

# p-Raf-1 (E-1): sc-271929



The Power to Question

## 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 3611 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.

## REFERENCES

1. Rapp, U.R., et al. 1983. Structure and biological activation of v-Raf, a unique oncogene transduced by a retrovirus. *Proc. Natl. Acad. Sci. USA* 80: 4218-4222.
2. Huleihel, M., et al. 1986. Characterization of murine A-Raf, a new oncogene related to the v-Raf oncogene. *Mol. Cell. Biol.* 6: 2655-2662.

## CHROMOSOMAL LOCATION

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

## SOURCE

p-Raf-1 (E-1) 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<sub>2b</sub> kappa light chain in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

p-Raf-1 (E-1) is available conjugated to agarose (sc-271929 AC), 500 µg/0.25 ml agarose in 1 ml, for IP; to HRP (sc-271929 HRP), 200 µg/ml, for WB, IHC(P) and ELISA; to either phycoerythrin (sc-271929 PE), fluorescein (sc-271929 FITC), Alexa Fluor® 488 (sc-271929 AF488), Alexa Fluor® 546 (sc-271929 AF546), Alexa Fluor® 594 (sc-271929 AF594) or Alexa Fluor® 647 (sc-271929 AF647), 200 µg/ml, for WB (RGB), IF, IHC(P) and FCM; and to either Alexa Fluor® 680 (sc-271929 AF680) or Alexa Fluor® 790 (sc-271929 AF790), 200 µg/ml, for Near-Infrared (NIR) WB, IF and FCM.

Blocking peptide available for competition studies, sc-271929 P, (100 µg peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% stabilizer protein).

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## RESEARCH USE

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

## APPLICATIONS

p-Raf-1 (E-1) 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).

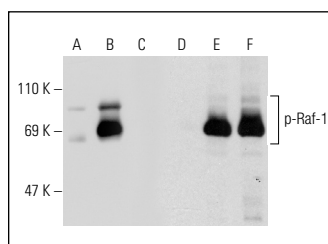
p-Raf-1 (E-1) is also recommended for detection of correspondingly phosphorylated Raf-1 in additional species, including equine and avian.

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.

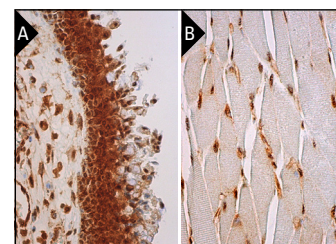
Molecular Weight of p-Raf-1: 74 kDa.

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

## 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 (E-1): sc-271929 (A,B,C) and Raf-1 (C-20): sc-227 (D,E,F).



p-Raf-1 (E-1): sc-271929. 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

1. Islam, S.U., et al. 2018. Decursinol angelate inhibits LPS-induced macrophage polarization through modulation of the NFκB and MAPK signaling pathways. *Molecules* 23: 1880.
2. Yin, Q., et al. 2019. K27-linked ubiquitination of BRAF by ITCH engages cytokine response to maintain MEK-ERK signaling. *Nat. Commun.* 10: 1870.
3. Dai, C., et al. 2020. Lactate dehydrogenase a governs cardiac hypertrophic growth in response to hemodynamic stress. *Cell Rep.* 32: 108087.
4. Shen, L., et al. 2022. DDX3 acts as a tumor suppressor in colorectal cancer as loss of DDX3 in advanced cancer promotes tumor progression by activating the MAPK pathway. *Int. J. Biol. Sci.* 18: 3918-3933.

## STORAGE

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