Anti-PI 3 Kinase p85 alpha Antibody [SU04-07] - BSA and Azide free

HA750162

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, FC
Molecular Wt:	Predicted band size: 84 kDa
Clone number:	SU04-07

Description: Phosphatidylinositol 3-kinase (PI 3-kinase) phosphorylates the 3' OH position of the inositol ring of inositol lipids and is composed of p85 and p110 subunits. PI 3-kinase p85 lacks PI 3-kinase activity and acts as an adapter, coupling p110 to activated protein tyrosine kinase. Two forms of p85 have been described (p85α and p85β), each possessing one SH3 and two SH2 domains. PI 3-kinase p85α, also known as GRB1, phosphatidylinositol 3-kinase regulatory 1 or p85, is a 724 amino acid protein that exists as four alternatively spliced isoforms. Involved in insulin metabolism, defects in the PI 3-kinase p85α gene have been linked to insulin resistance. PI 3-kinase p85α is polyubiquitinated in T-cells by Cbl-b, and has multiple phosphorylated amino acid residues, including a phosphorylated tyrosine residue at position 467.

Immunogen: Synthetic peptide within C-terminal human PI 3 Kinase p85 alpha.

Positive control: HepG2 cell lysate, Jurkat cell lysate, MCF7 cell lysate, NIH/3T3 cell lysate, C6 cell lysate, Mouse brain tissue lysate, Rat brain tissue lysate, Hela, MCF-7, HepG2, NIH/3T3, human brain tissue, human lung tissue, mouse lung tissue, rat lung tissue.

Subcellular location: Cytoplasm, membrane, nucleus.

Database links: SwissProt: P27986 Human | P26450 Mouse | Q63787 Rat

Recommended Dilutions:

WB	1:2,000-1:5,000
IF-Cell	1:50-1:200
IF-Tissue	1:50-1:200
IHC-P	1:50-1:1,000
FC	1:50-1:100

Storage Buffer: 1*PBS (pH7.4).

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃ or -80℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

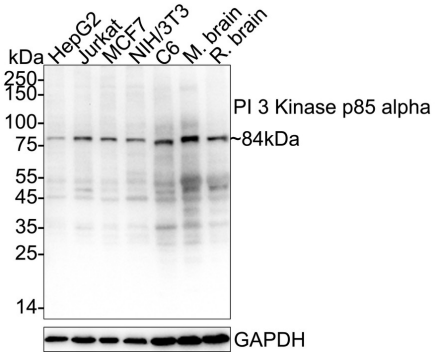

Fig1: Western blot analysis of PI 3 Kinase p85 alpha on different lysates with Rabbit anti-PI 3 Kinase p85 alpha antibody (HA750162) at 1/5,000 dilution.

Lane 1: HepG2 cell lysate (20 µg/Lane)
Lane 2: Jurkat cell lysate (20 µg/Lane)
Lane 3: MCF7 cell lysate (20 µg/Lane)
Lane 4: NIH/3T3 cell lysate (20 µg/Lane)
Lane 5: C6 cell lysate (20 µg/Lane)
Lane 6: Mouse brain tissue lysate (40 µg/Lane)
Lane 7: Rat brain tissue lysate (40 µg/Lane)

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

Exposure time: 1 minute; 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 (HA750162) at 1/5,000 dilution was used in primary antibody dilution (K1803) at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

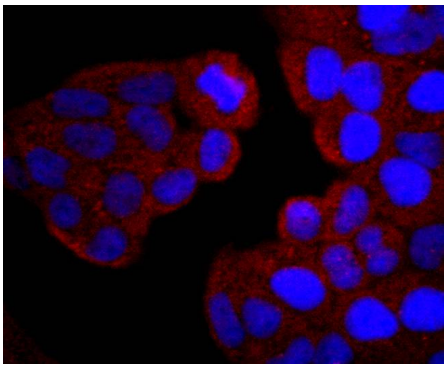

Fig2: ICC staining of PI 3 Kinase p85 alpha in Hela cells (red). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (HA750162, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®594 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

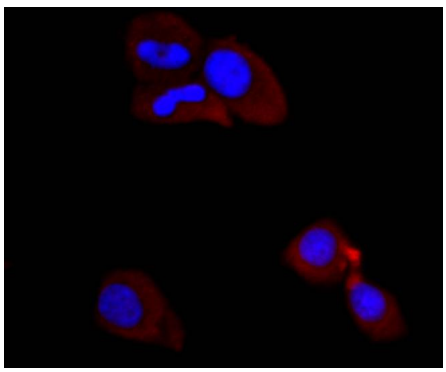

Fig3: ICC staining of PI 3 Kinase p85 alpha in MCF-7 cells (red). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (HA750162, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®594 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

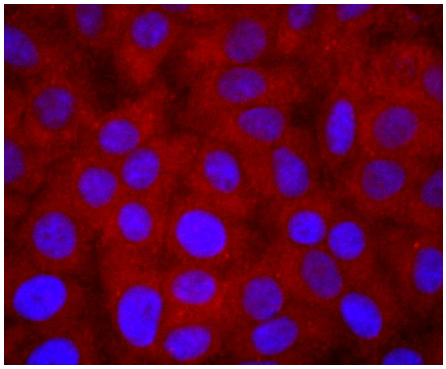

Fig4: ICC staining of PI 3 Kinase p85 alpha in HepG2 cells (red). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (HA750162, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®594 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

