abcam

Product datasheet

Anti-PDGFR alpha antibody [EPR22059-270] ab203491 敲除 验证 重组 RabMAb

★★★★★ 4 Abreviews 22 References 19 图像

概述

产 品名称	Anti-PDGFR alpha 抗体 [EPR22059-270]	
描述	兔单克隆抗体[EPR22059-270] to PDGFR alpha	
宿主	Rabbit	
特异性	PDGFR alpha is membrane protein, so enrichment of membrane could help increasing the detection level of PDGFR alpha.	
经测试应 用	适用于: WB, IHC-P, ICC/IF, Flow Cyt, IP, Indirect ELISA, IHC-Fr	
种属反应性	与反应: Mouse, Rat, Human	
免疫原	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.	
阳性 对照	WB: SH-SY5Y, A-204, MG-63, C6, and NIH/3T3 whole cell lysates. IHC-P: Mouse E14.5 lung, E14.5 intervertebral disc and uterus tissues; Rat E14.5 intervertebral disc tissue and Human endometrium tissue. ICC/IF: SH-SY5Y and A-204 cells. Flow Cyt: NIH/3T3 and A-204 cells. IP: A-204 whole cell lysate IHC-Fr: Human kidney tissue sections.	
常 规说 明	 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>. 	

性能	
形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
存储溶液	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol, 0.05% BSA
纯 度	Protein A purified

克隆	单 克隆
克 隆 编号	EPR22059-270
同种型	lgG

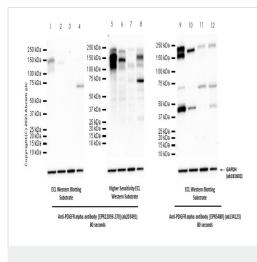
应用

The Abpromise guarantee _____ Abpromise ™承诺保证使用ab203491于以下的经测试应用

"应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应 用	Ab评论	说明
WB		1/1000. Detects a band of approximately 150, 180 kDa (predicted molecular weight: 122 kDa).
ІНС-Р	★★★★	Use a concentration of 0.26 µg/ml. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF	★★★★★ <u>(1)</u>	1/500.
Flow Cyt		1/50.
IP		1/30.
Indirect ELISA		Use a concentration of 1 µg/ml.
IHC-Fr	★ ★ ★ ★ ★ ★ (1)	1/500.

靶标 功能 Receptor that binds both PDGFA and PDGFB and has a tyrosine-protein kinase activity. 组织特异性 Expressed in primary and metastatic colon tumors and in normal colon tissue. Tumors may express a different isoform to that found in normal tissue. 疾病相关 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). 序列相似性 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. 细胞定位 Membrane.



Western blot - Anti-PDGFR alpha antibody [EPR22059-270] (ab203491) Lanes 1-8 : Anti-PDGFR alpha antibody [EPR22059-270] (ab203491) at 1/1000 dilution Lanes 9-12 : Anti-PDGFR alpha antibody [EPR5480] (<u>ab134123</u>) at 1/1000 dilution

Lanes 1 & 5 & 9 : SH-SY5Y Lanes 2 & 6 & 10 : Human brain tissue lysate at 20 µg Lanes 3 & 7 & 11 : Human heart tissue lysate at 20 µg Lanes 4 & 8 & 12 : Human lung tissue lysate at 20 µg

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.

We recommend using higher sensitivity ECL to improve results.

<u>ab134123</u> can be a good alternative when testing samples with low level of PDGFR alpha which detects stronger signal than ab203491 in western blot.

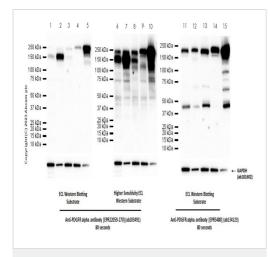
Lanes 1-10 : Anti-PDGFR alpha antibody [EPR22059-270] (ab203491) at 1/1000 dilution Lanes 11-15 : Anti-PDGFR alpha antibody [EPR5480] (ab134123) at 1/1000 dilution

Lanes 1 & 6 & 11 : Rat heart tissue lysate Lanes 2 & 7 & 12 : Rat lung tissue lysate Lanes 3 & 8 & 13 : Mouse brain tissue lysate Lanes 4 & 9 & 14 : Mouse heart tissue lysate Lanes 5 & 10 & 15 : 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.

