 minutes with gentle agitation. Do not shake cells.
5. Add 400 µL PBS containing 8% BSA and incubate 30 minutes at room temperature.
6. Wash cells with PBS for 15 min.
7. Incubate the cells with 10 µg/mL of Anti-Caspase 1 in PBS containing 1% BSA and incubate overnight at 4 °C in a humidified chamber.
8. Wash the cells 2 X with PBS for 15 minutes.
9. Incubate the cells with a 1:150 dilution of Anti-Rabbit IgG (whole molecule)–FITC, Catalog No. F9887, in PBS for 1 hour at room temperature in the dark.
10. Wash the cells 3 X with PBS for 5 minutes in the dark.
11. Mount coverslips with gel mount and allow gel mount to dry in the dark.
12. Examine the cells under a fluorescent microscope.

Product Profile

Immunoblotting: recommended working concentration is 0.5-2 µg/mL using a human HL-60 cell lysate, Anti-Rabbit IgG-Peroxidase and a chemiluminescent detection system. The immunoreactivity can be inhibited by the immunizing peptide.

Immunocytochemistry: minimum working concentration is 10 µg/mL using human HL-60 cells.

Note: In order to obtain best results and assay sensitivity in different techniques and preparations we recommend determining optimal working dilutions by titration test.

References

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