

Instruction Manual

Catalog Number	PK-RP577-1088-25
Description	Caspase-8 (also known as FLICE, MASH, Mch5) is a member of the caspase-family of cysteine proteases. Similar to other caspases, caspase-8 also exists in cells as an inactive proenzyme. During apoptosis procaspase-8 is processed at aspartate residues by self-proteolysis and/or cleavage by another caspase. The processed active form of caspase-8 consists of large and small subunits which associate to form the active enzyme. Active caspase-8 has been shown to activate caspase-3 leading to degradation of a variety of cellular target proteins during apoptosis. The recombinant active human caspase-8 was expressed in E. coli. The active caspase-8 is routinely tested for its ability to enzymatically cleave these two substrates Ac-IETD-pNA or Ac-IETD-AFC.
Quantity	25 units
Specific Activity	5000 units/mg
Unit Definition	One unit of the recombinant caspase-8 is the enzyme activity that cleaves 1 nmol of the caspase substrate IETD-pNA (pNA: p-nitroaniline) per hour at 37°C in a reaction solution containing 50 mM HEPES, pH 7.2, 50 mM NaCl, 0.1% CHAPS, 10 mM EDTA, 5% Glycerol, and 10 mM DTT.
Purity	>95% by SDS-PAGE
Formulation	Lyophilized powder
Reconstitution	The active recombinant caspases can be reconstituted to 0.1-1 unit per µl in PBS or - for longer stability - in PBS containing 15% glycerol or the Reaction Buffer described above (also available separately from PromoKine, Cat. No. PK-CA577-1068-20 and PK-CA577-1068-80). We recommend using 1 unit per assay for analyzing caspase activity.
Storage	The lyophilized caspase-8 is stable for 1 year at -70°C. Following reconstitution in PBS + 15% glycerol, the enzyme should be aliquoted and immediately stored at -70°C. Avoid multiple freeze/thaw cycles as activity might decrease.
Applications	Active caspase-8 is useful in studying enzyme regulation, determining target substrates, screening caspase inhibitors, or as a positive control in caspase activity assays. We recommend using 1 unit/assay for analyzing caspase activity. For a complete caspase-8 assay protocol, please refer to PromoKine's Caspase-8 Fluorometric or Colorimetric Assay Kits.

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