abcam

Product datasheet

Anti-PDGFR alpha antibody [EPR5480] ab134123

KO VALIDATED Recombinant RabMAb

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Overview

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Product name	Anti-PDGFR alpha antibody [EPR5480]
Description	Rabbit monoclonal [EPR5480] to PDGFR alpha
Host species	Rabbit
Specificity	Mouse and rat species are recommended based on WB result. We do not guarantee IHC-P for mouse and rat.
Tested applications	Suitable for: Flow Cyt (Intra), Indirect ELISA, WB, IHC-P
Species reactivity	Reacts with: Mouse, Rat, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	IHC-P: Human colonic carcinoma, human colon tissue. WB: SH SY5Y, Human skeletal muscle, Human spleen, and Human fetal brain lysates. Flow Cyt (intra): SH-SY5Y, L6 cells.
General notes	 This product is a recombinant monoclonal antibody, which offers several advantages including: High batch-to-batch consistency and reproducibility Improved sensitivity and specificity Long-term security of supply Animal-free production For more information <u>see here</u>. Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to <u>RabMAb[®] patents</u>.

Properties	
Form	Liquid
Storage instructions	Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C.
Storage buffer	pH: 7.2 Preservative: 0.05% Sodium azide Constituents: 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue culture supernatant
Purity	Protein A purified
Clonality	Monoclonal

Clone number	EPR5480
lsotype	lgG

Applications

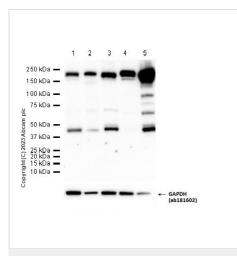
The Abpromise guarantee Our <u>Abpromise guarantee</u> covers the use of ab134123 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		1/20.
Indirect ELISA		Use at an assay dependent concentration.
WB		1/1000 - 1/10000. Detects a band of approximately 190 kDa (predicted molecular weight: 122 kDa).
IHC-P	★ ★ ☆ ☆ ☆ <u>(4)</u>	1/500. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See IHC antigen retrieval protocols .

Target	
Function	Receptor that binds both PDGFA and PDGFB and has a tyrosine-protein kinase activity.
Tissue specificity	Expressed in primary and metastatic colon tumors and in normal colon tissue. Tumors may express a different isoform to that found in normal tissue.
Involvement in disease	Note=A chromosomal aberration involving PDGFRA is found in some cases of hypereosinophilic syndrome. Interstitial chromosomal deletion del(4)(q12q12) causes the fusion of FIP1L1 and PDGFRA (FIP1L1-PDGFRA).
Sequence similarities	Belongs to the protein kinase superfamily. Tyr protein kinase family. CSF-1/PDGF receptor subfamily. Contains 5 lg-like C2-type (immunoglobulin-like) domains. Contains 1 protein kinase domain.
Cellular localization	Membrane.

Images



Western blot - Anti-PDGFR alpha antibody [EPR5480] (ab134123) **All lanes :** Anti-PDGFR alpha antibody [EPR5480] (ab134123) at 1/1000 dilution

Lane 1 : Rat heart tissue lysate

Lane 2 : Rat lung tissue lysate

Lane 3 : Mouse brain tissue lysate

Lane 4 : Mouse heart tissue lysate

Lane 5 : Mouse lung tissue lysate

Lysates/proteins at 20 µg per lane.

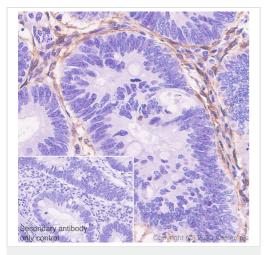
Secondary

All lanes : Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

Predicted band size: 122 kDa Observed band size: 150 kDa

Exposure time: 80 seconds

Blocking/dilution buffer: 5% NFDM/TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PDGFR alpha antibody [EPR5480] (ab134123) Immunohistochemical analysis of paraffin-embedded Human colonic carcinoma tissue labeling PDGFR alpha with ab134123 at 1/500 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Positive staining on stromal cells in human colonic carcinoma. The section was incubated with ab134123 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Western blot - Anti-PDGFR alpha antibody [EPR5480] (ab134123)

All lanes : Anti-PDGFR alpha antibody [EPR5480] (ab134123) at 1/1000 dilution

Lane 1 : Wild-type SH-SY5Y cell lysate Lane 2 : PDGFRA knockout SH-SY5Y cell lysate

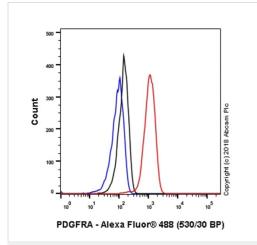
Lysates/proteins at 40 µg per lane.