We recommend using higher sensitivity ECL to improve results.

<u>ab134123</u> can be a good alternative when testing samples with low level of PDGFR alpha which detects stronger signal than ab203491 in western blot.

All lanes : Anti-PDGFR alpha antibody [EPR22059-270] (ab203491) at 1/1000 dilution

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

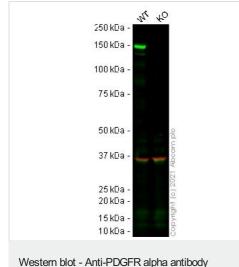
Lysates/proteins at 40 µg per lane.

Performed under reducing conditions.

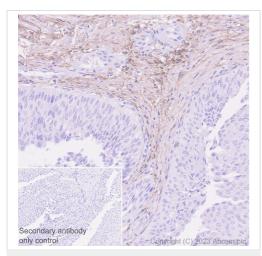
Predicted band size: 122 kDa Observed band size: 150 kDa

Lanes 1 - 2: Merged signal (red and green). Green - ab203491 observed at 150 kDa. Red - loading control <u>ab8245</u> (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

ab203491 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 ab203491 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)



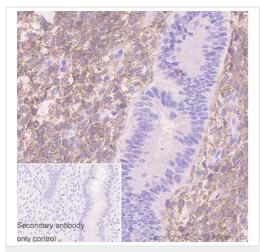
[EPR22059-270] (ab203491)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PDGFR alpha antibody [EPR22059-270] (ab203491)

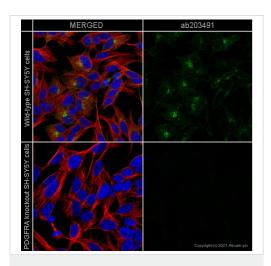
preabsorbed (<u>ab216776</u>) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.

Immunohistochemical analysis of paraffin-embedded human human bladder carcinoma labeling PDGFR alpha with ab203491 at 0.26 µg/mL followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Positive staining in human bladder carcinoma was observed. The section was incubated with ab203491 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 solution 2) for 20 mins was used.

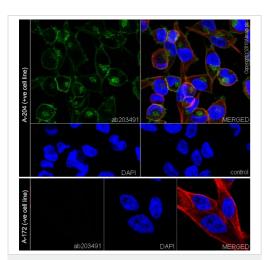


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PDGFR alpha antibody [EPR22059-270] (ab203491)

Immunohistochemical analysis of paraffin-embedded human endometrium tissue labeling PDGFR alpha with ab203491 at 0.26 µg/mL followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Positive staining in human endometrium was observed. The section was incubated with ab203491 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 solution 2) for 20 mins was used.



Immunocytochemistry/ Immunofluorescence - Anti-PDGFR alpha antibody [EPR22059-270] (ab203491)



Immunocytochemistry/ Immunofluorescence - Anti-PDGFR alpha antibody [EPR22059-270] (ab203491)

ab203491 staining PDGFR alpha in wild-type SH-SY5Y cells (top panel) and PDGFRA knockout SH-SY5Y cells (**ab275335**) (bottom panel). The cells were fixed with 100% methanol (5 min) then permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab203491 at 1µg/ml concentration and **ab7291** (Mouse monoclonal to alpha Tubulin) at 1/1000 dilution overnight at 4°C followed by a further incubation at room temperature for 1h with a goat secondary antibody to rabbit IgG (Alexa Fluor[®] 488) (**ab150081**) at 2 µg/ml (shown in green) and a goat secondary antibody to mouse IgG (Alexa Fluor[®] 594) (**ab150120**) at 2 µg/ml (shown in red). Nuclear DNA was labelled in blue with DAPI.

Image was taken with a confocal microscope (Leica-Microsystems TCS SP8).

