abcam

Product datasheet

Anti-MTCO1 antibody [1D6E1A8] ab14705

Recombinant

*** * * 42 Abreviews 474 References 12 Images

Overview

Product name Anti-MTCO1 antibody [1D6E1A8]

Description Mouse monoclonal [1D6E1A8] to MTCO1

Host species Mouse

Tested applications Suitable for: ICC, IHC-P, WB, Flow Cyt

Species reactivity Reacts with: Mouse, Rat, Human

Predicted to work with: Sheep, Goat, Cat, Dog, Pig, Caenorhabditis elegans, Zebrafish, Quail,

Rhesus monkey, Chinese hamster, Common marmoset ...

Immunogen Full length native protein (purified). This information is proprietary to Abcam and/or its suppliers.

Positive control WB: HeLa, MCF7, C6, PC-12, Neuro-2a, RAW 264.7 whole cell lysates. Human skeletal, Rat

heart and Rat liver tissue lysates. IHC-P: Human kidney, Mouse colon, and Rat colon tissue. ICC: Human cervical adenocarcinoma, Mouse neuroblastoma neuroblast, and rat glial tumor cells.

Flow Cyt: HeLa, Neuro-2a, and C6 cells.

General notes This product has switched from a hybridoma to recombinant production method on 24th May

2023.

Western blot protocol advice:

For best results with this antibody in Western blot, do not boil samples before loading onto the gel. Boiling of the sample will cause a loss of signal.

Hydrophobic intrinsic membrane proteins such as the core mtDNA-encoded proteins of the mitochondrial OXPHOS complexes tend to run faster in SDS-PAGE than predicted by their amino acid composition. This is likely due to incomplete unfolding of the protein and a more negative charge:mass ratio.

This antibody clone [1D6E1A8] is manufactured by Abcam. If you require a different buffer formulation or a particular conjugate for your experiments, please contact **orders@abcam.com**.

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.

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at -20°C.

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

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

Purity Protein A purified

ClonalityMonoclonalClone number1D6E1A8IsotypeIgG2aLight chain typekappa

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab14705 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
ICC	*** <u>*</u> (2)	1/100.
IHC-P	★★★★★ (13)	1/5000.
WB	★★★★ <u>(18)</u>	1/1000.
Flow Cyt	★★★★★ (1)	1/500.

Target

Function Cytochrome c oxidase is the component of the respiratory chain that catalyzes the reduction of

oxygen to water. Subunits 1-3 form the functional core of the enzyme complex. CO I is the catalytic subunit of the enzyme. Electrons originating in cytochrome c are transferred via the copper A center of subunit 2 and heme A of subunit 1 to the bimetallic center formed by heme A3 and

copper B.

Pathway Energy metabolism; oxidative phosphorylation.

Involvement in disease Defects in MT-CO1 are a cause of Leber hereditary optic neuropathy (LHON) [MIM:535000].

LHON is a maternally inherited disease resulting in acute or subacute loss of central vision, due to optic nerve dysfunction. Cardiac conduction defects and neurological defects have also been described in some patients. LHON results from primary mitochondrial DNA mutations affecting

the respiratory chain complexes.

Defects in MT-CO1 are a cause of anemia sideroblastic acquired idiopathic (AISA) [MIM:516030]; a disease characterized by inadequate formation of heme and excessive

accumulation of iron in mitochondria.

Defects in MT-CO1 are a cause of mitochondrial complex IV deficiency (MT-C4D) [MIM:220110]; also known as cytochrome c oxidase deficiency. A disorder of the mitochondrial respiratory chain

with heterogeneous clinical manifestations, ranging from isolated myopathy to severe multisystem disease affecting several tissues and organs. Features include hypertrophic cardiomyopathy, hepatomegaly and liver dysfunction, hypotonia, muscle weakness, excercise intolerance, developmental delay, delayed motor development and mental retardation. A subset of patients manifest Leigh syndrome.

Defects in MT-CO1 are associated with recurrent myoglobinuria mitochondrial (RM-MT) [MIM:550500]. Recurrent myoglobinuria is characterized by recurrent attacks of rhabdomyolysis (necrosis or disintegration of skeletal muscle) associated with muscle pain and weakness, and followed by excretion of myoglobin in the urine.

Defects in MT-CO1 are a cause of deafness sensorineural mitochondrial (DFNM) [MIM:500008]. DFNM is a form of non-syndromic deafness with maternal inheritance. Affected individuals manifest progressive, postlingual, sensorineural hearing loss involving high frequencies. Defects in MT-CO1 are a cause of colorectal cancer (CRC) [MIM:114500].

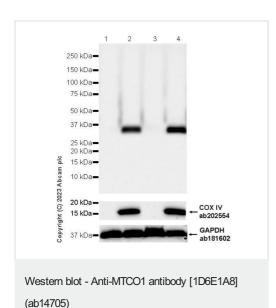
Sequence similarities

Cellular localization

Belongs to the heme-copper respiratory oxidase family.

