abcam

Product datasheet

Anti-pan-AKT (phospho T308) antibody ab8933

59 References 画像数 8

製品の概要

製品名 Anti-pan-AKT (phospho T308) antibody

製品の詳細 Rabbit polyclonal to pan-AKT (phospho T308)

由来種 Rabbit

特異性 This antibody was made against a peptide directed against the phosphorylated form of AKT1 at

T308, but due to a high degree of homology it is predicted to cross react with AKT2 and AKT3 if

they are phosphorylated at the corresponding residue. Weak cross reactivity with AKT2.

アプリケーション 適用あり: IHC-P, WB, Dot blot

種交差性 交差種: Human, Recombinant fragment

交差が予測される動物種: Mouse, Rat 4

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

ポジティブ・コントロール IHC-P: Human lung, testis and breast tissue. WB: Human spleen, small intestine, placenta,

skeletal muscle, lung, tonsil and thymus tissue lysate. GST-tagged AKT1 recombinant protein.

AKT1 and AKT3 recombinant protein.

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. Upon delivery aliquot and store at -20°C or -80°C. Avoid repeated freeze / thaw

cycles.

パッファー Preservative: 0.01% Sodium azide

Constituents: 0.42% Potassium phosphate, 0.87% Sodium chloride

精製度 Immunogen affinity purified

特記事項(精製) This product was prepared from monospecific antiserum by immunoaffinity chromatography using

phospho peptide coupled to agarose beads followed by solid phase adsorption(s) against non-

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phospho peptide and non-specific peptide to remove any unwanted reactivities. Assay by immunoelectrophoresis resulted in a single precipitin arc against anti-Rabbit Serum.

ポリモノ

ポリクローナル

アイソタイプ

ΙgG

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ab8933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
IHC-P		Use a concentration of 4 μ g/ml. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB		1/500 - 1/1000. Predicted molecular weight: 56 kDa.
Dot blot		Use a concentration of 5 µg/ml.

ターゲット情報

機能

Plays a role as a 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). General protein kinase capable of phosphorylating several known proteins. Phosphorylates TBC1D4. Signals downstream of phosphatidylinositol 3-kinase (Pl(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 I (IGF-I). Plays a role in glucose transport by mediating insulin-induced translocation of the GLUT4 glucose transporter to the cell surface. Mediates the antiapoptotic effects of IGF-I. Mediates insulin-stimulated protein synthesis by phosphorylating TSC2 at 'Ser-939' and 'Thr-1462', thereby activating mTORC1 signaling and leading to both phosphorylation of 4E-BP1 and in activation of RPS6KB1. Promotes glycogen synthesis by mediating the insulin-induced activation of glycogen synthase. The activated form can suppress FoxO gene transcription and promote cell cycle progression. Essential for the SPATA13-mediated regulation of cell migration and adhesion assembly and disassembly.

組織特異性

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

関連疾患

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].

Defects in AKT1 are associated with susceptibility to ovarian cancer [MIM:604370]; also called

susceptibility to familial breast-ovarian cancer type 1 (BROVCA1).

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.

ドメイン Binding of the PH domain to the phosphatidylinositol 3-kinase alpha (PI(3)K) results in its

targeting to the plasma membrane. The PH domain mediates interaction with TNK2 and Tyr-176

is also essential for this interaction.

The AGC-kinase C-terminal mediates interaction with THEM4.

翻訳後修飾 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.

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.

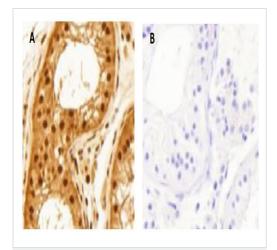
細胞内局在 Cytoplasm. Nucleus. Cell membrane. Nucleus after activation by integrin-linked protein kinase 1

(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 $% \left(1\right) =\left(1\right) \left(1\right)$

translocates to the nucleus.

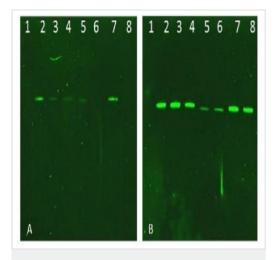
画像



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-pan-AKT (phospho T308) antibody (ab8933)

Panel A: ab8933 (1/200 dilution, 30mins at RT) staining AKT (phospho T308) in human testis tissue in immunohistochemical analysis. Secondary used was an anti-Rabbit polyclonal HRP conjugate (Ready to use) DAB staining. Heat induced antigen retrieval was performed using Leica Bond Epitope Retrieval Bu□er 1 (Citrate solution, pH6.0) for 20 minutes (ER1(20)). Counterstain is hemotoxylin.

Panel B: Secondary antibody only.



Western blot - Anti-pan-AKT (phospho T308) antibody (ab8933)

All lanes : Anti-pan-AKT (phospho T308) antibody (ab8933) at 1/2270 dilution

Lane 1: MW Protein ladder

Lane 2: Recombinant AKT1 protein, 50ng

Lane 3: Recombinant AKT1 protein, 50ng (phosphatase treated)

Lane 4: Recombinant AKT1 T308A/S473A mutant protein, 50ng

Lane 5: Recombinant AKT2 protein, 50ng

Lane 6: Recombinant AKT2 protein, 50ng (phosphatase treated)

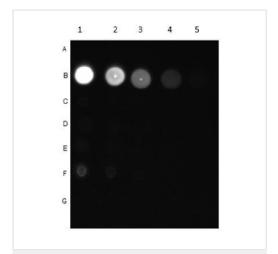
Lane 7: Recombinant AKT3 protein, 50ng

Lane 8: Recombinant AKT3 protein, 50ng (phosphatase treated)

Predicted band size: 56 kDa

Blot A: ab8933 used at 1/2270.

