

# Rabbit Anti-AKT1+2+3 antibody

## SL6951R

Product Name:	AKT1+2+3
Chinese Name:	蛋白激酶AKT1,2,3抗体
Alias:	pan-AKT; pan AKT; AKT1 + AKT2 + AKT3; AKT1+AKT2+AKT3; AKT1/AKT2/AKT3; AKT 1; AKT; AKT1; AKT-1; AKT1_HUMAN; AKT 2; AKT2; AKT-2; AKT2_HUMAN; AKT 3; AKT3; AKT-3; AKT3_HUMAN; C AKT; cAKT; MGC9965; MGC99656; Oncogene AKT1; PKB; PKB alpha; PKB-ALPHA; PKB beta; PKB gamma; PRKBA; Protein Kinase B Alpha; Protein kinase B; Proto-oncogene c-Akt; RAC Alpha; RAC alpha serine/threonine protein kinase; RAC; RAC PK Alpha; Rac protein kinase alpha; RAC Serine/Threonine Protein Kinase; RAC-alpha serine/threonine-protein kinase; RAC-PK-alpha; v akt murine thymoma viral oncogene homolog 1; vAKT Murine Thymoma Viral Oncogene Homolog 1.
Organism Species:	Rabbit
Clonality:	Polyclonal
React Species:	Human, Mouse, Rat, Chicken, Dog, Pig, Cow, Rabbit, Sheep,
Applications:	WB=1:500-2000ELISA=1:500-1000IHC-P=1:400-800IHC-F=1:400-800Flow-Cyt=1µg/TestICC=1:100-500IF=1:100-500 (Paraffin sections need antigen repair) not yet tested in other applications.  optimal dilutions/concentrations should be determined by the end user.
Molecular weight:	56kDa
Cellular localization:	cytoplasmicThe cell membrane
Form:	Lyophilized or Liquid
Concentration:	1mg/ml
immunogen:	KLH conjugated synthetic peptide derived from human AKT1/2/3:401-480/480
Lsotype:	IgG
Purification:	affinity purified by Protein A
Storage Buffer:	0.01M TBS(pH7.4) with 1% BSA, 0.03% Proclin300 and 50% Glycerol.
Storage:	Store at -20 °C for one year. Avoid repeated freeze/thaw cycles. The lyophilized antibody is stable at room temperature for at least one month and for greater than a year when kept at -20 °C. When reconstituted in sterile pH 7.4 0.01M PBS or diluent of antibody the antibody is stable for at least two weeks at 2-4 °C.

P	ubMed:

#### PubMed

AKT, also known as protein kinase B (PKB), is a 57 kDa serine/threonine protein kinase. There are three mammalian isoforms of Akt: AKT1 (PKB alpha), AKT2 (PKB beta) and AKT3 (PKB gamma) with AKT2 and AKT3 being approximately 82% identical with the AKT1 isoform. Each isoform has a pleckstrin homology (PH)domain, a kinase domain and a carboxy terminal regulatory domain. AKT was originally cloned from the retrovirus AKT8, and is a key regulator of many signal transduction pathways. Its tight control over cell proliferation and cell viability are manifold; overexpression or inappropriate activation of AKT has been seen in many types of cancer. AKT mediates many of the downstream events of phosphatidylinositol 3 kinase (a lipid kinase activated by growth factors, cytokines and insulin). PI3 kinase recruits AKT to the membrane, where it is activated by PDK1 phosphorylation. Once phosphorylated, AKT dissociates from the membrane and phosphorylates targets in the cytoplasm and the cell nucleus.

#### **Function:**

IGF-1 leads to the activation of AKT3, which may play a role in regulating cell survival. Capable of phosphorylating several known proteins. Truncated isoform 2/PKB gamma 1 without the second serine phosphorylation site could still be stimulated but to a lesser extent.

