

Product datasheet

Anti-Pan Trk antibody [EP1058Y] ab76291

Recombinant RabMAb

★★★★☆ 11 Abreviews 32 References 20 Images

Overview

Product name	Anti-Pan Trk antibody [EP1058Y]
Description	Rabbit monoclonal [EP1058Y] to Pan Trk
Host species	Rabbit
Specificity	This antibody detects both phosphorylated and unphosphorylated Pan Trk. Based on the WB and FC data, this antibody has relatively lower affinity to TrkC compared to TrkA and TrkB.
Tested applications	Suitable for: ELISA, ICC/IF, WB, IP, IHC-P, Flow Cyt
Species reactivity	Reacts with: Mouse, Rat, Human
Immunogen	Synthetic peptide within Human Pan Trk. The exact sequence is proprietary. A synthetic peptide corresponding to residues surrounding tyrosine 791 of human Pan Trk. Database link: P04629
Positive control	WB: Human, mouse and rat brain tissue lysates IHC-P: Human, mouse and rat cerebrum tissue ICC/IF: U87-MG and SH-SY5Y cells; Mouse DRG neurons; mouse primary neuron. Flow Cyt: SH-SY5Y cells. IP: Rat and mouse brain tissue lysate.
General notes	<p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"> - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production <p>For more information see here.</p> <p>Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents.</p> <p>We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.</p>

Properties

Form Liquid

Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EP1058Y
Isotype	IgG

Applications

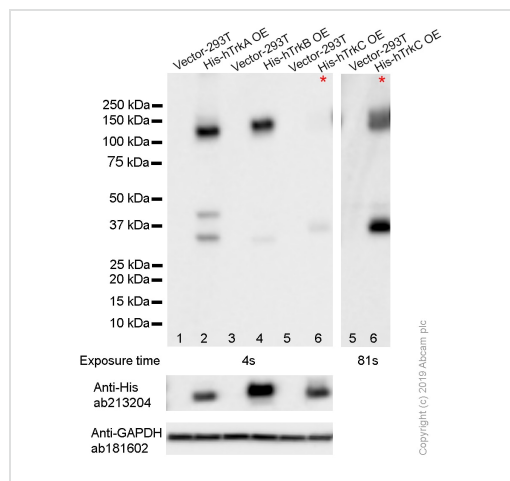
Our [Abpromise guarantee](#) covers the use of **ab76291** in the following tested applications.

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

Application	Abreviews	Notes
ELISA		Use at an assay dependent concentration.
ICC/IF	★★★★☆	1/100.
WB	★★★★★	1/1000. Predicted molecular weight: 87 kDa.
IP		1/40.
IHC-P	★★★★☆	1/1000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See IHC antigen retrieval protocols .
Flow Cyt	★★★★☆	1/130. Paraformaldehyde or methanol fixed cells, permeabilization recommended. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Target

Relevance	Family of neurotrophic tyrosine kinase (NTRK1/2/3) genes which encode TrkA, TrkB and TrkC protein kinases. The three family members are activated by different neurotrophins: TrkA is activated by Nerve growth factor (NGF), TrkB by Brain-derived neurotrophic factor (BDNF) or neurotrophin-4 (NT-4) and TrkC by NT-3. Neurotrophin signalling activates cellular pathways involved in the development and the maturation of the central and peripheral nervous systems through regulation of proliferation, differentiation and survival of sympathetic and nervous neurons. Localization TrkA: Cell membrane. Early endosome membrane. Late endosome membrane. Internalized to endosomes upon binding of NGF or NT-3 and further transported to the cell body via a retrograde axonal transport. Localized at cell membrane and early endosomes before nerve growth factor (NGF) stimulation. Recruited to late endosomes after NGF stimulation. Colocalized with RAPGEF2 at late endosomes (By similarity). TrkB: Membrane. TrkC: Membrane.
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Western blot - Anti-Pan Trk antibody [EP1058Y] (ab76291)

