

Anti-PKA R2/PKR2 (phospho S99) antibody [E151] ab32390

Recombinant RabMAb

★★★★★ 1 Abreviews 37 References 11 Images

Overview

Product name	Anti-PKA R2/PKR2 (phospho S99) antibody [E151]
Description	Rabbit monoclonal [E151] to PKA R2/PKR2 (phospho S99)
Host species	Rabbit
Specificity	<p>The antibody only detects PKA R2/PKR2 phosphorylated on Serine 99. Phosphoserine 99 in Human corresponds to phosphoserine 96 in mouse.</p> <p>The mouse and pig recommendation is based on the WB results. We do not guarantee IHC-P for mouse and pig.</p>
Tested applications	<p>Suitable for: WB, IHC-P, ICC/IF</p> <p>Unsuitable for: Flow Cyt</p>
Species reactivity	Reacts with: Mouse, Rat, Human, Pig
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: K562 cell lysate. HeLa starved for 3 hours and then treated with 100 nM Calyculin A for 30 minutes, NIH/3T3, NIH/3T3 treated with Calyculin A, and C6 treated with 100 ng/ml Calyculin A for 1 hour cell lysates. Pig heart lysates. ICC/IF: HeLa cells. IHC-P: Human lymph node metastatic tissue. Human kidney without Lambda Protein Phosphatase treatment (image A) and Rat kidney without Lambda Protein Phosphatase treatment
General notes	<p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"> - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production <p>For more information see here.</p> <p>Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents.</p>

Properties

Form Liquid

Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: PBS, 0.05% BSA, 40% Glycerol (glycerin, glycerine)
Purity	Protein A purified
Clonality	Monoclonal
Clone number	E151
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab32390 in the following tested applications.

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

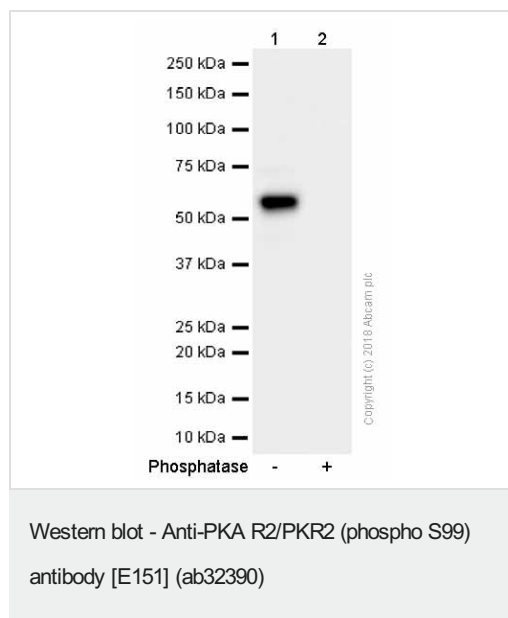
Application	Abreviews	Notes
WB	★★★★★ (1)	1/1000. Detects a band of approximately 51 kDa (predicted molecular weight: 45 kDa). For unpurified use at 1/2500
IHC-P		1/500. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. The mouse and pig recommendation is based on the WB results. We do not guarantee IHC-P for mouse and pig. See <u>IHC antigen retrieval protocols</u>. For unpurified use at 1/250
ICC/IF		1/500.

Application notes Is unsuitable for Flow Cyt.

Target

Function	Regulatory subunit of the cAMP-dependent protein kinases involved in cAMP signaling in cells. Type II regulatory chains mediate membrane association by binding to anchoring proteins, including the MAP2 kinase.
Tissue specificity	Four types of regulatory chains are found: I-alpha, I-beta, II-alpha, and II-beta. Their expression varies among tissues and is in some cases constitutive and in others inducible.
Sequence similarities	Belongs to the cAMP-dependent kinase regulatory chain family. Contains 2 cyclic nucleotide-binding domains.
Post-translational modifications	Phosphorylated by the activated catalytic chain.
Cellular localization	Cytoplasm. Cell membrane. Colocalizes with PJA2 in the cytoplasm and the cell membrane.

