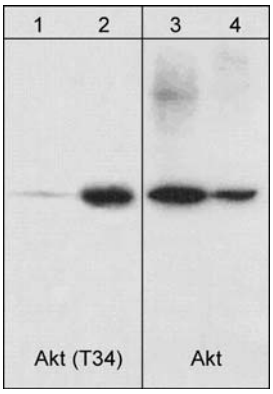
Anti-Akt (Thr-34), phospho-specific

Cat. #	**AP1001**
Host	**Rabbit Polyclonal**
Size	**100µl**

Background:
Akt, also known as Protein Kinase B (PKB) and Rac (Related to the A and C kinases), is a 60kDa serine/threonine kinase critical for controlling diverse cellular functions, including glucose metabolism, gene transcription, cell proliferation, and apoptosis. Akt phosphorylates a number of physiological substrates including MBP, glycogen synthetase, PKA RII subunit, and histone H1. Akt is activated in response to insulin and growth factors in a PI3-kinase dependent manner. Activation of PI3-Kinase generates phosphatidylinositol 3,4-bisphosphate which may induce the membrane translocation of Akt coincident with its phosphorylation and activation. Upon activation, Akt associates with members of the PKC family of kinases, such as PKCδ and PKCζ. Ceramide-activated PKCζ leads to phosphorylation of the Akt-Pleckstrin Homology (PH) domain on threonine 34. This phosphorylation inhibits PIP_3 binding to Akt, preventing activation of the kinase by insulin. Since Akt confers a prosurvival signal and regulates pathways in response to insulin, suppressing its activation may be one mechanism by which ceramide promotes cell death and induces insulin resistance.



Western blot analysis of A431 cells, serum starved overnight (20 µg/lanes 1 & 3) and calyculin A (100 nM) treated for 30 minutes (20 µg/lanes 2 & 4). The blot was probed with anti-Akt (Thr-34) (lanes 1 & 2) or anti-Akt1 (N-terminal region) (lanes 3 & 4).

References:

Jones, P.F. et al. (1991) Proc. Natl. Acad. Sci. USA 88:4171-4175.
Marte, B. & Downward. J. (1997) TIBS. 22:355-358.
Powell, D.J. et al. (2003) Mol. Cell Biol. 23:7794-7808.

Immunogen:
Phospho-Akt (Thr-34) synthetic peptide (coupled to KLH) corresponding to amino acid residues around threonine 34 of human Akt. This sequence is highly conserved in rat and mouse Akt.

Buffer and Storage:
Rabbit polyclonal, affinity-purified antibody is supplied in 100µl phosphate-buffered saline, 50% glycerol, 1 mg/ml BSA, and 0.05% sodium azide. Store at –20°C. Do not aliquot. Stable for 1 year.

Applications:

Western blotting	1:1000 dilution†
ELISA	1:2000 dilution

End user should determine optimal dilution for their particular applications and experiments.
†Membrane was incubated with diluted antibody in 5% non-fat milk, PBS, 0.04% Tween20 for 1hour at room temperature.

Specificity:
The antibody detects a 60 kDa* protein corresponding to the apparent molecular mass of phoshorylated Akt on SDS-PAGE immunoblots of A431 + calyculin A cell lysate. Similar results were seen in calyculin A treated human aortic endothelial and HeLa cells, rabbit spleen fibroblasts, and rat pituitary cells.
*All molecular weights (MW) are confirmed by comparison to Bio-Rad Rainbow Markers and to western blot mobilities of known proteins with similar MW.

Related Products:
AM1011 Akt1 (N-terminal Region) Mouse Monoclonal
AX1005 phospho-Akt (Thr-34) Peptide
AM1141 Akt (Ser-473), phospho-specific Mouse Monoclonal

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www.ecmbiosciences.com
telephone: 859-879-2075
toll-free: 1-800-859-8202
tech: info@ecmbiosciences.com

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