

Akt (1G1) Mouse mAb

✓ 800 µl
(40 immunoprecipitations)

Orders ■ 877-616-CELL (2355)
orders@cellsignal.com

Support ■ 877-678-TECH (8324)
info@cellsignal.com

Web ■ www.cellsignal.com

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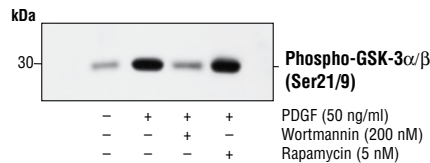
This product is intended for research purposes only. This product is not intended to be used for therapeutic or diagnostic purposes in humans or animals.

Applications	Species Cross-Reactivity	Molecular Wt.	Isotype
IP Endogenous	H, M, R, Hm	60 kDa	Mouse IgG2a

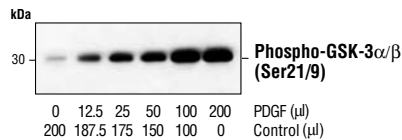
Background: Akt, also referred to as PKB or Rac, plays a critical role in controlling survival and apoptosis (1-3). This protein kinase is activated by insulin and various growth and survival factors to function in a wortmannin-sensitive pathway involving PI3 kinase (2,3). Akt is activated by phospholipid binding and activation loop phosphorylation at Thr308 by PDK1 (4) and by phosphorylation within the carboxy terminus at Ser473. The previously elusive PDK2 responsible for phosphorylation of Akt at Ser473 has been identified as mammalian target of rapamycin (mTOR) in a rapamycin-insensitive complex with rictor and Sin1 (5,6). Akt promotes cell survival by inhibiting apoptosis through phosphorylation and inactivation of several targets, including Bad (7), forkhead transcription factors (8), c-Raf (9) and caspase-9. PTEN phosphatase is a major negative regulator of the PI3 kinase/Akt signaling pathway (10). LY294002 is a specific PI3 kinase inhibitor (11). Another essential Akt function is the regulation of glycogen synthesis through phosphorylation and inactivation of GSK-3 α and β (12,13). Akt may also play a role in insulin stimulation of glucose transport (12). In addition to its role in survival and glycogen synthesis, Akt is involved in cell cycle regulation by preventing GSK-3 β mediated phosphorylation and degradation of cyclin D1 (14) and by negatively regulating the cyclin dependent kinase inhibitors p27 Kip (15) and p21 Waf1/CIP1 (16). Akt also plays a critical role in cell growth by directly phosphorylating mTOR in a rapamycin-sensitive complex containing raptor (17). More importantly, Akt phosphorylates and inactivates tuberlin (TSC2), an inhibitor of mTOR within the mTOR-raptor complex (18). Inhibition of mTOR stops the protein synthesis machinery by inactivating p70 S6 kinase and activating the eukaryotic initiation factor 4E binding protein 1 (4E-BP1), an inhibitor of translation (18,19).

Specificity/Sensitivity: Akt (1G1) Mouse mAb binds preferentially to Akt phosphorylated at Ser473 and is designed for use in *in vitro* kinase assays. The antibody will also bind some nonphospho-Akt. It is prepared by coupling carbohydrates on the Fc region of IgG to cross-linked hydrazide agarose beads.

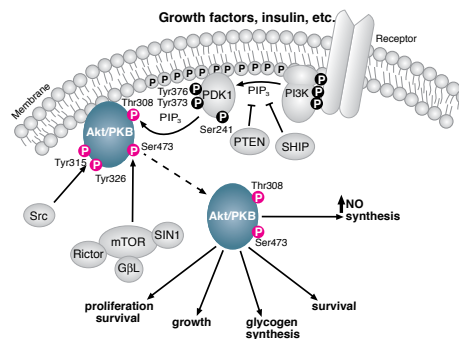
Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic phosphopeptide corresponding to residues around Ser473 of human Akt.



Analysis of Akt kinase activity in PDGF, wortmannin and rapamycin treated NIH/3T3 cells. Cell extracts (200 µl) were incubated for 2 hours with Akt (1G1) Mouse mAb. After extensive washing, the kinase reaction was performed in the presence of 200 µM cold ATP and GSK-3 substrate. Phosphorylation of GSK-3 was measured by Western blot using Phospho-GSK-3 α/β (Ser21/9) Antibody #9331.



Analysis of Akt kinase activity in PDGF treated NIH/3T3 cell extracts: Cell extracts (200 µl) were incubated for 2 hours with Akt (1G1) Mouse mAb. After extensive washing, the kinase reaction was performed in the presence of 200 µM cold ATP and GSK-3 substrate. Phosphorylation of GSK-3 was measured by Western blot using Phospho-GSK-3 α/β (Ser21/9) Antibody #9331.



Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at -20°C. Do not aliquot the antibody.

Recommended Antibody Dilutions:

Immunoprecipitation 1:100

For application specific protocols please see the web page for this product at www.cellsignal.com.

Please visit www.cellsignal.com for a complete listing of recommended companion products.

Background References:

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Applications Key: W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E-P—ELISA-Peptide

Species Cross-Reactivity Key: H—human M—mouse R—rat Hm—hamster Mk—monkey Mi—mink C—chicken Dm—D. melanogaster X—Xenopus Z—zebrafish B—bovine

Dg—dog Pg—pig Sc—S. cerevisiae Ce—C. elegans Hr—horse

All—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.