All lanes : Anti-Pan Trk antibody [EP1058Y] (ab76291) at 1/1000 dilution

Lanes 1 & 3 & 5 : Empty vector over expression 293T whole cell lysates

Lane 2 : His-human TrkA overexpression 293T whole cell lysates

Lane 4 : His-human TrkB overexpression 293T whole cell lysates

Lane 6 : His-human TrkC overexpression 293T whole cell lysates

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

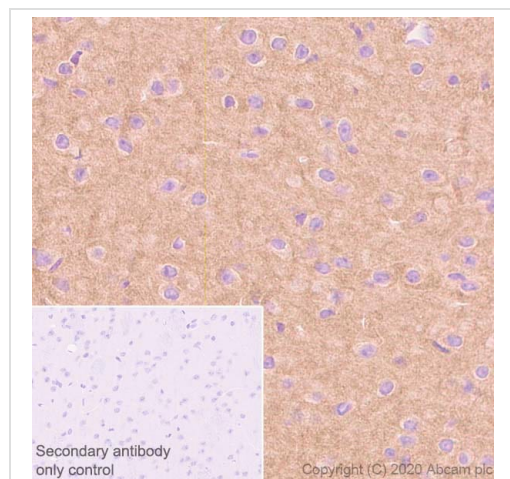
Predicted band size: 87 kDa

Observed band size: 120 kDa

[why is the actual band size different from the predicted?](#)

Blocking/Diluting buffer and concentration: 5% NFDm/TBST

This antibody has relatively lower affinity to TrkC compared to TrkA and TrkB.

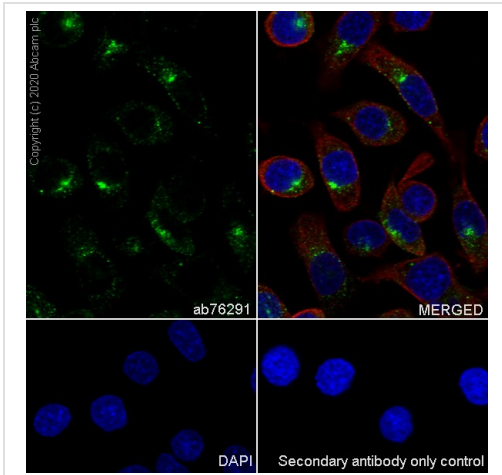


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Pan Trk antibody [EP1058Y] (ab76291)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse cerebrum tissue labelling Pan Trk with purified ab76291 at 1/1000. Rabbit specific IHC polymer detection kit HRP/DAB (ab209101) was used as the secondary antibody. Heat mediated antigen retrieval was performed using Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 minutes. Sections were counterstained with hematoxylin. Negative control using PBS instead of primary antibody.

Positive staining on mouse cerebrum.

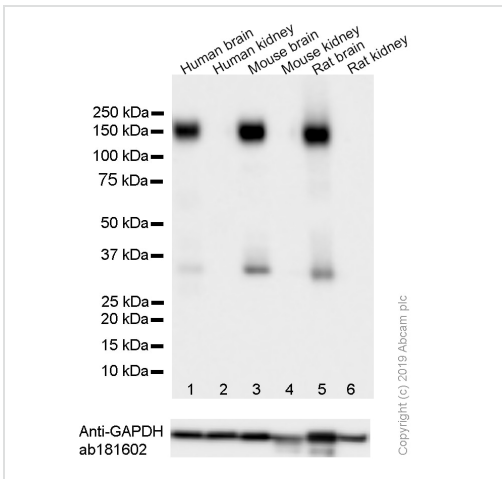
The section was incubated with ab76291 for 30 mins at room temperature. The immunostaining staining was performed on a Leica Biosystems BOND® RX instrument.



Immunocytochemistry/ Immunofluorescence - Anti-Pan Trk antibody [EP1058Y] (ab76291)

Immunocytochemistry/immunofluorescence analysis of SH-SY5Y (Human neuroblastoma epithelial cell) labeling pan Trk with ab76291 at 1/100 dilution (7 µg/mL). [ab150077](#), AlexaFluor®488 Goat anti-Rabbit secondary at 1/1000 (2 µg/mL) was used as the secondary antibody. Cells were fixed with 4% Paraformaldehyde and permeabilised with 0.1% TritonX-100. DAPI (blue) was used as nuclear counterstain. Cells were counterstained with [ab195889](#), anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) at 1/200 (2.5 µg/mL).

