abcam

ab185633

# Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671]

Recombinant | RabMAb | 20ul selling size

## Key facts

| | |
|---|---|
| **Isotype** | IgG |
| **Host species** | Rabbit |
| **Storage buffer** | pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA |
| **Form** | Liquid |
| **Clonality** | Monoclonal |
| **Immunogen** | The exact immunogen used to generate this antibody is proprietary information. |
| **Clone number** | EPR17671 |
| **Purification technique** | Affinity purification Protein A |
| **Concentration** | 0.24 mg/mL The concentration of this product may be batch-dependent<br>Batch concentration finder → |

## Reactivity data

## IP

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/50 |
| **Notes** | - |

### Expected

| | |
|---|---|
| **Species** | Mouse, Rat |
| **Dilution info** | Use at an assay dependent concentration. |
| **Notes** | - |

### Not recommended

| | |
|---|---|
| **Species** | Recombinant full length protein - Human |
| **Dilution info** | - |
| **Notes** | - |

## WB

### Tested

| | |
|---|---|
| **Species** | Mouse |
| **Dilution info** | 1/2000 |
| **Notes** | - |

| | |
|---|---|
| **Species** | Rat |
| **Dilution info** | 1/2000 |
| **Notes** | - |

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/2000 |
| **Notes** | - |

| | |
|---|---|
| **Species** | Recombinant full length protein - Human |
| **Dilution info** | 1/2000 |
| **Notes** | - |

## Flow Cyt (Intra)

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/50 |

| | |
|---|---|
| **Notes** | ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody. |

### Expected

| | |
|---|---|
| **Species** | Mouse, Rat |
| **Dilution info** | Use at an assay dependent concentration. |
| **Notes** | - |

### Not recommended

| | |
|---|---|
| **Species** | Recombinant full length protein - Human |
| **Dilution info** | - |
| **Notes** | - |

## IHC-P

### Tested

| | |
|---|---|
| **Species** | Mouse |
| **Dilution info** | 1/400 |
| **Notes** | Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. |

| | |
|---|---|
| **Species** | Rat |
| **Dilution info** | 1/400 |
| **Notes** | Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. |

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/400 |
| **Notes** | Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. |

### Not recommended

| | |
|---|---|
| **Species** | Recombinant full length protein - Human |

| | |
|---|---|
| **Dilution info** | - |
| **Notes** | - |

## Target data

See full target information AKT1

**Function**

AKT1 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 (PubMed:11882383, PubMed:15526160, PubMed:15861136, PubMed:21432781, PubMed:21620960, PubMed:31204173). This is mediated through serine and/or threonine phosphorylation of a range of downstream substrates (PubMed:11882383, PubMed:15526160, PubMed:21432781, PubMed:21620960, PubMed:29343641, PubMed:31204173). Over 100 substrate candidates have been reported so far, but for most of them, no isoform specificity has been reported (PubMed:11882383, PubMed:15526160, PubMed:21432781, PubMed:21620960). AKT is responsible of the regulation of glucose uptake by mediating insulin-induced translocation of the SLC2A4/GLUT4 glucose transporter to the cell surface (By similarity). Phosphorylation of PTPN1 at 'Ser-50' negatively modulates its phosphatase activity preventing dephosphorylation of the insulin receptor and the attenuation of insulin signaling (By similarity). Phosphorylation of TBC1D4 triggers the binding of this effector to inhibitory 14-3-3 proteins, which is required for insulin-stimulated glucose transport (PubMed:11994271). AKT also regulates the storage of glucose in the form of glycogen by phosphorylating GSK3A at 'Ser-21' and GSK3B at 'Ser-9', resulting in inhibition of its kinase activity (By similarity). Phosphorylation of GSK3 isoforms by AKT is also thought to be one mechanism by which cell proliferation is driven (By similarity). AKT also regulates cell survival via the phosphorylation of MAP3K5 (apoptosis signal-related kinase) (PubMed:11154276). Phosphorylation of 'Ser-83' decreases MAP3K5 kinase activity stimulated by oxidative stress and thereby prevents apoptosis (PubMed:11154276). AKT mediates insulin-stimulated protein synthesis by phosphorylating TSC2 at 'Ser-939' and 'Thr-1462', thereby activating the mTORC1 signaling pathway, and leading to both phosphorylation of 4E-BP1 and in activation of RPS6KB1 (PubMed:12150915, PubMed:12172553). Also regulates the mTORC1 signaling pathway by catalyzing phosphorylation of CASTOR1 and DEPDC5 (PubMed:31548394, PubMed:33594058). AKT plays a role as key modulator of the AKT-mTOR signaling pathway controlling the tempo of the process of newborn neurons integration during adult neurogenesis, including correct neuron positioning, dendritic development and synapse formation (By similarity). Part of a positive feedback loop of mTORC2 signaling by mediating phosphorylation of MAPKAP1/SIN1, promoting mTORC2 activation (By similarity). 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 (PubMed:10358075). In particular, FOXO1 is phosphorylated at 'Thr-24', 'Ser-256' and 'Ser-319' (PubMed:10358075). FOXO3 and FOXO4 are phosphorylated on equivalent sites (PubMed:10358075). 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) (PubMed:9829964). The phosphorylation of CREB1 induces the binding of accessory proteins that are necessary for the transcription of pro-survival genes