Images



All lanes : Anti-PKA R2/PKR2 (phospho S99) antibody [E151] (ab32390) at 1/1000 dilution (Purified)

Lane 1 : Pig heart lysates

Lane 2 : Pig heart. Then the membrane was incubated with phosphatase

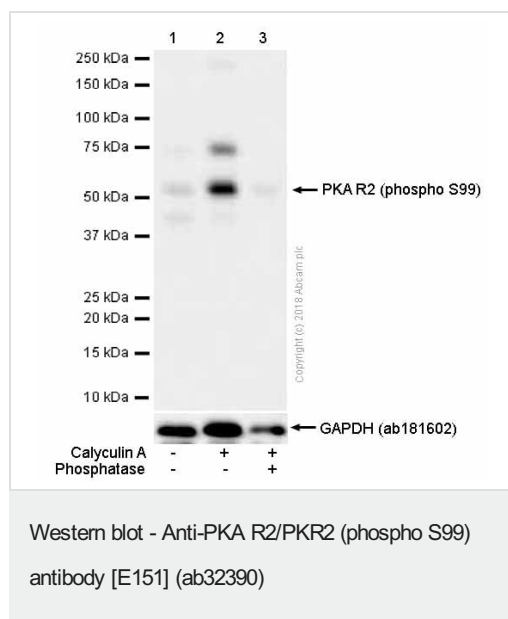
Lysates/proteins at 15 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/20000 dilution

Predicted band size: 45 kDa

Observed band size: 51 kDa



All lanes : Anti-PKA R2/PKR2 (phospho S99) antibody [E151] (ab32390) at 1/1000 dilution (Purified)

Lane 1 : C6 (Rat glial tumor glial cell) whole cell lysates

Lane 2 : C6 (Rat glial tumor glial cell) treated with 100ng/ml Calyculin A for 1 hour

Lane 3 : C6 (Rat glial tumor glial cell) treated with 100ng/ml Calyculin A for 1 hour. Then the membrane was incubated with phosphatase

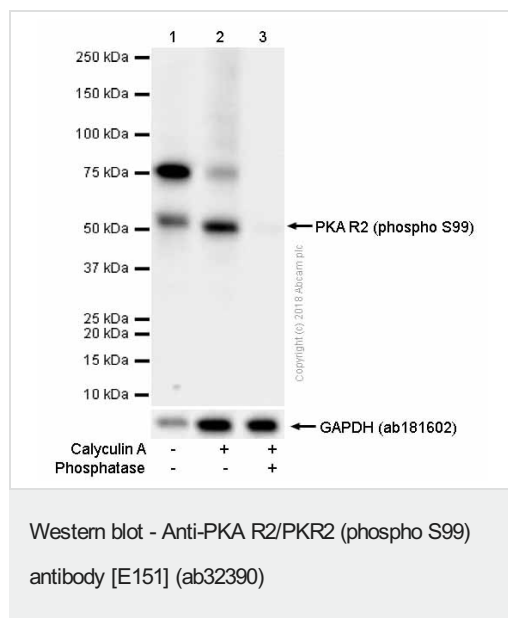
Lysates/proteins at 15 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/20000 dilution

Predicted band size: 45 kDa

Observed band size: 51 kDa



All lanes : Anti-PKA R2/PKR2 (phospho S99) antibody [E151] (ab32390) at 1/1000 dilution (Purified)

Lane 1 : NIH/3T3 (Mouse embryonic fibroblast) whole cell lysates

Lane 2 : NIH/3T3 (Mouse embryonic fibroblast) treated with Calyculin A

Lane 3 : NIH/3T3 (Mouse embryonic fibroblast) treated with Calyculin A. Then the membrane was incubated with phosphatase

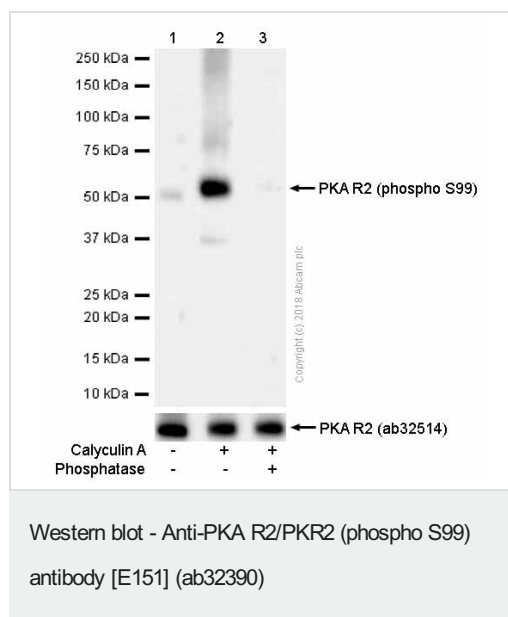
Lysates/proteins at 15 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/20000 dilution

