



# MAB-94125 Rabbit Anti Phospho-Akt1 (Ser473) Clone (D7F10)

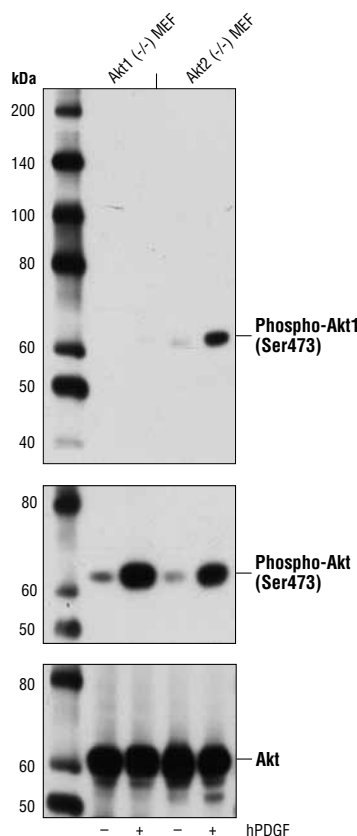
100 µl (10 western blots)

Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
W, IP, IF-IC, F Endogenous	H, M, R	60 kDa	Rabbit IgG**

**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 $\alpha$ -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 (18,19).

**Specificity/Sensitivity:** Phospho-Akt1 (Ser473) (D7F10) Rabbit mAb (Akt1 Specific) recognizes endogenous levels of Akt1 protein only when phosphorylated at Ser473. It does not detect Akt2 protein when phosphorylated at Ser474.

**Source/Purification:** Monoclonal antibody is produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Ser473 of human Akt1 protein.



Western blot analysis of extracts from Akt1 (-/-) mouse embryonic fibroblast (MEF) or Akt2 (-/-) MEF, untreated or stimulated with hPDGF (100 ng/ml, 15 min), using Phospho-Akt1 (Ser473) (D7F10) Rabbit mAb (Akt1 Specific) (upper), Phospho-Akt (Ser473) (D9E) Rabbit mAb (middle), or Akt (pan) (C67E7) Rabbit (lower).

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Entrez-Gene ID #207

UniProt ID #P31749

**Buffer:** Supplied in 20 mM Tris HCL pH 8.0 buffer and 10 mg/mL BSA as stabilizer and 0.5% Sodium Azide.

**Store:** At +4°C for short term, at -20°C for longer term. Avoid freezing and thawing cycles.

**Shipping:** At room Temperature

**Recommended Antibody Dilutions:**

Western blotting	1:1000
Immunoprecipitation	1:50
Immunofluorescence (IF-IC)	1:50

**Background References:**

- (1) Franke, T.F. et al. (1997) *Cell* 88, 435-7.
- (2) Burgering, B.M. and Coffey, P.J. (1995) *Nature* 376, 599-602.
- (3) Franke, T.F. et al. (1995) *Cell* 81, 727-36.
- (4) Alessi, D.R. et al. (1996) *EMBO J* 15, 6541-51.
- (5) Sarbassov, D.D. et al. (2005) *Science* 307, 1098.
- (6) Jacinto, E. et al. (2006) *Cell* 127, 125-37.
- (7) Cardone, M.H. et al. (1998) *Science* 282, 1318.
- (8) Brunet, A. et al. (1999) *Cell* 96, 857-68.
- (9) Zimmermann, S. and Moelling, K. (1999) *Science* 286, 1741-4.
- (10) Cantley, L.C. and Neel, B.G. (1999) *Proc Natl Acad Sci USA* 96, 4240-5.
- (11) Vlahos, C.J. et al. (1994) *J Biol Chem* 269, 5241.

**Important:**

For Western Blot solutions:

Wash buffer 1x Tris Buffered Saline (TBS);

0.1% Triton X-100 ;

Blocking buffer: 1 XTBS; 0.1% Triton X

For western blots, incubate membrane with diluted antibody in blocking buffer at Room Temperature for 2 hours.

Note: Anti Rabbit HRP secondary antibody must be used for Western Blot (cat. IS-1054P Gt Anti Rb IgG)