

Phospho-AKT1 (Ser473) Recombinant Monoclonal Antibody [4H12]

(Catalog #A74183)

Background

This gene encodes one of three closely related AKT serine-threonine protein kinases: protein kinase B alpha, beta, and gamma. These AKT proteins share a pleckstrin homology domain, a kinase domain, and a regulatory domain. They are phosphorylated by phosphoinositide 3-kinase (PI3K) and play pivotal roles in multiple signaling pathways involving membrane-bound ligands like receptor tyrosine kinases, G-protein coupled receptors, and integrin-linked kinase. AKT proteins regulate diverse cellular functions such as proliferation, survival, metabolism, and angiogenesis in normal and malignant cells. Activated by phosphatidylinositol 3,4,5-trisphosphate (PIP3), AKT proteins require phosphorylation at threonine 308 and serine 473 for full activation. They are involved in the PI3K/AKT and mTOR signaling pathways, which control translation initiation and are implicated in cancer and other diseases. Mutations in this gene are linked to various cancers and conditions like Proteus syndrome and Cowden syndrome 6. Multiple transcript variants of this gene exist due to alternative splicing.

Description

Phospho-AKT1 (Ser473) Recombinant Monoclonal Antibody [4H12]. Unconjugated. Raised in: HEK293F Cell.

Formulation

Buffer: Rabbit IgG in phosphate buffered saline, pH 7.4, 150mM NaCl, 0.02% sodium azide and 50% glycerol.

Specificity

Human

Isotype

Rabbit IgG

Uniprot ID

P31749

Purification

Affinity Chromatography

Immunogen

A synthesized peptide derived from human Phospho-AKT1 (Ser473)

Storage

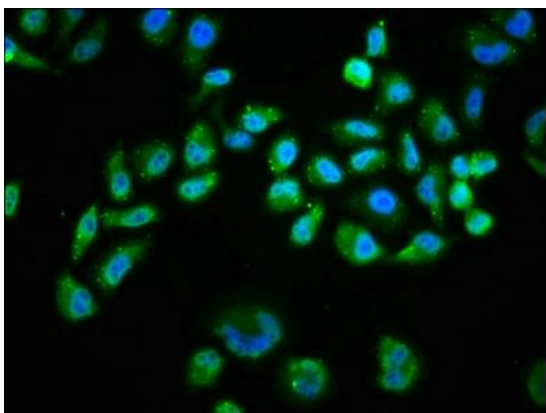
Upon receipt, store at -20°C or -80°C. Avoid repeated freeze.

Alternative Names

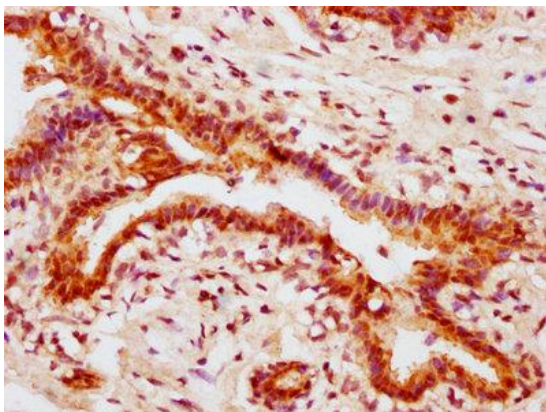
RAC-alpha serine/threonine-protein kinase, Protein kinase B, PKB, Protein kinase B alpha, PKB alpha, Proto-oncogene c-Akt, RAC-PK-alpha, AKT1, PKB, RAC

Application

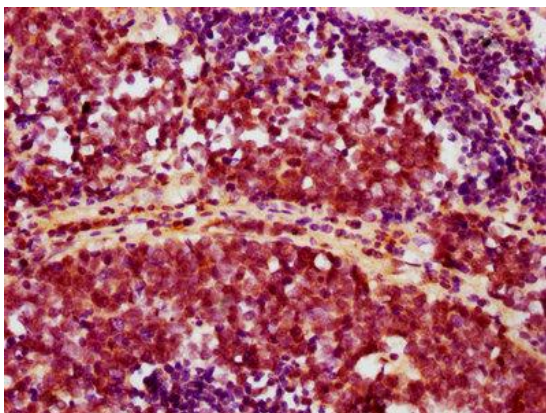
ELISA, WB, IHC, IF, IP; Recommended dilution: WB:1:500-1:5000, IHC:1:50-1:200, IF:1:20-1:200, IP:1:200-1:1000