Mitochondrion inner membrane.

Images



All lanes : Anti-MTCO1 antibody [1D6E1A8] (ab14705) at 1/1000 dilution

Lane 1: HeLa (human cervical adenocarcinoma epithelial cell) non-mitochondrial fraction

Lane 2: HeLa mitochondrial fraction

Lane 3: MCF7(human breast adenocarcinoma epithelial cell) non-mitochondrial fraction

Lane 4: MCF7 mitochondrial fraction

Lysates/proteins at 20 µg per lane.

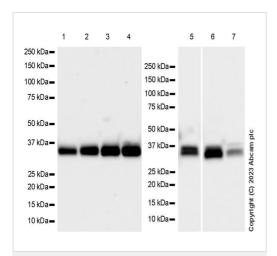
Secondary

All lanes : Peroxidase-Conjugated Goat anti-Mouse IgG (H+L) at 1/10000 dilution

Observed band size: 35 kDa

Exposure time: 5 seconds

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



Western blot - Anti-MTCO1 antibody [1D6E1A8] (ab14705)

All lanes : Anti-MTCO1 antibody [1D6E1A8] (ab14705) at 1/1000 dilution

Lane 1: C6 (rat glial tumor glial cell) whole cell lysate

Lane 2: PC-12 (rat adrenal gland pheochromocytoma cell) whole cell lysate

Lane 3: Neuro-2a (mouse neuroblastoma neuroblast) whole cell lysate

Lane 4: RAW 264.7 (mouse Abelson murine leukemia virusinduced tumor macrophage) whole cell lysate

Lane 5: Human skeletal muscle tissue lysate

Lane 6 : Rat heart tissue lysate

Lane 7 : Rat liver tissue lysate

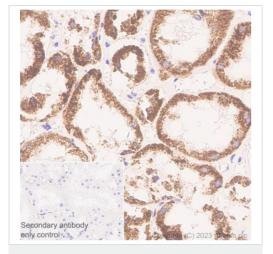
Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Peroxidase-Conjugated Goat anti-Mouse IgG (H+L) at 1/10000 dilution

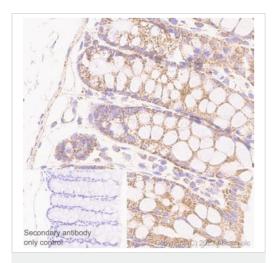
Observed band size: 35 kDa

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



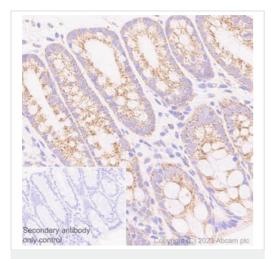
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MTCO1 antibody
[1D6E1A8] (ab14705)

Immunohistochemical analysis of paraffin-embedded Human kidney tissue labelling MTCO1 with ab14705 at 1/5000 dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on Human kidney. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Heat mediated antigen retrieval was performed with Citrate buffer (pH 6.0, epitope retrieval Solution2) for 20 mins. Counterstained with hematoxylin.



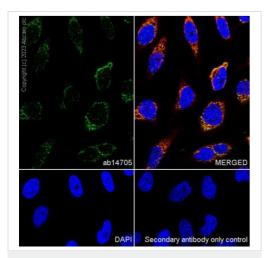
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MTCO1 antibody
[1D6E1A8] (ab14705)

Immunohistochemical analysis of paraffin-embedded Mouse colon tissue labelling MTCO1 with ab14705 at 1/5000 dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on Mouse colon. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Heat mediated antigen retrieval was performed with Citrate buffer (pH 6.0, epitope retrieval Solution2) for 20 mins. Counterstained with hematoxylin.



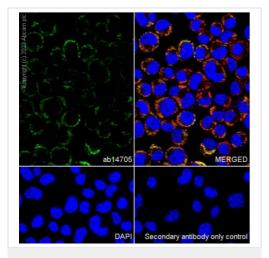
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MTCO1 antibody
[1D6E1A8] (ab14705)

Immunohistochemical analysis of paraffin-embedded Rat colon tissue labelling MTCO1 with ab14705 at 1/5000 dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on Rat colon. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Heat mediated antigen retrieval was performed with Citrate buffer (pH 6.0, epitope retrieval Solution2) for 20 mins. Counterstained with hematoxylin.



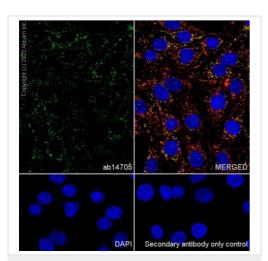
Immunocytochemistry/ Immunofluorescence - Anti-MTCO1 antibody [1D6E1A8] (ab14705)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized Human cervical adenocarcinoma epithelial cells labeling MTCO1 with ab14705 at 1/100 dilution, followed by ab150113 Goat Anti-Mouse IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 dilution. Confocal image showing cytoplasmic and membranous staining in subsets of Human cervical adenocarcinoma epithelial cells. ab186735 Anti-TOMM20 antibody [EPR15581-54] - Mitochondrial Marker was used to counterstain tubulin at 1/400 dilution. The Nuclear counterstain was DAPI (Blue). Secondary antibody only control: Secondary antibody is ab150080 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 594) preadsorbed at 1/1000 dilution.