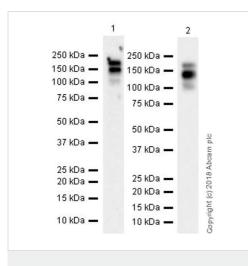
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized A-204 (human muscle rhabdomyosarcoma cell line) cells and A-172 (human brain glioblastoma cell line) labeling PDGFR alpha with ab203491 at 1/500 dilution followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) (**ab150077**) secondary antibody at 1/1000 dilution (green).

Confocal image showing membranous and cytoplasmic staining in A-204 cell line.

Negative control: A-172 (PMID:8425771.

The nuclear counter stain is DAPI (blue). Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor[®] 594) (**ab195889**) (red) at 1/200 dilution.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) (**ab150077**) secondary antibody at 1/1000 dilution.



Western blot - Anti-PDGFR alpha antibody [EPR22059-270] (ab203491) **All lanes :** Anti-PDGFR alpha antibody [EPR22059-270] (ab203491) at 1/1000 dilution

Lane 1 : C6 (rat glial tumor cell line) whole cell lysate Lane 2 : NIH/3T3 (mouse embyro fibroblast cell line) whole cell lysate

Lysates/proteins at 10 µg per lane.

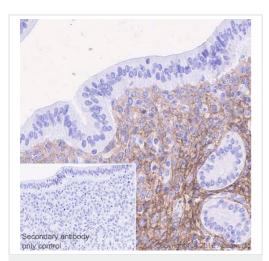
Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 122 kDa Observed band size: 150,180 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

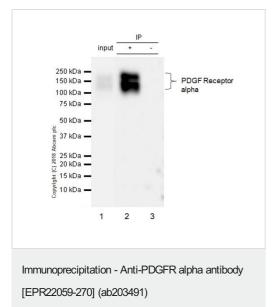
Exposure time : Lane 1: 3 minutes; Lane 2: 59 seconds.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PDGFR alpha antibody [EPR22059-270] (ab203491)

Immunohistochemical analysis of paraffin-embedded mouse uterus tissue labeling PDGFR alpha with ab203491 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Positive staining on interstitial cells of mouse uterus (PMID: 25788664). Counter stained with hematoxylin. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) Ready to use.



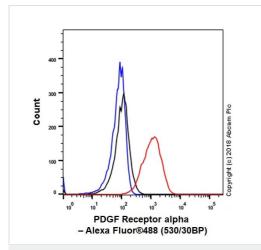
PDGFR alpha was immunoprecipitated from 0.35 mg of A-204 (human muscle rhabdomyosarcoma cell line) whole cell lysate with ab203491 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab203491 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (<u>ab131366</u>), was used for detection at 1/1000 dilution.

Lane 1: A-204 whole cell lysate 10 µg (Input).

Lane 2: ab203491 IP in A-204 whole cell lysate.

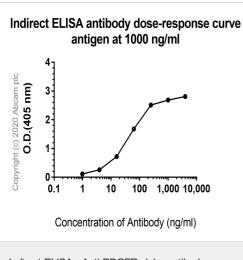
Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab203491 in A-204 whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST. Exposure time: 50 seconds.



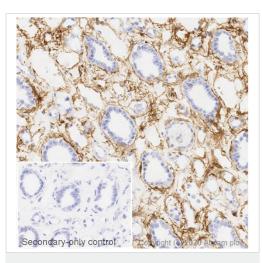
Flow Cytometry - Anti-PDGFR alpha antibody [EPR22059-270] (ab203491) Flow cytometric analysis of NIH/3T3 (mouse embyro fibroblast cell line) cell line labeling PDGFR alpha with ab203491 at 1/500 dilution (red) compared with a Rabbit IgG, monoclonal [EPR25A] - Isotype Control (<u>ab172730</u>) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) (<u>ab150077</u>) at 1/2000 dilution was used as the secondary antibody.

Gated on viable cells.



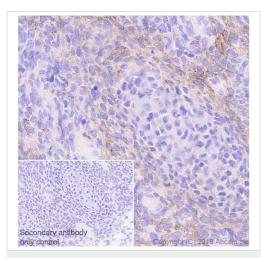
Indirect ELISA using ab203491 at varying antibody concentrations (4000~0 ng /ml) and Human PDGF Receptor alpha antigen at 1000 ng/ml. Alkaline Phosphatase-conjugated AffiniPure Goat Anti-Rabbit IgG (H+L) at 1/2500 dilution dilution was used as a secondary antibody.

