

## Product Information

### **ANTI-CASPASE 8 (GD-13)** Developed in Rabbit, IgG Fraction of Antiserum

Product Number **C 2976**

#### **Product Description**

Anti-Caspase 8 (GD-13) is developed in rabbit using a synthetic peptide (K-GDNYQKGIPVETD) corresponding to human caspase 8a/MACH $\alpha$ 1 (amino acids 362-374 with a N-terminal added lysine) conjugated to KLH as immunogen. This sequence corresponds to the C-terminal region of the enzyme p20 subunit and is identical in human caspase 8 isoforms (caspase 8b/MACH $\alpha$ 2, MACH $\alpha$ 3 and MACH $\beta$ ) and has limited homology (50%) in mouse caspase 8. Whole antiserum is fractionated and then further purified by ion-exchange chromatography to provide the IgG fraction of antiserum that is essentially free of other rabbit serum proteins.

Anti-Caspase 8 (GD-13) recognizes human caspase 8a (55 kDa) and caspase 8b (54kDa) by immunoblotting. Staining of caspase 8 in immunoblotting is specifically inhibited with caspase 8 immunizing peptide (human, amino acids 362-374).

Apoptosis or programmed cell death (PCD), plays an essential role in development, homeostasis and defense of multicellular organisms. Several cell surface receptors including Fas (APO-1/CD95), TNFR-1, DR3, DR4 belonging to the TNF/NGF receptor superfamily can trigger apoptosis upon binding their respective ligands. Among the many known effectors of apoptosis the ICE-related, cysteine aspartic-specific proteases or caspases play a crucial role in apoptosis in almost every cell type.<sup>1,2</sup> At least 14 different caspases have been identified which differ in their substrate specificities. Caspase 8 (also termed FLICE, MCH5, MACH),<sup>3-5</sup> is at the apex of the apoptotic pathway linking death signals to caspase activation. At least eight different caspase 8 isoforms (designated as caspase 8a-h also termed FLICE/MACH $\alpha$ 1, MACH $\alpha$ 2, MACH $\alpha$ 3, MACH $\beta$ 1-4 and MCH5) have been described at the mRNA level.<sup>6</sup> The predominant caspase 8 isoforms expressed in cells, caspase 8a/b (479 and 464 a.a.), are formed as inactive 55kDa and 54kDa

precursors, respectively. The N-terminal region of caspase 8 contains two death effector domains (DEDs) which facilitate the interaction with the adaptor protein FADD/MORT-1. The recruitment and activation of caspase 8 is inhibited by v-FLIPs and I-FLICE proteins.<sup>7</sup> Upon activation of Fas or TNFR-1, caspase 8 is recruited to FADD on the receptor complex to initiate the caspase cascade and induce apoptosis.<sup>3,4</sup> Caspase 8 is activated by oligomerization-induced processing resulting in removal of its death domain and cleavage into active 20kDa and 10kDa subunits which are released in the cytosol.<sup>8</sup> These subunits form a proteolytically active heterodimer that are capable to cleave other downstream caspase family members.<sup>9,10</sup>

#### **Reagents**

The product is supplied as IgG fraction in 0.01M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide as a preservative.

#### **Precautions and Disclaimer**

Due to the sodium azide content a material safety data 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 minimum working dilution of 1:1,000 is determined by immunoblotting using a whole cell extract of the human Burkitt lymphoma Raji cell line.

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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