abcam

Product datasheet

Anti-Pan Trk antibody [EPR24009-335] - BSA and Azide free ab284409



18 Images

Overview

Product name Anti-Pan Trk antibody [EPR24009-335] - BSA and Azide free

Description Rabbit monoclonal [EPR24009-335] to Pan Trk - BSA and Azide free

Host species Rabbit

Tested applications Suitable for: WB, IHC-P, IP, Flow Cyt (Intra)

Unsuitable for: ICC/IF or IHC-Fr

Species reactivity Reacts with: Mouse, Rat, Human

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: Whole cell lysates: 293T (human embryonic kidney) transfected with vectors containing a

> myc-His-tag® and: control, TrkB, TrkC, TrcA. Human, mouse and rat brain tissue lysates. IHC-P: Human glioblastoma and cerebrum, mouse and rat cerebrum, HEK-293T transfected with TrkC or TrkA or TrkB expression vector. Flow cyt. Intr.: ICR Mouse primary neuron cells, SD Rat primary

neuron. IP: Mouse and rat brain lysates.

General notes ab284409 is a carrier free version of ab267830.

> Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cellbased assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar[®] is a trademark of Fluidigm Canada Inc.

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 see here.

Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**[®] **patents**.

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C.

Storage buffer pH: 7.2

Constituent: 100% PBS

Carrier free Yes

Purity Protein A purified

Clonality Monoclonal

Clone number EPR24009-335

Isotype IgG

Applications

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab284409 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
WB		Use at an assay dependent concentration. Predicted molecular weight: 92 kDa.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
IP		Use at an assay dependent concentration.
Flow Cyt (Intra)		Use at an assay dependent concentration.

Application notes

Is unsuitable for ICC/IF or IHC-Fr.

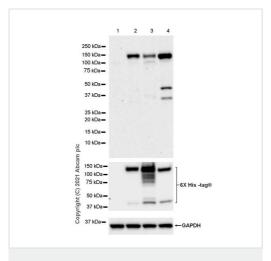
Target

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.

Images



Western blot - Anti-Pan Trk antibody [EPR24009-335] - BSA and Azide free (ab284409) **All lanes :** Anti-Pan Trk antibody [EPR24009-335] (<u>ab267830</u>) at 1/1000 dilution

Lane 1 : 293T (human embryonic kidney) transfected with an empty vector (vector control) containing a myc-His-tag®, whole cell lysate

Lane 2: 293T transfected with human TrkB expression vector containing a myc-His-tag®, whole cell lysate

Lane 3: 293T transfected with human TrkC expression vector containing a myc-His-tag®, whole cell lysate

Lane 4: 293T transfected with human TrkA expression vector containing a myc-His-tag®, whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

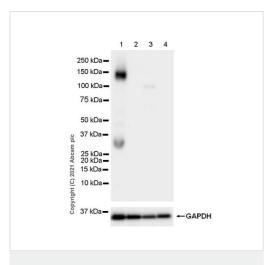
All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 92 kDa **Observed band size:** 140 kDa

Exposure time: 3 minutes

This data was developed using <u>ab267830</u>, the same antibody clone in a different buffer formulation.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.



Western blot - Anti-Pan Trk antibody [EPR24009-335] - BSA and Azide free (ab284409) **All lanes :** Anti-Pan Trk antibody [EPR24009-335] (<u>ab267830</u>) at 1/1000 dilution

Lane 1: Human brain tissue lysate

Lane 2: Human heart tissue lysate

Lane 3: Human kidney tissue lysate

Lane 4: Human spleen tissue lysate

Lysates/proteins at 1/20 dilution per lane.

Secondary

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

Predicted band size: 92 kDa

Observed band size: 140, 32 kDa

Exposure time: 15 seconds

This data was developed using <u>ab267830</u>, the same antibody clone in a different buffer formulation.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

The 32KDa band may be an intracellular fragment of TrkB (TrkB-ICD). The observed Mw of \sim 140KDa is higher than the predicted Mw due to glycosylation (PMID:24860020).

Negative control: heart, kidney, spleen (PMID: 7823156).



