Product datasheet

Anti-Transferrin Receptor antibody ab84036

Product name
Anti-Transferrin Receptor antibody

Description
Rabbit polyclonal to Transferrin Receptor

Host species
Rabbit

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

Species reactivity
Reacts with: Mouse, Human

Predicted to work with: Dog, Pig, Orangutan

Immunogen
Synthetic peptide conjugated to KLH derived from within residues 1 - 100 of Human Transferrin Receptor. Read Abcam's proprietary immunogen policy (Peptide available as ab101219.)

Positive control

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 or -80°C. Avoid freeze / thaw cycle.

Storage buffer
Preservative: 0.02% Sodium Azide
Constituents: 1% BSA, PBS, pH 7.4

Purity
Immunogen affinity purified

Clonality
Polyclonal

Isotype
IgG

Applications

Our Abpromise guarantee covers the use of ab84036 in the following tested applications.
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

<table>
<thead>
<tr>
<th>Application</th>
<th>Abreviews</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>WB</td>
<td>★★★★☆☆</td>
<td>Use a concentration of 1 µg/ml. Detects a band of approximately 98 kDa (predicted molecular weight: 84 kDa).</td>
</tr>
</tbody>
</table>
Function

Cellular uptake of iron occurs via receptor-mediated endocytosis of ligand-occupied transferrin receptor into specialized endosomes. Endosomal acidification leads to iron release. The apotransferrin-receptor complex is then recycled to the cell surface with a return to neutral pH and the concomitant loss of affinity of apotransferrin for its receptor. Transferrin receptor is necessary for development of erythrocytes and the nervous system (By similarity). A second ligand, the hereditary hemochromatosis protein HFE, competes for binding with transferrin for an overlapping C-terminal binding site. Positively regulates T and B cell proliferation through iron uptake (PubMed:26642240).

(Microbial infection) Acts as a receptor for new-world arenaviruses: Guanarito, Junin and Machupo virus.

Involvement in disease

Immunodeficiency 46

Sequence similarities

Belongs to the peptidase M28 family. M28B subfamily.
Contains 1 PA (protease associated) domain.

Post-translational modifications

N- and O-glycosylated, phosphorylated and palmitoylated. The serum form is only glycosylated.
Proteolytically cleaved on Arg-100 to produce the soluble serum form (sTfR).
Palmitoylated on both Cys-62 and Cys-67. Cys-62 seems to be the major site of palmitoylation.

Cellular localization

Secreted and Cell membrane. Melanosome. Identified by mass spectrometry in melanosome fractions from stage I to stage IV.

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<tbody>
<tr>
<td>IHC-P</td>
<td>⭐⭐⭐⭐</td>
<td>1/6000. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.</td>
</tr>
<tr>
<td>ICC/IF</td>
<td>⭐⭐⭐⭐</td>
<td>Use a concentration of 5 µg/ml.</td>
</tr>
<tr>
<td>IHC-FrFI</td>
<td>⭐⭐⭐⭐</td>
<td>1/200.</td>
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</table>

Images

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Analysis of co-localisation of FcRn with cellular markers.

HepG2 (Human liver hepatocellular carcinoma cell line) cells were fixed, permeabilized and co-stained with anti-FcRn humanised Fab’, followed by anti-human IgG conjugated Alexa Fluor® 488 (Top left panel. Bar 5 μm) and ab84036 followed by anti-rabbit/mouse conjugated Alexa Fluor® 568 (Top right panel). FcRn fluorescence is shown in green and marker fluorescence is shown in red, yellow fluorescence in the overlay images (Bottom left panel) indicates co-localization.

Images are magnifications of the white boxed area (Bottom right panel. Bar 20 μm).

All lanes: Anti-Transferrin Receptor antibody (ab84036) at 1 μg/ml

Lane 1: HeLa (Human epithelial carcinoma cell line) whole cell lysate

Lane 2: Jurkat (Human T cell lymphoblast-like cell line) whole cell lysate

Lane 3: U-2 OS (Human osteosarcoma cell line) whole cell lysate

Lysates/proteins at 10 μg per lane.

Secondary

All lanes: Goat Anti-Rabbit IgG H&L (HRP) preadsorbed (ab97080) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 84 kDa

Observed band size: 98 kDa

why is the actual band size different from the predicted?

Additional bands at: 37 kDa. We are unsure as to the identity of these extra bands.

Exposure time: 4 minutes
Transferrin Receptor contains a number of potential glycosylation sites (SwissProt) which may explain its migration at a higher molecular weight than predicted.

**ICC/IF image of ab84036 stained HeLa (Human epithelial cell line from cervix adenocarcinoma) cells.**

The cells were fixed in 4% formaldehyde (10 minutes) then permeabilized using 0.1% PBS-Triton and then incubated in 1% BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1 hour to further permeabilize the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody ab84036 at 1 µg/ml overnight at +4°C.

The secondary antibody (pseudo-colored green) was an Alexa-Fluor® 488 goat anti-rabbit (ab150081) IgG (H+L) preadsorbed, used at a 1/1000 dilution for 1 hour. Alexa-Fluor® 594 WGA was used to label plasma membranes (pseudo-colored red) at a 1/200 dilution for 1 hour at room temperature.

DAPI was used to stain the cell nuclei (pseudo-colored blue) at a concentration of 1.43 µM for 1 hour at room temperature.

**ab84036 staining mouse liver sections by IHC-FrFl.**

The animal was perfused with 4% paraformaldehyde, further post fixed with 4% paraformaldehyde overnight and prepared for free floating sectioning. Staining with ab84036 at a 1/200 dilution in 0.1% Triton X with 0.1x PBS and 10% donkey was performed for 24 hours at 24°C. A donkey anti-rabbit Alexa-Fluor®568 polyclonal antibody was used as the secondary antibody.
ab84036 immunohistochemical staining in the rat brain.

Samples were fixed with formaldehyde and heat-mediated antigen retrieval was performed with citric acid. Tissue sections were blocked in 1% BSA for 10 minutes at 21°C before incubation with ab84036 (1/6000) for 16 hours at 21°C. A biotin conjugated goat anti-rabbit IgG secondary was used at 1/250.

ab84036 staining Transferrin Receptor in canine kidney cells by Immunocytochemistry/ Immunofluorescence.

Cells were fixed in paraformaldehyde, permeabilized with 0.5% saponin then incubated with ab84036 at a 1/100 dilution for 1.5 hours at 20°C. The secondary used was an Alexa-Fluor®568 conjugated donkey anti-rabbit polyclonal, used at a 1/350 dilution.

Anti-Transferrin Receptor antibody (ab84036) at 1 µg/ml + Mouse spleen tissue lysate at 10 µg

Secondary

Goat Anti-Rabbit IgG H&L (HRP) preadsorbed (ab97080) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

**Predicted band size**: 84 kDa

**Observed band size**: 100,50 kDa

*why is the actual band size different from the predicted?*
Exposure time: 8 minutes

Transferrin Receptor contains a number of potential glycosylation sites (SwissProt) which may explain its migration at a higher molecular weight than predicted.

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