abcam

Product datasheet

Anti-KIF5A + KIF5B + KIF5C antibody [EPR24325-98] ab271049

Recombinant RabMAb

13 Images

Overview

Product name Anti-KIF5A + KIF5B + KIF5C antibody [EPR24325-98]

Description Rabbit monoclonal [EPR24325-98] to KIF5A + KIF5B + KIF5C

Host species Rabbit

Tested applications Suitable for: Flow Cyt (Intra), WB, IP, ICC/IF

Unsuitable for: IHC-P

Species reactivity Reacts with: Mouse, Rat, Human

Immunogen Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: Neuro-2a, C2C12, U-87 MG, SH-SY5Y, Rat brain cortex, C6, Mouse brain, Mouse cerebral

> cortex, Mouse cerebellum, Rat brain, Rat cerebellum, Human cerebellum, Human hypothalamus, HEK-293T transfected with KIF5A, KIF5B and KIF5C expression vector containing a myc-Histag® whole cell lysates. ICC/IF: mouse primary neuron and rat primary neuron, SH-SY5Y cells.

Flow Cyt: SH-SY5Y, Neuro-2a and C6 cells. IP: Neuro-2a, SH-SY5Y 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 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

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: 7.2

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

Purity Protein A purified

Clonality Monoclonal

Clone number EPR24325-98

Isotype IgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab271049 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)		1/500.
WB		1/1000. Predicted molecular weight: 117 kDa.
IP		1/30.
ICC/IF		1/50.

Application notes Is unsuitable for IHC-P.

Target

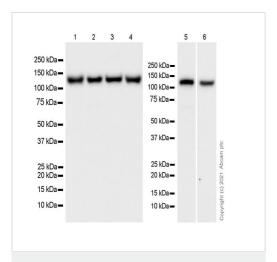
Relevance

The kinesin superfamily proteins (KIFs) are microtubule-dependent motors that slide along microtubules and transport cellular organelles and mRNA to different parts of the cell. In neurons, KIF5 performs this role in an anterograde fashion from the neuronal cell body to axonal or dendritic terminals. KIF5 was initially differentiated from the ubiquitously expressed kinesin as being highly enriched in subsets of neurons and selectively concentrated in the cell body. KIF5 has at least three known isoforms; this antibody recognizes all three. Mutations in one of the isoforms of KIF5 (KIF5A) are thought to be involved in hereditary spastic paraplegias.

Cellular localization

Uniformly distributed between soma and neurites in hippocampal neurons

Images



Western blot - Anti-KIF5A antibody [EPR24325-98] (ab271049)

All lanes: Anti-KIF5A + KIF5B + KIF5C antibody [EPR24325-98] (ab271049) at 1/1000 dilution

Lane 1 : Neuro-2a (mouse neuroblastoma neuroblast), whole cell lysate

Lane 2 : C2C12 (mouse myoblasts myoblast), whole cell lysate
Lane 3 : U-87 MG (human glioblastoma-astrocytoma epithelial cell), whole cell lysate

Lane 4: SH-SY5Y (human neuroblastoma epithelial cell), whole cell lysate

Lane 5: Rat brain cortex tissue lysate

Lane 6: C6 (rat glial tumor glial cell), whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated (ab97051) at 1/100000 dilution

Predicted band size: 117 kDa **Observed band size:** 117 kDa

Blocking and diluting buffer and concentration: 5% NFDM/TBST

Exposure time: 10 seconds

1 2 3 4 5

250 kDa—
150 kDa—
100 kDa—
75 kDa—
37 kDa—
25 kDa—
20 kDa—
115 kDa—
110 kDa—

Western blot - Anti-KIF5A antibody [EPR24325-98] (ab271049)

All lanes : Anti-KIF5A + KIF5B + KIF5C antibody [EPR24325-98] (ab271049) at 1/1000 dilution

Lane 1: Mouse brain tissue lysate

Lane 2 : Mouse cerebral cortex tissue lysate

Lane 3: Mouse cerebellum tissue lysate

Lane 4: Rat brain tissue lysate

Lane 5: Rat cerebellum tissue lysate

Lysates/proteins at 20 µg per lane.

Secondary

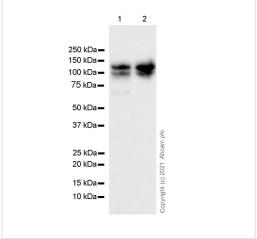
All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (**ab97051**) at 1/100000 dilution

Predicted band size: 117 kDa

Observed band size: 117,107 kDa

Blocking and diluting buffer and concentration: 5% NFDM/TBST Degraded fragment (107 KD) has also been described (PMID: 11144355)

Exposure time: 10 seconds



Western blot - Anti-KIF5A antibody [EPR24325-98]

(ab271049)

All lanes : Anti-KIF5A + KIF5B + KIF5C antibody [EPR24325-98] (ab271049) at 1/1000 dilution

Lane 1 : Human cerebellum tissue lysate

Lane 2 : Human hypothalamus tissue lysate

Lysates/proteins at 20 µg per lane.

