Product datasheet

Anti-PCSK9 antibody ab31762

Overview

Product name: Anti-PCSK9 antibody
Description: Rabbit polyclonal to PCSK9
Host species: Rabbit
Tested applications: Suitable for: WB, IP, ICC/IF, IHC-P
Species reactivity: Reacts with: Mouse, Rat
Immunogen: Synthetic peptide corresponding to Mouse PCSK9 aa 650 to the C-terminus (C terminal) conjugated to keyhole limpet haemocyanin.
(Peptide available as ab32727)
Positive control: This antibody gave a positive signal in the following tissue lysates: Liver (Mouse) Kidney (Mouse) Liver (Rat) Kidney (Rat) This antibody gave a positive signal in the following cell types: PC12. It also gave a positive signal in FFPE mouse large intestine tissue sections.

Properties

Form: Liquid
Storage instructions: Shipped at 4°C. Upon delivery aliquot and store at -20°C or -80°C. Avoid repeated freeze / thaw cycles.
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 ab31762 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>WB</td>
<td></td>
<td>Use a concentration of 1 µg/ml. Detects a band of approximately 62 kDa (predicted molecular weight: 74 kDa).</td>
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<tr>
<td>IP</td>
<td></td>
<td>Use a concentration of 5 µg/ml.</td>
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<tr>
<td>ICC/IF</td>
<td></td>
<td>Use a concentration of 5 µg/ml.</td>
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<tr>
<td>IHC-P</td>
<td></td>
<td>Use a concentration of 5 µg/ml. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.</td>
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**Target**

**Function**
May be implicated in the differentiation of cortical neurons and may play a role in cholesterol homeostasis.

**Tissue specificity**
Expressed in neuro-epithelioma, colon carcinoma, hepatic and pancreatic cell lines, and in Schwann cells.

**Involvement in disease**
Defects in PCSK9 are the cause of familial hypercholesterolemia 3 (FH3) [MIM:603776]. FH3 inheritance is autosomal dominant.

**Sequence similarities**
Belongs to the peptidase S8 family. Contains 1 peptidase S8 domain.

**Post-translational modifications**
The soluble zymogen undergoes autocatalytic intramolecular processing in the endoplasmic reticulum, resulting in the cleavage of its propeptide that remains associated with the secreted enzyme.

**Cellular localization**
Secreted.

**Images**

![Western blot - Anti-PCSK9 antibody (ab31762)](image)

**All lanes**: Anti-PCSK9 antibody (ab31762) at 1 µg/ml

**Lane 1**: Liver (Mouse) Tissue Lysate

**Lane 2**: Kidney (Mouse) Tissue Lysate

**Lane 3**: Liver (Rat) Tissue Lysate

**Lane 4**: Kidney (Rat) Whole Cell Lysate - normal tissue (ab29480)

Lysates/proteins at 10 µg per lane.

**Secondary**

**All lanes**: IRDye 680 Conjugated Goat Anti-Rabbit IgG (H+L) at 1/10000 dilution

Performed under reducing conditions.
Predicted band size: 74 kDa
Observed band size: 62 kDa

Why is the actual band size different from the predicted?

ab31762 detects a band of ~62 kDa in liver and kidney lysates from mouse and rat. This corresponds to the cleaved form of PCSK9. ab31762 has not detected the 74 kDa proprotein form in the lysates tested.

ICC/IF image of ab31762 stained PC12 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 ab31762 at 5µg/ml overnight at +4°C. The secondary antibody (green) was DyLight® 488 goat anti-rabbit (ab96899) 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.

IHC image of ab31762 staining in mouse large intestine formalin fixed paraffin embedded tissue section, performed on a Leica Bond™ system using the standard protocol B. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab31762, 5µg/ml, for 15 mins at room temperature. A goat anti-rabbit biotinylated secondary antibody was used to detect the primary, and visualized using an HRP conjugated ABC system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.
PCSK9 was immunoprecipitated using 0.5mg Mouse Liver tissue, 5µg of Rabbit polyclonal to PCSK9 and 50µl of protein G magnetic beads (+). No antibody was added to the control (-).

The antibody was incubated under agitation with Protein G beads for 10min, Mouse Liver tissue lysate diluted in RIPA buffer was added to each sample and incubated for a further 10min under agitation.

Proteins were eluted by addition of 40µl SDS loading buffer and incubated for 10min at 70°C; 10µl of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab31762.


Band: 62kDa; PCSK9

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