Confocal image showing cytoplasmic staining in SH-SY5Y cell line.



Western blot - Anti-Pan Trk antibody [EP1058Y] (ab76291)

All lanes : Anti-Pan Trk antibody [EP1058Y] (ab76291) at 1/1000 dilution

- Lane 1** : Human brain lysates
- Lane 2** : Human kidney lysates
- Lane 3** : Mouse brain lysates
- Lane 4** : Mouse kidney lysates
- Lane 5** : Rat brain lysates
- Lane 6** : Rat kidney lysates

Lysates/proteins at 20 µg per lane.

Secondary

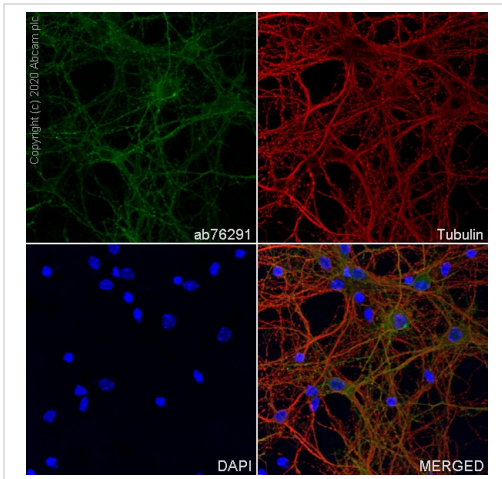
All lanes : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/20000 dilution

Predicted band size: 87 kDa

Observed band size: 120 kDa [why is the actual band size different from the predicted?](#)

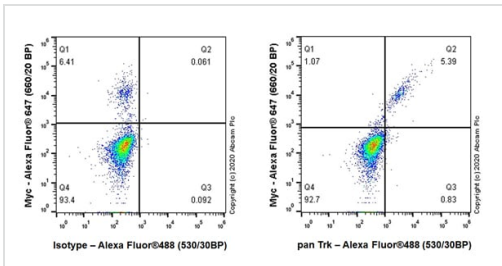
Exposure time: 4 seconds

Blocking/Diluting buffer and concentration: 5% NFD/MBST



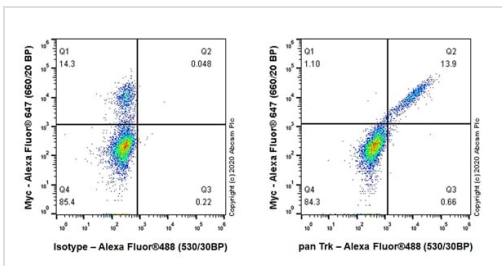
Immunocytochemistry/ Immunofluorescence - Anti-Pan Trk antibody [EP1058Y] (ab76291)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized mouse primary neuron cells labelling Pan Trk with ab76291 at 1/100 dilution, followed by ab150077 AlexaFluor®488 Goat anti-Rabbit secondary antibody at 1/1000 dilution (Green). Confocal scanning Z step was set as 0.3 µm followed by image processing with maximum Z projection. ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 dilution (Red). The Nuclear counterstain was DAPI (Blue).



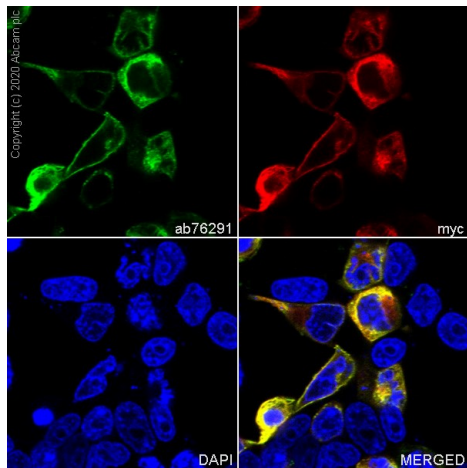
Flow Cytometry - Anti-Pan Trk antibody [EP1058Y] (ab76291)

Flow cytometry analysis of 293T (human embryonic kidney epithelial cell) transfected with TrkB overexpression construct labeling Pan Trk with ab76291 at 1/700 dilution (0.1 µg) (Right). The cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 dilution was used as the secondary antibody. Isotype control/colour: Rabbit monoclonal IgG (ab172730) (Left).



