## Overview

<table>
<thead>
<tr>
<th><strong>Product name</strong></th>
<th>Anti-NDUFA9 antibody [20C11B11B11]</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Description</strong></td>
<td>Mouse monoclonal [20C11B11B11] to NDUFA9</td>
</tr>
<tr>
<td><strong>Host species</strong></td>
<td>Mouse</td>
</tr>
<tr>
<td><strong>Tested applications</strong></td>
<td>Suitable for: IHC-P, WB, Flow Cyt</td>
</tr>
<tr>
<td><strong>Species reactivity</strong></td>
<td>Reacts with: Mouse, Rat, Cow, Human</td>
</tr>
<tr>
<td><strong>Immunogen</strong></td>
<td>Tissue, cells or virus corresponding to Cow NDUFA9.</td>
</tr>
<tr>
<td><strong>Positive control</strong></td>
<td>WB: WI38 and NIH 3T3 whole cell lysates, human testis tissue lysate and human, cow, rat and mouse heart mitochondria. IHC-P: Human spinal column tissue. Flow Cyt: HepG2 cells.</td>
</tr>
<tr>
<td><strong>General notes</strong></td>
<td>This antibody clone is manufactured by Abcam. This monoclonal antibody to NDUFA9 has been knockout validated in Western blot. The expected band for NDUFA9 was observed in wild type cells and the band was not seen in knockout cells. If you require this antibody in a particular buffer formulation or a particular conjugate for your experiments, please contact <a href="mailto:orders@abcam.com">orders@abcam.com</a> or you can find further information here.</td>
</tr>
</tbody>
</table>

## Properties

<table>
<thead>
<tr>
<th><strong>Form</strong></th>
<th>Liquid</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Storage instructions</strong></td>
<td>Shipped at 4°C. Store at +4°C.</td>
</tr>
<tr>
<td><strong>Storage buffer</strong></td>
<td>Preservative: 0.02% Sodium azide Constituent: HEPES buffered saline</td>
</tr>
<tr>
<td><strong>Purity</strong></td>
<td>IgG fraction</td>
</tr>
<tr>
<td><strong>Purification notes</strong></td>
<td>Near homogeneity as judged by SDS-PAGE. The antibody was produced in vitro using hybridomas grown in serum-free medium, and then purified by biochemical fractionation.</td>
</tr>
<tr>
<td><strong>Clonality</strong></td>
<td>Monoclonal</td>
</tr>
<tr>
<td><strong>Clone number</strong></td>
<td>20C11B11B11</td>
</tr>
<tr>
<td><strong>Isotype</strong></td>
<td>IgG1</td>
</tr>
<tr>
<td><strong>Light chain type</strong></td>
<td>kappa</td>
</tr>
</tbody>
</table>
Function

Accessory subunit of the mitochondrial membrane respiratory chain NADH dehydrogenase (Complex I), that is believed not to be involved in catalysis. Complex I functions in the transfer of electrons from NADH to the respiratory chain. The immediate electron acceptor for the enzyme is believed to be ubiquinone.

Sequence similarities

Belongs to the complex I NDUFA9 subunit family.

Cellular localization

Mitochondrion matrix.

Applications

Our Abpromise guarantee covers the use of ab14713 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>Abrevies</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>IHC-P</td>
<td><img src="https://www.abcam.com/ab14713" alt="ab14713" /></td>
<td>Use at an assay dependent concentration.</td>
</tr>
<tr>
<td>WB</td>
<td><img src="https://www.abcam.com/ab14713" alt="ab14713" /></td>
<td>Use a concentration of 1 µg/ml. Detects a band of approximately 36 kDa (predicted molecular weight: 40 kDa).</td>
</tr>
<tr>
<td>Flow Cyt</td>
<td></td>
<td>Use 1-2µg for 10⁶ cells. ab170190 - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.</td>
</tr>
</tbody>
</table>

Target

Function

Accessory subunit of the mitochondrial membrane respiratory chain NADH dehydrogenase (Complex I), that is believed not to be involved in catalysis. Complex I functions in the transfer of electrons from NADH to the respiratory chain. The immediate electron acceptor for the enzyme is believed to be ubiquinone.

Sequence similarities

Belongs to the complex I NDUFA9 subunit family.

Cellular localization

Mitochondrion matrix.

Images

Lane 1: Wild-type HAP1 cell lysate (20 µg)
Lane 2: NDUFA9 knockout HAP1 cell lysate (20 µg)
Lane 3: WI38 cell lysate (20 µg)
Lane 4: NIH3T3 cell lysate (20 µg)
Lanes 1 - 4: Merged signal (red and green). Green - ab14713 observed at 40 kDa. Red - loading control, ab176560, observed at 52 kDa.

ab14713 was shown to specifically react with NDUFA9 in wild-type HAP1 cells. No band was observed when NDUFA9 knockout HAP1 samples were used. Wild-type and NDUFA9 knockout samples were subjected to SDS-PAGE. ab14713 and ab176560 (loading control to alpha tubulin) were diluted at 1µg/mL and 1/10,000 respectively and incubated overnight at 4°C. Blots were developed with Goat anti-Mouse IgG H&L (IRDye® 800CW) preadsorbed (ab216772) and Goat Anti-Rabbit IgG H&L (IRDye® 680RD) preadsorbed (ab216777) secondary antibodies at 1/10,000 dilution for 1 hour at room temperature before imaging.
ab14713 a 1/100 dilution staining NDUFA9 in Human spinal column tissue by Immunohistochemistry (Formalin/PFA-Fixed paraffin-embedded sections). Antibody was incubated with the sample for 1 hour. Sections were incubated in peroxidase-conjugated rabbit anti-mouse secondary (diluted 1/100 in 4% BSA in PBST) for 1 hour at room temperature. Sections were washed x3 in PBST and peroxidase activity was demonstrated using kit. Antigen retrieval was performed by 1 minute of pressure cooking with 1 mmol EDTA pH 8.0.

Overlay histogram showing HepG2 cells stained with ab14713 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab14713, 2µg/1x10^6 cells) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-mouse IgG (H+L) (ab96879) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was Mouse IgG1 [ICIGG1] (ab91353, 2µg/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed.

All lanes: Anti-NDUFA9 antibody [20C11B11B11] (ab14713) at 1 µg/ml

Lane 1: WI38 (Human lung fibroblast cell line) Whole Cell Lysate
Lane 2: Human testis tissue lysate - total protein (ab30257)
Lane 3: NIH 3T3 (Mouse embryonic fibroblast cell line) Whole Cell Lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes: Goat polyclonal to Mouse IgG - H&L - Pre-Adsorbed (HRP) at 1/3000 dilution

Developed using the ECL technique.

Performed under reducing conditions.
Predicted band size: 40 kDa
Observed band size: 36 kDa

why is the actual band size different from the predicted?

Additional bands at: 58 kDa. We are unsure as to the identity of these extra bands.

Exposure time: 20 minutes

The band observed at 36 kDa could potentially be a cleaved form of NDUFA9 due to the presence of a 35 amino acid transit peptide.

**All lanes**: Anti-NDUFA9 antibody [20C11B11B11] (ab14713)

**Lane 1**: Isolated mitochondria from Human heart at 5 µg

**Lane 2**: Isolated mitochondria from Bovine heart at 1 µg

**Lane 3**: Isolated mitochondria from Rat heart at 10 µg

**Lane 4**: Isolated mitochondria from Mouse Heart at 10 µg

**Secondary**

**All lanes**: Goat anti-Mouse IgG

Predicted band size: 40 kDa
Observed band size: 37 kDa

why is the actual band size different from the predicted?

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