abcam

Product datasheet

Anti-PKC delta (phospho S299) antibody [EPNCI119] ab133456





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Overview

Product name Anti-PKC delta (phospho S299) antibody [EPNCI119]

Description Rabbit monoclonal [EPNCI119] to PKC delta (phospho S299)

Host species Rabbit

Tested applications Suitable for: WB, IHC-P

Unsuitable for: Flow Cyt or ICC/IF

Species reactivity Reacts with: Human

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

Positive control WB: MCF7 treated with 100nM Calyculin A; IHC-P: Human bladder cancer tissue.

General notes This product has switched from a hybridoma to recombinant production method on 29th January

2024.

This antibody was developed as part of a collaboration between Epitomics, the National Cancer Institutes Center for Cancer Research and the lab of Peter Blumberg. View antibodies from NCI

Center for Cancer Research Collaboration.

The auto-phosphorylation of PKC delta at pS299 targeted by ab133456 was identified in the following paper: FEBS Lett. 2007 Jul 24;581(18):3377-81. Novel phosphorylation site markers of protein kinase C delta activation. Durgan J1, Michael N, Totty N, Parker PJ.

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

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.

Rat: We have preliminary internal testing data to indicate this antibody may not react with this species. Please contact us for more information.

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C.

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

Purity Protein A purified

Clonality Monoclonal
Clone number EPNCI119

Isotype IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab133456 in the following tested applications.

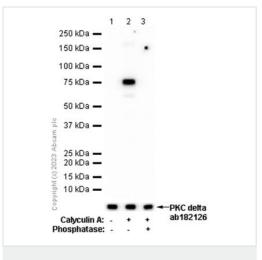
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB	* * * * * * * (3)	1/5000. Detects a band of approximately 78 kDa (predicted molecular weight: 78 kDa).
IHC-P		1/5000. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Application notes Is unsuitable for Flow Cyt or ICC/IF.

Target

Function	This is calcium-independent, phospholipid-dependent, serine- and threonine-specific enzyme. PKC is activated by diacylglycerol which in turn phosphorylates a range of cellular proteins. PKC also serves as the receptor for phorbol esters, a class of tumor promoters. May play a role in antigen-dependent control of B-cell function. Phosphorylates MUC1 in the C-terminal and regulates the interaction between MUC1 and beta-catenin.
Sequence similarities	Belongs to the protein kinase superfamily. AGC Ser/Thr protein kinase family. PKC subfamily. Contains 1 AGC-kinase C-terminal domain. Contains 1 C2 domain. Contains 2 phorbol-ester/DAG-type zinc fingers. Contains 1 protein kinase domain.
Domain	The C1 domain, containing the phorbol ester/DAG-type region 1 (C1A) and 2 (C1B), is the diacylglycerol sensor. The C2 domain is a non-calcium binding domain. It binds proteins containing phosphotyrosine in a sequence-specific manner.
Post-translational modifications	Phosphorylated on Thr-507, within the activation loop. Autophosphorylated and/or phosphorylated. Although the Thr-507 phosphorylation occurs it is not a prerequisite for enzymatic activity.
Cellular localization	Cytoplasm. Membrane.



Western blot - Anti-PKC delta (phospho S299) antibody [EPNCI119] (ab133456)

All lanes : Anti-PKC delta (phospho S299) antibody [EPNCI119] (ab133456) at 1/5000 dilution

Lane 1 : Untreated MCF7 (Human breast adenocarcinoma epithelial cell) whole cell lysate

Lane 2: MCF7 treated with 100nM Calyculin A for 30min whole cell lysate

Lane 3: MCF7 treated with 100nM Calyculin A for 30min whole cell lysate, then the membrane was incubated with phosphatase

Lysates/proteins at 15 µg per lane.

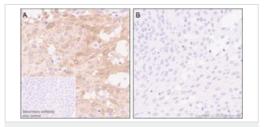
Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 78 kDa **Observed band size:** 78 kDa

Exposure time: 20 seconds

Blocking and diluting buffer and concentration: 5% NFDM/TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PKC delta (phospho S299) antibody [EPNCI119] (ab133456)

Immunohistochemical analysis of paraffin-embedded Human bladder cancer tissue labeling PKC delta with ab133456 at 1/5000 (0.077 µg/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on human bladder cancer without alkaline phosphatase treatment (image A). No signal was detected when tissues were treated with alkaline phosphatase (image B). The human bladder cancer sections were performed by Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). The sections were using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins, then labeling ab133456 at 1/5000 dilution for 30 mins at room temperature. Counterstained with hematoxylin. The immunostaining was performed on a Leica Biosystems BOND® RX instrument.

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