Akt Phospho-Regulation
Antibody Sampler Kit

Catalog # AK6340

Kit Components:

Catalog#	Description	Host	Size	Applications	Species Reactivity	MW (kDa)
AM1141	Akt (Ser-473), phospho-specific	Mouse mAb	50 µl	WB, E, IP	H, R, M	60
AP1001	Akt (Thr-34), phospho-specific	Rabbit pAb	50 µl	WB, E	H, R, M	60
AM1011	Akt (N-terminal region)	Mouse mAb	50 µl	WB, E, ICC	H, R, M	60
MS3001	Anti-Mouse Ig:HRP	Donkey pAb	100 µl	WB, E		
RS3251	Anti-Rabbit Ig Light-Chain Specific:HRP	Mouse mAb	100 µl	WB, E		

Applications: WB = western blot, E = ELISA, ICC = Immunocytochemistry, IP = immunoprecipitation. Species: H = Human, R = Rat, M = Mouse

Kit Summary:
The Akt phospho-regulation antibody sampler kit can be used to detect Akt activation via Ser-473 phosphorylation, as well as Akt inhibition through Thr-34 phosphorylation. The kit also includes a monoclonal antibody to monitor the total Akt expression levels and secondary reagents for detection of these antibodies.

Buffers and Storage:
Mouse monoclonal Akt and rabbit polyclonal Akt (Thr-34) antibodies are supplied in 50µl phosphate-buffered saline, 50% glycerol, 1 mg/ml BSA, and 0.05% sodium azide. Mouse monoclonal Akt (Ser-473) is supplied in 50µl phosphate-buffered saline with 0.02% sodium azide. Secondary reagents are supplied in phosphate-buffered saline, 50% glycerol, and 1 mg/ml BSA. Store at –20°C. Stable for 1 year.

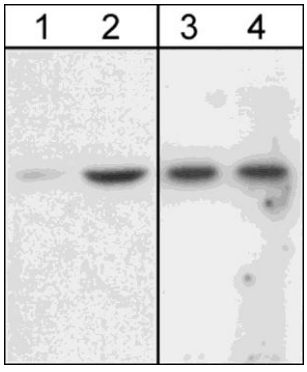
Background:
Akt (PKB, Rac kinase) is a 60kDa ser/thr kinase critical for controlling diverse cellular functions, including glucose metabolism, gene transcription, cell proliferation, and apoptosis. Akt phosphorylates a number of substrates including MBP, glycogen synthetase, PKA RII subunit, and histone 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 induces membrane translocation of Akt coincident with its phosphorylation at Thr-308 and Ser-473. Upon activation, Akt associates with members of the PKC family of kinases, such as PKCδ and PKCζ. Ceramide-activated PKCζ leads to phosphorylation of Thr-34 within the pleckstrin homology domain of Akt. This phosphorylation inhibits PIP3 binding to Akt preventing activation of the kinase and may lead to ceramide-induced cell death.

References:

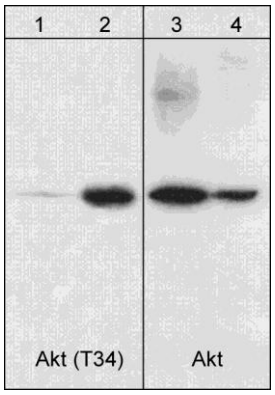
Jones, P.F. et al. (1991) Proc. Natl. Acad. Sci. USA 88:4171-4175.
Marte, B. & Downward. J. (1997) TIBS. 22:355-358.
Powell, D.J. et al. (2003) Mol. Cell Biol. 23:7794-7808.

Product References (AM1011):

Smith, M.A. et al. (2007) Am. J. Phys. Cell. Phys. 293:1947. (WB: mouse C2C12)
Moylan, J.S. et al. (2008) Am. J. Phys. Cell. Phys. 295:986. (WB: mouse C2C12)



Western blot analysis of A431 cells untreated (lanes 1 & 3) or treated with 100 ng/ml EGF for 60 min. (lanes 2 & 4). The blots were probed with monoclonal anti-Akt (Ser-473) (lanes 1 & 2) or monoclonal anti-Akt1 (N-terminal region) (lanes 3 &4).



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).

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