#### Subunit:

Interacts (via the C-terminus) with CCDC88A (via its C-terminus). Interacts with GRB10; the interaction leads to GRB10 phosphorylation thus promoting YWHAE-binding. Interacts with AGAP2 (isoform 2/PIKE-A); the interaction occurs in the presence of guanine nucleotides. Interacts with AKTIP. Interacts (via PH domain) with MTCP1, TCL1A AND TCL1B. Interacts with CDKN1B; the interaction phosphorylates CDKN1B promoting 14-3-3 binding and cell-cycle progression. Interacts with MAP3K5 and TRAF6. Interacts with BAD, PPP2R5B, STK3 and STK4. Interacts (via PH domain) with SIRT1. Interacts with SRPK2 in a phosphorylation-dependent manner. Interacts with RAF1. Interacts with TRIM13; the interaction ubiquitinates AKT1 leading to its proteasomal degradation. Interacts with TNK2 and CLK2. Interacts (via the C-terminus) with THEM4 (via its C-terminus). Interacts with and phosphorylated by PDPK1.

### **Subcellular Location:**

Cytoplasm. Nucleus. Cell membrane. Note=Nucleus after activation by integrin-linked protein kinase 1 (ILK1). Nuclear translocation is enhanced by interaction with TCL1A. Phosphorylation on Tyr-176 by TNK2 results in its localization to the cell membrane where it is targeted for further phosphorylations on Thr-308 and Ser-473 leading to its activation and the activated form translocates to the nucleus.

#### Tissue Specificity:

Expressed in prostate cancer and levels increase from the normal to the malignant state (at protein level). Expressed in all human cell types so far analyzed. The Tyr-176 phosphorylated form shows a significant increase in expression in breast cancers during the progressive stages i.e. normal to hyperplasia (ADH), ductal carcinoma in situ

### Product Detail:

(DCIS), invasive ductal carcinoma (IDC) and lymph node metastatic (LNMM) stages.

#### Post-translational modifications:

O-GlcNAcylation at Thr-305 and Thr-312 inhibits activating phosphorylation at Thr-308 via disrupting the interaction between AKT1 and PDPK1. O-GlcNAcylation at Ser-473 also probably interferes with phosphorylation at this site.

Phosphorylation on Thr-308, Ser-473 and Tyr-474 is required for full activity. Activated TNK2 phosphorylates it on Tyr-176 resulting in its binding to the anionic plasma membrane phospholipid PA. This phosphorylated form localizes to the cell membrane, where it is targeted by PDPK1 and PDPK2 for further phosphorylations on Thr-308 and Ser-473 leading to its activation. Ser-473 phosphorylation by mTORC2 favors Thr-308 phosphorylation by PDPK1. Ser-473 phosphorylation is enhanced by interaction with AGAP2 isoform 2 (PIKE-A). Ser-473 phosphorylation is enhanced in focal cortical dysplasias with Taylor-type balloon cells. Ser-473 phosphorylation is enhanced by signaling through activated FLT3. Dephosphorylated at Thr-308 and Ser-473 by PP2A phosphatase. The phosphorylated form of PPP2R5B is required for bridging AKT1 with PP2A phosphatase.

Ubiquitinated via 'Lys-48'-linked polyubiquitination by ZNRF1, leading to its degradation by the proteasome. Ubiquitinated; undergoes both 'Lys-48'- and 'Lys-63'-linked polyubiquitination. TRAF6-induced 'Lys-63'-linked AKT1 ubiquitination is critical for phosphorylation and activation. When ubiquitinated, it translocates to the plasma membrane, where it becomes phosphorylated. When fully phosphorylated and translocated into the nucleus, undergoes 'Lys-48'-polyubiquitination catalyzed by TTC3, leading to its degradation by the proteasome. Also ubiquitinated by TRIM13 leading to its proteasomal degradation.

Acetylated on Lys-14 and Lys-20 by the histone acetyltransferases EP300 and KAT2B. Acetylation results in reduced phosphorylation and inhibition of activity. Deacetylated at Lys-14 and Lys-20 by SIRT1. SIRT1-mediated deacetylation relieves the inhibition.

#### DISEASE:

Defects in AKT1 are a cause of susceptibility to breast cancer (BC) [MIM:114480]. A common malignancy originating from breast epithelial tissue. Breast neoplasms can be distinguished by their histologic pattern. Invasive ductal carcinoma is by far the most common type.

