# Anti-PI3 Kinase p110α Antibody [SJ0186] ET1606-36

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse
Applications:	WB, IF-Cell, IF-Tissue, IP, IHC-P
Molecular Wt:	Predicted band size: 124 kDa
Clone number:	SJ0186
Description:	Phosphoinositide-3-kinase (PI3K) phosphorylates phosphatidylinositol (PI) and its phosphorylated derivatives at position 3 of the inositol ring to produce 3-phosphoinositides. Uses ATP and PtdIns(4,5)P2 (phosphatidylinositol 4,5-bisphosphate) to generate phosphatidylinositol 3,4,5-trisphosphate (PIP3). PIP3 plays a key role by recruiting PH domain-containing proteins to the membrane, including AKT1 and PDPK1, activating signaling cascades involved in cell growth, survival, proliferation, motility and morphology. Participates in cellular signaling in response to various growth factors. Involved in the activation of AKT1 upon stimulation by receptor tyrosine kinases ligands such as EGF, insulin, IGF1, VEGFA and PDGF. Involved in signaling via insulin-receptor substrate (IRS) proteins. Essential in endothelial cell migration during vascular development through VEGFA signaling, possibly by regulating RhoA activity. Required for lymphatic vasculature development, possibly by binding to RAS and by activation by EGF and FGF2, but not by PDGF. Regulates invadopodia formation through the PDPK1-AKT1 pathway. Participates in cardiomyogenesis in embryonic stem cells through a AKT1 pathway. Participates in vasculogenesis in embryonic stem cells through pDK1 and protein kinase C pathway. In addition to its lipid kinase activity, it displays a serine-protein kinase activity that results in the autophosphorylation of the p85alpha regulatory subunit as well as phosphorylation of other proteins such as 4EBP1, H-Ras, the IL-3 beta c receptor and possibly others. Plays a role in the positive regulation of phagocytosis and pinocytosis.
lmmunogen:	Synthetic peptide within Human PIK3CA aa 1,001-1,050 / 1,068.
Positive control:	HepG2 cell lysate, Hela cell lysate, Jurkat cell lysate, NIH/3T3 cell lysate, mouse brain tissue lysate, mouse liver tissue lysate, HepG2 cells, Hela cells, MCF-7 cells, human brain tissue, human liver tissue, mouse liver tissue.
Subcellular location:	Cytosol, plasma membrane, cytoplasm, lamellipodium, membrane, phosphatidylinositol 3-kinase complex, phosphatidylinositol 3-kinase complex, class IA.
Database links:	SwissProt: P42336 Human   P42337 Mouse
Recommended Dilutions: WB IF-Cell IF-Tissue IP IHC-P	1:1,000 1:50-1:200 1:50-1:200 1-2µg/sample 1:50-1:200
Storage Buffer:	1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Shipped at 4°C. Store at +4°C short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20°C long term.
Purity:	Protein A affinity purified.

## Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880

Technical:0086-571-89986345

Service mail:support@huabio.cn



11.

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 PI3 Kinase p110 $\alpha$  on different lysates with Rabbit anti-PI3 Kinase p110 $\alpha$  antibody (ET1606-36) at 1/1,000 dilution.

Lane 1: HepG2 cell lysate Lane 2: Hela cell lysate Lane 3: Jurkat cell lysate Lane 4: NIH/3T3 cell lysate Lane 5: mouse brain tissue lysate (20 µg/Lane) Lane 6: mouse liver tissue lysate (20 µg/Lane)

Lysates/proteins at 10 µg/Lane.

Predicted band size: 124 kDa Observed band size: 110 kDa

Exposure time: 3 minutes;

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 (ET1606-36) at 1/1,000 dilution was used in 5% NFDM/TBST at  $4^{\circ}$ C 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 PI3 Kinase  $p110\alpha$  in HepG2 cells (green). 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 (ET1606-36, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 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 PI3 Kinase p110 $\alpha$  in Hela cells (green). 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 (ET1606-36, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

#### Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880

Technical:0086-571-89986345

Service mail:support@huabio.cn



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





Fig4: ICC staining of PI3 Kinase p110α in MCF-7 cells (green). 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 (ET1606-36, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

Fig5: Immunohistochemical analysis of paraffin-embedded human brain tissue with Rabbit anti-PI3 Kinase p110α antibody (ET1606-36) at 1/50 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<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1606-36) at 1/50 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.

Fig6: Immunohistochemical analysis of paraffin-embedded human liver tissue with Rabbit anti-PI3 Kinase p110a antibody (ET1606-36) at 1/50 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<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1606-36) at 1/50 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 mouse liver tissue with Rabbit anti-PI3 Kinase p110a antibody (ET1606-36) 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<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1606-36) 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.

## Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880

Technical:0086-571-89986345

Service mail:support@huabio.cn

www.huabio.cr

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



**Fig8:** PI3 Kinase p110 $\alpha$  was immunoprecipitated from 0.2 mg HeLa cell lysate with ET1606-36 at 2 µg/25 µl agarose. Western blot was performed from the immunoprecipitate using ET1606-36 at 1/1,000 dilution. Anti-Rabbit IgG for IP Nano-secondary antibody (NBI01H) at 1/5,000 dilution was used for 1 hour at room temperature.

Lane 1: HeLa cell lysate (input) Lane 2: ET1606-36 IP in HeLa cell lysate Lane 3: Rabbit IgG instead of ET1606-36 in HeLa cell lysate

Blocking/Dilution buffer: 5% NFDM/TBST Exposure time: 2 minutes; ECL: K1802

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

#### **Background References**

- 1. Salm, F. et al. 2015. The Phosphoinositide 3-Kinase p110α Isoform Regulates Leukemia Inhibitory Factor Receptor Expression via c-Myc and miR-125b to Promote Cell Proliferation in Medulloblastoma. PloS one. 10: e0123958.
- 2. Akula, M.K. et al. 2016. Control of the innate immune response by the mevalonate pathway. Nature immunology. 17: 922-9.

## Hangzhou Huaan Biotechnology Co., Ltd.



**Orders:**0086–571–88062880

Technical:0086-571-89986345

Service mail:support@huabio.cn

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