Anti-RSK4 Antibody [PSH02-04]

HA721763



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IHC-P, IF-Cell, FC
Molecular Wt:	Predicted band size: 84 kDa
Clone number:	PSH02-04
Description:	This gene encodes a member of ribosomal S6 kinase family, serine-threonine protein kinases which are regulated by growth factors. The encoded protein may be distinct from other members of this family, however, as studies suggest it is not growth factor dependent and may not participate in the same signaling pathways.
lmmunogen:	Recombinant protein within human RSK4 aa 31-420 / 745.
Positive control:	HEK-293 cell lysate, Caco-2 cell lysate, MDA-MB-231 cell lysate, MDA-MB-468 cell lysate, Raji cell lysate, Ramos cell lysate, NIH/3T3 cell lysate, PC-12 cell lysate, human kidney tissue, mouse kidney tissue, rat kidney tissue, PC-12, Caco-2.
Subcellular location:	Cytoplasm, cytosol, Nucleus.
Database links:	SwissProt: Q9UK32 Human Q7TPS0 Mouse Entrez Gene: 317203 Rat
Recommended Dilutions: WB IHC-P IF-Cell FC	1:1,000 1:200-1:1,000 1:100 1:1,000
Storage Buffer:	PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Store at +4 $^\circ\!\!\mathbb{C}$ after thawing. Aliquot store at -20 $^\circ\!\!\mathbb{C}$. Avoid repeated freeze / thaw cycles.
Purity:	Protein A affinity purified.

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Orders:0086-571-88062880

Technical:0086-571-89986345

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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

Images



Fig1: Western blot analysis of RSK4 on different lysates with Rabbit anti-RSK4 antibody (HA721763) at 1/1,000 dilution.

Lane 1: HEK-293 cell lysate Lane 2: Caco-2 cell lysate Lane 3: MDA-MB-231 cell lysate Lane 4: MDA-MB-468 cell lysate Lane 5: Raji cell lysate Lane 6: Ramos cell lysate Lane 7: NIH/3T3 cell lysate Lane 8: PC-12 cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 84 kDa Observed band size: 84 kDa

Exposure time: 1 minute 14 seconds;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA721763) at 1/1,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.



Fig2: Immunohistochemical analysis of paraffin-embedded human kidney tissue with Rabbit anti-RSK4 antibody (HA721763) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA721763) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

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Fig3: Immunohistochemical analysis of paraffin-embedded mouse kidney tissue with Rabbit anti-RSK4 antibody (HA721763) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA721763) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



Fig4: Immunohistochemical analysis of paraffin-embedded rat kidney tissue with Rabbit anti-RSK4 antibody (HA721763) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA721763) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

Fig5: Immunocytochemistry analysis of PC-12 cells labeling RSK4 with Rabbit anti-RSK4 antibody (HA721763) at 1/100 dilution.



Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-RSK4 antibody (HA721763) at 1/100 dilution in 1% BSA in PBST overnight at 4 $^{\circ}$ C. Goat Anti-Rabbit IgG H&L (iFluorTM 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at $+4^{\circ}$ C. Goat Anti-Mouse IgG H&L (iFluor 1594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

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Fig6: Flow cytometric analysis of Caco-2 cells labeling RSK4.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA721763, 1µg/mL) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4 °C for an hour, the cells were stained with a iFluor TM 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4 °C. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

- 1. Chrysostomou S et al. Repurposed floxacins targeting RSK4 prevent chemoresistance and metastasis in lung and bladder cancer. Sci Transl Med. 2021 Jul 14
- 2. Xu J et al. Prominent roles of ribosomal S6 kinase 4 (RSK4) in cancer. Pathol Res Pract. 2021 Mar

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