Anti-PDGFR alpha Antibody [JF104-6]

ET1702-49

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IHC-P, FC, IHC-Fr
Molecular Wt:	Predicted band size: 123 kDa
Clone number:	JF104-6
Description:	Platelet-derived growth factor (PDGF) is a mitogen for mesenchyme- and glia-derived cells. PDGF consists of two chains, A and B, which dimerize to form functionally distinct isoforms, PGDF-AA, PDGF-AB and PDGF-BB. These three isoforms bind with different affinities to two receptor types, PDGFR- α and - β , which are endowed with protein tyrosine kinase domains. PDGFR- α can bind to both A and B subunits of PDGF, while PDGFR- β can only bind the B subunit. Ligand binding promotes either homo- or heterodimerization of the PDGF receptors in a specific manner. PDGF-AA induces the dimerization of two α receptors, PDGF-AB induces dimerization of $\alpha\alpha$ and $\alpha\beta$ and PDGF-BB induces the formation of three types of dimers, $\alpha\alpha$, $\alpha\beta$ and $\beta\beta$. Translocation of the PDGFR- β gene with the Tel gene is linked to chronic myelomonocytic leukemia (CMML), a myelodysplastic syndrome, and demonstrates the oncogenic potential of the PDGF receptors.
lmmunogen:	Synthetic peptide within C-terminal human PDGFR alpha.
Positive control:	Mouse embryonic femur tissue, mouse embryonic intervertebral disc tissue, mouse embryonic lung tissue, rat embryonic femur tissue, rat embryonic intervertebral disc tissue, rat embryonic lung tissue, rat endometrium tissue, human colon tissue, human colon cancer tissue, human glioblastoma tissue, NIH/3T3 cell lysates, SHG-44 cell lysates, A549, NIH/3T3.
Subcellular location:	Golgi apparatus, Cell membrane, cilium.
Database links:	SwissProt: P16234 Human P26618 Mouse P20786 Rat
Recommended Dilutions: WB IHC-P IHC-Fr FC	1:1,000 1:500-1:2,000 1:200 1:50-1:100
Storage Buffer:	1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Store at +4 $^\circ\!{\rm C}$ after thawing. Aliquot store at -20 $^\circ\!{\rm C}$ or -80 $^\circ\!{\rm C}$. Avoid repeated freeze / thaw cycles.
Purity:	Protein A affinity purified.

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Images



Fig1: Immunofluorescence analysis of frozen mouse P0 brain tissue with Rabbit anti-PDGFR alpha antibody (ET1702-49) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for about 2 minutes in microwave oven. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (ET1702-49, green) at 1/200 dilution overnight at 4 $^{\circ}$ C, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor TM 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).



Fig2: Immunohistochemical analysis of paraffin-embedded mouse embryonic femur tissue with Rabbit anti-PDGFR alpha antibody (ET1702-49) at 1/2,000 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 ddH₂O and PBS, and then probed with the primary antibody (ET1702-49) at 1/2,000 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.



Fig3: Immunohistochemical analysis of paraffin-embedded mouse embryonic intervertebral disc tissue with Rabbit anti-PDGFR alpha antibody (ET1702-49) at 1/2,000 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 ddH₂O and PBS, and then probed with the primary antibody (ET1702-49) at 1/2,000 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.

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Fig4: Immunohistochemical analysis of paraffin-embedded mouse embryonic lung tissue with Rabbit anti-PDGFR alpha antibody (ET1702-49) at 1/2,000 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 ddH₂O and PBS, and then probed with the primary antibody (ET1702-49) at 1/2,000 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: Immunohistochemical analysis of paraffin-embedded rat embryonic femur tissue with Rabbit anti-PDGFR alpha antibody (ET1702-49) at 1/2,000 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 ddH₂O and PBS, and then probed with the primary antibody (ET1702-49) at 1/2,000 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.



Fig6: Immunohistochemical analysis of paraffin-embedded rat embryonic intervertebral disc tissue with Rabbit anti-PDGFR alpha antibody (ET1702-49) at 1/500 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 ddH₂O and PBS, and then probed with the primary antibody (ET1702-49) at 1/500 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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Fig7: Immunohistochemical analysis of paraffin-embedded rat embryonic lung tissue with Rabbit anti-PDGFR alpha antibody (ET1702-49) at 1/2,000 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 ddH₂O and PBS, and then probed with the primary antibody (ET1702-49) at 1/2,000 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 rat endometrium tissue with Rabbit anti-PDGFR alpha antibody (ET1702-49) 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 ddH₂O and PBS, and then probed with the primary antibody (ET1702-49) 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 human colon tissue with Rabbit anti-PDGFR alpha antibody (ET1702-49) at 1/500 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 ddH₂O and PBS, and then probed with the primary antibody (ET1702-49) at 1/500 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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Fig10: Immunohistochemical analysis of paraffin-embedded human colon cancer tissue with Rabbit anti-PDGFR alpha antibody (ET1702-49) at 1/500 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 ddH₂O and PBS, and then probed with the primary antibody (ET1702-49) at 1/500 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.



Fig11: Immunohistochemical analysis of paraffin-embedded human glioblastoma tissue with Rabbit anti-PDGFR alpha antibody (ET1702-49) at 1/500 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 ddH₂O and PBS, and then probed with the primary antibody (ET1702-49) at 1/500 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.

Fig12: Western blot analysis of PDGFR alpha on NIH/3T3 cell lysates with Rabbit anti-PDGFR alpha antibody (ET1702-49) at 1/1,000 dilution.

Lysates/proteins at 10 µg/Lane.

Predicted band size: 123 kDa Observed band size: 180 kDa

Exposure time: 2 minutes;

6% 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 (ET1702-49) 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 1:200,000 dilution was used for 1 hour at room temperature.



kDa WH¹³¹³ 250- PDGFR alpha ~180kDa 150-100-75-



Fig13: Western blot analysis of PDGFR alpha on different lysates with Rabbit anti-PDGFR alpha antibody (ET1702-49) at 1/500 dilution.

Lane 1: NIH/3T3-si NT cell lysate Lane 2: NIH/3T3-si PDGFR alpha cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 123 kDa Observed band size: 190 kDa

Exposure time: 2 minutes;

4-20% SDS-PAGE gel.

ET1702-49 was shown to specifically react with PDGFR alpha in Hela-si NT cells. Weakened band was observed when Hela-si PDGFR alpha sample was tested. Hela-si NT and Hela-si PDGFR alpha samples were subjected to SDS-PAGE. Proteins were transferred to a PVDF membrane and blocked with 5% NFDM in TBST for 1 hour at room temperature. The primary antibody (ET1702-49, 1/500) and Loading control antibody (Rabbit anti-GAPDH, ET1601-4, 1/10,000) were used in 5% BSA at room temperature for 2 hours. Goat Anti-rabbit IgG-HRP Secondary Antibody (HA1001) at 1:300,000 dilution was used for 1 hour at room temperature.



Fig14: Flow cytometric analysis of PDGFR alpha was done on NIH/3T3 cells. The cells were fixed, permeabilized and stained with the primary antibody (ET1702-49, 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. Benedykcinska A et al. Generation of brain tumours in mice by Cre-mediated recombination of neural progenitors in situ with the tamoxifen metabolite endoxifen. Dis Model Mech 9:211-20 (2016).
- 2. Rondahl V et al. Lrig2-deficient mice are protected against PDGFB-induced glioma. PLoS One 8:e73635 (2013).

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