abcam

Product datasheet

Anti-AKT2 antibody [4H7] ab175354

16 References 画像数9

製品の概要

製品名 Anti-AKT2 antibody [4H7]

製品の詳細 Mouse monoclonal [4H7] to AKT2

由来種 Mouse

アプリケーション 適用あり: ICC/IF, ChIP, IP, IHC-P, WB

種交差性 交差種: Mouse, Rat, Human, African green monkey

交差が予測される動物種: Non human primates 4

免疫原 Recombinant full length protein corresponding to Human AKT2 aa 1-481. (NP 001617) produced

in HEK293T cell.

Sequence:

MNEVSVIKEGWLHKRGEYIKTWRPRYFLLKSDGSFIGYKERP

EAPDQTLP

PLNNFSVAECQLMKTERPRPNTFVIRCLQWTTVIERTFHVDS

PDEREEWM

RAIQMVANSLKQRAPGEDPMDYKCGSPSDSSTTEEMEVAVSK

ARAKVTMN

DFDYLKLLGKGTFGKVILVREKATGRYYAMKILRKEVIIAKD

EVAHTVTE

SRVLQNTRHPFLTALKYAFQTHDRLCFVMEYANGGELFFHLS

RERVFTEE

RARFYGAEIVSALEYLHSRDVVYRDIKLENLMLDKDGHIKIT

DFGLCKEG

ISDGATMKTFCGTPEYLAPEVLEDNDYGRAVDWWGLGVVMYE

MMCGRL

PFYNQDHERLFELILMEEIRFPRTLSPEAKSLLAGLLKKDPK

ORL GGGPS

DAKEVMEHRFFLSINWQDVVQKKLLPPFKPQVTSEVDTRYFD DEFTAQSI TITPPDRYDSLGLLELDQRTHFPQFSYSASIRE

Database link: P31751

Run BLAST with Run BLAST with

ポジティブ・コントロール MCF7, HeLa, HepG2, A549, 293T, Jurkat, A431, U2OS, COS7, 3T3 L1 and NRK whole celll

lysates; AKT2 transfected U2OS cells; Human Medulla Oblongata tissue; Human Esophageal

cancer tissue.

特記事項 The Life Science industry has been in the grips of a reproducibility crisis for a number of years.

Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

製品の特性

製品の状態 Liquid

保存方法 Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long

term. Avoid freeze / thaw cycle.

パッファー Preservative: 0.05% Sodium azide

Constituents: 0.1% BSA, 69% PBS, 30% Glycerol (glycerin, glycerine)

精製度 Protein A purified

ポリ/モノ モノクローナル

ウローン名 4H7 **アイソタイプ** lgG1

アプリケーション

The Abpromise guarantee <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab175354の使用に適用されます アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
ICC/IF		1/10 - 1/100.
ChIP		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration. Use 2µg.
IHC-P		1/200. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
WB		1/1000. Predicted molecular weight: 55 kDa.

ターゲット情報

機能 General protein kinase capable of phosphorylating several known proteins.

組織特異性 Expressed in all human cell types so far analyzed.

配列類似性 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.

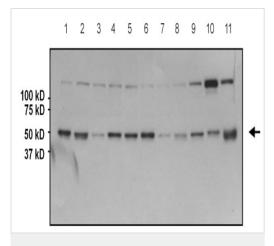
Contains 1 protein kinase domain.

翻訳後修飾

Phosphorylation on Thr-309 and Ser-474 is required for full activity.

Ubiquitinated; undergoes both 'Lys-48'- and 'Lys-63'-linked polyubiquitination. TRAF6-induced 'Lys-63'-linked AKT2 ubiquitination. When fully phosphorylated and translocated into the nucleus, undergoes 'Lys-48'-polyubiquitination catalyzed by TTC3, leading to its degradation by the proteasome.

画像



Western blot - Anti-AKT2 antibody [4H7] (ab175354)

All lanes: Anti-AKT2 antibody [4H7] (ab175354) at 1/1000 dilution

Lane 1: MCF7 whole cell lysate

Lane 2: HeLa whole cell lysate

Lane 3: HepG2 whole cell lysate

Lane 4: A549 whole cell lysate

Lane 5: 293T whole cell lysate

Lane 6: Jurkat whole cell lysate

Lane 7: A431 whole cell lysate

Lane 8: U2OS whole cell lysate

Lane 9: COS7 whole cell lysate

Lane 10: 3T3 L1 whole cell lysate

Lane 11: NRK whole cell lysate

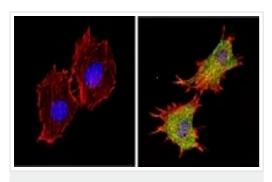
Lysates/proteins at 25 µg per lane.

Secondary

All lanes: goat anti-mouse-HRP at 1/20000 dilution

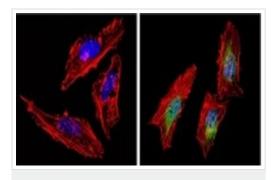
Developed using the ECL technique.

