Product name: Anti-TrkA antibody [6B2] ab86474

Description: Mouse monoclonal [6B2] to TrkA

Host species: Mouse

Tested applications: Suitable for: Flow Cyt, WB, ELISA, ICC/IF

Species reactivity: Reacts with: Rat, Human, Recombinant fragment

Immunogen: Purified recombinant extracellular fragment of Human TrkA (aa33-423) fused with hlgGFc tag expressed in HEK293 cell line.


Form: Liquid

Storage instructions: Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.

Storage buffer: Preservative: 0.05% Sodium azide
Constituent: PBS

Purity: Protein G purified

Purification notes: Purified from tissue culture supernatant.

Clonality: Monoclonal

Clone number: 6B2

Isotype: IgG1

Applications:

Our Abpromise guarantee covers the use of ab86474 in the following tested applications.

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

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<th>Application</th>
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<td>Flow Cyt</td>
<td>ab170190</td>
<td>Use 1µg for 10^6 cells. Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.</td>
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</table>
Function
Receptor tyrosine kinase 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. High affinity receptor for NGF which is its primary ligand, it can also bind and be activated by NT3/neurotrophin-3. However, NT3 only supports axonal extension through NTRK1 but has no effect on neuron survival. Upon dimeric NGF ligand-binding, undergoes homodimerization, autophosphorylation and activation. Recruits, phosphorylates and/or activates several downstream effectors including SHC1, FRS2, SH2B1, SH2B2 and PLCG1 that regulate distinct overlapping signaling cascades driving cell survival and differentiation. Through SHC1 and FRS2 activates a GRB2-Ras-MAPK cascade that regulates cell differentiation and survival. Through PLCG1 controls NF-Kappa-B activation and the transcription of genes involved in cell survival. Through SHC1 and SH2B1 controls a Ras-PI3 kinase-AKT1 signaling cascade that is also regulating survival. In absence of ligand and activation, may promote cell death, making the survival of neurons dependent on trophic factors.

Isoform TrkA-III is resistant to NGF, constitutively activates AKT1 and NF-kappa-B and is unable to activate the Ras-MAPK signaling cascade. Antagonizes the anti-proliferative NGF-NTRK1 signaling that promotes neuronal precursors differentiation. Isoform TrkA-III promotes angiogenesis and has oncogenic activity when overexpressed.

Tissue specificity
Isoform TrkA-I is found in most non-neuronal tissues. Isoform TrkA-II is primarily expressed in neuronal cells. TrkA-III is specifically expressed by pluripotent neural stem and neural crest progenitors.

Involvement in disease
Congenital insensitivity to pain with anhidrosis
Chromosomal aberrations involving NTRK1 are found in papillary thyroid carcinomas (PTCs) (PubMed:2869410, PubMed:7565764, PubMed:1532241). Translocation t(1;3)(q21;q11) with TFG generates the TRKT3 (TRK-T3) transcript by fusing TFG to the 3'-end of NTRK1 (PubMed:7565764). A rearrangement with TPM3 generates the TRK transcript by fusing TPM3 to the 3'-end of NTRK1 (PubMed:2869410). An intrachromosomal rearrangement that links the protein kinase domain of NTRK1 to the 5'-end of the TPR gene forms the fusion protein TRK-T1. TRK-T1 is a 55 kDa protein reacting with antibodies against the C-terminus of the NTRK1 protein (PubMed:1532241).

Sequence similarities
Belongs to the protein kinase superfamily. Tyr protein kinase family. Insulin receptor subfamily.
Contains 2 Ig-like C2-type (immunoglobulin-like) domains.
Contains 2 LRR (leucine-rich) repeats.
Contains 1 LRRCT domain.
Contains 1 protein kinase domain.

Domain
The transmembrane domain mediates interaction with KIDINS220.
The extracellular domain mediates interaction with NGFR.

Post-translational modifications
Ligand-mediated autophosphorylation. Interaction with SQSTM1 is phosphotyrosine-dependent.
Autophosphorylation at Tyr-496 mediates interaction and phosphorylation of SHC1.

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<td>ELISA</td>
<td>1/10000</td>
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<tr>
<td>ICC/IF</td>
<td>1/200 - 1/1000.</td>
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Target
N-glycosylated (Probable). Isoform TrkA-I is N-glycosylated. Ubiquitinated. Undergoes polyubiquitination upon activation; regulated by NGFR. Ubiquitination regulates the internalization of the receptor.

**Cellular localization**

Cell membrane. Early endosome membrane. Late endosome membrane. Internalized to endosomes upon binding of NGF or NTF3 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).

**Images**

Anti-TrkA antibody [6B2] (ab86474) at 1/500 dilution + extracellular domain of Human TrkA (aa33-423).

**Predicted band size:** 87 kDa

The observed band size may not correspond to the predicted protein molecular weight as the immunogen (recombinant fragment) was used for the test lane.

Confocal immunofluorescence analysis of PC-12 cells using ab86474 at a 1/200 dilution (green), showing membrane and cytoplasmic localization. Blue: DRAQ5 fluorescent DNA dye.
Overlay histogram showing SH-SY5Y cells stained with ab86474 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab86474, 1µg/1x10^6 cells) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-mouse IgG (H+L) (ab96879) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse IgG1 [ICIGG1] (ab91353, 2µg/1x10^6 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 4% paraformaldehyde (10 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.

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