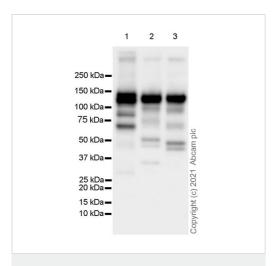
Secondary

All lanes : VeriBlot for IP secondary antibody(HRP)(<u>ab131366</u>) at 1/1000 dilution

Predicted band size: 117 kDa **Observed band size:** 117,107 kDa

Blocking and diluting buffer and concentration: 5% NFDM/TBST Degraded fragment (107 KD) has also been described (PMID: 11144355)

Exposure time: 10 seconds



Western blot - Anti-KIF5A antibody [EPR24325-98] (ab271049)

All lanes : Anti-KIF5A + KIF5B + KIF5C antibody [EPR24325-98] (ab271049) at 1/5000 dilution

Lane 1 : HEK-293T (human embryonic kidney) transfected with KIF5A expression vector containing a myc-His-tag®, whole cell lysate

Lane 2: HEK-293T transfected with KIF5B expression vector containing a myc-His-tag®, whole cell lysate

Lane 3: HEK-293T transfected with KIF5C expression vector containing a myc-His-tag®, whole cell lysate

Lysates/proteins at 2 µg per lane.

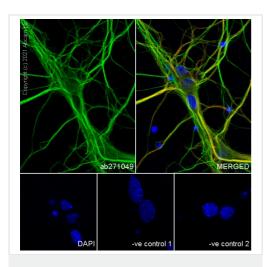
Secondary

All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 117 kDa

Blocking and diluting buffer and concentration: 5% NFDM/TBST

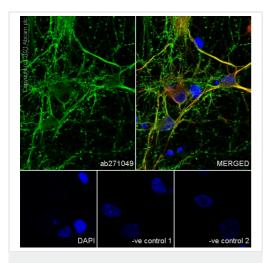
Exposure time: 3 seconds



Immunocytochemistry/ Immunofluorescence - Anti-KIF5A antibody [EPR24325-98] (ab271049)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized mouse primary neuron cells labelling KIF5A + KIF5B + KIF5C with ab271049 at 1/50 (9.72 ug/ml) dilution, followed by **ab150080** Goat Anti-Rabbit lgG H&L (Alexa Fluor® 594) antibody at 1/500 (4 ug/ml) dilution (Green). Confocal image showing cytoplasmic staining in mouse primary neuron. Confocal scanning Z step was set as 0.3 µm followed by image processing with maximum Z projection. **ab195889** Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 (2.5 ug/ml) dilution (Red). The Nuclear counterstain was DAPI (Blue).

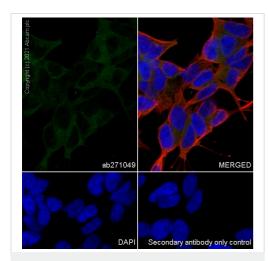
Secondary antibody only control: Secondary antibody is <u>ab150080</u> Goat Anti-Rabbit IgG H&L (Alexa Fluor® 594) at 1/500 (4 ug/ml) dilution.



Immunocytochemistry/ Immunofluorescence - Anti-KIF5A antibody [EPR24325-98] (ab271049)

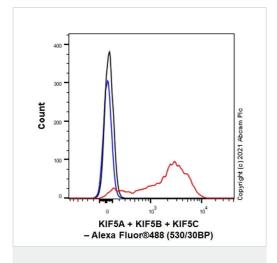
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized rat primary neuron cells labelling KIF5A + KIF5B + KIF5C with ab271049 at 1/50 (9.72 ug/ml) dilution, followed by ab150080 Goat Anti-Rabbit lgG H&L (Alexa Fluor® 594) antibody at 1/500 (4 ug/ml) dilution (Green). Confocal image showing cytoplasmic staining in rat primary neuron. Confocal scanning Z step was set as 0.3 µm followed by image processing with maximum Z projection ab11267 Anti-MAP2 mouse monoclonal antibody was used to counterstain tubulin at 1/500 (4 ug/ml) dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is **ab150080** Goat Anti-Rabbit lgG H&L (Alexa Fluor® 594) at 1/500 (4 ug/ml) dilution.



