abcam

Product datasheet

Anti-Mitofusin 2 antibody [EPR19796] ab205236



Recombinant RabMAb

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Overview

Product name Anti-Mitofusin 2 antibody [EPR19796]

Rabbit monoclonal [EPR19796] to Mitofusin 2 **Description**

Host species Rabbit

Tested applications Suitable for: Flow Cyt (Intra), WB, IP, ICC/IF Species reactivity Reacts with: Human, Recombinant fragment

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: Human Mitofusin 2 recombinant protein fragment; Human fetal heart, fetal kidney and fetal

liver lysates; HeLa and HEK-293 whole cell lysates. ICC/IF: HeLa cells. Flow Cyt (intra): HeLa

cells. IP: HeLa whole cell lysate.

General notes This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility

- Improved sensitivity and specificity - Long-term security of supply - Animal-free production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

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 long

term. Avoid freeze / thaw cycle.

Storage buffer pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

Purity Protein A purified

Clonality Monoclonal Clone number EPR19796

Isotype IgG

Applications

The Abpromise guarantee

Our Abpromise guarantee covers the use of ab205236 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		1/700.
WB		1/2000. Detects a band of approximately 86 kDa (predicted molecular weight: 86 kDa).
IP		1/30.
ICC/IF		1/250.

Target

Function

Essential transmembrane GTPase, which mediates mitochondrial fusion. Fusion of mitochondria occurs in many cell types and constitutes an important step in mitochondria morphology, which is balanced between fusion and fission. MFN2 acts independently of the cytoskeleton. It therefore plays a central role in mitochondrial metabolism and may be associated with obesity and/or apoptosis processes. Overexpression induces the formation of mitochondrial networks. Plays an important role in the regulation of vascular smooth muscle cell proliferation. Involved in the clearance of damaged mitochondria via selective autophagy (mitophagy). Is required for PARK2 recruitment to dysfunctional mitochondria. Involved in the control of unfolded protein response (UPR) upon ER stress including activation of apoptosis and autophagy during ER stress. Acts as an upstream regulator of EIF2AK3 and suppresses EIF2AK3 activation under basal conditions.

Tissue specificity

Ubiquitous; expressed at low level. Highly expressed in heart and kidney.

Involvement in disease

Charcot-Marie-Tooth disease 2A2

Neuropathy, hereditary motor and sensory, 6A

Sequence similarities

Belongs to the TRAFAC class dynamin-like GTPase superfamily. Dynamin/Fzo/YdjA family.

Mitofusin subfamily.

Contains 1 dynamin-type G (guanine nucleotide-binding) domain.

Post-translational modifications

Phosphorylated by PINK1.

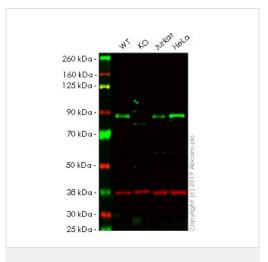
Ubiquitinated by non-degradative ubiquitin by PARK2, promoting mitochondrial fusion;

deubiquitination by USP30 inhibits mitochondrial fusion.

Cellular localization

Mitochondrion outer membrane. Colocalizes with BAX during apoptosis.

Images



Western blot - Anti-Mitofusin 2 antibody [EPR19796] (ab205236)

All lanes : Anti-Mitofusin 2 antibody [EPR19796] (ab205236) at 1/2000 dilution

Lane 1 : Wild-type HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate

Lane 2: MFN2 knockout HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate

Lane 3 : Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate

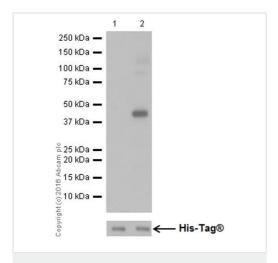
Lane 4: HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

Lysates/proteins at 20 µg per lane.

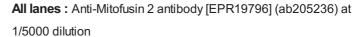
Predicted band size: 86 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab205236 observed at 86 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab205236 was shown to recognize MFN2 (Mitofusin 2) in wild-type HEK-293 cells as signal was lost at the expected MW in MFN2 knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and MFN2 knockout samples were subjected to SDS-PAGE. The membrane was blocked with 3% Milk. Ab205236 and ab8245 (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/2000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed ab216776 secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-Mitofusin 2 antibody [EPR19796] (ab205236)



Lane 1 : Human Mitofusin 1 recombinant protein fragment
Lane 2 : Human Mitofusin 2 recombinant protein fragment

Lysates/proteins at 0.01 µg per lane.

Secondary

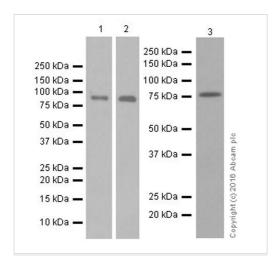
All lanes : Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 86 kDa

Exposure time: 1 second

Blocking/Dilution buffer: 5% NFDM/TBST.