Western blot - Anti-Pan Trk antibody [EPR24009-335] - BSA and Azide free (ab284409) **All lanes :** Anti-Pan Trk antibody [EPR24009-335] (<u>ab267830</u>) at 1/1000 dilution

Lane 1 : Mouse brain tissue lysate
Lane 2 : Mouse heart tissue lysate

Lane 3 : Mouse kidney tissue lysate
Lane 4 : Mouse spleen tissue lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 92 kDa **Observed band size:** 140, 32 kDa

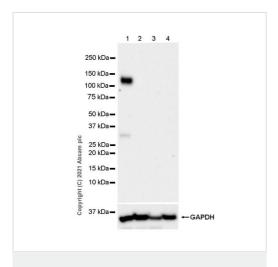
Exposure time: 37 seconds

This data was developed using <u>ab267830</u>, the same antibody clone in a different buffer formulation.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

The 32KDa band may be an intracellular fragment of TrkB (TrkB-ICD). The observed Mw of \sim 140KDa is higher than the predicted Mw due to glycosylation (PMID:24860020).

Negative control: heart, kidney, spleen (PMID: 7823156).



Western blot - Anti-Pan Trk antibody [EPR24009-335] - BSA and Azide free (ab284409) **All lanes :** Anti-Pan Trk antibody [EPR24009-335] ($\underline{ab267830}$) at 1/1000 dilution

Lane 1 : Rat brain tissue lysate

Lane 2: Rat heart tissue lysate

Lane 3: Rat kidney tissue lysate

Lane 4: Rat spleen tissue lysate

Lysates/proteins at 1/20 dilution per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 92 kDa

Observed band size: 140, 32 kDa

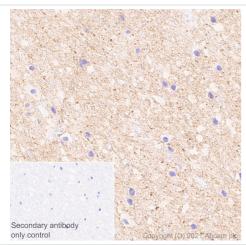
Exposure time: 59 seconds

This data was developed using <u>ab267830</u>, the same antibody clone in a different buffer formulation.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

The 32KDa band may be an intracellular fragment of TrkB (TrkB-ICD). The observed Mw of ~140KDa is higher than the predicted Mw due to glycosylation (PMID:24860020).

Negative control: heart, kidney, spleen (PMID: 7823156).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Pan Trk antibody [EPR24009-335] - BSA and Azide free (ab284409)

Hematoxylin. Secondary antibody only control: PBS was used instead of primary antibody followed by ready to use secondary antibody

The section was incubated with ab267830 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with

This data was developed using ab267830, the same antibody

Immunohistochemical analysis of paraffin-embedded sections of human cerebrum labeling Trk with ab267830 at 1/200 dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine

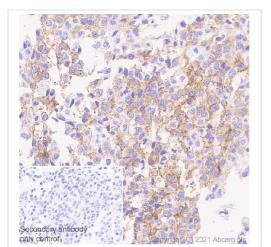
Detection). Positive staining on human cerebrum (PMID:

clone in a different buffer formulation.

26171003).

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

LeicaDS9800 (BOND™ Polymer Refine Detection).

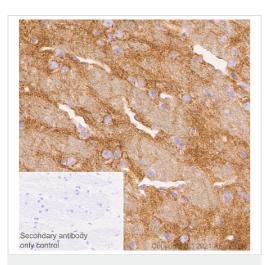


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Pan Trk antibody [EPR24009-335] - BSA and Azide free (ab284409)

This data was developed using **ab267830**, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded sections of human glioblastoma labeling Trk with ab267830 at 1/200 dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on human glioblastoma (PMID: 26171003). The immunostaining was performed on a Leica Biosystems BOND® RX instrument. The section was incubated with ab267830 for 30 mins at room temperature. Counterstained with Hematoxylin.

Secondary antibody only control: PBS was used instead of primary antibody followed by ready to use secondary antibody LeicaDS9800 (BOND™ Polymer Refine Detection).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Pan Trk antibody

[EPR24009-335] - BSA and Azide free (ab284409)

Secondary antibody

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Pan Trk antibody

[EPR24009-335] - BSA and Azide free (ab284409)

This data was developed using <u>ab267830</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded sections of mouse cerebrum labeling Trk with <u>ab267830</u> at 1/200 dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on mouse cerebrum. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. The section was incubated with <u>ab267830</u> for 30 mins at room temperature. Counterstained with Hematoxylin.