Blot B: Anti-Akt used 1/1000.



Dot Blot - Anti-pan-AKT (phospho T308) antibody (ab8933)

Dot Blot - Anti-pan-AKT (phospho T308) antibody (ab8933, 5 µg/ml).

Secondary antibody is an anti-rabbit lgG HRP used at a 1/70,000 dilution.

Exposure time 60 secs.

Columns 1 – 5, Left to Right 100, 33.33, 11.11, 3.70, 1.23 ng

Row A: AKT1-BSA peptide

Row B: AKT1 pT308 – BSA peptide

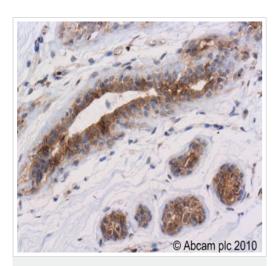
Row C: AKT1 S473 – BSA peptide

Row D: AKT1 pS473 - BSA peptide

Row E: CDC27 T244 -BSA peptide

Row F: CDC27 pT244 - BSA peptide

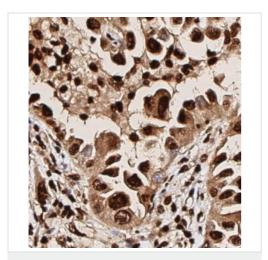
Row G: BSA control



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-pan-AKT (phospho T308) antibody (ab8933)

ab8933 (4µg/ml) staining AKT (phospho T308) in human breast using an automated system (DAKO Autostainer Plus). Using this protocol there is strong staining of membrane and cytoplasmic compartment within the breast ductal regions.

Sections were rehydrated and antigen retrieved with the Dako 3 in 1 AR buffer EDTA pH 9.0 in a DAKO PT link. Slides were peroxidase blocked in 3% H2O2 in methanol for 10 mins. They were then blocked with Dako Protein block for 10 minutes (containing casein 0.25% in PBS) then incubated with primary antibody for 20 min and detected with Dako envision flex amplification kit for 30 minutes. Colorimetric detection was completed with Diaminobenzidine for 5 minutes. Slides were counterstained with Haematoxylin and coverslipped under DePeX. Please note that, for manual staining, optimization of primary antibody concentration and incubation time is recommended.

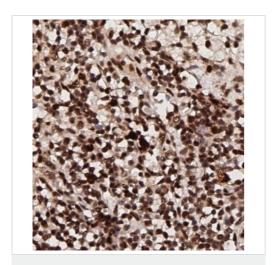


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-pan-AKT (phospho T308) antibody (ab8933)

ab8933 (1/200 dilution, 30mins at RT) staining AKT (phospho T308) in human lung tissue in immunohistochemical analysis.

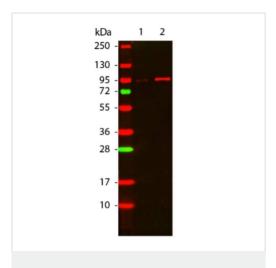
Secondary used was an anti-Rabbit polyclonal HRP conjugate (Ready to use, 8mins at RT) DAB staining. HIER using citrate buffer for 20mins. Counterstain is hemotoxylin.

Signal amplification may be required.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-pan-AKT (phospho T308) antibody (ab8933)

ab8933 (1/200 dilution, 30mins at RT) staining AKT (phospho T308) in human lymph node in breast tissue in immunohistochemical analysis. Secondary used was an anti-Rabbit polyclonal HRP conjugate (Ready to use, 8mins at RT) DAB staining. HIER using citrate buffer for 20mins. Counterstain is hemotoxylin.



Western blot - Anti-pan-AKT (phospho T308) antibody (ab8933)

All lanes : Anti-pan-AKT (phospho T308) antibody (ab8933) at 1/1000 dilution (overnight at 4degC)

Lane 1 : GST-tagged AKT1 recombinant protein, 50ng

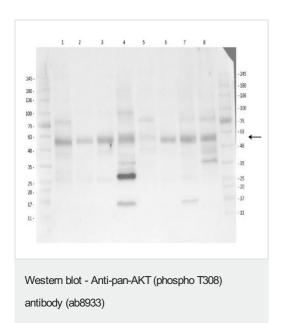
Lane 2 : GST-tagged AKT1 active recombinant protein, 50ng

Secondary

All lanes : DyLight™ 649 rabbit secondary antibody, (30mins at RT) at 1/20000 dilution

Predicted band size: 56 kDa

Recombinant protein expected to run ~80-100 kDa.



All lanes : Anti-pan-AKT (phospho T308) antibody (ab8933) at 1/1000 dilution

Lane 1: Human spleen whole tissue lysate

Lane 2: Human small intestine whole tissue lysate

Lane 3: Human placenta whole tissue lysate

Lane 4: Human skeletal muscle whole tissue lysate

Lane 5: Human brain cerebellum whole tissue lysate

Lane 6: Human lung whole tissue lysate

Lane 7: Human tonsil whole tissue lysate

Lane 8: Human thymus whole tissue lysate

Secondary

All lanes: Goat anti-Rabbit lg HRP at 1/40000 dilution

Predicted band size: 56 kDa

Exposure time: 8 seconds

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