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# anti-ASK1 antibody (pSer83)

2 Images



Publication



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Quantity:	200 μg
Target:	ASK1 (MAP3K5)
Binding Specificity:	AA 76-87, pSer83
Reactivity:	Human
Host:	Rabbit
Clonality:	Polyclonal
Conjugate:	This ASK1 antibody is un-conjugated
Application:	Western Blotting (WB), Immunohistochemistry (IHC), ELISA, Immunoprecipitation (IP), Radioimmunoassay (RIA)

#### **Product Details**

Immunogen:	This purified antibody was prepared from rabbit serum after repeated immunizations with a	
	KLH conjugated peptide corresponding to amino acids 76-87 of human ASK-1 protein.	
Isotype:	IgG	
Characteristics:	Concentration Definition: by UV absorbance at 280 nm	

## **Target Details**

Target:	ASK1 (MAP3K5)	
Alternative Name:	ASK-1 (MAP3K5 Products)	
Background:	ASK-1 (apoptosis signal-regulating kinase 1 - also referred to as MEK Kinase-5 or MAPKKK5) is	
	a novel serine/threonine MAP kinase kinase kinase (MAPKKK) component of the mitogen -	

activated protein (MAP) cascade that is activated in response to extracellular stimuli by cytokines, growth factors and environmental stresses and other factors. Overexpression of ASK-1 induces apoptotic cell death. ASK-1 is expressed in a variety of human and mouse tissues. The overall amino acid sequence identity between the mouse and human ASK1 is 91.9%. ASK-1 interacts with CDKN1A (also known as p21, WAF1, CIP1). Please refer to the reference list at the end of this document for further information.

Synonyms: Apoptosis signal regulating kinase 1 antibody, ASK 1 antibody, ASK1 antibody,

MAP/ERK kinase kinase 5 antibody, MAP3K5 antibody, MAPK/ERK kinase kinase 5 antibody

Gene ID: 4217

UniProt: Q99683

Pathways: MAPK Signaling, Positive Regulation of Endopeptidase Activity, Unfolded Protein Response

### **Application Details**

Application Notes:

This phospho specific polyclonal antibody reacts human pS83 ASK1 and shows minimal reactivity by western blot, ELISA and competitive ELISA with non-phosphorylated ASK1. Although not tested, this antibody is likely functional in RIA, immunohistochemistry and immunoprecipitation. For immunoblotting a 1:1,000 dilution is recommended. A 155 kDa band corresponding to human ASK-1 is detected. Whole cell lysates from SW1353 can be used as a positive control. For ELISA a 1:5,000 to 1:10,000 dilution is recommended. Researchers should determine optimal titers for other applications.

Restrictions:

For Research Use only

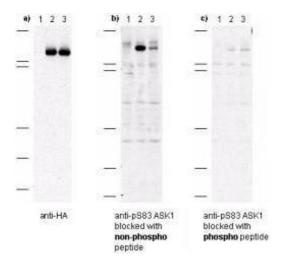
### Handling

Format:	Liquid
Concentration:	1.0 mg/mL
Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	Sodium azide
Precaution of Use:	This product contains sodium azide: a POISONOUS AND HAZARDOUS SUBSTANCE which should be handled by trained staff only.
Storage:	-20 °C

Product cited in:

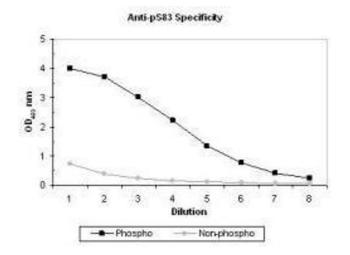
Miller, Tejada, Gazzano-Santoro: "Development of an ELISA based bridging assay as a surrogate measure of ADCC." in: **Journal of immunological methods**, (2012) (PubMed).

#### **Images**



#### **Western Blotting**

Image 1. Western blot of anti-pS83 ASK1 antibodies shows specificity for phosphorylated human ASK1. Anti-pS83 (aa 76-87) antibody was tested by western blot against Cos-7 cell lysates after transient transfection with 1) vector only, 2) recombinant HA-ASK1, and 3) recombinant human HA-ASK1 where S83 was substituted with an alanine residue. Cells were lysed 24 h post-transfection in 200 ?L of 1x SDSsample buffer, heated at 96?C for 5', and vortexed for 30 sec. Samples (10 ?L each) were separated on a 12% SDS-PAGE gel and transferred to PVDF (Millipore) followed by blocking for 45' using TTBS supplemented with 5% non-fat dry milk. All incubations were performed at room temperature. In panel a) all samples were incubated with anti-HA antibody. This blot demonstrates both recombinant transfections express rASK1. In panel b) all samples were incubated with anti-pS83 ASK1. Lane 2 shows strong specific staining of ASK1. Lane 3, where S83 was replaced with alanine, shows greatly diminished staining. In panel c) all samples were incubated with anti-pS83 ASK1 antibody as before except the antibody was pre-incubated with phospho peptide prior to membrane incubation. No staining is observed after phospho peptide blocking occurs.



#### **ELISA**

**Image 2.** ELISA results of purified polyclonal anti-pS83 ASK-1 (aa 76-87) antibody tested against BSA conjugates of non-phospho and phospho forms of immunizing peptide. Each well was coated with 0.1 mg of conjugate. The starting dilution of antibody was 1:1,000 and each point on the X-axis represents a 2-fold dilution. HRP conjugated Gt-a-Rabbit IgG H&L and TMB substrate were used for detection.