

Product datasheet

Anti-RSK1 p90 antibody [E4] ab32114

KO VALIDATED Recombinant RabMAB

★★★★★ 4 Abreviews 12 References 10 Images

Overview

Product name	Anti-RSK1 p90 antibody [E4]
Description	Rabbit monoclonal [E4] to RSK1 p90
Host species	Rabbit
Specificity	The mouse and rat recommendation is only based on the WB results.
Tested applications	Suitable for: WB, IHC-P, IP, Flow Cyt (Intra), ICC/IF
Species reactivity	Reacts with: Mouse, Rat, Human Predicted to work with: Pig 
Immunogen	Synthetic peptide within Human RSK1 p90 (N terminal). The exact sequence is proprietary.
Positive control	WB: MCF7, K-562, PC-12 whole cell lysate, mouse lung tissue lysate. IHC-P: Human colon tissue, Human breast cancer tissue IP: K-562 cell lysate Flow Cyt (intra): HeLa cells ICC/IF: K-562 cells
General notes	This product is a recombinant monoclonal antibody, which offers several advantages including: <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 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 short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol, 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal

Clone number E4
Isotype IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab32114 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	★★★★★ (2)	1/1000. Detects a band of approximately 90 kDa (predicted molecular weight: 83 kDa).
IHC-P	★★★★★ (2)	1/100. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See IHC antigen retrieval protocols . The mouse, rat and pig recommendation is based on the WB results. We do not guarantee IHC-P for mouse, rat and pig.
IP		1/70. For unpurified format use at 1/10 dilution.
Flow Cyt (Intra)		1/100 - 1/1000. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
ICC/IF		1/150 - 1/200.

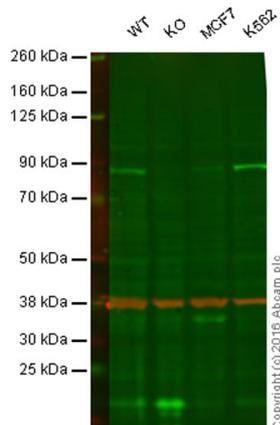
Target

Function Serine/threonine kinase that may play a role in mediating the growth-factor and stress induced activation of the transcription factor CREB.

Sequence similarities Belongs to the protein kinase superfamily. AGC Ser/Thr protein kinase family. S6 kinase subfamily.
Contains 1 AGC-kinase C-terminal domain.
Contains 2 protein kinase domains.

Post-translational modifications Autophosphorylated on Ser-380, as part of the activation process.

Images



Western blot - Anti-RSK1 p90 antibody [E4]
(ab32114)

Lane 1: Wild-type HAP1 cell lysate (20 µg)

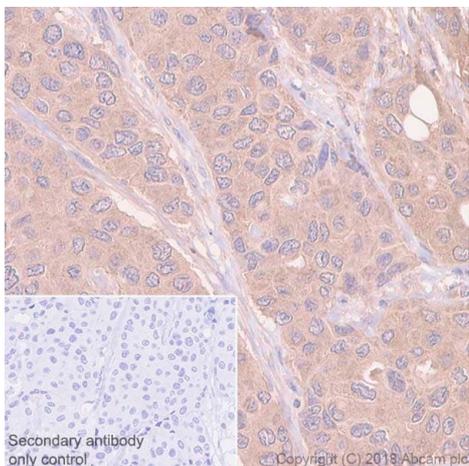
Lane 2: RSK1 p90 knockout HAP1 cell lysate (20 µg)

Lane 3: MCF7 cell lysate (20 µg)

Lane 4: K562 cell lysate (20 µg)

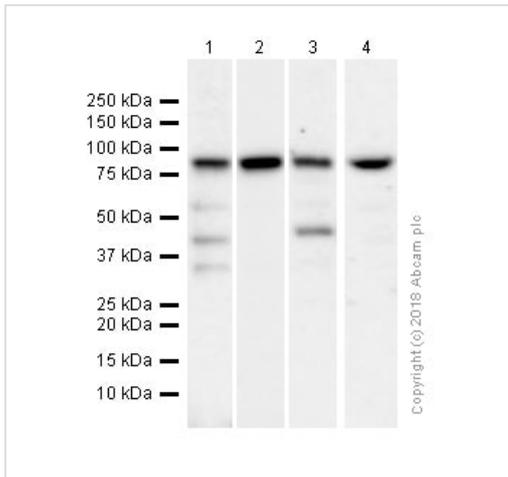
Lanes 1 - 4: Merged signal (red and green). Green - ab32114 (unpurified) observed at 88 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab32114 was shown to recognize RSK1 p90 when RSK1 p90 knockout samples were used, along with additional cross-reactive bands. Wild-type and RSK1 p90 knockout samples were subjected to SDS-PAGE. ab32114 and **ab8245** (loading control to GAPDH) were diluted at 1/500 and 1/10 000 respectively and incubated overnight at 4°C. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1/10 000 dilution for 1 h at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-RSK1 p90 antibody [E4]
(ab32114)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human breast cancer tissue sections labeling RSK1 p90 with purified ab32114 at 1/100 dilution (15.4 µg/ml). Heat mediated antigen retrieval was performed using 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.



