Anti-Phospho-PI3K p85 (Y467) + PI3K p55 (Y199) Antibody [PSH01-38] HA721672

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse
Applications:	WB, IF-Cell
Molecular Wt:	Predicted band size: 84 kDa/55 kDa
Clone number:	PSH01-38
Description:	Phosphatidylinositol 3-kinase regulatory subunit alpha is an enzyme that in humans is encoded by the PIK3R1 gene. Phosphatidylinositol 3-kinase phosphorylates the inositol ring of phosphatidylinositol at the 3-prime position. The enzyme comprises a 110 kD catalytic subunit and a regulatory subunit of either 85, 55, or 50 kD. This gene encodes the 85 kD regulatory subunit. Phosphatidylinositol 3-kinase plays an important role in the metabolic actions of insulin, and a mutation in this gene has been associated with insulin resistance. Alternative splicing of this gene results in three transcript variants encoding different isoforms.
Immunogen:	Synthetic peptide within human PIK3R1 aa 451-500 / 724.
Positive control:	NIH/3T3 treated with 1mM sodium orthovanadate for 30 minutes cell lysate, NIH/3T3 cells treated with 1mM Sodium orthovanadate for 30 minutes, 293T overexpress Src cell lysate, 293T overexpress Src treated with 1mM sodium orthovanadate for 30 minutes cell lysate.
Subcellular location:	Cell-cell junction, cis-Golgi network, cytoplasm, cytosol, membrane, nucleus.
Database links:	SwissProt: P27986 Human P26450 Mouse
Recommended Dilutions: WB IF-Cell	1:1,000 1:100
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

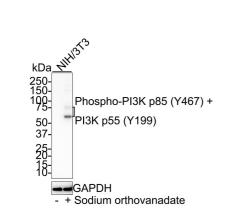
Technical:0086-571-89986345

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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



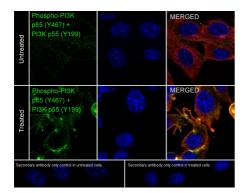


Fig1: Western blot analysis of Phospho-PI3K p85 (Y467) + PI3K p55 (Y199) on different lysates with Rabbit anti-Phospho-PI3K p85 (Y467) + PI3K p55 (Y199) antibody (HA721672) at 1/1,000 dilution.

Lane 1: NIH/3T3 cell lysate Lane 2: NIH/3T3 treated with 1mM sodium orthovanadate for 30 minutes cell lysate

Lysates/proteins at 30 µg/Lane.

Predicted band size: 84 kDa Observed band size: 55/85 kDa

Exposure time: 3 minutes 10 seconds; ECL: K1801;

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 (HA721672) 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 1/100,000 dilution was used for 1 hour at room temperature.

Fig2: Immunocytochemistry analysis of NIH/3T3 cells treated with or without 1mM Sodium orthovanadate for 30 minutes labeling Phospho-PI3K p85 (Y467) + PI3K p55 (Y199) with Rabbit anti-Phospho-PI3K p85 (Y467) + PI3K p55 (Y199) antibody (HA721672) 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-Phospho-PI3K p85 (Y467) + PI3K p55 (Y199) antibody (HA721672) at 1/100 dilution in 1% BSA in PBST overnight at 4 $^{\circ}$ C. Goat Anti-Rabbit IgG H&L (iFluor m 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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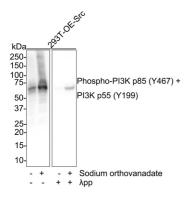


Fig3: Western blot analysis of Phospho-PI3K p85 (Y467) + PI3K p55 (Y199) on different lysates with Rabbit anti-Phospho-PI3K p85 (Y467) + PI3K p55 (Y199) antibody (HA721672) at 1/1,000 dilution.

Lane 1: 293T overexpress Src cell lysate Lane 2: 293T overexpress Src treated with 1mM sodium orthovanadate for 30 minutes cell lysate Lane 3: 293T overexpress Src cell lysate, the membrane treated with λ pp for 1 hour Lane 4: 293T overexpress Src treated with 1mM sodium

orthovanadate for 30 minutes cell lysate, then the membrane treated with λpp for 1 hour

Lysates/proteins at 20 µg/Lane.

Predicted band size: 84 kDa Observed band size: 55 kDa

Exposure time: 28 seconds; ECL: K1801;

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 (HA721672) 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 1/100,000 dilution was used for 1 hour at room temperature.

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

Background References

- 1. Koch PA et al. The molecular mechanisms mediating class II PI 3-kinase function in cell physiology. FEBS J. 2021 Dec
- 2. Acosta-Martinez M et al. The PI3K/Akt Pathway in Meta-Inflammation. Int J Mol Sci. 2022 Dec

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