such as BCL2 and MCL1 (PubMed:9829964). AKT phosphorylates 'Ser-454' on ATP citrate lyase (ACLY), thereby potentially regulating ACLY activity and fatty acid synthesis (By similarity). Activates the 3B isoform of cyclic nucleotide phosphodiesterase (PDE3B) via phosphorylation of 'Ser-273', resulting in reduced cyclic AMP levels and inhibition of lipolysis (By similarity). Phosphorylates PIKFYVE on 'Ser-318', which results in increased PI(3)P-5 activity (By similarity). The Rho GTPase-activating protein DLC1 is another substrate and its phosphorylation is implicated in the regulation cell proliferation and cell growth (By similarity). Signals downstream of phosphatidylinositol 3-kinase (PI(3)K) to mediate the effects of various growth factors such as platelet-derived growth factor (PDGF), epidermal growth factor (EGF), insulin and insulin-like growth factor 1 (IGF1) (PubMed:12176338, PubMed:12964941). AKT mediates the antiapoptotic effects of IGF1 (By similarity). Essential for the SPATA13-mediated regulation of cell migration and adhesion assembly and disassembly (PubMed:19934221). May be involved in the regulation of the placental development (By similarity). Phosphorylates STK4/MST1 at 'Thr-120' and 'Thr-387' leading to inhibition of its: kinase activity, nuclear translocation, autophosphorylation and ability to phosphorylate FOXO3 (PubMed:17726016). Phosphorylates STK3/MST2 at 'Thr-117' and 'Thr-384' leading to inhibition of its: cleavage, kinase activity, autophosphorylation at Thr-180, binding to RASSF1 and nuclear translocation (PubMed:20086174). Phosphorylates SRPK2 and enhances its kinase activity towards SRSF2 and ACIN1 and promotes its nuclear translocation (PubMed:19592491). Phosphorylates RAF1 at 'Ser-259' and negatively regulates its activity (PubMed:10576742). Phosphorylation of BAD stimulates its pro-apoptotic activity (PubMed:10926925). Phosphorylates KAT6A at 'Thr-369' and this phosphorylation inhibits the interaction of KAT6A with PML and negatively regulates its acetylation activity towards p53/TP53 (PubMed:23431171). Phosphorylates palladin (PALLD), modulating cytoskeletal organization and cell motility (PubMed:20471940). Phosphorylates prohibitin (PHB), playing an important role in cell metabolism and proliferation (PubMed:18507042). Phosphorylates CDKN1A, for which phosphorylation at 'Thr-145' induces its release from CDK2 and cytoplasmic relocalization (PubMed:16982699). These recent findings indicate that the AKT1 isoform has a more specific role in cell motility and proliferation (PubMed:16139227). Phosphorylates CLK2 thereby controlling cell survival to ionizing radiation (PubMed:20682768). Phosphorylates PCK1 at 'Ser-90', reducing the binding affinity of PCK1 to oxaloacetate and changing PCK1 into an atypical protein kinase activity using GTP as donor (PubMed:32322062). Also acts as an activator of TMEM175 potassium channel activity in response to growth factors: forms the lysoK(GF) complex together with TMEM175 and acts by promoting TMEM175 channel activation, independently of its protein kinase activity (PubMed:32228865). Acts as a regulator of mitochondrial calcium uptake by mediating phosphorylation of MICU1 in the mitochondrial intermembrane space, impairing MICU1 maturation (PubMed:30504268). Acts as an inhibitor of tRNA methylation by mediating phosphorylation of the N-terminus of METTL1, thereby inhibiting METTL1 methyltransferase activity (PubMed:15861136). In response to LPAR1 receptor pathway activation, phosphorylates Rabin8/RAB3IP which alters its activity and phosphorylates WDR44 which induces WDR44 binding to Rab11, thereby switching Rab11 vesicular function from preciliary trafficking to endocytic recycling (PubMed:31204173).

