

**Caspase-9/p35 (CASP-9/MHC-6) Antibodies**

Cat. # CASP91-P	Human Casp-9/p35 Control Peptide # 1	<b>SIZE:</b> 100 ug
Cat. # CASP91-S	Rabbit Anti-Human Casp-9/p35 antiserum # 1	<b>SIZE:</b> 100 ul
Cat. # CASP91-A	Rabbit Anti-Human Casp-9/p35 IgG # 1 (aff pure)	<b>SIZE:</b> 100 ug

Apoptosis or programmed cell death is a fundamental cellular process that is essential for normal tissue development and abnormal growth such as cancer, neurodegeneration, autoimmune diseases, and angiogenesis, etc. Apoptosis is driven by specialized proteases known as **caspases**. After the initial discovery of the first mammalian caspase 1 or ICE (interleukin 1 beta converting enzyme), a growing family of **caspases 1-14** have been cloned and characterized. Caspases are synthesized as inactive zymogen or proenzyme forms (30-55 kDa), which upon apoptotic stimulation are proteolytically processed (self or by other proteins) in a sequential manner into their active heterotetrameric forms. The processed form consists of large subunit (17-20 kDa) and a small (10-12 kDa) subunits, which may associate to form an active enzyme. Functionally active caspases initiate a proteolytic cascade, capable of cleaving and activating numerous cellular targets including PARP, G4-GDI, DFF, MEKK, etc. On a functional basis, two categories of caspases have been defined: the **initiator caspases** (caspases-8, -9, and -10) are activated in the earlier phases of apoptosis, whereas the **executioner caspases** (caspases-3, -6, and -7) are activated by initiator caspases and are responsible for dismantling cellular components. Caspases are widely distributed in various tissues and cells.

**Caspase-9 (rat/mouse 454 aa; human 416-aa**, also known as **apoptotic protease activating factor 3 (APAF-3)/ICE-like apoptotic protease 6 or ICE-LAP6 or MCH-6**, is involved in the activation cascade of caspases responsible for apoptosis execution. Binding of Caspase-9 to Apaf-1 leads to activation of the protease which then cleaves and activates caspase-3. Casp-9 proteolytically cleaves poly ADP-ribose polymerase (PARP). It is a heterodimer of a ~35 kDa (p35) and 10 kDa (p10) subunit. Proteolytic cleavages at Asp315 by granzyme B and at Asp330 by CPP32 generate the two active subunits. Caspase-8 and -10 may also be involved in these processing events. Caspase-9 is ubiquitous with highest expression in the heart, and moderate expression in liver and skeletal muscle.

**Source of Antigen and Antibodies**

<b>Antigen</b>	25-aa peptide of <b>human Caspase-9 (1); Designation (CASP91-P, control peptide)</b> . Epitope location ~ CT of p35 subunit
<b>Ab Host/type</b>	Rabbit, Polyclonal Unpurified antiserum (cat # CASP91-S) and aff pure IgG (cat # CASP91-A) purified over the antigen column
<b>2ab</b>	Cat # 20320, goat anti-rabbit IgG-HRP (AP, biotin, FITC conjugates also available)
<b>-ve control</b>	# 20009-1, Rabbit (non-immune) IgG, purified, suitable for ELISA, Western, IHC as -ve control

**Form & Storage of Antibodies/Peptide Control**

**Antiserum (unpurified)**

100ul solution lyophilized powder  
Supplied in Buffer: 0.05% azide

**Reconstitute** powder in 100 ul PBS

**Affinity pure IgG**

100 ug/100ul solution lyophilized powder  
Supplied in **Buffer:** PBS+0.1% BSA  
**Reconstitute powder** in PBS at 1 mg/ml

**Control/blocking peptide**

100 ug/100 ul solution lyophilized powder  
Supplied in **Buffer:** PBS pH 7.5,  
**Reconstitute powder in PBS at 1 mg/ml.**

**Storage**

**Short-term:** unopened, undiluted liquid vials at 20°C and powder at 4°C or -20°C..

**Long-term:** at -20°C or below in suitable aliquots after reconstitution. Do not freeze and thaw and store working, diluted solutions.

**Stability:** 6-12 months at -20°C or below.

**Shipping:** 4°C for solutions and room temp for powder

**Recommended Usage**

**Western Blotting** (1:1K-5K for neat serum and 1-10 ug/ml for affinity pure antibody using ECL technique). Caspase -9 p35 subunit is ~35 Kda.

**ELISA:** Control peptide can be used to coat ELISA plates at 1 ug/ml and detected with antibodies (1:10-50K for neat serum and 0.5-1 ug/ml for affinity pure).

**Histochemistry & Immunofluorescence:** Not tested.

**Specificity & Cross-reactivity**

Human CASP91-P control peptide is 68% conserved in rat, 65% in mouse, 64% in xenopus, and 62% in chicken caspase-9. No significant sequence homology is detected with other caspases. Since CASP91-P is located at the C-terminus of p35 subunit, the antibodies will not detect the p10 subunit. Antibody cross-reactivity in various species has not been studied. Control peptide, because of its low mol. Wt (<3 kDa), is not suitable for Western. It should be used for ELISA or antibody blocking experiments (use 5-10 ug control peptide per 1 ug of aff pure IgG or 1 ul antiserum) to confirm antibody specificity (detailed protocol is available at the web site).

**General References:** (1) Duan H et al (196) JBC 271, 16720; Srinivasula SM et al (1996) JBC 271, 27099; Angelastro JM et al (2001) JBC 276, 12190

\*This product is for In vitro research use only.

**Related material available from ADI**

Antibodies AIF, Apaf-1, Cytochrome-C, Caspases, IAPs, Survivin, EPR-1, CARD, Flash, Aven, Livin, Iceberg

CASP91-S-A-P 71214S



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