Indirect ELISA - Anti-PDGFR alpha antibody [EPR22059-270] (ab203491)



Immunohistochemistry (Frozen sections) - Anti-PDGFR alpha antibody [EPR22059-270] (ab203491) IHC image of PDGFR alpha staining in a section of frozen normal human kidney performed on a Leica BONDTM system using the standard protocol. The section was fixed in 10% paraformaldehyde (10 min) prior to staining. The section was incubated with ab203491, 1/500 dilution, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. The inset secondary-only control image is taken from an identical assay without primary antibody.

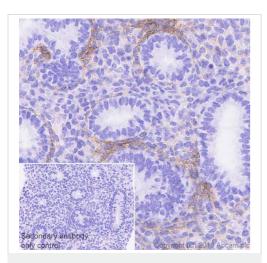
For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PDGFR alpha antibody [EPR22059-270] (ab203491)

Immunohistochemical analysis of paraffin-embedded rat E14.5 intervertebral disc tissue labeling PDGFR alpha with ab203491 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Positive staining in mesenchymal cells of rat E14.5 intervertebral disc (PMID: 9199674). Counter stained with hematoxylin. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

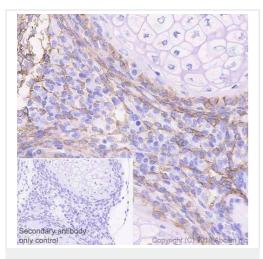
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) Ready to use.



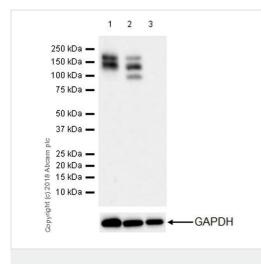
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PDGFR alpha antibody [EPR22059-270] (ab203491)

Immunohistochemical analysis of paraffin-embedded mouse E14.5 lung tissue labeling PDGFR alpha with ab203491 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Positive staining in the mesenchyme of mouse E14.5 lung (PMID: 8681381). Counter stained with hematoxylin. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) Ready to use.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PDGFR alpha antibody [EPR22059-270] (ab203491)



Western blot - Anti-PDGFR alpha antibody [EPR22059-270] (ab203491)

Immunohistochemical analysis of paraffin-embedded mouse E14.5 intervertebral disc tissue labeling PDGFR alpha with ab203491 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Positive staining in mesenchymal cells of mouse E14.5 intervertebral disc (PMID: 9199674). Counter stained with hematoxylin. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

All lanes : Anti-PDGFR alpha antibody [EPR22059-270] (ab203491) at 1/1000 dilution

Lane 1 : A-204 (Human muscle rhabdomyosarcoma cell line) whole cell lysate

Lane 2 : MG-63 (human osteosarcoma fibroblast cell line) whole cell lysate

Lane 3 : A-172 (human brain glioblastoma cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

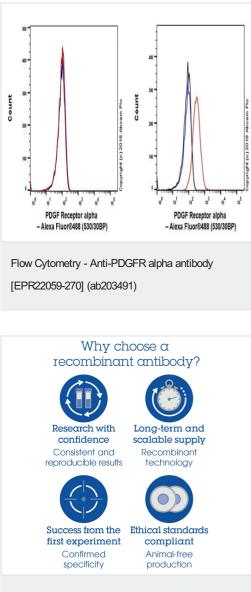
Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 122 kDa Observed band size: 150,180 kDa

Exposure time: 92 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.



Anti-PDGFR alpha antibody [EPR22059-270] (ab203491)

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Flow cytometric analysis of A-172 (human brain glioblastoma cell line, left) and A-204 (human muscle rhabdomyosarcoma cell line, right) cell lines labeling PDGFR alpha with ab203491 at 1/50 dilution (red) compared with a Rabbit IgG, monoclonal [EPR25A] -Isotype Control (<u>ab172730</u>) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) (<u>ab150077</u>) at 1/2000 dilution was used as the secondary antibody.

Gated on viable cells.

• We investigate all quality concerns to ensure our products perform to the highest standards

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