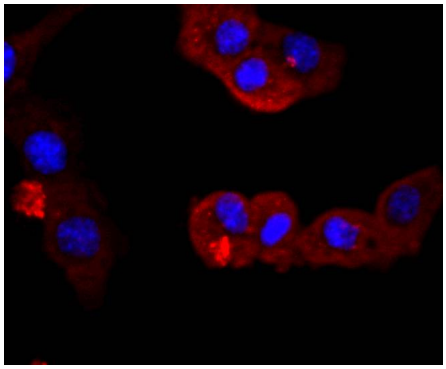

Fig5: ICC staining of PI 3 Kinase p85 alpha in NIH/3T3 cells (red). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (HA750162, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®594 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

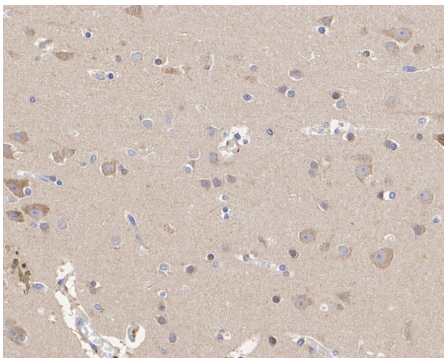

Fig6: Immunohistochemical analysis of paraffin-embedded human brain tissue with Rabbit anti-PI 3 Kinase p85 alpha antibody (HA750162) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750162) 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.

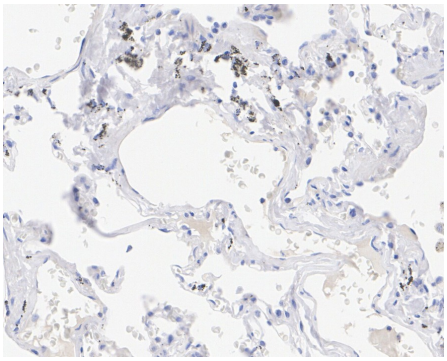

Fig7: Immunohistochemical analysis of paraffin-embedded human lung tissue with Rabbit anti-PI 3 Kinase p85 alpha antibody (HA750162) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750162) 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.

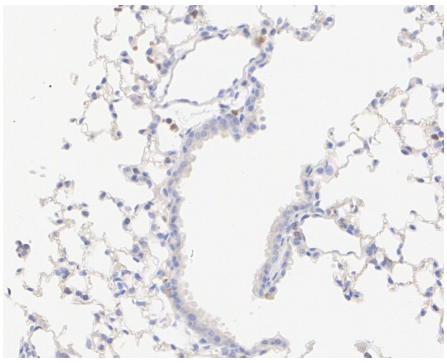

Fig8: Immunohistochemical analysis of paraffin-embedded mouse lung tissue with Rabbit anti-PI 3 Kinase p85 alpha antibody (HA750162) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750162) 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.

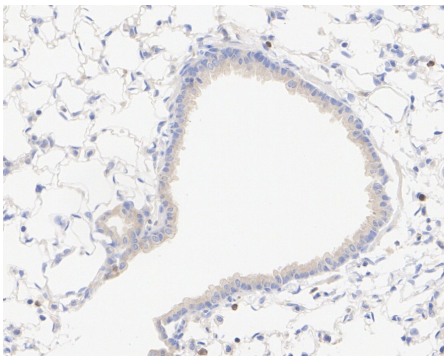

Fig9: Immunohistochemical analysis of paraffin-embedded rat lung tissue with Rabbit anti-PI 3 Kinase p85 alpha antibody (HA750162) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750162) 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.

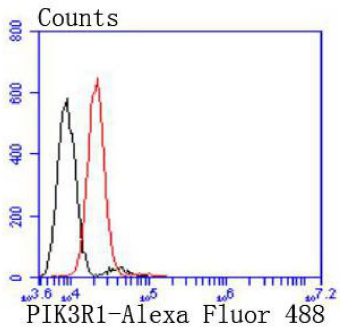

Fig10: Flow cytometric analysis of PI 3 Kinase p85 alpha was done on HepG2 cells. The cells were fixed, permeabilized and stained with the primary antibody (HA750162, 1/50) (red). After incubation of the primary antibody at room temperature for an hour, the cells were stained with a Alexa Fluor 488-conjugated Goat anti-Rabbit IgG Secondary antibody at 1/1000 dilution for 30 minutes.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. Yan LX et al. PIK3R1 targeting by miR-21 suppresses tumor cell migration and invasion by reducing PI3K/AKT signaling and reversing EMT, and predicts clinical outcome of breast cancer. Int J Oncol 48:471-84 (2016).
2. Hu J et al. Filamin B regulates chondrocyte proliferation and differentiation through Cdk1 signaling. PLoS One 9:e89352 (2014).