SANTA CRUZ BIOTECHNOLOGY, INC.

p-Akt1/2/3 (B-5): sc-271966



BACKGROUND

The serine/threonine kinase Akt family contains several members, including Akt1 (also designated PKB or RacPK), Akt2 (also designated PKBß or RacPK-ß) and Akt 3 (also designated PKBy or thyoma viral proto-oncogene 3), which exhibit sequence homology with the protein kinase A and C families and are encoded by the c-Akt proto-oncogene. All members of the Akt family have a pleckstrin homology domain. Akt1 and Akt2 are activated by PDGF stimulation. This activation is dependent on PDGFR-β tyrosine residues 740 and 751, which bind the subunit of the phosphatidylinositol 3-kinase (PI 3-kinase) complex. Activation of Akt1 by Insulin or Insulin-growth factor-1 (IGF-1) results in phosphorylation of both Thr 308 and Ser 473. Akt proteins become phosphorylated and activated in Insulin/IGF-1-stimulated cells by an upstream kinase(s), and the activation of Akt1 and Akt2 is inhibited by the PI kinase inhibitor Wortmannin. Taken together, this data strongly suggests that the protein signals downstream of the PI kinases. Akt3 is phosphorylated on a serine residue in response to Insulin. However, the activation of Akt3 by Insulin is inhibited by prior activation of protein kinase C via a mechanism that does not require the presence of the PH domain. Akt3 is expressed in 3T3-L1 fibroblasts, adipocytes and skeletal muscle and may be involved in various biological processes, including adipocyte and muscle differentiation, glycogen synthesis, glucose uptake, apoptosis and cellular proliferation.

SOURCE

p-Akt1/2/3 (B-5) is a mouse monoclonal antibody epitope corresponding to a short amino acid sequence containing Thr 308 phosphorylated Akt2 of human origin.

PRODUCT

Each vial contains 200 $\mu g~lg G_{2b}$ kappa light chain in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

p-Akt1/2/3 (B-5) is available conjugated to agarose (sc-271966 AC), 500 µg/ 0.25 ml agarose in 1 ml, for IP; to HRP (sc-271966 HRP), 200 µg/ml, for WB, IHC(P) and ELISA; to either phycoerythrin (sc-271966 PE), fluorescein (sc-271966 FITC), Alexa Fluor[®] 488 (sc-271966 AF488), Alexa Fluor[®] 546 (sc-271966 AF546), Alexa Fluor[®] 594 (sc-271966 AF594) or Alexa Fluor[®] 647 (sc-271966 AF647), 200 µg/ml, for WB (RGB), IF, IHC(P) and FCM; and to either Alexa Fluor[®] 680 (sc-271966 AF680) or Alexa Fluor[®] 790 (sc-271966 AF790), 200 µg/ml, for Near-Infrared (NIR) WB, IF and FCM.

Blocking peptide available for competition studies, sc-271966 P, (100 μ g peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% stabilizer protein).

Alexa Fluor® is a trademark of Molecular Probes, Inc., Oregon, USA

STORAGE

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

PROTOCOLS

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

APPLICATIONS

p-Akt1/2/3 (B-5) is recommended for detection of Thr 308 phosphorylated Akt1 and correspondingly Thr 309 phosphorylated Akt2 and correspondingly Thr 305 phosphorylated Akt3 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)], immuno-fluorescence (starting dilution 1:50, dilution range 1:50-1:500), immunohis-tochemistry (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-Akt1/2/3 (B-5) is also recommended for detection of correspondingly phosphorylated Akt1, Akt2 and Akt3 in additional species, including equine and avian.

Molecular Weight of p-Akt1/p-Akt2/p-Akt3: 62/56/60 kDa.

Positive Controls: A-431 + EGF whole cell lysate: sc-2202, HeLa + heat shock cell lysate: sc-2272 or Jurkat whole cell lysate: sc-2204.

DATA





Western blot analysis of Akt1 phosphorylation in nontransfected: sc-11752 (A,D), untreated human Akt1 transfected: sc-158248 (**B**,**E**) and lambda protein phosphatase (sc-200312A) treated human Akt1 transfected: sc-158248 (**C**,**F**) 293T whole cell lysates. Antibodies tested include p-Akt1/2/3 (B-5): sc-271966 (**A-C**) and Akt1 (C-20): sc-1618 (**D-F**). p-Akt1/2/3 (B-5): sc-271966. Immunoperoxidase staining of formalin fixed, paraffin-embedded human pancreas tissue showing nuclear staining of exocrine glandular cells and nuclear and cytoplasmic staining of Islets of Langerhans.

SELECT PRODUCT CITATIONS

- Pen, Y., et al. 2016. Membrane-tethered Akt kinase regulates basal synaptic transmission and early phase LTP expression by modulation of post-synaptic AMPA receptor level. Hippocampus 26: 1149-1167.
- Ye, Y., et al. 2018. MicroRNA-495 suppresses cell proliferation and invasion of hepatocellular carcinoma by directly targeting Insulin-like growth factor receptor-1. Exp. Ther. Med. 15: 1150-1158.
- Takino, J.I., et al. 2019. The inhibition of Bax activation-induced apoptosis by RasGRP2 via R-Ras-PI3K-Akt signaling pathway in the endothelial cells. Sci. Rep. 9: 16717.
- Medlej, A., et al. 2020. A novel miRNA located in the GATA4 gene regulates the expression of IGF-1R and Akt1/2 genes and controls cell proliferation. J. Cell. Biochem. 121: 3438-3450.

RESEARCH USE

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