Breast cancer is etiologically and genetically heterogeneous. Important genetic factors have been indicated by familial occurrence and bilateral involvement. Mutations at more than one locus can be involved in different families or even in the same case. Defects in AKT1 are associated with colorectal cancer (CRC) [MIM:114500]. Note=Genetic variations in AKT1 may play a role in susceptibility to ovarian cancer. Defects in AKT1 are a cause of Proteus syndrome (PROTEUSS) [MIM:176920]. A highly variable, severe disorder of asymmetric and disproportionate overgrowth of body parts, connective tissue nevi, epidermal nevi, dysregulated adipose tissue, and vascular malformations. Many features of Proteus syndrome overlap with other overgrowth syndromes.

#### Similarity:

Belongs to the protein kinase superfamily. AGC Ser/Thr protein kinase family.

RAC subfamily.

Contains 1 AGC-kinase C-terminal domain.

Contains 1 PH domain.

Contains 1 protein kinase domain.

**SWISS:** P31749

Gene ID:

207

Database links:

Entrez Gene: 207 Human

Entrez Gene: 11651 Mouse

Entrez Gene: 24185 Rat

Omim: 164730 Human

SwissProt: O57513 Chicken

SwissProt: P31749 Human

SwissProt: P31750 Mouse

SwissProt: P47196 Rat

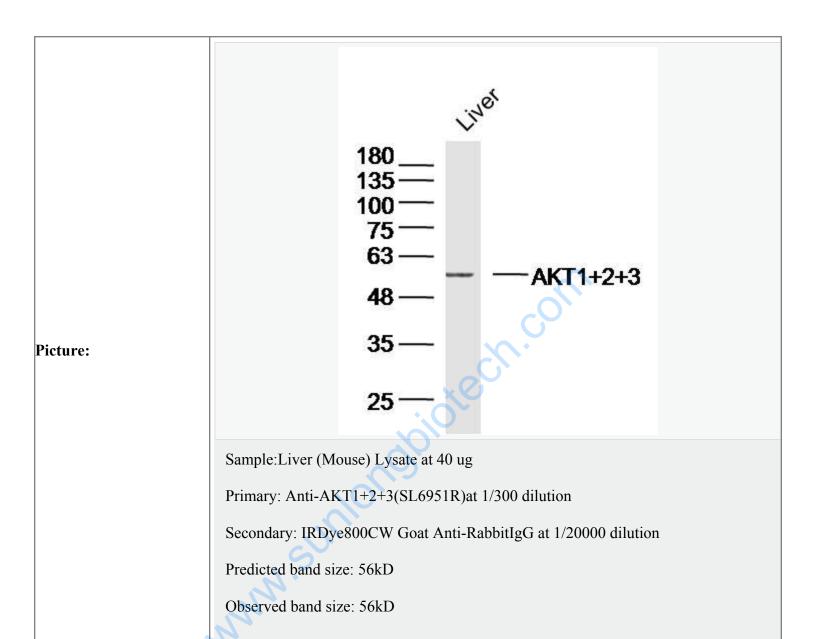
Unigene: 525622 Human

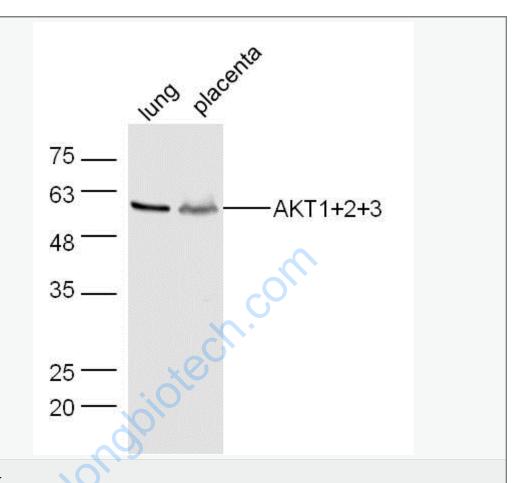
Unigene: 6645 Mouse

Unigene: 11422 Rat

### Important Note:

This product as supplied is intended for research use only, not for use in human, therapeutic or diagnostic applications.





