Recombinant AKT1 protein

Catalog No: 81145, 81845
Lot No: 11418001
Expressed In: Baculovirus

Quantity: 20, 1000 µg
Concentration: 0.4 µg/µl
Source: Human

Buffer Contents: Recombinant AKT1 protein is supplied in 25 mM HEPES-NaOH pH 7.5, 300 mM NaCl, 10% glycerol, 0.04% Triton X-100, and 0.5 mM TCEP.

Background: AKT1 (AKT Serine/Threonine Kinase 1), also known as Protein Kinase B Alpha, PRKBA or PKB Alpha, is one of 3 closely related serine/threonine-protein kinases (AKT1, AKT2 and AKT3) called the AKT kinase, and which regulate many processes including metabolism, proliferation, cell survival, growth and angiogenesis. This is mediated through serine and/or threonine phosphorylation of a range of downstream substrates. Over 100 substrate candidates have been reported so far, but for most of them, no isoform specificity has been reported. AKT is responsible of the regulation of glucose uptake and the storage of glucose. It also regulates cell survival via the phosphorylation of MAP3K5 (apoptosis signal-related kinase), mediates insulin-stimulated protein synthesis by phosphorylating TSC2. AKT is involved in the phosphorylation of members of the FOXO factors (Forkhead family of transcription factors), leading to binding of 14-3-3 proteins and cytoplasmic localization. AKT has an important role in the regulation of NF-kappa-B-dependent gene transcription and positively regulates the activity of CREB1 (cyclic AMP (cAMP)-response element binding protein). The Rho GTPase-activating protein DLC1 is another substrate and its phosphorylation is implicated in the regulation cell proliferation and cell growth. AKT plays a role as key modulator of the AKT-mTOR signaling pathway controlling the correct neuron positioning, dendritic development and synapse formation.

AKT1-specific substrates have been recently identified, including palladin (PALLD), which phosphorylation modulates cytoskeletal organization and cell motility; prohibitin (PHB), playing an important role in cell metabolism and proliferation; and CDKN1A, for which phosphorylation at Thr-145 induces its release from CDK2 and cytoplasmic relocalization. These recent findings indicate that the AKT1 isoform has a more specific role in cell motility and proliferation.

Protein Details: Recombinant AKT1 protein was expressed in a baculovirus expression system as the full length protein (accession number NP_005154.2) with an N-terminal 6A-His and FLAG tag. The molecular weight of the protein is 60.5 kDa.

Application Notes: Recombinant AKT1 protein is suitable for use in the study of enzyme kinetics, inhibitor screening, and selectivity profiling.

Kinase Activity Assay Conditions: 1 µM STK S3 substrate was incubated with different concentrations AKT1 protein in a 10 µl reaction system containing 1× Enzymatic Buffer, 5 mM MgCl2, 1 mM DTT, 5 nM SEB and 100 µM ATP for 1 hour. The 10 µl detection reagents containing TK antibody and SA-XL665, each of which was 1:100 diluted with 1× Detection Buffer were added and incubated with the reactions for 30 min. All the operations and reactions were performed at room temperature, and HTRF KinEASE STK assay was used to detect the enzymatic activity.

Storage and Guarantee: Recombinant proteins in solution are temperature sensitive and must be stored at -80°C to prevent degradation. Avoid repeated freeze/thaw cycles and keep on ice when not in storage. This product is for research use only and is not for use in diagnostic procedures. This product is guaranteed for 6 months from date of arrival.
Recombinant AKT1 protein gel
10% SDS-PAGE Coomassie staining
MW: 60.5 kDa
Purity: ≥90%

HTRF assay for recombinant AKT1 protein activity
1 µM STK S3 substrate was incubated with different concentrations AKT1 protein in a 10 µl reaction system for 1 hour. The 10 µl detection reagents were added and incubated with the reactions for 30 min. All the operations and reactions were performed at room temperature, and HTRF KinEASE STK assay was used to detect the enzymatic activity.