SANTA CRUZ BIOTECHNOLOGY, INC.

αPAK (YY08): sc-100360

SANTA CRUZ BIOTECHNOLOGY

The Power to Question

BACKGROUND

Three isoforms of serine/threonine kinases, designated αPAK p68, βPAK p65 and γPAK 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 γPAK isoforms complex specifically with Rac 1 and Cdc42 in their active GTP-bound state, inhibiting their intrinsic GTPase activity leading to their autophosphorylation. There are eight sites of autophosphorylation on γPAK, including Ser 19, Ser 141 and Thr 402, and phosphorylation of Ser 141 and Thr 402 is correlated with γPAK activation. 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 Rac 1 and Cdc42, they do not interact with Rho.

REFERENCES

1. Didsbury, J., et al. 1989. Rac, a novel Ras-related family of proteins that are botulinum toxic substrates. J. Biol. Chem. 264: 16378-16382.
2. 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 87: 9853-9857.
3. Boguski, M.S., et al. 1993. Proteins regulating Ras and its relatives. Nature 366: 643-654.
4. Manser, E., et al. 1994. A brain serine/threonine protein kinase activated by Cdc42 and Rac 1. Nature 367: 40-46.
5. Yan, M., et al. 1994. Activation of stress-activated protein kinase by MEKK1 phosphorylation of its activator SEK1. Nature 372: 798-800.
6. Minden, A., et al. 1994. Differential activation of ERK and JNK mitogen-activated protein kinases by Raf-1 and MEKK. Science 266: 1719-1723.
7. Martin, G.A., et al. 1995. A novel serine kinase activated by Rac 1/Cdc42Hs-dependent autophosphorylation is related to PAK65 and Ste20. EMBO J. 14: 1970-1978.
8. Coso, O.A., et al. 1995. The small GTP-binding proteins Rac 1 and Cdc42 regulate the activity of the JNK/SAPK signaling pathway. Cell 81: 1137-1146.

CHROMOSOMAL LOCATION

Genetic locus: PAK1 (human) mapping to 11q13-q14.

SOURCE

αPAK (YY08) is a mouse monoclonal antibody raised against recombinant αPAK of human origin.

PRODUCT

Each vial contains 100 µg IgG_1 in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

APPLICATIONS

αPAK (YY08) is recommended for detection of αPAK of human origin by Western Blotting (starting dilution 1:200, 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).

Suitable for use as control antibody for αPAK siRNA (h): sc-29700, αPAK shRNA Plasmid (h): sc-29700-SH and αPAK shRNA (h) Lentiviral Particles: sc-29700-V.

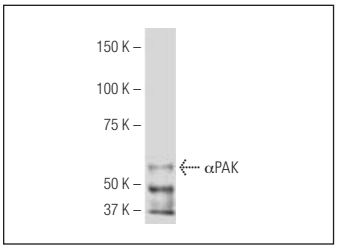
Molecular Weight of αPAK: 65 kDa.

Positive Controls: HeLa nuclear extract: sc-2120, HeLa whole cell lysate: sc-2200 or human stomach tissue.

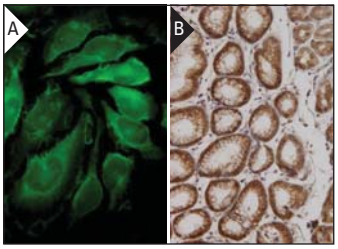
RECOMMENDED SECONDARY REAGENTS

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml). 3) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941. 4) Immunohistochemistry: use ImmunoCruz™: sc-2050 or ABC: sc-2017 mouse IgG Staining Systems.

DATA



αPAK (YY08): sc-100360. Western blot analysis of αPAK expression in HeLa nuclear extract.



αPAK (YY08): sc-100360. Immunofluorescence staining of paraformaldehyde-fixed HeLa cells showing cytoplasmic localization (**A**). Immunoperoxidase staining of formalin-fixed, paraffin-embedded human stomach tissue showing membrane and cytoplasmic localization (**B**).

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.