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ProductInformation

CASPASE 2, HUMAN RECOMBINANT
expressed in *E. coli*

Product Code **C 2854**
Storage Temperature –20 °C

Synonyms: ICH-1, Nedd2

Product Description
To create this product the sequences corresponding to amino acids 153 - 316 and 331 - 435 of human caspase-2 were expressed in *E. coli*. Purified caspase-2 subunits were mixed and refolded. Characterization by size exclusion chromatography indicates that the proenzyme is approximately 56 kDa. On SDS polyacrylamide gels, the recombinant active form migrates as two major polypeptides of 17 kDa and 10 kDa as detected by immunoblotting.

Caspase-2 (ICH-1, Nedd2) is a member of the caspase family of cysteine proteases that play an essential role in the execution phase of apoptosis. These enzymes share a dominant primary specificity for cleaving bonds following aspartic acid residues. "Initiator" caspases (such as caspase-8) activate "effector" caspases (e.g. caspase-3 and –7). The effector caspases then cleave cellular substrates ultimately leading to the morphological changes of apoptosis.[1-3]

Caspases are synthesized as inactive proenzymes. The precursor proteins contain N-terminal pro-sequences of various lengths followed by the p20 and p10 subunits. Caspases are activated by cleavage at specific Asp residues to produce two subunits of approximately 20 kDa (p20) and 10 kDa (p10), which together form the heterodimeric active protease.[2-5] In some cases, these subunits are separated by a linker that may be involved in regulation of the activation of the caspase. All caspases contain an active-site pentapeptide of general structure QACXG (where X is R, Q or G). The amino acids Cys-285 and His-237 involved in catalysis, and those involved in forming the P1 carboxylate binding pocket (Arg-179, Gln-283, Arg-341 and Ser-347) are conserved in all caspases, except for the substitution of Thr for Ser-347 in caspase-8. This explains the absolute requirement for an Asp in the P1 position. Residues forming the P2–P4 binding pocket are not well conserved. This suggests they may determine the substrate specificities of the caspases. Evidence suggests that not all caspases are required for cell death, and some caspases appear to be more important than others.[2]

Procaspase-2 is believed to localize in the mitochondria and to be released into the cytosol upon activation triggered by a variety of physiological or pathological stimuli.[6] Caspase-2 is an initiator upstream (apical) caspase that is recruited through its prodomain CARD by the death adaptor protein RAIDD/CRADD and functions as an early trigger for apoptosis.[7] Procaspase-2 is cleaved *in vitro* by the active caspases 1 and 3 and granzyme B, and to a lesser extent by the mature caspases -2 and -6.[4] Activated caspase-2 is a heterodimer of the small and large subunits, both required for full proteolytic activity.

Caspase-2 exists as two splice variants, ICH-1L (long) and ICH-1S (short). Functionally, the two forms differ in that overexpression of the long form induces apoptosis, while over-expression of the short form suppresses it.[8] During embryogenesis, caspase-2 is expressed at relatively high levels in various tissues including liver, kidneys, lungs and central nervous. It is also expressed in several adult cell types such as post-mitotic neurons system.[9]

Reagent
The product is supplied as a solution in 25 mM HEPES, pH 7.5, 0.15 M NaCl, 5 mM dithiothreitol, 10% sucrose with bovine serum albumin (0.5 mg per 1 mg of enzyme).

Storage/Stability
Store at –20 °C. Enzyme is stable for 4 hours at 4 °C but loses activity at room temperature. Repeated freezing and thawing is not recommended. Storage in "frost-free freezers is not recommended.

Product Profile
Purity: ≥95% (SDS-PAGE)
Activity: >1000 units/mg protein
Unit Definition: One unit will hydrolyze one nmol of Ac-Val-Asp-Val-Ala-Asp-AFC per minute at pH 7.5 at 25 °C.

References
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4. Harvey, N., et al., Genes Cells, **1,** 673-685 (1996).
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7. Duan, H. and Dixit, V.M., Nature, **385,** 86-89 (1997).
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Related Products

Substrates: N-Acetyl-Val-Asp-Val-Ala-Asp 7-amido-4-trifluoromethylcoumarin (Ac-VDVAD-AFC), Product Code A 5345;
N-Acetyl-Val-Asp-Val-Ala-Asp p-nitroanilide (Ac-VDVAD-pNA), Product Code A 5470
Inhibitor: N-CBZ-Val-Asp(OMe)-Val-Ala-Asp(OMe) fluoromethyl ketone (Z-VDVAD-FMK), Product Code C 1605

AC 10/13/00