

## Product Specification Sheet

### Ice Protease-Activating Factor (IPAF) or CARD12 Antibodies

Cat. # IPAF11-P	Human IPAF Control blocking/Peptide	<b>SIZE:</b> 100 ug
Cat. # IPAF11-S	Rabbit Anti-Human IPAF antiserum	<b>SIZE:</b> 100 ul
Cat. # IPAF11-A	Rabbit Anti-Human IPAF IgG (affinity 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. Apoptosis is driven by two classes of specialized proteases known as caspases (**Cysteine Aspartase**). Several key factors are released from the mitochondria that regulate apoptosis. The first such factor (Cytochrome-C) to be described binds to a cytoplasmic scaffolding protein called **Apaf-1** (Apoptosis Protease activating factor-1), a homolog of *C. elegans Ced-4*. Both Apaf-1 and Ced-4 are composed of an N-terminal Caspase Recruitment domain (CARD) linked to a Nucleotide-binding domain (NBD), also known as NB-ARC or NOD domain. Ced-4 and Apaf-1 self-associate via the NBD and activate Casp-3 and -9. In response to certain apoptotic stimuli, Cytochrome-C is released from the mitochondria and binds to Apaf-1 to form a ternary complex with, and activate, the initiator pro-caspase-9. Active caspase-9 then turns on downstream effector caspases, initiating apoptosis. Recently, Additional members of Apaf-1 family, **NOD1/CARD4** and **NOD2** have been cloned and characterized.

**Ice Protease-Activating Factor (IPAF) or CARD12** is a member of the Ced4 family and can induce apoptosis. Full length **IPAF** or **CLANA**, (human 1,024-aa) contains the CARD, LRR, and NACHT domains, including 4 LRRs, whereas the 359-residue protein, termed **CLANB**, has the CARD spliced directly to the LRRs. The 156- and 92-amino acid proteins, termed **CLANC** and **CLAND**, contain only the CARD and regions with no homology to known motifs. CARD is ubiquitous expressed in lung and spleen, CLANA was detected primarily in lung. **CLANC** and **CLAND** were detected only in an embryonic kidney cell line and brain, respectively.

#### Source of Antigen and Antibodies

<b>Antigen</b>	16-aa peptide from <b>human IPAF (1)</b> ; <b>Designation (IPAF11-P, control peptide)</b> Epitope location ~ N-terminus
<b>Ab Host/type</b>	Rabbit, Polyclonal Unpurified antiserum (cat # IPAF11-S) and aff pure IgG (cat # IPAF11-A) purified over antigen-agarose column
<b>2-Ab`</b>	<b>Goat Anti-rabbit IgG-HRP</b> cat # 20320 (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** Antiserum at 1:1K-1:3K; Aff pure at 1-5 ug/ml using ECL technique).

**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 IAPF11-P control peptide sequence is 100% conserved in all 4 isoforms of IpaF/Card12. It has no significant homology with ced-3/4, Apaf-1, NOD1/2 or other proteins. 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) Geddes BJ et al (2001) BBRC 284, 77-82; Damiano JS et al (2001) Genomics 75, 77-83; Poyet JL et al (2001) JBC 276, 28309-28313

\*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, Aven, Livin, Iceberg, IPAF

**Recycle Immuno blots in Just 5-10 min. (use the same blot for various proteins).** (no boiling or pungent mercaptoethanol).

IPAF11-S-A-P 71215S

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