Performed under reducing conditions.

Predicted band size: 122 kDa Observed band size: 180 kDa

Lanes 1 - 2: Merged signal (red and green). Green - ab134123 observed at 180 kDa. Red - loading control <u>ab8245</u> (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

ab134123 was shown to react with PDGFR alpha in wild-type SH-SY5Y cells in Western blot with loss of signal observed in PDGFRA knockout cell line <u>ab275335</u> (knockout cell lysate <u>ab275522</u>). Wildtype SH-SY5Y and PDGFRA knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween[®]) before incubation with ab134123 and <u>ab8245</u> (Mouse anti-GAPDH antibody [6C5]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye[®] 800CW) preabsorbed (<u>ab216773</u>) and Goat anti-Mouse IgG H&L (IRDye[®] 680RD)



Flow Cytometry (Intracellular) - Anti-PDGFR alpha

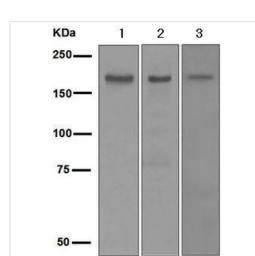
antibody [EPR5480] (ab134123)

Intracellular Flow Cytometry analysis of L6 (Rat skeletal muscle myoblast) cells labelling PDGFR alpha with ab134123 at 1/20 dilution (14.65 µg/ml) (Red). Cells were fixed with 4%

preabsorbed (ab216776) secondary antibodies at 1 in 20000

dilution for 1 h at room temperature before imaging.

paraformaldehyde . Goat anti rabbit lgG (Alexa Fluor[®] 488, <u>ab150077</u>) was used as the secondary antibody at 1/2000 dilution. lsotype control - Rabbit monoclonal lgG (<u>ab172730</u>) (Black). Unlabeled control - Unlabelled cells (Blue).



Western blot - Anti-PDGFR alpha antibody [EPR5480] (ab134123) **All lanes :** Anti-PDGFR alpha antibody [EPR5480] (ab134123) at 1/1000 dilution

Lane 1 : Human skeletal muscle lysate Lane 2 : Human spleen lysate

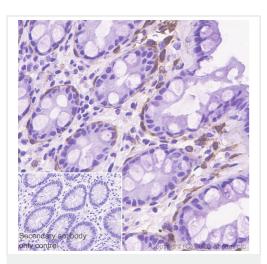
Lane 3 : Human fetal brain lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat anti-rabbit HRP conjugated antibody at 1/2000 dilution

Predicted band size: 122 kDa Observed band size: 190 kDa

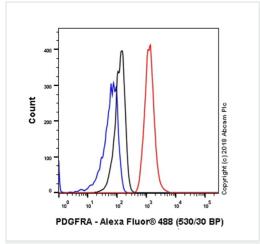


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PDGFR alpha antibody [EPR5480] (ab134123)

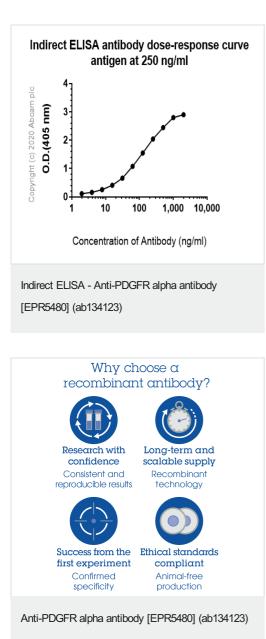
Immunohistochemical analysis of paraffin-embedded Human colon tissue labeling PDGFR alpha with ab134123 at 1/500 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Positive staining on stromal cells in human colon (PMID: 23429582). The section was incubated with ab134123 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Flow Cytometry (Intracellular) - Anti-PDGFR alpha antibody [EPR5480] (ab134123) Intracellular Flow Cytometry analysis of SH-SY5Y (Human neuroblastoma epithelial cell) cells labelling PDGFR alpha with ab134123 at 1/20 dilution (14.65µg/ml) (Red). Cells were fixed with 4% paraformaldehyde . Goat anti rabbit IgG (Alexa Fluor[®] 488, **ab150077**) was used as the secondary antibody at 1/2000 dilution. Isotype control - Rabbit monoclonal IgG (**ab172730**) (Black). Unlabeled control - Unlabelled cells (Blue).



ELISA analysis of Human PDGFR alpha recombinant protein at 250 ng/mL with ab134123. An Alkaline Phosphatase-conjugated AffiniPure Goat Anti-Rabbit lgG (H+L) at 1/2500 dilution was used as the secondary antibody.

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