3050 Spruce Street, St. Louis, MO 63103 USA
Tel: (800) 521-8956 (314) 771-5765 Fax: (800) 325-5052 (314) 771-5757
email: techservice@sial.com sigma-aldrich.com

Product Information

Akt1, active, GST-tagged, human recombinant, expressed in insect cells

Catalog Number **A8729**
Storage Temperature –70 °C

Synonyms: Protein Kinase B-α, PKB-α, RAC-PKα

Product Description
Akt consists of three highly conserved isoforms, which are designated in humans as Akt1, Akt2, and Akt3. Each isoform consists of an N-terminus pleckstrin homology (PH) domain, which mediates lipid-protein or protein-protein interactions, and a C-terminus kinase catalytic domain. Although each kinase is expressed differentially in a tissue-specific manner, they respond in a similar fashion to various stimuli. Akt can be activated by a diverse array of growth factors and physiologic stimuli in a PI3-K-dependent manner. Activation of Akt kinase involves both membrane translocation and phosphorylation. Activated PI3-K generates 3′ phosphoinositide products, 3,4,5-triphosphates (PI-3,4,5-P3) and 3,4-diphosphates (PI-3,4-P2).[1]

Akt is recruited from the cytosol to the plasma membrane through the interaction of its PH domain with these phosphoinositides. Upon membrane localization, Akt undergoes a conformational change, which makes it accessible to phosphorylation at Thr^{308} and Ser^{473} in the kinase domain by PDK-1 and related kinases. Activated Akt is involved in glucose metabolism, transcription, survival, cell proliferation, angiogenesis, cell motility, and a number of metabolic effects of insulin. The effects of Akt activation may be mediated by modulation of expression or activity of various molecules including Bcl-2, BAD, caspase-9, endothelial nitric oxide synthase (eNOS), glycogen synthase, and transcription factors (NF-κB, Forkhead, CREB, and Mdm2).[2-4]

Because of its growth-promoting effects, Akt plays a central role in tumorigenesis. A number of oncogenes and tumor suppressor genes act upstream of Akt to influence cancer progression. Deletion of PTEN, a tumor suppressor gene that encodes a phosphatase, correlates with increased Akt activity in several cancers. Similarly, overexpression of active Ras, Her/Neu, or Akt genes causes hyperactivation of Akt in many cancers including pancreatic, gastric, breast, ovarian, and prostate adenocarcinomas. Small-molecule therapeutics that block PI3K signaling might inhibit cancer cells by blocking many aspects of the tumor-cell phenotype.[5-7]

The product is active recombinant, full-length human Akt1 containing an N-terminal GST-tag. It is supplied in a solution of 50 mM Tris-HCl, pH 7.5, 150 mM NaCl, 0.25 mM DTT, 0.1 mM EGTA, 0.1 mM EDTA, 0.1 mM PMSF and 25% glycerol.

Purity: ≥ 90% (SDS-PAGE)

Molecular mass: ~85 kDa

Specific Activity: ≥50 units/mg protein

Unit Definition: One unit will incorporate one nanomole of phosphate into the Akt/SGK substrate peptide (RPRAATF) per minute at 30 °C at pH 7.2 using a final concentration of 50 μM [^{32}P] ATP.

Precautions and Disclaimer
This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Preparation Instructions
For maximum product recovery, centrifuge the vial before removing the cap after thawing.

Storage/Stability
The product remains active for at least 12 months when stored at –70 °C.

After initial thawing, store in smaller, working aliquots at –70 °C. Use the working aliquots immediately upon thawing. Avoid repeated freeze-thaw cycles to prevent denaturing of the protein. Do not store in a frost-free freezer.

References

1. Vanhaesebroeck, B., and Alessi, D.R., The PI3K-PDK1 connection: more than just a road to PKB. Biochem. J., **346**, 561-576 (2000).
2. Datta, S.R. et al., Akt phosphorylation of BAD couples survival signals to the cell intrinsic death machinery. Cell, **91**, 231-241 (1997).
3. Fulton, D. et al., Regulation of endothelium-derived nitric oxide production by the protein kinase Akt. Nature, **399**, 597-601 (1999).
4. Kops, G.J. et al., Control of cell cycle exit and entry by protein kinase B-regulated forkhead transcription factors. Mol. Cell Biol., **22**, 2025-2036 (2002).
5. Sun, M. et al., Akt1/PKBα kinase is frequently elevated in human cancers and its constitutive activation is required for oncogenic transformation in NIH3T3 cells. Am. J. Pathol., **159**, 431-437 (2001).
6. Stiles, B. et al., Essential role of AKT-1/protein kinase B-α in PTEN-controlled tumorigenesis. Mol. Cell Biol., **22**, 3842-3851 (2002).
7. Vivanco, I., and Sawyers, C.L., The phosphatidylinositol 3-Akt pathway in human cancer. Nat. Rev. Cancer, **2**, 489-501 (2002).

ADM,SH,PHC,MAM 06/11-1