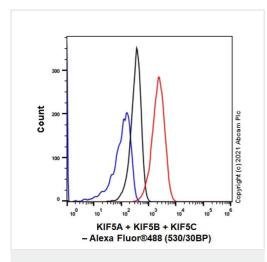
Immunocytochemistry/ Immunofluorescence - Anti-KIF5A antibody [EPR24325-98] (ab271049)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized SH-SY5Y cells labelling KIF5A + KIF5B + KIF5C with ab271049 at 1/50 (9.72 ug/ml) dilution, followed by ab150080 Goat Anti-Rabbit lgG H&L (Alexa Fluor® 594) antibody at 1/500 (4 ug/ml) dilution (Green). Confocal image showing cytoplasmic staining in SH-SY5Y cell line. is observed. ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 (2.5 ug/ml) dilution (Red). The Nuclear counterstain was DAPI (Blue). Secondary antibody only control: Secondary antibody is ab150080 Goat Anti-Rabbit lgG H&L (Alexa Fluor® 594) at 1/500 (4 ug/ml) dilution.



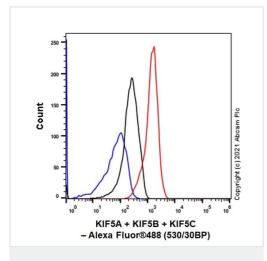
Flow Cytometry (Intracellular) - Anti-KIF5A + KIF5B + KIF5C antibody [EPR24325-98] (ab271049)

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized SH-SY5Y (Human neuroblastoma epithelial cell) cells labelling KIF5A + KIF5B + KIF5C with ab271049 at 1/500 dilution (0.1ug) (Red) compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat F(ab')2 Anti-Rabbit IgG(DyLight® 488, ab98507) at 1/500 dilution was used as the secondary antibody.



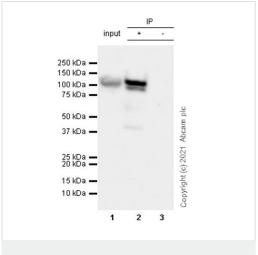
Flow Cytometry (Intracellular) - Anti-KIF5A + KIF5B + KIF5C antibody [EPR24325-98] (ab271049)

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized Neuro-2a (Mouse neuroblastoma neuroblast) cells labelling KIF5A + KIF5B + KIF5C with ab271049 at 1/500 dilution (0.1ug) (Red) compared with a Rabbit monoclonal lgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat Anti-Rabbit lgG (Alexa Fluor® 488, ab150081) at 1/2000 dilution was used as the secondary antibody.



Flow Cytometry (Intracellular) - Anti-KIF5A + KIF5B + KIF5C antibody [EPR24325-98] (ab271049)

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized C6 (Rat glial tumor glial cell) cells labelling KIF5A + KIF5B + KIF5C with ab271049 at 1/500 dilution (0.1ug) (Red) compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/2000 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-KIF5A antibody [EPR24325-98] (ab271049)

KIF5A + KIF5B + KIF5C was immunoprecipitated from 0.35 mg
Neuro-2a (mouse neuroblastoma neuroblast) whole cell lysate 10
ug with ab271049 at 1/30 dilution (2ug in 0.35mg lysates). Western
blot was performed on the immunoprecipitate using ab271049 at
1/1000 dilution. VeriBlot for IP secondary antibody(HRP)
(ab131366) was used at 1/5000 dilution.

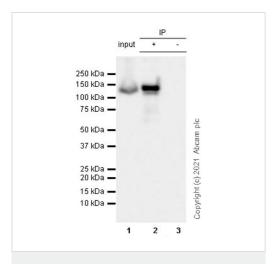
Lane 1: Neuro-2a (mouse neuroblastoma neuroblast) whole cell lysate 10 ug

Lane 2: ab271049 IP in Neuro-2a whole cell lysate

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab271049 in Neuro-2a whole cell lysate

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

Exposure time: 3 seconds



Immunoprecipitation - Anti-KIF5A antibody [EPR24325-98] (ab271049)

KIF5A + KIF5B + KIF5C was immunoprecipitated from 0.35 mg SH-SY5Y (human neuroblastoma epithelial cell) whole cell lysate 10 ug with ab271049 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab271049 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP) (ab131366) was used at 1/5000 dilution.

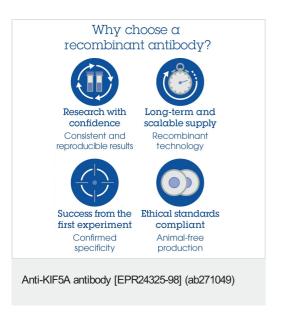
Lane 1: SH-SY5Y (human neuroblastoma epithelial cell) whole cell lysate 10 ug

Lane 2: ab271049 IP in SH-SY5Y whole cell lysate

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab271049 in SH-SY5Y whole cell lysate

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

Exposure time: 3 seconds



Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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