Product datasheet

Anti-TMEM16A antibody ab53212

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Overview

Product name  Anti-TMEM16A antibody
Description  Rabbit polyclonal to TMEM16A
Host species  Rabbit
Tested applications  Suitable for: IHC-P, ICC/IF, IHC-Fr
Species reactivity  Reacts with: Mouse, Rat, Human
Immunogen  Synthetic peptide within Human TMEM16A aa 400-500. The exact sequence is proprietary. Database link: Q5XXA6
Positive control  GIST

Properties

Form  Liquid
Storage instructions  Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.
Storage buffer  pH: 7.6
Preservative: 0.1% Sodium azide
 Constituents: PBS, 1% BSA
Purity  Immunogen affinity purified
Clonality  Polyclonal
Isotype  IgG

Applications

Our Abpromise guarantee covers the use of ab53212 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>
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<tbody>
<tr>
<td>IHC-P</td>
<td>★★★★☆</td>
<td>1/100.</td>
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<tr>
<td>ICC/IF</td>
<td></td>
<td>Use a concentration of 2 µg/ml. PubMed: 19819874</td>
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### Function
Acts as a calcium-activated chloride channel. Required for normal tracheal development.

### Tissue specificity
Broadly expressed with higher levels in liver and skeletal muscle.

### Sequence similarities
Belongs to the anoctamin family.

### Domain
The region spanning the fifth and sixth transmembrane domains probably forms the pore-forming region.

### Cellular localization
Cell membrane. Cytoplasm.

### Images

![Immunocytochemistry/ Immunofluorescence analysis of Wt1<sup>cre-YFP</sup> mouse interstitial cell of Cajal labelling TMEM16A with ab53212 at 1/20 dilution. Cells from the muscular layer, especially the circular one, show TMEM16A (ANO1 - red) immunoreactivity. This immunostaining becomes strong by E16.5. Colocalization of YFP with ANO1 is observed in many cells of both, the circular and the longitudinal layer.](image-url)

*Image from Carmona, Rita et al. PLoS ONE 8.2 (2013): e55890. doi: 10.1371/journal.pone.0055890. Fig 4A. Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/*
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-TMEM16A antibody (ab53212)

ab53212 (2µg/ml) staining TMEM16A in human liver (left panel) using an automated system (DAKO Autostainer Plus). Using this protocol there is strong staining of hepatocyte cell membrane. Sections were rehydrated and antigen retrieved with the Dako 3 in 1 AR buffer EDTA pH 9.0 in a DAKO PT link. Slides were peroxidase blocked in 3% H2O2 in methanol for 10 mins. They were then blocked with Dako Protein block for 10 minutes (containing casein 0.25% in PBS) then incubated with primary antibody for 20 min and detected with Dako envision flex amplification kit for 30 minutes. Colorimetric detection was completed with Diaminobenzidine for 5 minutes. Slides were counterstained with Haematoxylin and coverslipped under DePeX. Please note that, for manual staining, optimization of primary antibody concentration and incubation time is recommended. Signal amplification may be required.

Analysis of TMEM16A expression was performed in frozen liver sections (10 µm) from normal and 7-day bile duct-ligated rats and mice. Sections were defrosted at room temperature and fixed in 4% paraformaldehyde (1× PBS), permeabilized in PBST (1× PBS with 0.2% Triton), and blocked in 4% BSA (in PBST) for 1 hour at room temperature. Samples were then incubated with ab53212 at a 1/100 dilution, diluted in 1% BSA. Sections were incubated overnight at 4 °C and washed three times for 10 minutes each with 1× PBST at room temperature. Sections were incubated with Dylight 488 conjugated donkey anti-rabbit secondary at a 1/600 dilution for 2 hours at room temperature protected from light. Following incubation, the slides were washed in PBST at room temperature and coverslipped with Antifade gold and DAPI. Images were visualized using an Olympus IX-71 confocal microscope.

Scale bar, 10 µm.
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human gastrointestinal stromal tumor labeling TMEM16A with ab53212.

ICC/IF image of ab53212 stained HepG2 cells. The cells were 4% formaldehyde fixed (10 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab53212, 1µg/ml) overnight at +4°C. The secondary antibody (green) was Alexa Fluor® 488 goat anti-rabbit IgG (H+L) used at a 1/1000 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.

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