## Storage

| | |
|---|---|
| **Shipped at conditions** | Conditional Ambient |

| | |
|---|---|
| **Appropriate short-term storage duration** | 1-2 weeks |
| **Appropriate short-term storage conditions** | +4°C |
| **Appropriate long-term storage conditions** | -20°C |
| **Aliquoting information** | Upon delivery aliquot |
| **Storage information** | Avoid freeze / thaw cycle |

## Notes

**Patented technology**
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

**What are the advantages of a recombinant monoclonal antibody?**
This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity
- Long-term security of supply
- Animal-free batch production

For more information, read more on recombinant antibodies.

## Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

| | |
|---|---|
| **Activity summary** | AKT1 AKT2 and AKT3 also known as Protein Kinase B (PKB) isoforms are serine/threonine-specific protein kinases with critical roles in cellular processes. AKT1 has a molecular weight of about 55.8 kDa AKT2 weighs approximately 56.1 kDa and AKT3 typically has a similar mass. These proteins are expressed in many tissues including brain and heart with AKT1 ubiquitously present AKT2 focused in insulin-responsive tissues and AKT3 mainly in the brain. The molecular weight of AKT plays an important role in their functionality and specificity in tissues. |
| **Biological function summary** | AKT proteins regulate cell cycle growing cell survival proliferation and metabolism. They participate as core components of the PI3K/AKT/mTOR signaling pathway forming complexes with other proteins to transmit signals. They bind to phosphoinositide lipids on the cell membrane facilitating their activation and downstream signaling. Through these activities the AKT transporter proteins maintain cellular homeostasis and play a part in stress response. |
| **Pathways** | AKT proteins engage in important signaling networks including the PI3K/AKT pathway and mTOR pathway. They work closely with PI3K and mTOR proteins |

| | |
|---|---|
| | coordinating cellular growth and energy metabolism. In particular the AKT pathway responds to growth factors and insulin influencing glucose uptake and glycolysis regulation through interaction with proteins such as glycogen synthase kinase 3 (GSK3). |
| **Associated diseases and disorders** | Dysregulation of AKT signaling can lead to cancer and diabetes. High AKT activation correlates with various cancers by promoting cell survival and growth. In diabetes impaired AKT2 regulation disrupts glucose uptake affecting insulin response. AKT's relationship with mTOR is significant as it often influences tumor growth and progression in cancerous tissues. |

## Product promise

**Tested**

We have tested this species and application combination and it works. It is covered by our product promise.

**Expected**

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

**Predicted**

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

**Not recommended**

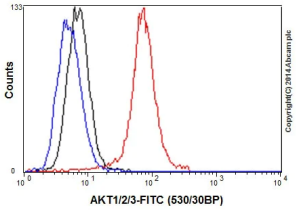
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

## 10 product images

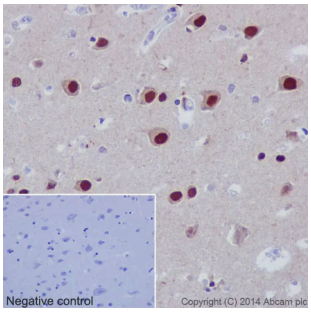



### Flow Cytometry (Intracellular) - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] (ab185633)

AKT1 + AKT2 + AKT3 Flow Cytometry (Intracellular) staining using rabbit Anti-AKT1 + AKT2 + AKT3 antibody