Human Mitofusin 1 recombinant protein fragment contains aa130-485 with a His-Tag®. Human Mitofusin 2 recombinant protein fragment contains aa151-506 with a His-Tag®.



Western blot - Anti-Mitofusin 2 antibody [EPR19796] (ab205236)

All lanes : Anti-Mitofusin 2 antibody [EPR19796] (ab205236) at 1/5000 dilution

Lane 1: Human fetal heart lysate

Lane 2: Human fetal kidney lysate

Lane 3: Human fetal liver lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/100000 dilution

Predicted band size: 86 kDa

Observed band size: 86 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure times: Lane 1: 15 seconds; Lane 2 and 3: 30 seconds.

The expression profile is consistent with the literature (PMID

14561718; PMID 25574749).

All lanes : Anti-Mitofusin 2 antibody [EPR19796] (ab205236) at 1/2000 dilution

Lane 1 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

Lane 2: HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate

Lysates/proteins at 20 µg per lane.

2 250 kDa -250 kDa -150 kDa 🕳 150 kDa -100 kDa -100 kDa -75 kDa 🕳 75 kDa 🕳 50 kDa -50 kDa -37 kDa -Copyright (c) 2016 Abcam plo 37 kDa -25 kDa **—** 20 kDa **—** 25 kDa -20 kDa -15 kDa 🕳 15 kDa 🕳 10 kDa 🕳 10 kDa 🕳

Western blot - Anti-Mitofusin 2 antibody [EPR19796] (ab205236)

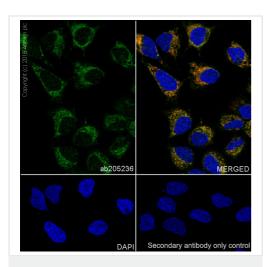
Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 86 kDa **Observed band size:** 86 kDa

Exposure time: 10 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.



Immunocytochemistry/ Immunofluorescence - Anti-Mitofusin 2 antibody [EPR19796] (ab205236)

ab205236 MERGED

DAPI

Secondary antibody only control

Immunocytochemistry/ Immunofluorescence - Anti-Mitofusin 2 antibody [EPR19796] (ab205236)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling Mitofusin 2 with ab205236 at 1/250 dilution, followed by Goat anti-rabbit lgG (Alexa Fluor[®] 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing mitochondrial staining on HeLa cell line.

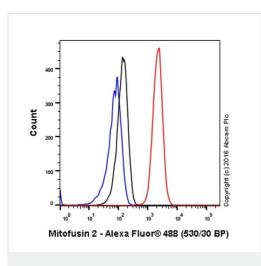
The nuclear counter stain is DAPI (blue). COX IV is detected with ab33985 (Anti-COX IV antibody [mAbcam33985] - Mitochondrial Marker), at 1/200 dilution, followed by secondary detection using ab150120 Alexa Fluor Boat anti-Mouse (red).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat anti-rabbit lgG (Alexa Fluor[®] 488) (ab150077) at 1/1000 dilution.

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling Mitofusin 2 with ab205236 at 1/250 dilution, followed by Goat anti-rabbit lgG (Alexa Fluor[®] 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing cytoplasmic staining on HeLa cell line.

The nuclear counter stain is DAPI (blue). Tubulin is detected with <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/200 dilution, followed by secondary detection using <u>ab150120</u> Alexa Fluor[®] 594 Goat anti-Mouse (red).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat anti-rabbit lgG (Alexa Fluor[®] 488) (ab150077) at 1/1000 dilution.



Flow Cytometry (Intracellular) - Anti-Mitofusin 2 antibody [EPR19796] (ab205236) Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling Mitofusin 2 with ab205236 at 1/700 dilution (red) compared with a rabbit monoclonal lgG isotype control (ab172730; black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti rabbit lgG (Alexa Fluorr[®] 488) at 1/2000 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-Mitofusin 2 antibody [EPR19796] (ab205236)

Mitofusin 2 was immunoprecipitated from 0.35 mg of HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate with ab205236 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab205236 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/10000 dilution.

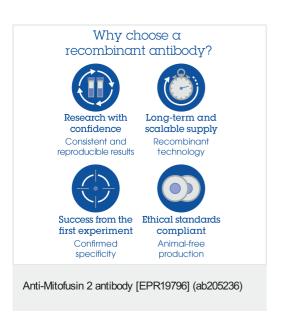
Lane 1: HeLa whole cell lysate, 10 µg (Input).

Lane 2: ab205236 IP in HeLa whole cell lysate.

Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab205236 in HeLa whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 3 seconds.



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