

p-ASK1 (Thr 845): sc-109911

BACKGROUND

Mitogen-activated protein (MAP) kinase cascades are activated by various extracellular stimuli including growth factors. The MEK kinases (also designated MAP kinase kinase kinases, MKKKs, MAP3Ks or MEKKs) phosphorylate and thereby activate the MEKs (also called MAP kinase kinases or MKKs), including ERK, JNK and p38. These activated MEKs in turn phosphorylate and activate the MAP kinases. The MEK kinases include Raf-1, Raf-B, Mos, MEK kinase-1, MEK kinase-2, MEK kinase-3, MEK kinase-4, ASK1 and MAP3K6 (MEK kinase-6). ASK1 (apoptosis signal-regulating kinase 1), also known as Map3k5 (mitogen-activated protein kinase kinase kinase 5) or Mekk5 (MAPK/ERK kinase kinase 5), is a 1,380 amino acid protein belonging to the protein kinase superfamily, STE Ser/Thr protein kinase family and MAP kinase kinase superfamily. Containing one protein kinase domain, ASK1 is phosphorylated at Thr-845 through autophosphorylation by MAP3K6/ASK2, leading to activation, and dephosphorylated by PP5.

REFERENCES

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2. Tobiume, K., Matsuzawa, A., Takahashi, T., Nishitoh, H., Morita, K., Takeda, K., Minowa, O., Miyazono, K., Noda, T. and Ichijo, H. 2001. ASK1 is required for sustained activations of JNK/p38 MAP kinases and apoptosis. *EMBO Rep.* 2: 222-228.
3. Tobiume, K., Saitoh, M. and Ichijo, H. 2002. Activation of apoptosis signal-regulating kinase 1 by the stress-induced activating phosphorylation of pre-formed oligomer. *J. Cell. Physiol.* 191: 95-104.
4. Takeda, K., Matsuzawa, A., Nishitoh, H., Tobiume, K., Kishida, S., Ninomiya-Tsuji, J., Matsumoto, K. and Ichijo, H. 2004. Involvement of ASK1 in Ca²⁺-induced p38 MAP kinase activation. *EMBO Rep.* 5: 161-166.
5. Matsuzawa, A., Saegusa, K., Noguchi, T., Sadamitsu, C., Nishitoh, H., Nagai, S., Koyasu, S., Matsumoto, K., Takeda, K. and Ichijo, H. 2005. ROS-dependent activation of the TRAF6-ASK1-p38 pathway is selectively required for TLR4-mediated innate immunity. *Nat. Immunol.* 6: 587-592.
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7. Liu, Q., Wilkins, B.J., Lee, Y.J., Ichijo, H. and Molkentin, J.D. 2006. Direct interaction and reciprocal regulation between ASK1 and calcineurin-NFAT control cardiomyocyte death and growth. *Mol. Cell. Biol.* 26: 3785-3797.
8. Saito, J., Toriumi, S., Awano, K., Ichijo, H., Sasaki, K., Kobayashi, T. and Tamura, S. 2007. Regulation of apoptosis signal-regulating kinase 1 by protein phosphatase 2C ϵ . *Biochem. J.* 405: 591-596.

CHROMOSOMAL LOCATION

Genetic locus: MAP3K5 (human) mapping to 6q22.33; Map3k5 (mouse) mapping to 10 A3.

SOURCE

p-ASK1 (Thr 845) is a rabbit polyclonal antibody raised against a short amino acid sequence containing Thr 845 phosphorylated ASK1 of mouse origin.

PRODUCT

Each vial contains 100 μ g IgG in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

Blocking peptide available for competition studies, sc-109911 P, (100 μ g peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% BSA).

APPLICATIONS

p-ASK1 (Thr 845) is recommended for detection of Thr 845 phosphorylated ASK1 of mouse origin and correspondingly phosphorylated Thr 838 of human origin by Western Blotting (starting dilution 1:100, dilution range 1:50-1:500), immunofluorescence (starting dilution 1:25, dilution range 1:25-1:250) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

p-ASK1 (Thr 845) is also recommended for detection of correspondingly phosphorylated ASK1 in additional species, including equine, canine, bovine, porcine and avian.

Molecular Weight of p-ASK1: 155 kDa.

RECOMMENDED SECONDARY REAGENTS

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto B Blocking Reagent: sc-2335 (use 50 mM NaF, sc-24988, as diluent), Western Blotting Luminol Reagent: sc-2048 and Lambda Phosphatase: sc-200312A. 2) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

SELECT PRODUCT CITATIONS

1. Shen, K., Ji, L., Lu, B., Xu, C., Gong, C., Morahan, G. and Wang, Z. 2014. Andrographolide inhibits tumor angiogenesis via blocking VEGFA/VEGFR2-MAPKs signaling cascade. *Chem. Biol. Interact.* 218: 99-106.

STORAGE

Store at 4° C, **DO NOT FREEZE**. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

See our web site at www.scbt.com or our catalog for detailed protocols and support products.