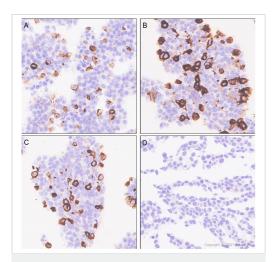
Secondary antibody only control: PBS was used instead of primary antibody followed by ready to use secondary antibody LeicaDS9800 (BOND™ Polymer Refine Detection).

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

This data was developed using <u>ab267830</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded sections of rat cerebrum labeling Trk with <u>ab267830</u> at 1/200 dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on rat cerebrum. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. The section was incubated with <u>ab267830</u> for 30 mins at room temperature. Counterstained with Hematoxylin.

Secondary antibody only control: PBS was used instead of primary antibody followed by ready to use secondary antibody
LeicaDS9800 (BOND™ Polymer Refine Detection).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Pan Trk antibody

[EPR24009-335] - BSA and Azide free (ab284409)

Immunohistochemical analysis of paraffin-embedded sections detailed below and labeling Trk with <u>ab267830</u> at 1/200 dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). The immunostaining was performed on a Leica Biosystems BOND[®] RX instrument. Counterstained with Hematoxylin. Sections included:

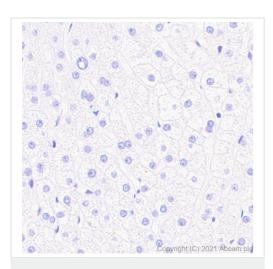
Panel A: HEK-293T (human epithelial cell line from embryonic kidney transformed with large T antigen) transfected with TrkC expression vector containing a myc-His-tag[®].

Panel B: HEK-293T transfected with TrkA vector containing a myc-His-tag $^{\rm B}$.

Panel C: HEK-293T transfected with TrkB vector containing a myc-His-tag $^{\rm I\!R}$.

Panel D: HEK-293T transfected with empty vector containing a myc-His-tag[®].

Positive staining on (A) HEK-293T transfected with TrkC expression vector, (B) HEK-293T transfected with a TrkA expression vector, and (C) HEK-293T transfected with TrkB expression vector. Almost no staining on (D) HEK-293T transfected with empty vector.

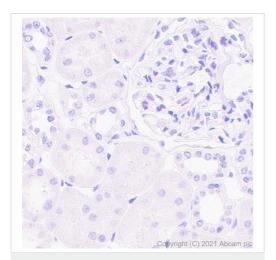


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Pan Trk antibody

[EPR24009-335] - BSA and Azide free (ab284409)

Immunohistochemical analysis of paraffin-embedded sections of human liver labeling Trk with <u>ab267830</u> at 1/200 dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Negative control: almost no staining on human liver. The immunostaining was performed on a Leica Biosystems BOND[®] RX instrument. The section was incubated with <u>ab267830</u> for 30 mins at room temperature. Counterstained with Hematoxylin.

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

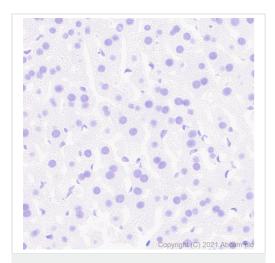


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Pan Trk antibody

[EPR24009-335] - BSA and Azide free (ab284409)

This data was developed using <u>ab267830</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded sections of human kidney labeling Trk with <u>ab267830</u> at 1/200 dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Negative control: almost no staining on human kidney The immunostaining was performed on a Leica Biosystems BOND® RX instrument. The section was incubated with <u>ab267830</u> for 30 mins at room temperature. Counterstained with Hematoxylin.

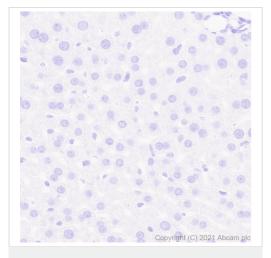


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Pan Trk antibody

[EPR24009-335] - BSA and Azide free (ab284409)

Immunohistochemical analysis of paraffin-embedded sections of mouse liver labeling Trk with <u>ab267830</u> at 1/200 dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Negative control: almost no staining on mouse liver. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. The section was incubated with <u>ab267830</u> for 30 mins at room temperature. Counterstained with Hematoxylin.