Predicted band size: 55 kDa



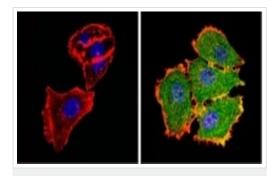
Immunocytochemistry/ Immunofluorescence - Anti-AKT2 antibody [4H7] (ab175354)

Immunofluorescent analysis of AKT2 (green) showing staining in the cytoplasm and nucleus of C2C12 cells (right) compared to a negative control without primary antibody (left). Formalin-fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 5-10 minutes and blocked with 3% BSA-PBS for 30 minutes at room temperature. Cells were probed with an AKT2 monoclonal antibody (ab175354) in 3% BSA-PBS at a dilution of 1:20 and incubated overnight at 4 °C in a humidified chamber. Cells were washed with PBST and incubated with a DyLight-conjugated secondary antibody in PBS at room temperature in the dark. F-actin (red) was stained with a fluorescent red phalloidin and nuclei (blue) were stained with Hoechst or DAPI. Images were taken at a magnification of 60x.



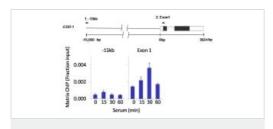
Immunocytochemistry/ Immunofluorescence - Anti-AKT2 antibody [4H7] (ab175354)

Immunofluorescent analysis of AKT2 (green) showing staining in the cytoplasm and nucleus of Hela cells (right) compared to a negative control without primary antibody (left). Formalin-fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 5-10 minutes and blocked with 3% BSA-PBS for 30 minutes at room temperature. Cells were probed with an AKT2 monoclonal antibody (ab175354) in 3% BSA-PBS at a dilution of 1:20 and incubated overnight at 4 °C in a humidified chamber. Cells were washed with PBST and incubated with a DyLight-conjugated secondary antibody in PBS at room temperature in the dark. F-actin (red) was stained with a fluorescent red phalloidin and nuclei (blue) were stained with Hoechst or DAPI. Images were taken at a magnification of 60x.



Immunocytochemistry/ Immunofluorescence - Anti-AKT2 antibody [4H7] (ab175354)

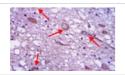
Immunofluorescent analysis of AKT2 (green) showing staining in the cytoplasm and nucleus of MCF-7 cells (right) compared to a negative control without primary antibody (left). Formalin-fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 5-10 minutes and blocked with 3% BSA-PBS for 30 minutes at room temperature. Cells were probed with an AKT2 monoclonal antibody (ab175354) in 3% BSA-PBS at a dilution of 1:20 and incubated overnight at 4 °C in a humidified chamber. Cells were washed with PBST and incubated with a DyLight-conjugated secondary antibody in PBS at room temperature in the dark. F-actin (red) was stained with a fluorescent red phalloidin and nuclei (blue) were stained with Hoechst or DAPI. Images were taken at a magnification of 60x.



ChIP - Anti-AKT2 antibody [4H7] (ab175354)

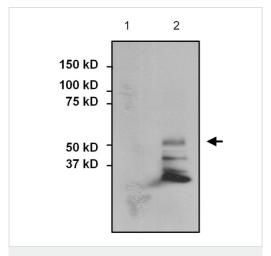
Chromatin immunoprecipitation analysis of Akt1 and Akt2 was performed using cross-linked chromatin from 1 x 10⁶ HCT116 colon carcinoma cells treated with serum for 0, 15, 30, and 60 minutes. Immunoprecipitation was performed with 1.0ul/100ul well volume of an Atk1 monoclonal antibody and an Akt2 monoclonal antibody (ab175354). Chromatin aliquots from ~1 x 10⁵ cells were used per ChIP pull-down. Quantitative PCR data were done in quadruplicate using 1ul of eluted DNA in 2ul SYBR real-time PCR reactions containing primers to amplify -15kb upstream of the Egr1 gene or exon-1 of Egr1. PCR calibration curves were generated for each primer pair from a dilution series of sheared total genomic DNA. Quantitation of immunoprecipitated chromatin is presented as signal relative to the total amount of input chromatin. Results represent the mean +/- SEM for three experiments. A schematic representation of the Egr-1 locus is shown above the data where boxes represent exons (black boxes = translated regions, white boxes = untranslated regions); the zigzag line represents an intron;

and the straight line represents upstream sequence. Regions amplified by Egr-1 primers are represented by black bars.



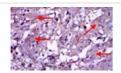
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-AKT2 antibody [4H7] (ab175354)

Immunohistochemical analysis of deparaffinized Human
Esophageal cancer tissue labeling AKT2 with ab175354 at 1/200
dilution. Detection was performed using a goat anti-mouse HRP
secondary antibody followed by colorimetric detection using DAB
substrate.



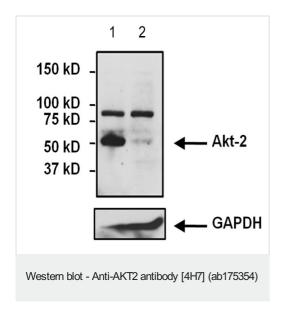
Immunoprecipitation - Anti-AKT2 antibody [4H7] (ab175354)

Immunoprecipitation of AKT2 was performed on HeLa cells. The antigen:antibody complex was formed by incubating 750 μ g whole cell lysate with 2 μ g of ab175354. WB detection used ab175354 at 1/1000 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-AKT2 antibody [4H7] (ab175354)

Immunohistochemical analysis of deparaffinized normal Human Medulla Oblongata tissue labeling AKT2 with ab175354 at 1/200 dilution. Detection was performed using a goat anti-mouse HRP secondary antibody followed by colorimetric detection using DAB substrate.



All lanes: Anti-AKT2 antibody [4H7] (ab175354) at 1/1000 dilution

Lane 1: Non-transfected U2OS cells

Lane 2: U2OS cells transfected with AKT2 siRNA

Secondary

All lanes: goat anti-mouse-HRP at 1/20000 dilution

Predicted band size: 55 kDa

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