Intracellular flow cytometric analysis of 2% paraformaldehyde-fixedA549 (Human lung carcinoma) cells labeling AKT1 + AKT2 + AKT3 with ab185633 at 1/50 dilution (red) compared with a rabbit monoclonal IgG isotype control (black) and an unlabelled control (cells without incubation with

primary antibody and secondary antibody; blue). Goat anti rabbit IgG (FITC) at 1/150 dilution was used as the secondary antibody.

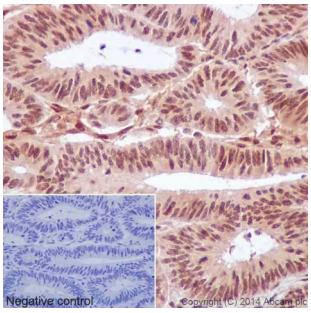



## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] (ab185633)

AKT1 + AKT2 + AKT3 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-AKT1 + AKT2 + AKT3 antibody

Immunohistochemical analysis of paraffin-embedded Human cerebral cortex tissue labeling AKT1 + AKT2 + AKT3 with ab185633 at 1/400 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution. Nuclear and weak cytoplasmic staining on neurons of the Human cerebral cortex is observed. Counter stained with Hematoxylin.
Negative control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

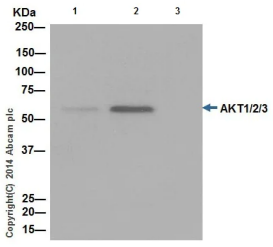



## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] (ab185633)

AKT1 + AKT2 + AKT3 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-AKT1 + AKT2 + AKT3 antibody

Immunohistochemical analysis of paraffin-embedded Human adenocarcinoma of colon tissue labeling AKT1 + AKT2 + AKT3 with ab185633 at 1/400 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution. Nuclear and weak cytoplasmic staining on Human adenocarcinoma of colon is observed. Counter stained with Hematoxylin.
Negative control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

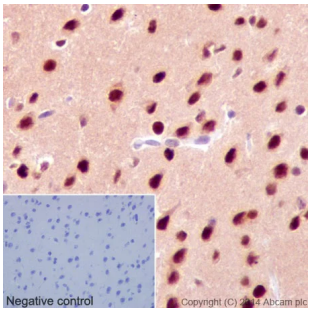



## Immunoprecipitation - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] (ab185633)

AKT1 + AKT2 + AKT3 was immunoprecipitated from 1mg of A549 (Human lung carcinoma) whole cell extract with ab185633 at 1/50 dilution. Western blot was performed from the immunoprecipitate using ab185633 at 1/1000 dilution. Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG, was used as secondary antibody at 1/1500 dilution.
Lane 1: A549 whole cell extract 10 µg (Input). Lane 2: ab185633 IP in A549 whole cell extract. Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab185633 in A549 whole cell extract.
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

All lanes:
Immunoprecipitation - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] (ab185633)

Predicted band size: 56 kDa

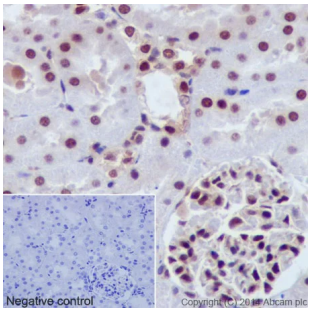



## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] (ab185633)

AKT1 + AKT2 + AKT3 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-AKT1 + AKT2 + AKT3 antibody

Immunohistochemical analysis of paraffin-embedded Mouse cerebral cortex tissue labeling AKT1 + AKT2 + AKT3 with ab185633 at 1/400 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution. Nuclear and weak cytoplasmic staining on neurons of the mouse cerebral cortex is observed. Counter stained with Hematoxylin.
Negative control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

