Product datasheet

Anti-MiTF antibody [C5] - ChIP Grade ab12039

4 Abreviews  45 References  3 Images

Overview

Product name Anti-MiTF antibody [C5] - ChIP Grade
Description Mouse monoclonal [C5] to MiTF - ChIP Grade
Host species Mouse
Specificity The antibody recognizes serine phosphorylated and non-phosphorylated melanocytic isoforms of microphthalmia.

Tested applications Suitable for: IHC-Fr, IHC-P, ICC/IF, ChIP, Flow Cyt, WB, IP
Species reactivity Reacts with: Mouse, Rat, Chicken, Human
Immunogen Recombinant fragment corresponding to Human MiTF (N terminal).
Positive control This antibody gave a positive signal in Malme-3M whole cell lysate. In Flow Cytometry, this antibody gave a positive signal in methanol fixed/Tween permeabilised Malme-3M cells.

General notes This antibody clone is manufactured by Abcam.
If you require this antibody in a particular buffer formulation or a particular conjugate for your experiments, please contact orders@abcam.com or you can find further information here.

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 pH: 7.40
Preservative: 0.02% Sodium azide
Constituents: PBS, 6.97% L-Arginine
Purity IgG fraction
Clonality Monoclonal
Clone number C5
Isotype IgG1

Applications

Our Abpromise guarantee covers the use of ab12039 in the following tested applications.
Function

Transcription factor for tyrosinase and tyrosinase-related protein 1. Binds to a symmetrical DNA sequence (E-boxes) (5'-CACGTG-3') found in the tyrosinase promoter. Plays a critical role in the differentiation of various cell types as neural crest-derived melanocytes, mast cells, osteoclasts and optic cup-derived retinal pigment epithelium.

Tissue specificity

Isoform M is exclusively expressed in melanocytes and melanoma cells. Isoform A and isoform H are widely expressed in many cell types including melanocytes and retinal pigment epithelium (RPE). Isoform C is expressed in many cell types including RPE but not in melanocyte-lineage cells.

Involvement in disease

Defects in MITF are the cause of Waardenburg syndrome type 2A (WS2A) [MIM:193510]. It is a dominant inherited disorder characterized by sensorineural hearing loss and patches of depigmentation. The features show variable expression and penetrance. Defects in MITF are a cause of Waardenburg syndrome type 2 with ocular albinism (WS2-OA) [MIM:103470]. It is an ocular albinism with sensorineural deafness. Defects in MITF are the cause of Tietz syndrome (TIETZS) [MIM:103500]. It is an autosomal dominant disorder characterized by generalized hypopigmentation and profound, congenital, bilateral deafness. Penetrance is complete.

Sequence similarities

Belongs to the MiT/TFE family.
Contains 1 basic helix-loop-helix (bHLH) domain.

Post-translational modifications

Phosphorylation at Ser-405 significantly enhances the ability to bind the tyrosinase promoter.

Cellular localization

Nucleus.

Images

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-Fr</td>
<td></td>
<td>Use at an assay dependent concentration.</td>
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<tr>
<td>IHC-P</td>
<td></td>
<td>Use a concentration of 1 µg/ml. Antigen retrieval is not essential but may optimise staining.</td>
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<tr>
<td>ICC/IF</td>
<td></td>
<td>Use at an assay dependent concentration. PubMed: 16648630 Fix with methanol/acetone, block with 1% BSA/PBS (see Wellbrock et al).</td>
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<tr>
<td>ChIP</td>
<td></td>
<td>Use at an assay dependent concentration. PubMed: 21990371</td>
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<tr>
<td>EMSA</td>
<td></td>
<td>Use at an assay dependent concentration.</td>
</tr>
<tr>
<td>Flow Cyt</td>
<td></td>
<td>Use 2µg for 10^6 cells. ab170190 - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.</td>
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<tr>
<td>WB</td>
<td></td>
<td>Use a concentration of 1 µg/ml. Detects a band of approximately 52-56 kDa (predicted molecular weight: 59 kDa).</td>
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<tr>
<td>IP</td>
<td></td>
<td>Use at 2 µg/mg of lysate.</td>
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Western blot - Anti-MiTF antibody [C5] - ChIP Grade (ab12039) at 1 µg/ml + Malme 3m (Human melanoma cells) Whole Cell Lysate at 10 µg

**Secondary**

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:** 59 kDa  
**Observed band size:** 40, 55, 70 kDa  
*why is the actual band size different from the predicted?*

**Exposure time:** 20 minutes

ab12039 staining MiTF in embryonic day 4 chick eye tissue by Immunohistochemistry (Frozen sections).  
Tissue was fixed with 4% paraformaldehyde, permeabilized using 1% saponin, blocked with 10% serum for 30 minutes at 37°C, then incubated with ab12039 at a 1/1000 dilution for 24 hours at 4deg.C. The secondary used was an Alexa-Fluor 488 conjugated mouse monoclonal, used at a 1/100 dilution. Counterstained with DAPI (blue).

Overlay histogram showing Malme 3m cells stained with ab12039 (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 (ab12039, 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.

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