Product datasheet

Anti-Myogenin antibody [F5D] ab1835

Product name Anti-Myogenin antibody [F5D]
Description Mouse monoclonal [F5D] to Myogenin
Host species Mouse
Specificity The immunogen used to raise this antibody has 100% homology with the Rat Myogenin protein. Some customers have successfully used ab1835 on rat samples, however we have not been successful detecting Myogenin in this species in our own testing and therefore cannot guarantee rat reactivity. Please contact Abcam Scientific Support for more information.
Tested applications Suitable for: IHC-FoFr, IHC-P, ICC/IF, WB
Species reactivity Reacts with: Mouse, Human
Predicted to work with: Rat, Pig
Immunogen Recombinant full length protein corresponding to Rat Myogenin aa 1-250.
Database link: P20428
Positive control IHC-P: Human Rhabdomyosarcoma; ICC/IF: 2 days differentiated C2C12 cells, ioSkeletal Myocytes - Human iPSC-Derived Skeletal Myocytes (ab277612);
General notes This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or conjugation for your experiments, please contact orders@abcam.com.

The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As.

Properties

Form Liquid
Storage buffer Preservative: 0.02% Sodium azide
          Constituents: PBS, 6.97% L-Arginine
Function
- Involved in muscle differentiation (myogenic factor). Induces fibroblasts to differentiate into myoblasts. Probable sequence specific DNA-binding protein.

Sequence similarities
- Contains 1 basic helix-loop-helix (bHLH) domain.

Cellular localization
- Nucleus.

Images
ab1835 staining Myogenin in undifferentiated C2C12 cells (top panel) and 2 days differentiated C2C12 cells (bottom panel).

The cells were fixed with 100% methanol (5 min), permeabilized with 0.1% Triton X-100 for 5 mins and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab1835 at 1 μg/ml and ab6046. Rabbit polyclonal to beta Tubulin - Loading Control, at 1/1000 dilution overnight at 4ºC. Cells were then incubated with ab150117. Goat Anti-Mouse IgG H&L (Alexa Fluor® 488) at 1/1000 dilution (shown in green) and ab150084, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 594) at 1/1000 dilution (shown in pseudocolor red). Nuclear DNA was labeled with DAPI (shown in blue).

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

All lanes:

Lane 1: Differentiated C2C12 whole cell lysate - Day 0 control
Lane 2: Differentiated C2C12 whole cell lysate - Day 1
Lane 3: Differentiated C2C12 whole cell lysate - Day 2
Lane 4: Differentiated C2C12 whole cell lysate - Day 3
Lane 5: Differentiated C2C12 whole cell lysate - Day 4
Lane 6: Differentiated C2C12 whole cell lysate - Day 5
Lane 7: Differentiated C2C12 whole cell lysate - Day 6

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**Predicted band size:** 25 kDa
**Observed band size:** 34 kDa

C2C12 cells were differentiated into myotubes as previously described in PMID: 26563778.

Lanes 1-7: Merged signal (red and green). Green - ab1835 observed at 30kDa. Red - loading control ab181602 observed at 37kDa.

This blot was produced using a 4-12% Bis-tris under the MOPS buffer system. The gel was run at 200V for 55 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes.
The membrane was blocked for an hour using 3% milk before ab1835 and ab181602 (Rabbit anti GAPDH loading control), were incubated overnight at 4°C at a 1 in 400 dilution and 1/20000 dilution respectively. Antibody binding was detected using Goat anti-Mouse IgG H&L (IRDye® 800CW) preadsorbed (ab216772) and Goat Anti-Rabbit IgG H&L (IRDye® 680RD) preadsorbed (ab216777) secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.

Immunofluorescence staining of Myogenin using ab1835 in ioSkeletal Myocytes - Human iPSC-Derived Skeletal Myocytes (ab277612), which were differentiated for 3 days post induction.

The cells were fixed with 4% formaldehyde (10 min), permeabilized with 0.1% PBS-Tween for 5 mins and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with ab1835 at 1 µg/mL and ab6046, rabbit polyclonal to beta Tubulin, at 1/1000 dilution. Cells were then incubated with ab150117, Goat Anti-Mouse IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (shown in green) and ab150088, Goat Anti-Rabbit IgG H&L (Alexa Fluor® 594) preadsorbed at 1/1000 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).

Image was acquired with a high-content analyser (Operetta CLS, Perkin Elmer) and a maximum intensity projection of confocal sections is shown. Gamma is adjusted to 1.5 in all channels.

The antibody ab1835 gave comparable results using MeOH fixation (100%, 5 min).
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Myogenin antibody [F5D] (ab1835)

IHC image of Myogenin staining in a section of formalin-fixed paraffin-embedded normal Human Rhabdomyosarcoma performed on a Leica BOND™ system using the standard Protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20mins. The section was then incubated with ab1835, 5µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. The inset secondary-only control image is taken from an identical assay without primary antibody. 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. *Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre.

Immunohistochemistry (PFA perfusion fixed frozen sections) - Anti-Myogenin antibody [F5D] (ab1835)

Adult mouse muscle section stained with ab1835. The animals were perfused with 4% PFA. The sections were incubated in 5% normal donkey serum in 0.1% PBS- and triton X100 for 1h to permeabilise the cells and block non-specific protein-protein interactions. The sections were then incubated with the antibody (ab1835, 1µg/ml) overnight at +4°C. The secondary antibody Alexa Fluor® 568 donkey anti-mouse IgG (H+L) (red) was used at a 1/1000 dilution for 1h.

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