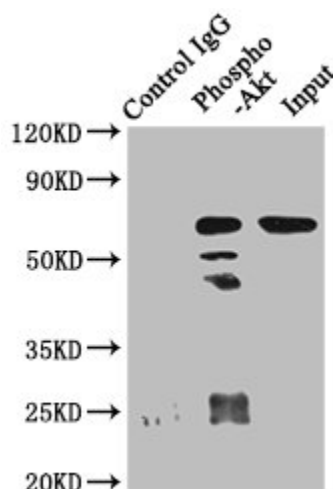
Immunofluorescence staining of HeLa cells (treated with 100mM EGF for 20min) with p-AKT1 rMAb at 1:68, counter-stained with DAPI. The cells were fixed in 4% formaldehyde, permeabilized using 0.2% Triton X-100 and blocked in 10% normal Goat Serum. The cells were then incubated with the antibody overnight at 4°C. The secondary antibody was Alexa Fluor 488-conjugated AffiniPure Goat Anti-Rabbit IgG (H+L).



IHC image of p-AKT1 rMAb diluted at 1:100 and staining in paraffin-embedded human breast cancer performed on a Leica Bond™ system. After dewaxing and hydration, antigen retrieval was mediated by high pressure in a citrate buffer (pH 6.0). Section was blocked with 10% normal goat serum 30min at RT. Then primary antibody (1% BSA) was incubated at 4°C overnight. The primary is detected by a biotinylated secondary antibody and visualized using an HRP conjugated SP system.



IHC image of p-AKT1 rMAb diluted at 1:100 and staining in paraffin-embedded human lung cancer performed on a Leica Bond™ system. After dewaxing and hydration, antigen retrieval was mediated by high pressure in a citrate buffer (pH 6.0). Section was blocked with 10% normal goat serum 30min at RT. Then primary antibody (1% BSA) was incubated at 4°C overnight. The primary is detected by a biotinylated secondary antibody and visualized using an HRP conjugated SP system.

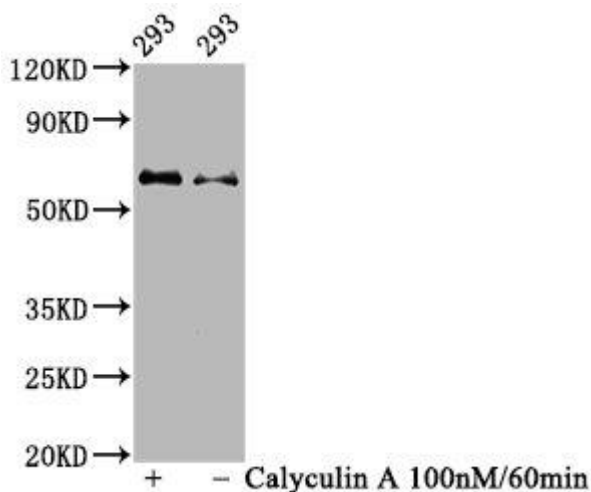


Immunoprecipitating Phospho-AKT1 in 293 whole cell lysate treated with Calyculin A

Lane 1: Rabbit control IgG (1ug) instead of p-AKT1 rMAb in 293 whole cell lysate treated with Calyculin A. For western blotting, a HRP-conjugated Protein G antibody was used as the secondary antibody (1/2000)

Lane 2: p-AKT1 rMAb (3ug) + 293 whole cell lysate treated with Calyculin A (1mg)

Lane 3: 293 whole cell lysate treated with Calyculin A (20ug)



Western Blot

Positive WB detected in 293 whole cell lysate (treated with Calyculin A or not)

All lanes Phospho-AKT1 antibody at 1.08ug/ml

Secondary

Goat polyclonal to rabbit IgG at 1/50000 dilution

Predicted band size: 60 KDa

Observed band size: 60 KDa