

# $\alpha/\beta/\gamma$ PAK (M-83): sc-48826

## BACKGROUND

Three isoforms of serine/threonine kinases, designated  $\alpha$ PAK p68,  $\beta$ PAK p65 and  $\gamma$ PAK p62, have been shown to exhibit a high degree of sequence homology with the *S. cerevisiae* kinase Ste20, involved in pheromone signaling. The  $\alpha$ ,  $\beta$  and  $\gamma$ PAK isoforms complex specifically with Rac1 and Cdc42 in their active GTP-bound state, inhibiting their intrinsic GTPase activity leading to their autophosphorylation. There are eight sites of autophosphorylation on  $\gamma$ PAK, including Ser 19, Ser 141 and Thr 402, and phosphorylation of Ser 141 and Thr 402 is correlated with  $\gamma$ PAK activation. Once phosphorylated and their affinity for Rac/Cdc42 reduced, the PAK isoforms dissociate 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.

## REFERENCES

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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* 98: 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 Rac1. *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 Rac1/Cdc42 Hs-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 Rac1 and Cdc42 regulate the activity of the JNK/SAPK signaling pathway. *Cell* 81: 1137-1146.

## SOURCE

$\alpha/\beta/\gamma$ PAK (M-83) is a rabbit polyclonal antibody raised against amino acids 263-345 mapping within an internal region of  $\beta$ PAK of mouse origin.

## PRODUCT

Each vial contains 200  $\mu$ g IgG in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

## STORAGE

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

## APPLICATIONS

$\alpha/\beta/\gamma$ PAK (M-83) is recommended for detection of  $\alpha$ PAK,  $\beta$ PAK and  $\gamma$ PAK of mouse, rat and human origin by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000), immunoprecipitation [1-2  $\mu$ g per 100-500  $\mu$ g of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

$\alpha/\beta/\gamma$ PAK (M-83) is also recommended for detection of  $\alpha$ PAK,  $\beta$ PAK and  $\gamma$ PAK in additional species, including equine, canine, bovine, porcine and avian.

Molecular Weight of  $\alpha/\beta/\gamma$ PAK: 67/68/62 kDa.

Positive Controls: HeLa whole cell lysate: sc-2200, Jurkat whole cell lysate: sc-2204 or SK-N-MC cell lysate: sc-2237.

## 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 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-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.

## RESEARCH USE

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

## PROTOCOLS

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Try  $\alpha/\beta/\gamma$ PAK (D-8): sc-166174, our highly recommended monoclonal alternative to  $\alpha/\beta/\gamma$ PAK (M-83).