

p-Raf-1 (H-8): sc-271928

BACKGROUND

Raf-1 is a ubiquitously expressed cytoplasmic protein with intrinsic serine/threonine kinase activity. Raf-1, or c-Raf, is the cellular homolog of v-Raf, the product of the transforming gene of the 3,611 strain of murine sarcoma virus. The unregulated kinase activity of the v-Raf protein is associated with cellular transformation and mitogenesis. Raf-1 is normally suppressed by its regulatory N-terminal domain. Raf-1 is activated in response to a variety of tyrosine kinase receptors as well as in response to pp60v-Src expression. Specifically, Raf-1 is phosphorylated in the catalytic domain at Ser 338 and, to a lesser extent, Ser 339. This phosphorylation requires the co-activation of PI 3-kinase and the Ras signaling pathway. Raf-1 is also phosphorylated on Tyr 340 and 341, which induces the phosphorylation of MEK. Phosphorylation of Ser 621 is essential for the catalytic activity of Raf-1 and downregulation by c-AMP-dependent protein kinase A (PKA). PKA also phosphorylates Raf-1 on Ser 43 and Ser 259. PKA phosphorylation of Ser 259 inhibits Raf-1 and decreases the phosphorylation necessary for Raf-1 activation at Ser 338.

CHROMOSOMAL LOCATION

Genetic locus: RAF1 (human) mapping to 3p25.2; Raf1 (mouse) mapping to 6 E3.

SOURCE

p-Raf-1 (H-8) is a mouse monoclonal antibody raised against a short amino acid sequence containing Ser 621 phosphorylated Raf-1 of human origin.

PRODUCT

Each vial contains 200 µg IgG_{2b} kappa light chain in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

Blocking peptide available for competition studies, sc-271928 P, (100 µg peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% stabilizer protein).

APPLICATIONS

p-Raf-1 (H-8) is recommended for detection of Ser 621 phosphorylated Raf-1 of mouse, rat and human origin by Western Blotting (starting dilution 1:100, dilution range 1:100-1:1000), immunoprecipitation [1-2 µg per 100-500 µg of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500), immunohistochemistry (including paraffin-embedded sections) (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

p-Raf-1 (H-8) is also recommended for detection of correspondingly phosphorylated Raf-1 in additional species, including equine and avian.

Suitable for use as control antibody for Raf-1 siRNA (h): sc-29462, Raf-1 siRNA (m): sc-29463, Raf-1 shRNA Plasmid (h): sc-29462-SH, Raf-1 shRNA Plasmid (m): sc-29463-SH, Raf-1 shRNA (h) Lentiviral Particles: sc-29462-V and Raf-1 shRNA (m) Lentiviral Particles: sc-29463-V.

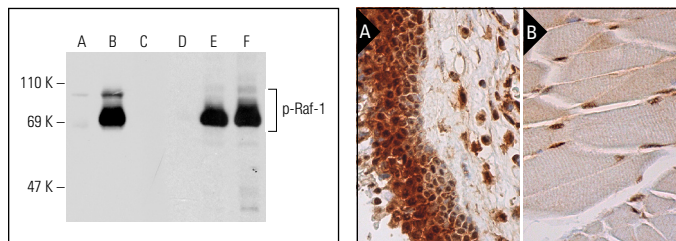
Molecular Weight of p-Raf-1: 74 kDa.

Positive Controls: Raf-1 (m): 293T Lysate: sc-122942, HeLa + UV cell lysate: sc-2221 or HeLa whole cell lysate: sc-2200.

STORAGE

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

DATA



Western blot analysis of Raf-1 phosphorylation in non-transfected: sc-117752 (A,D), untreated mouse Raf-1 transfected: sc-122942 (B,E) and lambda protein phosphatase (sc-200312A) treated mouse Raf-1 transfected: sc-122942 (C,F) 293T whole cell lysates. Antibodies tested include p-Raf-1 (H-8): sc-271928 (A,B,C) and Raf-1 (C-20): sc-227 (D,E,F).

p-Raf-1 (H-8): sc-271928. Immunoperoxidase staining of formalin fixed, paraffin-embedded human nasopharynx tissue showing nuclear and cytoplasmic staining of respiratory epithelial cells (A). Immunoperoxidase staining of formalin fixed, paraffin-embedded human skeletal muscle tissue showing nuclear staining of myocytes (B).

SELECT PRODUCT CITATIONS

- Wang, Z., et al. 2014. Tissue kallikrein protects rat hippocampal CA1 neurons against cerebral ischemia/reperfusion-induced injury through the B2R-Raf-MEK1/2-ERK1/2 pathway. *J. Neurosci. Res.* 92: 651-657.
- Cheng, D., et al. 2019. miR-142-5p suppresses proliferation and promotes apoptosis of human osteosarcoma cell line, HOS, by targeting PLA2G16 through the ERK1/2 signaling pathway. *Oncol. Lett.* 17: 1363-1371.
- Luan, X., et al. 2021. Cyclophilin A is a key positive and negative feedback regulator within interleukin-6 *trans*-signaling pathway. *FASEB J.* 35: e21958.
- Park, H.B., et al. 2022. USP7 regulates the ERK1/2 signaling pathway through deubiquitinating Raf-1 in lung adenocarcinoma. *Cell Death Dis.* 13: 698.
- Wang, J., et al. 2022. PHLDA1 promotes glioblastoma cell growth via sustaining the activation state of Ras. *Cell. Mol. Life Sci.* 79: 520.

RESEARCH USE

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

PROTOCOLS

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