

## Product Information

### Monoclonal Anti-Caspase 7

#### Clone 11E4

Purified Rat Immunoglobulin

Product Number **C 1104**

#### Product Description

Monoclonal Anti-Caspase 7 (rat IgG2a isotype) is derived from the 11E4 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from a rat immunized with a recombinant p20 subunit of human caspase 7. The antibody is purified from culture supernatant of hybridoma cells grown in a bioreactor.

Monoclonal Anti-Caspase 7 reacts specifically with human caspase 7.<sup>1</sup> The antibody may be used for immunoprecipitation and immunoblotting<sup>1</sup> to detect the full length caspase 7 (35 kDa), the p20 subunit of caspase 7,<sup>1</sup> and possibly an additional band of approximately 60 kDa.

Apoptosis, an evolutionary conserved form of cell suicide, requires specialized machinery. The central component of this machinery is a proteolytic system involving a family of proteases called caspases. These enzymes participate in a cascade that is triggered in response to proapoptotic signals and culminates in cleavage of a set of proteins, resulting in disassembly of the cell.

Caspases (**C**ysteine-requiring **A**spartate protease) are a family of proteases that share similarities in amino acid sequences, structure, and substrate specificity.<sup>2-5</sup> Caspases can be grouped into three subfamilies based on their amino acid sequence homology. The caspase 1 (ICE-type caspases) subfamily contains caspases 1, 4, 5, 11, and 13. This subfamily along with caspase 12, has a role in inflammation as well as in apoptosis; these proteases may also be indirectly involved in apoptosis as activators of other caspases (upstream activity). Caspase-8 and -10 are involved in death receptor mediated apoptosis. The caspase 2 subfamily contains caspases 2 and 9, while the caspase 3 subfamily contains caspases 3, 6 and 7, and are effectors of apoptosis (downstream activity). Effector caspases may cleave and activate cytosolic substrates which then promote cytochrome c release from mitochondria. Cytochrome c release acts to amplify a caspase during apoptosis.<sup>6</sup> Caspases are normally present in the cell as inactive procaspases. The proenzymes (30-50 kDa)

contain three domains: an NH<sub>2</sub>-terminal prodomain, a large subunit (17-22 kDa), and a small subunit (10-12 kDa). Proteolytic cleavage at Asp residues removes the regulatory N-terminal prodomain and cleaves the proenzyme into the large and small subunits. The subunits self-associate into heterodimers that in turn form the active caspase, a tetramer consisting of two large and two small subunits. The active caspases continue the cascade by autocleaving, cleaving other procaspases, or cleaving other key proteins such as (but not limited to) poly(ADP-ribose) polymerase (PARP), DNA-dependent protein kinase (DNA-PK), lamins, nuclear mitotic apparatus protein (NuMA), and sterol regulatory element binding proteins (SREBPs).

Caspase 7 is constitutively expressed in many fetal and adult tissues, with the lowest expression observed in brain.<sup>3</sup> It is localized diffusely to the cytoplasm and juxtamembrane structures (almost exclusively in the mitochondrial and microsomal fractions).<sup>8,10</sup> The gene for caspase 7 (also known as Mch3, CMH-1 and ICE-LAP3) encodes a "short" prodomain protein product of 35 kDa that belongs to the Ced3/ICE family of cysteine proteases. An alternatively spliced isoform of caspase 7, which may act as a negative regulator of apoptosis, has also been described.<sup>3,7</sup> Caspase 7 undergoes a cleavage upon activation by caspase-3, -9 and -10, and granzyme B (e.g., during Fas- and TNFR1-induced apoptosis), leading to p20 and p12 subunits.<sup>8,9</sup> Thus, caspase 7 is an important intracellular effector of granzyme B-mediated apoptosis and cytotoxic T-lymphocyte-induced cell killing *in vivo*.<sup>3</sup> Monoclonal antibodies reacting specifically with caspase 7 are useful tools for the study of protease networks involved in development and regulation and governing the life and death of cells and tissues.

#### Reagent

Monoclonal Anti-Caspase 7 is supplied as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 1 % bovine serum albumin and 15 mM sodium azide.

Antibody Concentration: Approx. 1 mg/ml.

### Precautions and Disclaimer

Due to the sodium azide content a material safety sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution.

Consult the MSDS for information regarding hazardous and safe handling practices.

### Storage/Stability

For continuous use, store at 2-8 °C for up to one month.

For extended storage, freeze in working aliquots.

Repeated freezing and thawing is not recommended.

Storage in "frost-free" freezers is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilution samples should be discarded if not used within 12 hours.

### Product Profile

A working concentration of 2-4 µg/ml is determined by immunoblotting using a whole extract of cultured human acute T cell leukemia Jurkat cells.

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

### References

1. Li, H., et al., *Cell*, **94**, 491-501 (1998).
2. Thornberry, N.A., and Lazebnik, Y., *Science*, **281**, 1312-1316 (1998).
3. Cohen, G.M., *Biochem J.*, **326**, 1-16 (1997).
4. Cryns, V., and Yuan, J., *Genes Develop.*, **12**, 1551-1570 (1998).
5. Kidd, V.J., *Annu. Rev. Physiol.*, **60**, 533-573 (1998).
6. Bossy-Wetzell, E., and Green, D.R., *J. Biol. Chem.*, **274**, 17484-17490 (1999).
7. Fernandes-Alnemri, T., et al., *Cancer Res.*, **55**, 6045-6052 (1995).
8. Duan, H., et al., *J. Biol. Chem.*, **271**, 1621-1625 (1996).
9. Chinnaiyan, A.M., et al., *Curr. Biol.*, **6**, 897-899 (1996).
10. Chandler, J.M., et al., *J. Biol. Chem.*, **273**, 10815-10818 (1998).

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