Predicted band size: 45 kDa

Observed band size: 51 kDa



All lanes : Anti-PKA R2/PKR2 (phospho S99) antibody [E151] (ab32390) at 1/1000 dilution (Purified)

Lane 1 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysates

Lane 2 : HeLa (Human cervix adenocarcinoma epithelial cell) were starved for 3 hours and then treated with 100 nM Calyculin A for 30 minutes

Lane 3 : HeLa (Human cervix adenocarcinoma epithelial cell) treated with 100 nM Calyculin A for 30 minutes. Then the membrane was incubated with phosphatase

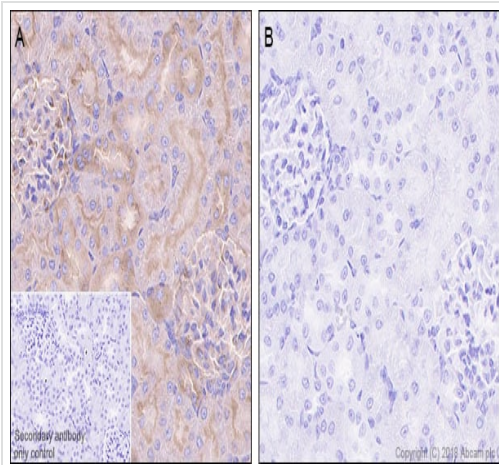
Lysates/proteins at 15 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/20000 dilution

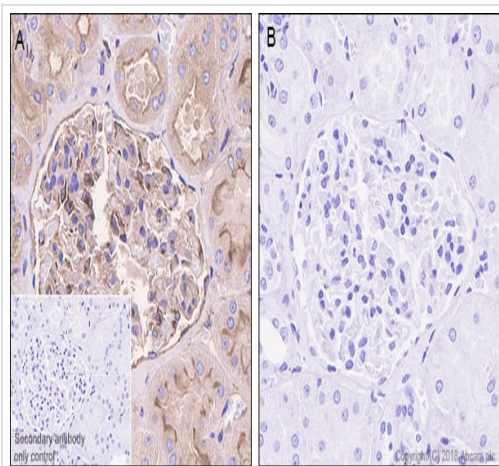
Predicted band size: 45 kDa

Observed band size: 51 kDa



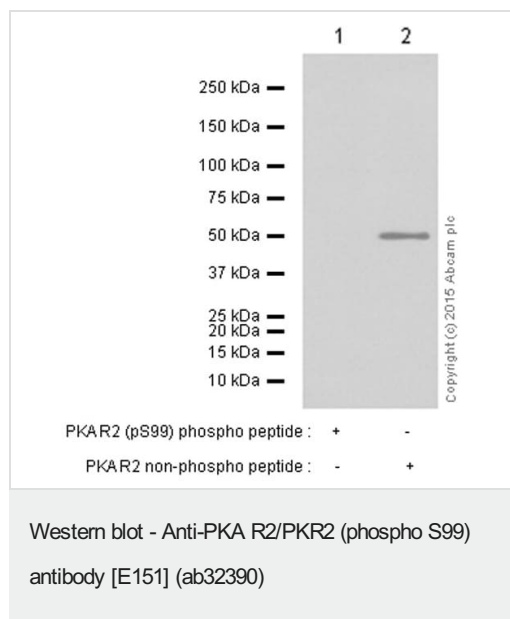
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PKA R2/PKR2 (phospho S99) antibody [E151] (ab32390)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Rat kidney without Lambda Protein Phosphatase treatment (image A) and treated with Lambda Protein Phosphatase (image B). tissue sections labeling PKA R2/PKR2 with purified ab32390 at 1:500 dilution (0.75 µg/ml). Heat mediated antigen retrieval was performed Perform heat mediated antigen retrieval using [ab93684](#) (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PKA R2/PKR2 (phospho S99) antibody [E151] (ab32390)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human kidney without Lambda Protein Phosphatase treatment (image A) and treated with Lambda Protein Phosphatase (image B). tissue sections labeling PKA R2/PKR2 with purified ab32390 at 1:500 dilution (0.75 µg/ml). Heat mediated antigen retrieval was performed Perform heat mediated antigen retrieval using [ab93684](#) (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



All lanes : Anti-PKA R2/PKR2 (phospho S99) antibody [E151] (ab32390) at 1/2000 dilution