Western blot - Anti-RSK1 p90 antibody [E4] (ab32114)

All lanes : Anti-RSK1 p90 antibody [E4] (ab32114) at 1/1000 dilution (Purified)

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

Lane 2 : K-562 (Human chronic myelogenous leukemia lymphoblast) whole cell lysate

Lane 3 : Mouse lung lysates

Lane 4 : PC-12 (Rat adrenal gland pheochromocytoma) whole cell lysates

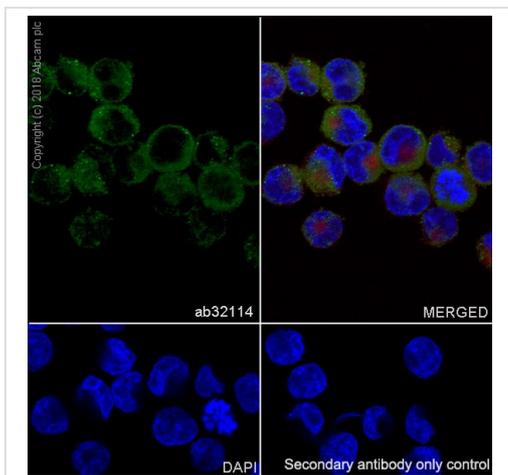
Lysates/proteins at 20 µg per lane.

Secondary

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

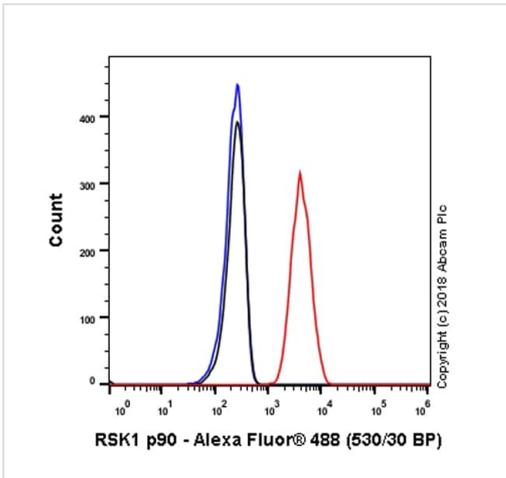
Predicted band size: 83 kDa

Observed band size: 83 kDa



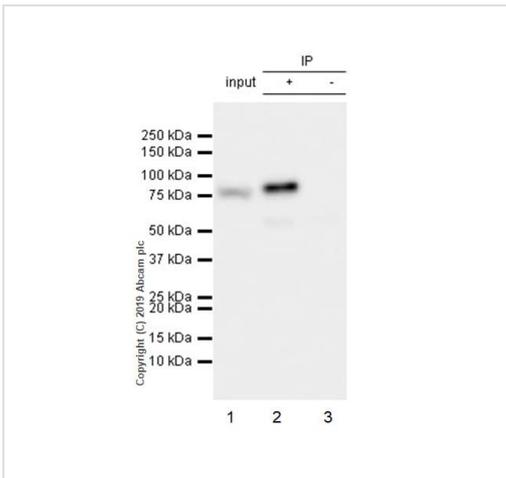
Immunocytochemistry/ Immunofluorescence - Anti-RSK1 p90 antibody [E4] (ab32114)

Immunocytochemistry/ Immunofluorescence analysis of K-562 (Human chronic myelogenous leukemia lymphoblast) cells labeling RSK1 p90 with purified ab32114 at 1/150 dilution (10 µg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor[®] 594) at 1:200 (2.5 µg/ml). Goat anti rabbit IgG (Alexa Fluor[®] 488, **ab150077**) was used as the secondary antibody at 1/1000 (2 µg/ml) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Flow Cytometry (Intracellular) - Anti-RSK1 p90 antibody [E4] (ab32114)

Intracellular Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling RSK1 p90 with purified ab32114 at 1/150 dilution (10µg/ml) (red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).