Flow Cytometry - Anti-Pan Trk antibody [EP1058Y] (ab76291)

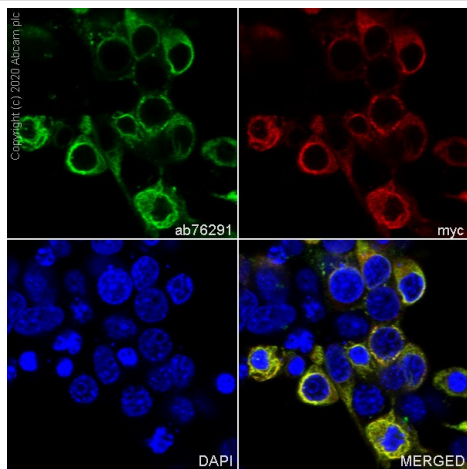
Flow cytometry analysis of 293T (human embryonic kidney epithelial cell) transfected with TrkA overexpression construct labeling Pan Trk with ab76291 at 1/700 dilution (0.1 µg) (Right). The cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 dilution was used as the secondary antibody. Isotype control/colour: Rabbit monoclonal IgG (ab172730) (Left).



Immunocytochemistry/ Immunofluorescence - Anti-Pan Trk antibody [EP1058Y] (ab76291)

Immunocytochemistry/immunofluorescence analysis of TrkA-overexpressed 293T (human embryonic kidney epithelial cell) labelled with the pan Trk antibody ab76291 at 1/1000 dilution (0.7 µg/mL). [ab150077](#), AlexaFluor®488 Goat anti-Rabbit secondary at 1/1000 (2 µg/mL) was used as the secondary antibody. Cells were fixed with 4% Paraformaldehyde and permeabilised with 0.1% TritonX-100. DAPI (blue) was used as nuclear counterstain. Cells were counterstained with Myc-Tag (9B11) Mouse mAb (Alexa Fluor® 647 Conjugate) at 1/200 (2.5 µg/mL).

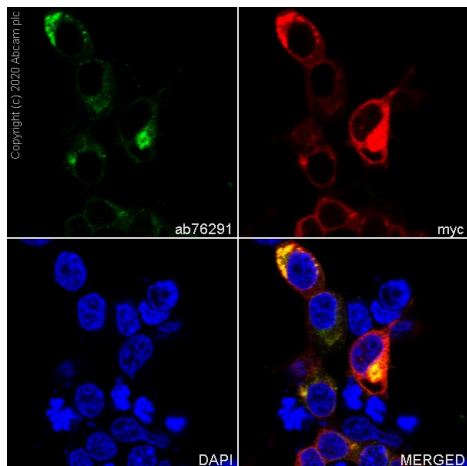
Confocal image showing cytoplasmic staining in 293T cells transfected with a myc-tagged hTrkA expression vector.



Immunocytochemistry/ Immunofluorescence - Anti-Pan Trk antibody [EP1058Y] (ab76291)

Immunocytochemistry/immunofluorescence analysis of TrkB-overexpressed 293T (human embryonic kidney epithelial cell) labelled with the pan Trk antibody ab76291 at 1/1000 dilution (0.7 µg/mL). [ab150077](#), AlexaFluor®488 Goat anti-Rabbit secondary at 1/1000 (2 µg/mL) was used as the secondary antibody. Cells were fixed with 4% Paraformaldehyde and permeabilised with 0.1% TritonX-100. DAPI (blue) was used as nuclear counterstain. Cells were counterstained with Myc-Tag (9B11) Mouse mAb (Alexa Fluor® 647 Conjugate) at 1/200 (2.5 µg/mL).

Confocal image showing cytoplasmic staining in 293T cells transfected with a myc-tagged hTrkB expression vector.