Lane 1 : K562 whole cell lysate with PKA R2/PKR2 (pS99) phospho peptide

Lane 2 : K562 whole cell lysate with PKA R2/PKR2 non-phospho peptide

Secondary

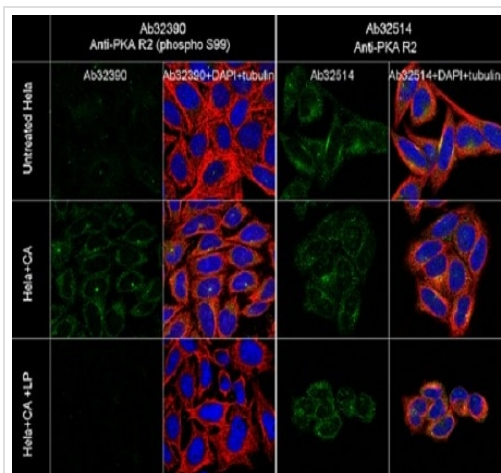
All lanes : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/20000 dilution

Predicted band size: 45 kDa

Observed band size: 51 kDa

Exposure time: 3 minutes

Blocking and dilution buffer: 5% NFDM/TBST.



Immunocytochemistry/ Immunofluorescence - Anti-
PKA R2/PKR2 (phospho S99) antibody [E151]
(ab32390)

Cell line: HeLa (human cervix adenocarcinoma)

Target AbID: Ab32390 anti-PKA R2/PKR2 (phospho S99)

Target Secondary: **ab150077** AlexaFluor®488 goat anti-rabbit

Counterstain AbID: **ab7291** anti-Tubulin (mouse mAb)

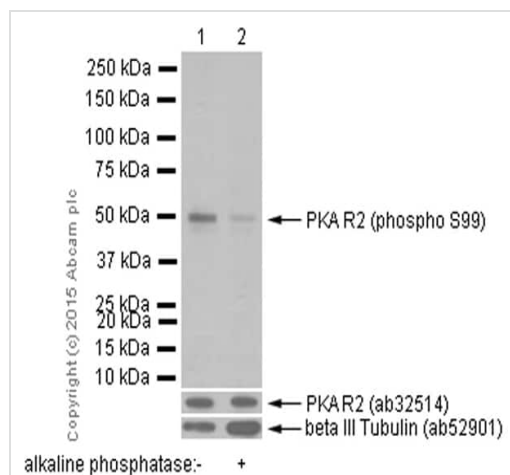
Target Secondary: **ab150120** AlexaFluor®594 goat anti-mouse

Fixative: 4% PFA

Permeabilization: 0.1% Triton-X

Nuclear counter stain: DAPI

Other comments: Confocal image showed increased cytoplasmic staining after CA (100ng/ml, 10min) treatment on HeLa cells. The LP treatment decreased the increased cytoplasmic staining caused by CA. Ab32514 was used as a Pan control for ab32390. The results showed cytoplasmic staining on untreated, CA and CA+LP treated HeLa cells.



Western blot - Anti-PKA R2/PKR2 (phospho S99)
antibody [E151] (ab32390)

All lanes : Anti-PKA R2/PKR2 (phospho S99) antibody [E151]
(ab32390) at 1/10000 dilution

Lane 1 : Untreated K562 whole cell lysate

Lane 2 : K562 whole cell lysate - treated with alkaline phosphatase

Lysates/proteins at 10 µg per lane.

Secondary

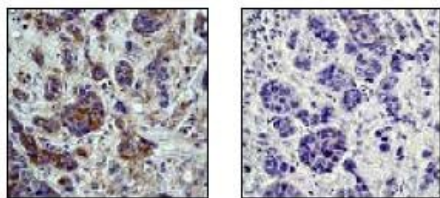
All lanes : Peroxidase-conjugated goat anti-rabbit IgG (H+L) at
1/1000 dilution

Predicted band size: 45 kDa

Observed band size: 51 kDa

Exposure time: 3 minutes

Blocking and dilution buffer: 5% NFDM/TBST.



A

B

ab32390, at a 1/250 dilution, staining (A) untreated (B) Phosphatase treated lymph node metastatic by immunohistochemistry, Paraffin embedded human tissue

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PKA R2/PKR2 (phospho S99) antibody [E151] (ab32390)

Why choose a recombinant antibody?



Research with confidence
Consistent and reproducible results



Long-term and scalable supply
Recombinant technology



Success from the first experiment
Confirmed specificity



Ethical standards compliant
Animal-free production

Anti-PKA R2/PKR2 (phospho S99) antibody [E151] (ab32390)

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