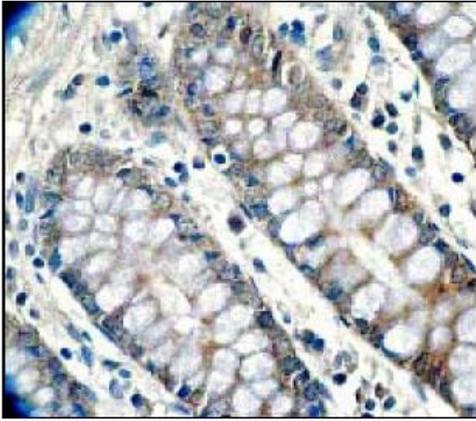


Immunoprecipitation - Anti-RSK1 p90 antibody [E4] (ab32114)

ab32114 (purified) at 1/70 dilution (2ug) immunoprecipitating RSK1 p90 in K-562 whole cell lysate. K-562 (Human chronic myelogenous leukemia lymphoblast) whole cell lysate 10ug
 Lane 2 (+): ab32114 & K-562 whole cell lysate
 Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of ab32114 in K-562 whole cell lysate

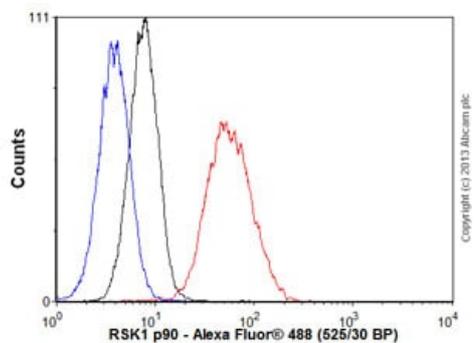
For western blotting, VeriBlot for IP secondary antibody (HRP) (**ab131366**) was used at 1/1000 dilution.

Blocking and diluting buffer: 5% NFDm/TBST.



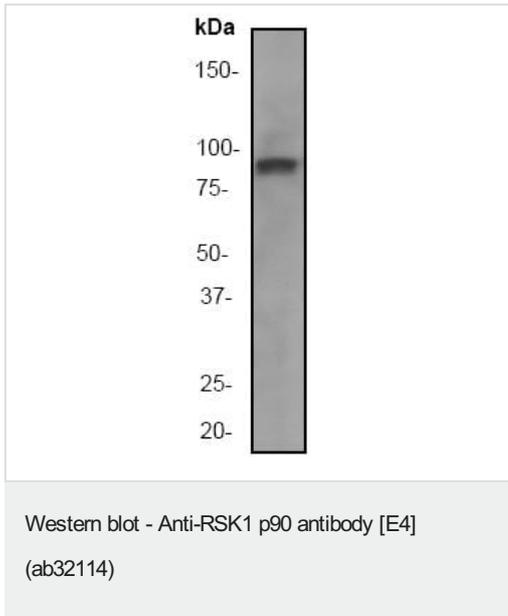
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-RSK1 p90 antibody [E4] (ab32114)

Ab32114 (unpurified), at a 1/100 dilution, staining RSK1 p90 in paraffin embedded human colon tissue by immunohistochemistry.



Flow Cytometry (Intracellular) - Anti-RSK1 p90 antibody [E4] (ab32114)

Overlay histogram showing HeLa cells stained with (unpurified)ab32114 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab32114, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor[®] 488 goat anti-rabbit IgG (H&L) (**ab150077**) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (0.1µg/1x10⁶ cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This antibody gave a positive signal in HeLa cells fixed with 4% paraformaldehyde (10 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.



Anti-RSK1 p90 antibody [E4] (ab32114) at 1/1000 dilution
(unpurified) + MCF7 cell lysate at 10 µg

Predicted band size: 83 kDa

Observed band size: 90 kDa

Why choose a recombinant antibody?

 <p>Research with confidence Consistent and reproducible results</p>	 <p>Long-term and scalable supply Recombinant technology</p>
 <p>Success from the first experiment Confirmed specificity</p>	 <p>Ethical standards compliant Animal-free production</p>

Anti-RSK1 p90 antibody [E4] (ab32114)

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