# Sample:

Lung (Mouse) Lysate at 40 ug

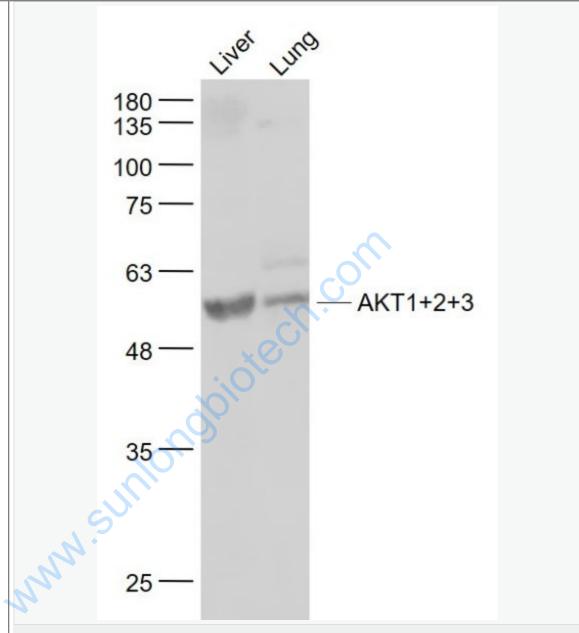
Placenta (Mouse) Lysate at 40 ug

Primary: Anti-AKT1+2+3 (SL6951R) at 1/300 dilution

Secondary: IRDye800CW Goat Anti-Rabbit IgG at 1/20000 dilution

Predicted band size: 56 kD

Observed band size: 56 kD



### Sample:

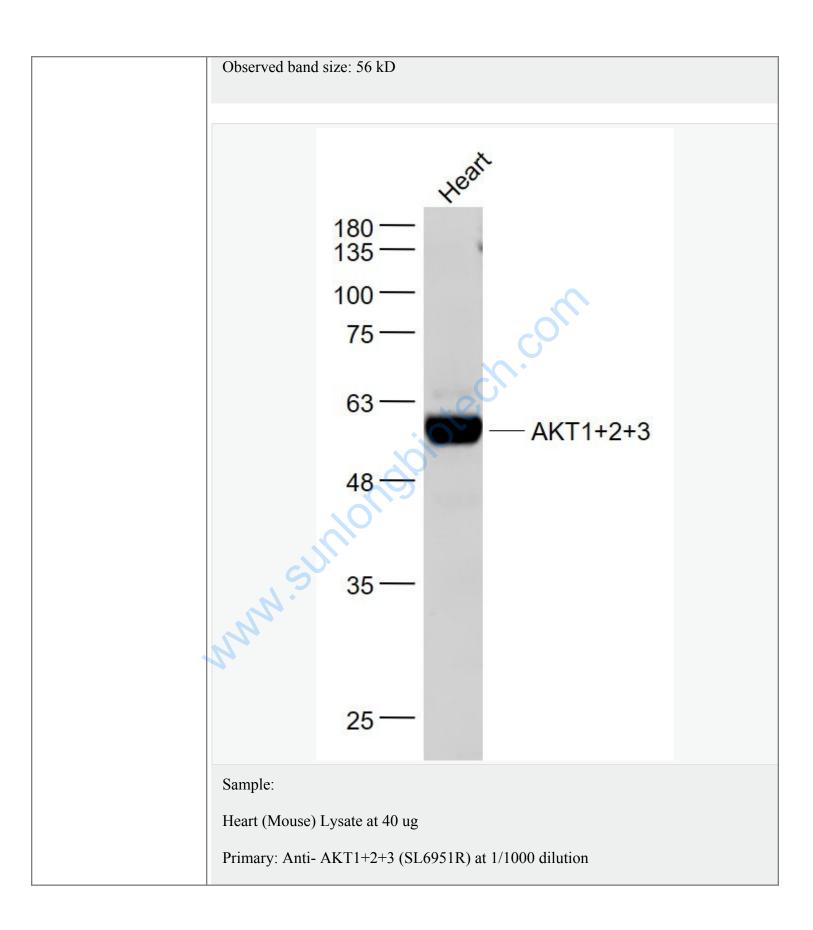
Liver (Mouse) Lysate at 40 ug

Lung (Mouse) Lysate at 40 ug

Primary: Anti- AKT1+2+3 (SL6951R) at 1/1000 dilution

Secondary: IRDye800CW Goat Anti-Rabbit IgG at 1/20000 dilution

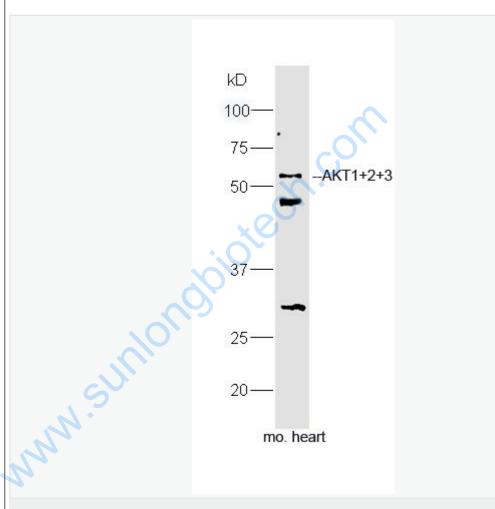
Predicted band size: 56 kD



Secondary: IRDye800CW Goat Anti-Rabbit IgG at 1/20000 dilution

Predicted band size: 56 kD

Observed band size: 56 kD



Sample:

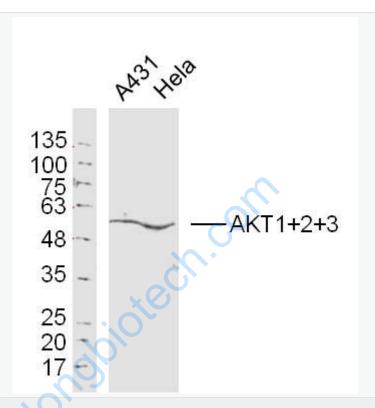
heart (Mouse) Lysate at 40 ug

Primary: Anti-AKT1+2+3 (SL6951R) at 1/300 dilution

Secondary: IRDye800CW Goat Anti-Rabbit IgG at 1/20000 dilution

