

**#2966** Store at **-20°C**

# Akt (5G3) Mouse mAb



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rev. 01/04/16

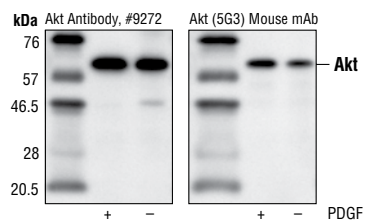
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Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
IP, IF-IC, F Endogenous	H, M, R, Hm	60 kDa	Mouse IgG1**

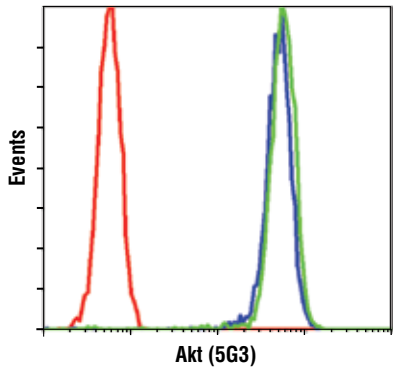
**Background:** Akt, also referred to as PKB or Rac, plays a critical role in controlling survival and apoptosis (1-3). This protein kinase is activated by insulin and various growth and survival factors to function in a wortmannin-sensitive pathway involving PI3 kinase (2,3). Akt is activated by phospholipid 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 $\alpha$  and  $\beta$  (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 $\beta$ -mediated phosphorylation and degradation of cyclin D1 (14) and by negatively regulating the cyclin dependent kinase inhibitors p27 Kip1 (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 tuberin (TSC2), an inhibitor of mTOR within the mTOR-raptor complex (18,19).

**Specificity/Sensitivity:** Akt (5G3) Mouse mAb detects endogenous levels of Akt1 and Akt3. This antibody does not cross-react with other related proteins.

**Source/Purification:** Monoclonal antibody is produced by immunizing animals with an Akt1 recombinant protein containing human Akt1 residues 140-480.



Immunoprecipitation of Akt from PDGF (100 ng/ml for 11 minutes) treated and untreated NIH/3T3 cell lysates, using control Akt Antibody #9272 (left) and Akt (5G3) Mouse mAb (right). Western blot detection was performed using Akt Antibody #9272.



Akt (5G3) mAb #2966 staining of untreated (blue) or LY294002-treated (green) jurkat cells compared to a nonspecific negative control antibody (red).

**Entrez-Gene ID** #207  
**Swiss-Prot Acc.** #P31749

**Storage:** Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100  $\mu$ g/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at  $-20^{\circ}\text{C}$ . Do not aliquot the antibody.

**\*Species cross-reactivity is determined by western blot.**  
**\*\*Anti-mouse secondary antibodies must be used to detect this antibody.**

**Recommended Antibody Dilutions:**

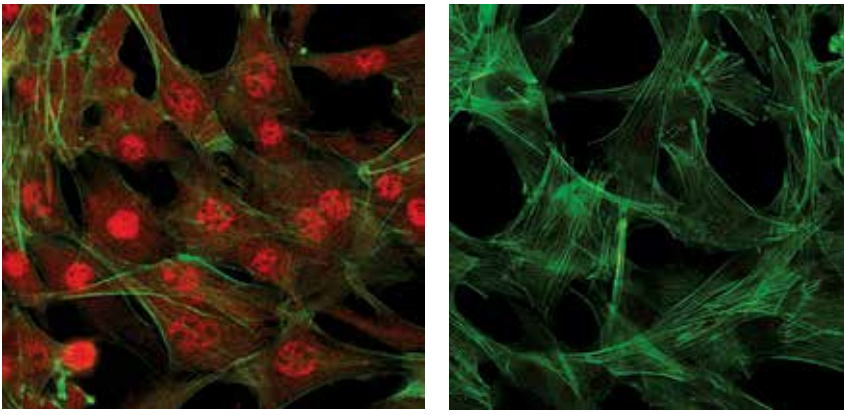
Immunoprecipitation	1:100
Immunofluorescence (IF-IC)	1:50
Flow Cytometry	1:50

**For product specific protocols please see the web page for this product at [www.cellsignal.com](http://www.cellsignal.com).**

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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.



Confocal immunofluorescent analysis of C2C12 cells showing nuclear and cytoplasmic localization with Akt (5G3) Mouse mAb (left, red) compared to an isotype control (right). Actin filaments have been labeled with fluorescein phalloidin.

#### Background References:

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