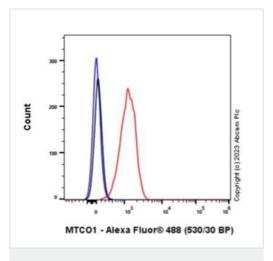
Immunocytochemistry/ Immunofluorescence - Anti-MTCO1 antibody [1D6E1A8] (ab14705)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized Mouse neuroblastoma neuroblast cells labeling MTCO1 with ab14705 at 1/100 dilution, followed by ab150113 Goat Anti-Mouse IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 dilution. Confocal image showing cytoplasmic and membranous staining in subsets of Mouse neuroblastoma neuroblast cells. ab186735 Anti-TOMM20 antibody [EPR15581-54] - Mitochondrial Marker was used to counterstain tubulin at 1/400 dilution. The Nuclear counterstain was DAPI (Blue). Secondary antibody only control: Secondary antibody is ab150080 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 594) preadsorbed at 1/1000 dilution.

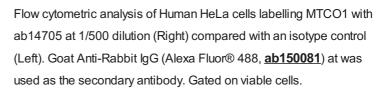


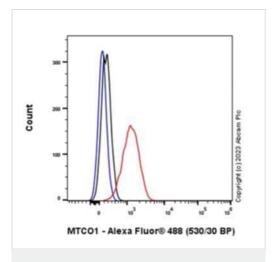
Immunocytochemistry/ Immunofluorescence - Anti-MTCO1 antibody [1D6E1A8] (ab14705)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized Rat glial tumor cells labeling MTCO1 with ab14705 at 1/100 dilution, followed by ab150113 Goat Anti-Mouse IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 dilution. Confocal image showing cytoplasmic and membranous staining in subsets of Rat glial tumor cells. ab186735 Anti-TOMM20 antibody [EPR15581-54] - Mitochondrial Marker was used to counterstain tubulin at 1/400 dilution. The Nuclear counterstain was DAPI (Blue). Secondary antibody only control: Secondary antibody is ab150080 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 594) preadsorbed at 1/1000 dilution.



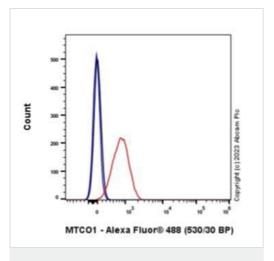
Flow Cytometry - Anti-MTCO1 antibody [1D6E1A8] (ab14705)





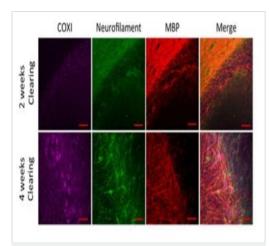
Flow Cytometry - Anti-MTCO1 antibody [1D6E1A8] (ab14705)

Flow cytometric analysis of Neuro-2a cells labelling MTCO1 with ab14705 at 1/500 dilution (Right) compared with an isotype control (Left). Goat Anti-Rabbit IgG (Alexa Fluor® 488, **ab150081**) at was used as the secondary antibody. Gated on viable cells.



Flow Cytometry - Anti-MTCO1 antibody [1D6E1A8] (ab14705)

Flow cytometric analysis of C6 cells labelling MTCO1 with ab14705 at 1/500 dilution (Right) compared with an isotype control (Left). Goat Anti-Rabbit lgG (Alexa Fluor® 488, <u>ab150081</u>) at was used as the secondary antibody. Gated on viable cells.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MTCO1 antibody
[1D6E1A8] (ab14705)

Image from Phillips J et al., Sci Rep. 2016 May 16;6:26013. Fig 3b doi: 10.1038/srep26013. Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/.

This image was generated using a previous batch manufactured using hybridoma production method.

 $250\,\mu m$ human cerebellar sections from control individuals and a patient with mitochondrial disease underwent passive clearing at $37\,^{\circ}\text{C}$ for 2 or 4 weeks.

The quality of immunofluorescent staining is determined by duration of passive clearing; 2 weeks of passive clearing produced minimal labelling of the white matter in the granule cell layer (NF-H; green; 488 nm and MBP; red, 546 nm) with an absence of labelling of mitochondria (MTCO1 (COXI) (ab14705, 1/100); purple; 647 nm; Extending passive clearing to 4 weeks improved the quality of stain with identifiable Purkinje cells and their axons (NF-H, green; 488 nm) and their myelin sheaths (MBP; red, 546 nm) and mitochondria (MTCO1 (COXI) (ab14705, 1/100); purple; 647 nm.

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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