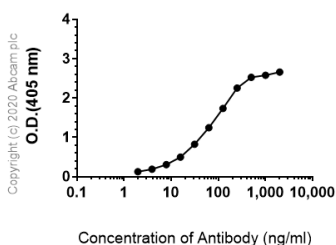


Immunocytochemistry/ Immunofluorescence - Anti-Pan Trk antibody [EP1058Y] (ab76291)

Immunocytochemistry/immunofluorescence analysis of TrkC-overexpressed 293T (human embryonic kidney epithelial cell) labelled with the pan Trk antibody ab76291 at 1/1000 dilution (0.7 µg/mL). ab150077, AlexaFluor®488 Goat anti-Rabbit secondary at 1/1000 (2 µg/mL) was used as the secondary antibody. Cells were fixed with 4% Paraformaldehyde and permeabilised with 0.1% TritonX-100. DAPI (blue) was used as nuclear counterstain. Cells were counterstained with Myc-Tag (9B11) Mouse mAb (Alexa Fluor® 647 Conjugate) at 1/200 (2.5 µg/mL).

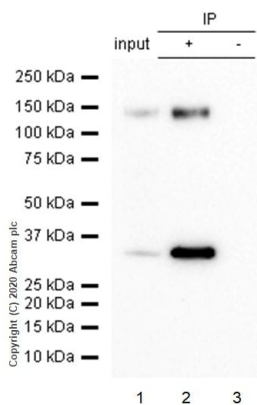
Confocal image showing cytoplasmic staining in 293T cells transfected with a myc-tagged hTrkC expression vector.

Indirect ELISA antibody dose-response curve antigen at 250 ng/ml



ELISA - Anti-Pan Trk antibody [EP1058Y] (ab76291)

ELISA analysis of Human TrkA recombinant protein at 250 ng/mL with ab76291. An Alkaline Phosphatase-conjugated AffiniPure Goat Anti-Rabbit IgG (H+L) at 1/2500 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-Pan Trk antibody [EP1058Y] (ab76291)

ab76291 at 1/40 immunoprecipitating Pan Trk in human brain tissue lysate observed at 145 kDa.

Lane 1 (input): Human brain tissue lysate (10µg)

Lane 2 (+): ab76291+ human brain tissue lysate.

Lane 3 (-): Rabbit monoclonal IgG ([ab172730](#)) instead of ab76291 in human brain lysate

For western blotting, ab76291 at 1/1000 dilution (0.7 µg/mL) and VeriBlot for IP Detection Reagent (HRP) ([ab131366](#)) at 1/5000 were used.

The 30 kDa band is an intracellular fragment, and the 140 kDa observed MW which is higher than the predicted one is due to the glycosylation modification. (refer to [ab189903](#)).

Blocking/Diluting buffer and concentration: 5% NFDm/TBST.



Immunoprecipitation - Anti-Pan Trk antibody [EP1058Y] (ab76291)

ab76291 at 1/40 immunoprecipitating Pan Trk in mouse brain tissue lysate observed at 145 kDa.

Lane 1 (input): Mouse brain tissue lysate (10µg)

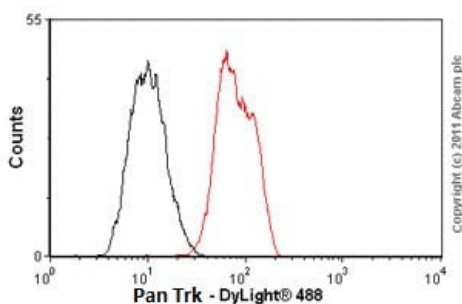
Lane 2 (+): ab76291+ mouse brain tissue lysate.

Lane 3 (-): Rabbit monoclonal IgG ([ab172730](#)) instead of ab76291 in Mouse brain lysate

For western blotting, ab76291 at 1/1000 dilution (0.7 µg/mL) and VeriBlot for IP Detection Reagent (HRP) ([ab131366](#)) at 1/5000 were used.

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Blocking/Diluting buffer and concentration: 5% NFDm/TBST.

