abcam

Product datasheet

Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] - BSA and Azide free ab214166

Recombinant

RabMAb

7 Images

Overview

Product name Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] - BSA and Azide free

Description Rabbit monoclonal [EPR17671] to AKT1 + AKT2 + AKT3 - BSA and Azide free

Host species Rabbit

Tested applications Suitable for: Flow Cyt (Intra), IP, IHC-P, WB

Species reactivity Reacts with: Mouse, Rat, Human

Immunogen Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: AKT3 recombinant protein fragment (His-Tag®): aa351-479; AKT2 recombinant protein

fragment (His-Tag®): aa282-481; AKT1 recombinant protein fragment (His-Tag®): aa281-480; A549 whole cell lysate; Human fetal brain and fetal kidney lysates; Mouse brain lysate; Rat brain and heart lysates. IHC-P: Human cerebral cortex, Human adenocarcinoma of colon, mouse cerebral cortex and rat kidney tissues. ICC/IF: HeLa cells. Flow Cyt (intra): A549 cells. IP: A549

whole cell extract.

General notes ab214166 is the carrier-free version of **ab185633**.

Our <u>carrier-free</u> antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for

increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our **conjugation kits** for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

This product is compatible with the Maxpar[®] Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar[®] is a trademark of Fluidigm Canada Inc.

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 production

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For more information see here.

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

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C. Do Not Freeze.

Storage buffer pH: 7.2

Constituent: PBS

Carrier free Yes

Purity Protein A purified

ClonalityMonoclonalClone numberEPR17671

Isotype IgG

Applications

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab214166 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		Use at an assay dependent concentration. ab199376 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
IP		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB		Use at an assay dependent concentration. Detects a band of approximately 56 kDa (predicted molecular weight: 56 kDa).

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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.

Tissue specificity In adult tissues, it is highly expressed in brain, lung and kidney, but weakly in heart, testis and liver.

In fetal tissues, it is highly expressed in heart, liver and brain and not at all in kidney.

Sequence similarities 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.

DomainBinding of the PH domain to the phosphatidylinositol 3-kinase alpha (Pl(3)K) results in its

targeting to the plasma membrane.

Post-translational Phosphorylation on Thr-305 and Ser-472 is required for full activity (By similarity). Phosphorylated

upon DNA damage, probably by ATM or ATR.

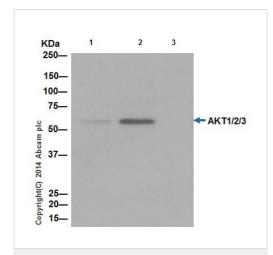
Ubiquitinated. When fully phosphorylated and translocated into the nucleus, undergoes 'Lys-48'-

polyubiquitination catalyzed by TTC3, leading to its degradation by the proteasome.

Cellular localization Cytoplasm. Membrane. Membrane-associated after cell stimulation leading to its translocation.

Images

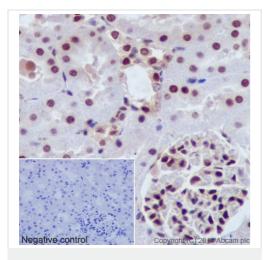
modifications



Immunoprecipitation - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] - BSA and Azide free (ab214166) 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 lgG (HRP), specific to the non-reduced form of lgG, was used as secondary antibody at 1/1500 dilution.

Lane 1: A549 whole cell extract 10 µg (Input). Lane 2: <u>ab185633</u> IP in A549 whole cell extract. Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of <u>ab185633</u> in A549 whole cell extract. Blocking and dilution buffer and concentration: 5% NFDM/TBST.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab185633</u>).



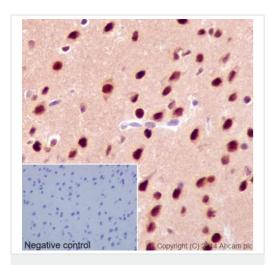
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] - BSA and Azide free (ab214166)

Immunohistochemical analysis of paraffin-embedded Rat kidney tissue labeling AKT1 + AKT2 + AKT3 with <u>ab185633</u> at 1/400 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) 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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab185633).

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



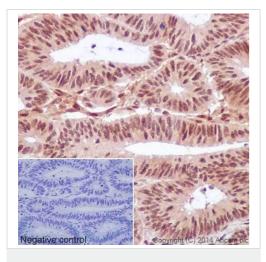
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] - BSA and Azide free (ab214166)

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 lgG H&L (HRP) (ab97051) at 1/500 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab185633).

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



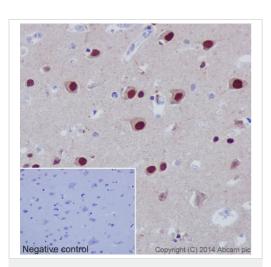
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] - BSA and Azide free (ab214166)

Immunohistochemical analysis of paraffin-embedded
Human adenocarcinoma of colon tissue labeling AKT1 + AKT2 +
AKT3 with <u>ab185633</u> at 1/400 dilution, followed by Goat AntiRabbit lgG H&L (HRP) (<u>ab97051</u>) 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 lgG H&L (HRP) (<u>ab97051</u>) at 1/500 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab185633</u>).

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



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] - BSA and Azide free (ab214166)

Immunohistochemical analysis of paraffin-embedded
Human cerebral cortex tissue labeling AKT1 + AKT2 + AKT3 with

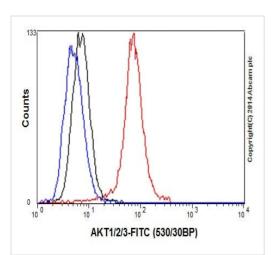
ab185633 at 1/400 dilution, followed by Goat Anti-Rabbit IgG H&L

(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 lgG H&L (HRP) (ab97051) at 1/500 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab185633).

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



Flow Cytometry (Intracellular) - Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] - BSA and Azide free (ab214166)

Intracellular flow cytometric analysis of 2% paraformaldehyde-fixedA549 (Human lung carcinoma) cells labeling AKT1 + AKT2 + AKT3 with <u>ab185633</u> at 1/50 dilution (red) compared with a rabbit monoclonal lgG isotype control (black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti rabbit lgG (FITC) at 1/150 dilution was used as the secondary antibody.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab185633</u>).



Anti-AKT1 + AKT2 + AKT3 antibody [EPR17671] - BSA and Azide free (ab214166)

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