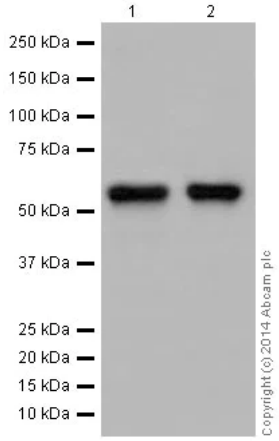



## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] (ab185633)

AKT1 + AKT2 + AKT3 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-AKT1 + AKT2 + AKT3 antibody

Immunohistochemical analysis of paraffin-embedded Rat kidney tissue labeling AKT1 + AKT2 + AKT3 with ab185633 at 1/400 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution. Nuclear and weak cytoplasmic staining on rat kidney is observed. Counter stained with Hematoxylin.
Negative control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.




## Western blot - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] (ab185633)

Blocking/Dilution buffer: 5% NFDM/TBST.
All lanes:
Western blot - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] (ab185633) at 1/2000 dilution

Lane 1:
Human fetal brain lysates at 10 µg

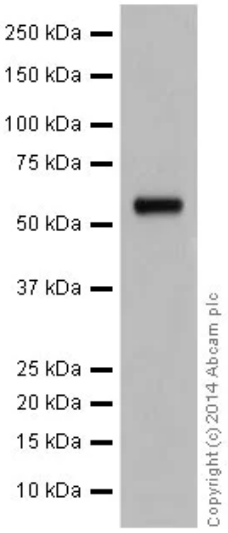
Lane 2:
Human fetal kidney lysates at 10 µg

Secondary

All lanes:
Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/1000 dilution

Predicted band size: 56 kDa

Observed band size: 56 kDa




## Western blot - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] (ab185633)

Blocking/Dilution buffer: 5% NFDM/TBST.
All lanes:
Western blot - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] (ab185633) at 1/2000 dilution

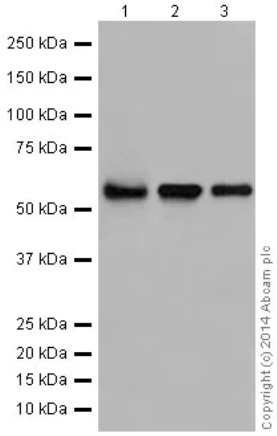
All lanes:
A549 (Human lung carcinoma) whole cell lysates at 20 µg

Secondary

All lanes:
Goat Anti-Rabbit IgG, (H+L),Peroxidase conjugated at 1/1000 dilution

Predicted band size: 141 kDa, 56 kDa

Observed band size: 141 kDa, 56 kDa




## Western blot - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] (ab185633)

AKT1 + AKT2 + AKT3 Western blot staining using rabbit Anti-AKT1 + AKT2 + AKT3 antibody

Blocking/Dilution buffer: 5% NFDM/TBST.
All lanes:
Western blot - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] (ab185633) at 1/2000 dilution

Lane 1:
Mouse brain lysates at 10 µg

Lane 2:
Rat brain lysates at 10 µg

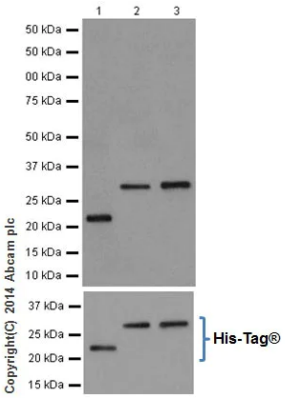
Lane 3:
Rat heart lysates at 10 µg

Secondary

All lanes:
Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/1000 dilution

Predicted band size: 56 kDa

Observed band size: 56 kDa




# Western blot - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] (ab185633)

Blocking/Dilution buffer: 5% NFDM/TBST.
The bottom panel is probed with an anti-His tag antibody.

All lanes:
Western blot - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] (ab185633) at 1/5000 dilution

Lane 1:
AKT3 recombinant protein fragment (His-Tag®): aa351-479 at 0.01 µg

Lane 2:
AKT2 recombinant protein fragment (His-Tag®): aa282-481 at 0.01 µg

Lane 3:
AKT1 recombinant protein fragment (His-Tag®): aa281-480 at 0.01 µg

Secondary

All lanes:
Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/1000 dilution

Predicted band size: 56 kDa

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.