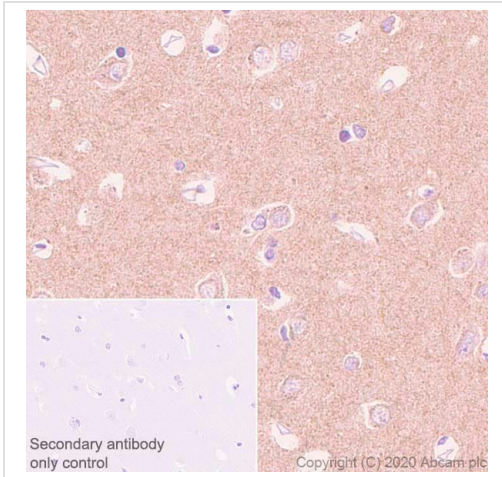


Flow Cytometry - Anti-Pan Trk antibody [EP1058Y] (ab76291)

Overlay histogram showing SH-SY5Y (human neuroblastoma cell line from bone marrow) cells stained with unpurified ab76291 (red line). The cells were fixed with 4% paraformaldehyde (10 min) and incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab76291, 1/50 dilution) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit IgG (H+L) ([ab96899](#)) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit monoclonal IgG (1µg/1x10⁶ cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in SH-SY5Y cells fixed with methanol (5 min) used under the same conditions.

Please note that Abcam do not have data for use of this antibody on

non-fixed cells. We welcome any customer feedback.

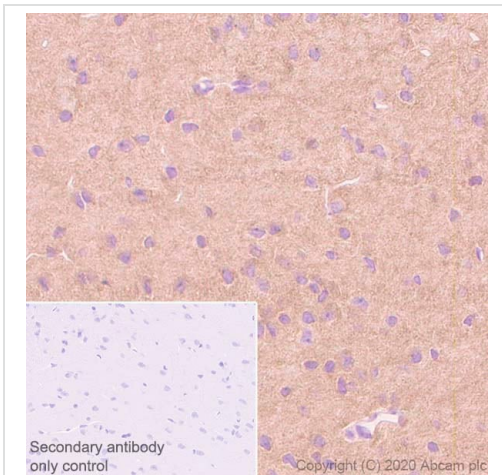


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Pan Trk antibody [EP1058Y] (ab76291)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human cerebrum tissue labelling Pan Trk with purified ab76291 at 1/1000. Rabbit specific IHC polymer detection kit HRP/DAB ([ab209101](#)) was used as the secondary antibody. Heat mediated antigen retrieval was performed using Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 minutes. Sections were counterstained with hematoxylin. Negative control using PBS instead of primary antibody.

Positive staining on human cerebrum.

The section was incubated with ab76291 for 30 mins at room temperature. The immunostaining staining was performed on a Leica Biosystems BOND[®] RX instrument.

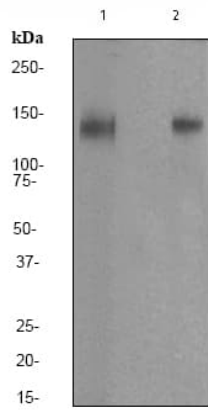


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Pan Trk antibody [EP1058Y] (ab76291)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of rat cerebrum tissue labelling Pan Trk with purified ab76291 at 1/1000. Rabbit specific IHC polymer detection kit HRP/DAB ([ab209101](#)) was used as the secondary antibody. Heat mediated antigen retrieval was performed using Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 minutes. Sections were counterstained with hematoxylin. Negative control using PBS instead of primary antibody.

Positive staining on rat cerebrum.

The section was incubated with ab76291 for 30 mins at room temperature. The immunostaining staining was performed on a Leica Biosystems BOND[®] RX instrument.



Western blot - Anti-Pan Trk antibody [EP1058Y] (ab76291)

All lanes : Anti-Pan Trk antibody [EP1058Y] (ab76291) at 1/5000 dilution (unpurified)

Lane 1 : Rat brain tissue lysate (untreated)

Lane 2 : Rat brain tissue lysate (treated with AP)

Lysates/proteins at 10 µg per lane.

