abcam

Product datasheet

Anti-TGN46 antibody [EPR12676] ab174280



Recombinant RabMAb

1 References 8 Images

Overview

Product name Anti-TGN46 antibody [EPR12676]

Rabbit monoclonal [EPR12676] to TGN46 **Description**

Host species Rabbit

Tested applications Suitable for: WB, IP, IHC-P, ICC/IF

Unsuitable for: Flow Cyt

Species reactivity Reacts with: Human

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

Positive control WB: A549, HepG2 and HeLa lysates (HeLa shows to be a low expressor). IHC-P: Human brain

and kidney tissues. IP: Immunoprecipitation pellet from fetal liver lysate. ICC/IF: Hep G2 and A549

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.

Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with

these species. Please contact us for more information.

Properties

Form Liquid

Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long Storage instructions

term. Avoid freeze / thaw cycle.

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 9% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA, 50% Tissue culture

supernatant

1

Purity Protein A purified

Clonality Monoclonal
Clone number EPR12676

Isotype IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab174280 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		1/1000 - 1/5000. Predicted molecular weight: 51 kDa.
IP		1/10 - 1/100.
IHC-P		1/50 - 1/100. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
ICC/IF		Use a concentration of 1 µg/ml.

Application notes Is unsuitable for Flow Cyt.

Target

Function May be involved in regulating membrane traffic to and from trans-Golgi network.

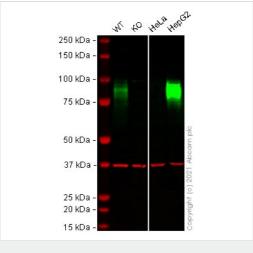
Tissue specificity Isoform TGN46 is widely expressed. Isoform TGN51 is more abundant in fetal lung and kidney.

Isoform TGN48 is barely expressed in embryonic kidney and promyelocytic cells.

Cellular localization Cell membrane. Golgi apparatus > trans-Golgi network membrane. Primarily in trans-Golgi

network. Cycles between the trans-Golgi network and the cell surface returning via endosomes.

Images



Western blot - Anti-TGN46 antibody [EPR12676] (ab174280)

All lanes : Anti-TGN46 antibody [EPR12676] (ab174280) at 1/1000 dilution

Lane 1 : Wild-type A549 (Human lung carcinoma cell line) whole cell lysate

Lane 2: TGOLN2 knockout A549 (Human lung carcinoma cell line) whole cell lysate

Lane 3: HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate

Lane 4: Hep G2 (Human liver hepatocellular carcinoma cell line) whole cell lysate

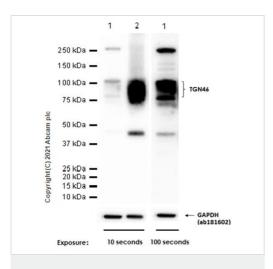
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 51 kDa Observed band size: 80 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab174280 observed at 80 kDa. Red - loading control <u>ab8245</u> (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

ab174280 was shown to react with TGN46 in wild-type A549 cells in Western blot with loss of signal observed in TGOLN2 knockout cell line ab269510 (knockout cell lysate ab269672). Wild-type A549 and TGOLN2 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with ab174280 and ab8245 (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 lgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preabsorbed (ab216773) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.



Western blot - Anti-TGN46 antibody [EPR12676] (ab174280)

All lanes : Anti-TGN46 antibody [EPR12676] (ab174280) at 1/1000 dilution

Lane 1 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate

Lane 2: HepG2 (Human hepatocellular carcinoma epithelial cell) whole cell 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: 51 kDa **Observed band size:** 75-100 kDa

Blocking buffer: 5% NFDM/TBST.

Exposure: 10 seconds, 100 seconds.

MERGED ab174280

MERGED ab174280

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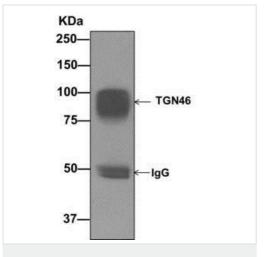
MERGED ab174280

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Immunocytochemistry/ Immunofluorescence - Anti-TGN46 antibody [EPR12676] (ab174280)

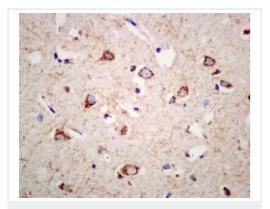
ab174280 staining TGN46 in wild-type A549 cells (top panel) and TGOLN2 knockout A549 cells (bottom panel) (ab269510). The cells were fixed with 100% methanol (5 min) then permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab174280 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 lgG (Alexa Fluor® 488) (ab150081) at 2 µg/ml (shown in green) and a goat secondary antibody to mouse lgG (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).



Immunoprecipitation. ab174280 at 1/1000 labeling TGN46 immunoprecipitated from Human fetal liver lysate using ab174280 at 1/10.

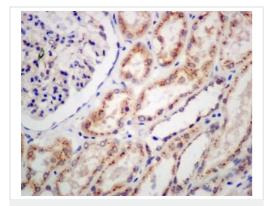




Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TGN46 antibody
[EPR12676] (ab174280)

Immunohistochemical analysis of paraffin embedded Human brain tissue labeling TGN46 with ab174280 at 1/50.

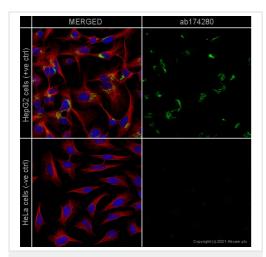
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TGN46 antibody
[EPR12676] (ab174280)

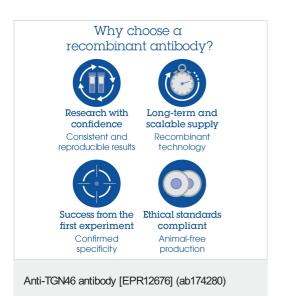
Immunohistochemical analysis of paraffin embedded Human kidney tissue labeling TGN46 with ab174280 at 1/50.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-TGN46 antibody [EPR12676] (ab174280)

ab174280 staining TGN46 in HepG2 cells (top panel - positive control) and HeLa cells (bottom panel - negative control, low TGN46 expression). 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 ab174280 at 1 μ g/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 lgG (Alexa Fluor[®] 488) (**ab150081**) at 2 μ g/ml (shown in green) and a goat secondary antibody to mouse lgG (Alexa Fluor[®] 594) (**ab150120**) at 2 μ g/ml (shown in red). Nuclear DNA was labelled in blue with DAPI. Image was taken with a confocal microscope (Leica-Microsystems TCS SP8).



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