SANTA CRUZ BIOTECHNOLOGY, INC.

p-PAK4 (71.Ser 474): sc-135774



BACKGROUND

Three recently identified isoforms of serine/threonine kinases, designated α PAK p68, β PAK p65 and γ PAK p62, have been shown to exhibit a high degree of sequence homology with the S. cerevisiae kinase STE20, involved in pheromone signaling. The α , β and γ PAK isoforms complex specifically with Rac 1 and Cdc42 in their active GTP bound state, inhibiting their intrinsic GTPase activity leading to their autophosphorylation. Once phosphorylated and their affinity for Rac/Cdc42 reduced, the PAK isoforms disassociate from the complex to seek downstream substrates. One such putative substrate is MEK kinase, an upstream effector of MEK4 which is involved in the JNK signaling pathway. While the PAK isoforms interact in a GTP-dependent manner with Rac1 and Cdc42, they do not interact with Rho. PAK4 is highly expressed in prostate, testis and colon. PAK4 interacts tightly with GTP-bound but not GDPbound Cdc42 and weakly with Rac. PAK4 phosphorylates and autophosphorylates and also activates the JNK pathway. Coexpression of PAK4 and activated Cdc42 induces the sustained formation of Actin-enriched filopodia protrusions and causes PAK4 to co-localize with polymerized Actin clusters and with β coat protein in the Golgi. The gene which encodes PAK4 maps to human chromosome 19.

REFERENCES

- Didsbury, J., et al. 1989. Rac, a novel Ras-related family of proteins that are *botulinum* toxic substrates. J. Biol. Chem. 264: 16378-16382.
- Shinjo, K., et al. 1990. Molecular cloning of the gene for the human placental GTP-binding protein Gp (G25K): identification of this GTP-binding protein as the human homolog of the yeast cell-division-cycle protein Cdc42. Proc. Natl. Acad. Sci. USA 98: 9853-9857.
- Boguski, M.S., et al. 1993. Proteins regulating Ras and its relatives. Nature 366: 643-654.
- Lange-Carter, C.A., et al. 1993. A divergence in the MAP kinase regulatory network defined by MEK kinase and Raf. Science 260: 315-319.
- Manser, E., et al. 1994. A brain serine/threonine protein kinase activated by Cdc42 and Rac 1. Nature 367: 40-46.

CHROMOSOMAL LOCATION

Genetic locus: PAK4 (human) mapping to 19q13.2; Pak4 (mouse) mapping to 7 A3.

SOURCE

p-PAK4 (71.Ser 474) is a mouse monoclonal antibody raised against a short amino acid sequence containing Ser 474 phosphorylated PAK4 of human origin.

PRODUCT

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

STORAGE

Store at 4° C, **D0 NOT FREEZE**. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

APPLICATIONS

p-PAK4 (71.Ser 474) is recommended for detection of Ser 474 phosphorylated PAK4 of human origin and Ser 476 phosphorylated PAK4 of mouse and rat origin by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000), immunoprecipitation [1-2 μ g per 100-500 μ g of total protein (1 ml of cell lysate)] and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

Suitable for use as control antibody for PAK4 siRNA (h): sc-39060, PAK4 siRNA (m): sc-39061, PAK4 shRNA Plasmid (h): sc-39060-SH, PAK4 shRNA Plasmid (m): sc-39061-SH, PAK4 shRNA (h) Lentiviral Particles: sc-39060-V and PAK4 shRNA (m) Lentiviral Particles: sc-39061-V.

Molecular Weight of p-PAK4: 68 kDa.

Positive Controls: PAK4 (h): 293 Lysate: sc-111101.

RECOMMENDED SUPPORT REAGENTS

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgGκ BP-HRP: sc-516102 or m-IgGκ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker[™] Molecular Weight Standards: sc-2035, TBS Blotto B Blocking Reagent: sc-2335 (use 50 mM NaF, sc-24988, as diluent), Lambda Phosphatase: sc-200312A and Western Blotting Luminol Reagent: sc-2048. 2) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml).

DATA



Western blot analysis of PAK4 phosphorylation in nontransfected: sc-110760 (**A**,**D**), untreated human PAK4 transfected: sc-111101 (**B**,**E**) and lambda protein phosphatase (sc-200312A) treated human PAK4 transfected: sc-111101 (**C**,**F**) 293 whole cell lysates. Antibodies tested include p-PAK4 (71.Ser 474): sc-135774 (**A**,**B**,**C**) and PAK4 (6C1): sc-81532 (**D**,**E**,**F**).

SELECT PRODUCT CITATIONS

 Li, N., et al. 2018. Dual PAK4-NAMPT inhibition impacts growth and survival, and increases sensitivity to DNA-damaging agents in Waldenstrom Macroglobulinemia. Clin. Cancer Res. pii: clincanres.

RESEARCH USE

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

PROTOCOLS

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