Predicted band size: 56 kD

Observed band size: 52 kD



# Sample:

A431Cell (Human) Lysate at 30 ug

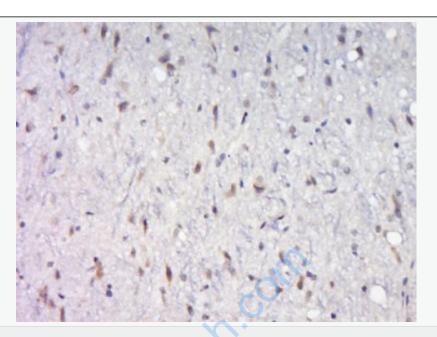
Hela Cell(Human)Lysate at 30 ug

Primary: Anti-AKT1+2+3(SL6951R)at 1/300 dilution

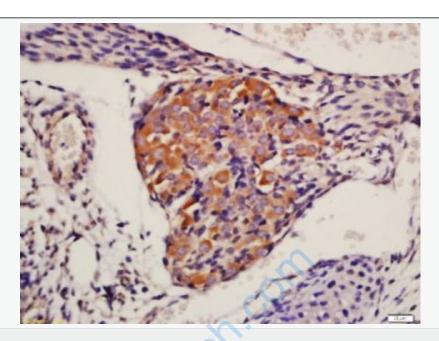
Secondary: IRDye800CW Goat Anti-Rabbit IgG at 1/20000 dilution

Predicted band size: 50 kD

Observed band size: 50 kD



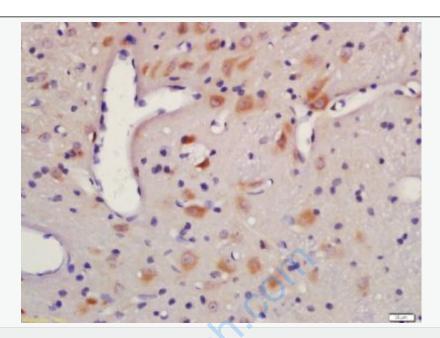
Paraformaldehyde-fixed, paraffin embedded (rat brain); Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15min; Block endogenous peroxidase by 3% hydrogen peroxide for 20 minutes; Blocking buffer (normal goat serum) at 37°C for 30min; Antibody incubation with (AKT1+2+3) Polyclonal Antibody, Unconjugated (SL6951R) at 1:400 overnight at 4°C, followed by a conjugated secondary (sp-0023) for 20 minutes and DAB staining.