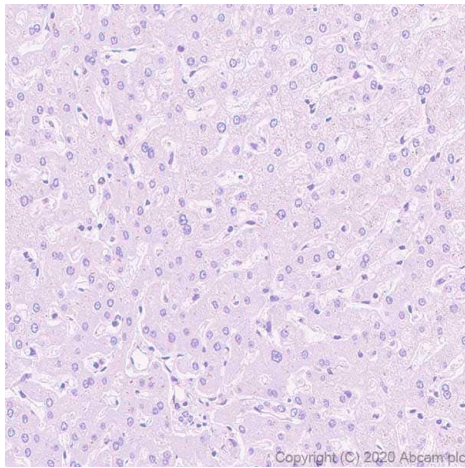
Secondary

Lane 1 : HRP-conjugated goat anti-rabbit IgG at 1/2000 dilution

Lane 2 : HRP-conjugate goat anti-rabbit IgG at 1/2000 dilution

Predicted band size: 87 kDa

Observed band size: 145 kDa [why is the actual band size different from the predicted?](#)

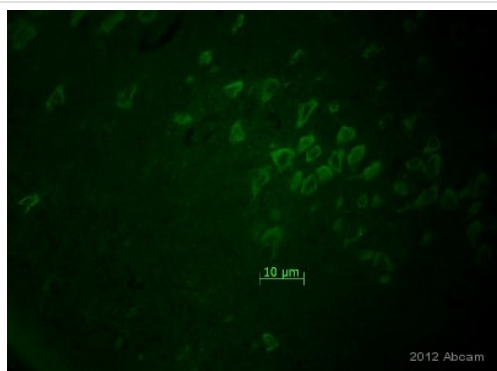


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Pan Trk antibody [EP1058Y] (ab76291)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human liver tissue labelling Pan Trk with purified ab76291 at 1/1000. Rabbit specific IHC polymer detection kit HRP/DAB ([ab209101](#)) was used as the secondary antibody. Heat mediated antigen retrieval was performed using Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 minutes. Sections were counterstained with hematoxylin. Negative control using PBS instead of primary antibody.

Negative control: No staining on human liver.

The section was incubated with ab76291 for 30 mins at room temperature. The immunostaining staining was performed on a Leica Biosystems BOND® RX instrument.



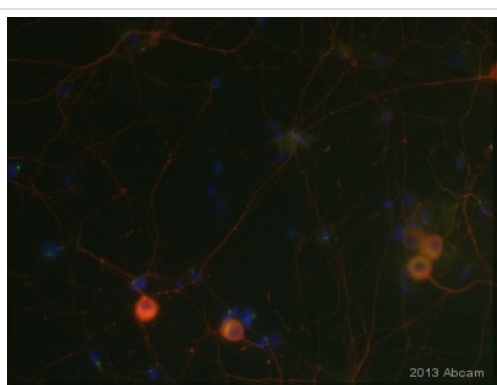
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Pan Trk antibody [EP1058Y] (ab76291)

This image is courtesy of an anonymous Abreview

Unpurified ab76291 staining Pan Trk in murine brain tissue by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections).

Tissue was fixed with formaldehyde, permeabilized with 0.1% Saponin/PBS and blocked with 4% serum for 30 minutes at 25°C, antigen retrieval was by heat mediation with a citrate buffer.

Samples were incubated with primary antibody (1/150 in blocking buffer) for 16 hours at 4°C. A FITC-conjugated goat anti-rabbit polyclonal IgG (1/100) was used as the secondary antibody.



Immunocytochemistry/ Immunofluorescence - Anti-Pan Trk antibody [EP1058Y] (ab76291)

This image is courtesy of an abreview submitted by Dr. Franziska Denk (KCL, United Kingdom)

ICC/IF image of Pan Trk staining on culture of mouse DRG neurons using unpurified ab76291 (1/100). The cells were fixed using formaldehyde and permeabilized using 0.2% Triton X-100. The cells were blocked using 10% Goat serum for 1 hour at 22°C. Unpurified ab76291 was diluted 1/100 using PBS and incubated with the cells for 30 mins at 22°C. The secondary antibody used was Goat polyclonal to Rabbit IgG conjugated to Alexa Fluor® 488 (1/1000). Neuron was stained using Beta III tubulin antibody (Alexa Fluor® 647)

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