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

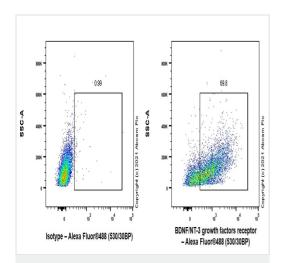


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Pan Trk antibody

[EPR24009-335] - BSA and Azide free (ab284409)

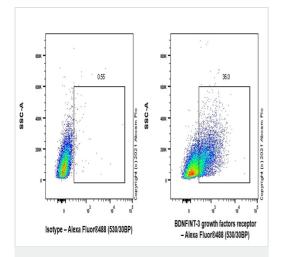
This data was developed using <u>ab267830</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded sections of rat liver labeling Trk with <u>ab267830</u> at 1/200 dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Negative control: almost no staining on rat liver. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. The section was incubated with <u>ab267830</u> for 30 mins at room temperature. Counterstained with Hematoxylin.



Flow Cytometry (Intracellular) - Anti-Pan Trk antibody [EPR24009-335] - BSA and Azide free (ab284409)

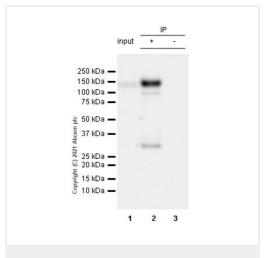
Flow cytometric analysis of 4% paraformaldehyde fixed and 90% methanol permeabilized ICR Mouse primary neuron cells labelling Trk with <u>ab267830</u> at 1/500 dilution (Right) compared with a Rabbit monoclonal IgG (<u>ab172730</u>) (Left) isotype control. Goat Anti-Rabbit IgG (Alexa Fluor[®] 488, <u>ab150097</u>) at 1/2000 dilution was used as the secondary antibody.



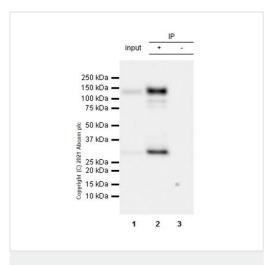
Flow Cytometry (Intracellular) - Anti-Pan Trk antibody [EPR24009-335] - BSA and Azide free (ab284409)

This data was developed using <u>ab267830</u>, the same antibody clone in a different buffer formulation.

Flow cytometric analysis of 4% paraformaldehyde fixed and 90% methanol permeabilized SD Rat primary neuron cells labelling Trk with <u>ab267830</u> at 1/500 dilution (Right) compared with a Rabbit monoclonal IgG (<u>ab172730</u>) (Left) isotype control. Goat Anti-Rabbit IgG (Alexa Fluor[®] 488, <u>ab150097</u>) at 1/2000 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-Pan Trk antibody
[EPR24009-335] - BSA and Azide free (ab284409)



Immunoprecipitation - Anti-Pan Trk antibody

[EPR24009-335] - BSA and Azide free (ab284409)

This data was developed using <u>ab267830</u>, the same antibody clone in a different buffer formulation.

Trk was immunoprecipitated from mouse brain tissue lysate 10 μ g with <u>ab267830</u> at 1/30 dilution (2 μ g in 0.35 mg lysates). Western blot was performed on the immunoprecipitate using <u>ab267830</u> at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP) (<u>ab131366</u>) was used at 1/5000 dilution.

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

Lane 2: ab267830 IP in mouse brain tissue lysate

Lane 3: Rabbit monoclonal $\lg G \left(\underline{ab172730} \right)$ instead of $\underline{ab267830}$ in mouse brain tissue lysate

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 8 seconds.

Observed molecular weight [kDa]: 140, 32.

This data was developed using <u>ab267830</u>, the same antibody clone in a different buffer formulation.

Trk was immunoprecipitated from rat brain tissue lysate 10 µg with ab267830 at 1/30 dilution (2µg in 0.35 mg lysates). Western blot was performed on the immunoprecipitate using ab267830 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP) (ab131366) was used at 1/5000 dilution.

Lane 1: Rat brain tissue lysate 10 µg (Inset)

Lane 2: ab267830 IP in rat brain tissue lysate

Lane 3: Rabbit monoclonal $\lg G$ ($\underline{ab172730}$) instead of $\underline{ab267830}$ in rat brain tissue lysate

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 8 seconds.

Observed molecular weight [kDa]: 140, 32.



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