Tissue/cell: mouse embryo tissue; 4% Paraformaldehyde-fixed and paraffinembedded;

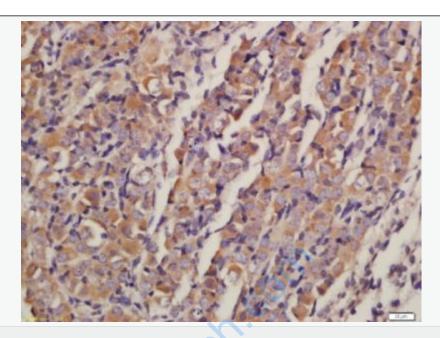
Antigen retrieval: citrate buffer (0.01M, pH 6.0), Boiling bathing for 15min; Block endogenous peroxidase by 3% Hydrogen peroxide for 30min; Blocking buffer (normal goat serum, C-0005) at 37°C for 20 min;

Incubation: Anti-AKT1+2+3 Polyclonal Antibody, Unconjugated(SL6951R) 1:200, overnight at 4°C, followed by conjugation to the secondary antibody(SP-0023) and DAB(C-0010) staining



Tissue/cell: rat brain tissue; 4% Paraformaldehyde-fixed and paraffin-embedded; Antigen retrieval: citrate buffer ( 0.01M, pH 6.0 ), Boiling bathing for 15min; Block endogenous peroxidase by 3% Hydrogen peroxide for 30min; Blocking buffer (normal goat serum,C-0005) at 37°C for 20 min;

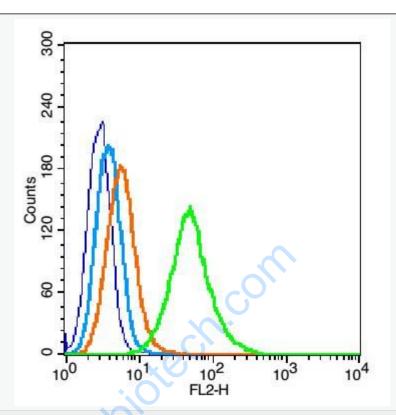
Incubation: Anti-AKT1+2+3 Polyclonal Antibody, Unconjugated(SL6951R) 1:200, overnight at 4°C, followed by conjugation to the secondary antibody(SP-0023) and DAB(C-0010) staining



Tissue/cell: Mouse embryos tissue; 4% Paraformaldehyde-fixed and paraffinembedded;

Antigen retrieval: citrate buffer (0.01M, pH 6.0), Boiling bathing for 15min; Block endogenous peroxidase by 3% Hydrogen peroxide for 30min; Blocking buffer (normal goat serum, C-0005) at 37°C for 20 min;

Incubation: Anti-AKT1 + 2 + 3 Polyclonal Antibody, Unconjugated(SL6951R) 1:200, overnight at 4°C, followed by conjugation to the secondary antibody(SP-0023) and DAB(C-0010) staining



Blank control(blue): TM4 cells(fixed with 2% paraformaldehyde (10 min), then permeabilized with 90% ice-cold methanol for 30 min on ice).

Primary Antibody:Rabbit Anti-AKT1+2+3 antibody(SL6951R), Dilution: 1 $\mu$ g in 100  $\mu$ L 1X PBS containing 0.5% BSA;

Isotype Control Antibody: Rabbit IgG(orange) ,used under the same conditions ); Secondary Antibody: Goat anti-rabbit IgG-PE(white blue), Dilution: 1:200 in 1 X PBS containing 0.5% BSA.