abcam

Product datasheet

Anti-COX IV antibody [EPR9442(ABC)] - Mitochondrial Loading Control ab202554

Recombinant RabMAb

**** 2 Abreviews 27 References 11 Images

Overview

Product name Anti-COX IV antibody [EPR9442(ABC)] - Mitochondrial Loading Control

Description Rabbit monoclonal [EPR9442(ABC)] to COX IV - Mitochondrial Loading Control

Host species Rabbit

Tested applications Suitable for: Flow Cyt (Intra), ICC/IF, IP, IHC-P, WB

Species reactivity Reacts with: Mouse, Rat, Human

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

Positive control WB: Human fetal heart lysate; HepG2 whole cell lysate; Mouse and rat heart lysates. IHC-P:

Human hepatocellular carcinoma, Human cervix carcinoma, mouse kidney and rat cardiac muscle tissues. ICC/IF: HeLa and HepG2 cells. Flow Cyt (intra): MCF7 cells. IP: Human fetal heart whole

cell lysate.

General notesThis 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

1

Clonality Monoclonal

Clone number EPR9442(ABC)

Isotype IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab202554 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)		1/20. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
ICC/IF		1/1000.
IP		1/20.
IHC-P		1/500. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB	**** <u>(2)</u>	1/2000. Detects a band of approximately 17 kDa (predicted molecular weight: 20 kDa).

Target

Function This protein is one of the nuclear-coded polypeptide chains of cytochrome c oxidase, the terminal

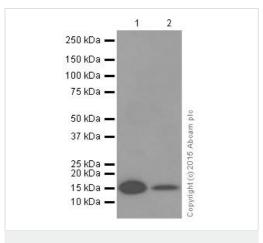
oxidase in mitochondrial electron transport.

Tissue specificity Ubiquitous.

Sequence similarities Belongs to the cytochrome c oxidase IV family.

Cellular localization Mitochondrion inner membrane.

Images



Western blot - Anti-COX IV antibody

[EPR9442(ABC)] - Mitochondrial Loading Control
(ab202554)

All lanes : Anti-COX IV antibody [EPR9442(ABC)] - Mitochondrial Loading Control (ab202554) at 1/2000 dilution

Lane 1: Human fetal heart lysate

Lane 2 : HepG2 (Human liver hepatocellular carcinoma) whole cell lysate

Lysates/proteins at 20 µg per lane.

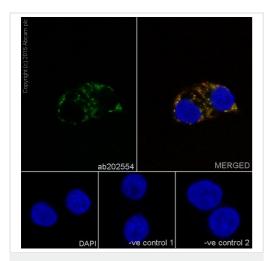
Secondary

All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 20 kDa **Observed band size:** 17 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.



Immunocytochemistry/ Immunofluorescence - Anti-COX IV antibody [EPR9442(ABC)] - Mitochondrial Loading Control (ab202554)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HepG2 (Human liver hepatocellular carcinoma) cells labeling COX IV with ab202554 at 1/1000 dilution, followed by Goat anti-rabbit lgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/500 dilution (green).

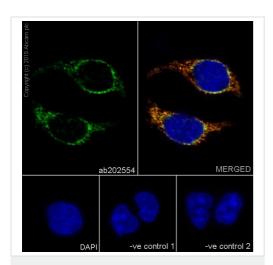
Cytoplasmic staining on HepG2 cells is observed.

The nuclear counter stain is DAPI (blue).

Tubulin is detected with <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution and <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).

The negative controls are as follows:

-ve control 1: ab202554 at 1/1000 dilution followed by <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution. -ve control 2: <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution followed by <u>ab150077</u> (Alexa Fluor®488 Goat Anti-Rabbit lgG H&L) at 1/500 dilution.



Immunocytochemistry/ Immunofluorescence - Anti-COX IV antibody [EPR9442(ABC)] - Mitochondrial Loading Control (ab202554)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cells from cervix adenocarcinoma) cells labeling COX IV with ab202554 at 1/1000 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/500 dilution (green).

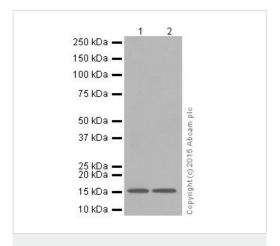
Cytoplasmic staining on HeLa cells is observed.

The nuclear counter stain is DAPI (blue).

Tubulin is detected with <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution and <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).

The negative controls are as follows:

-ve control 1: ab202554 at 1/1000 dilution followed by <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution. -ve control 2: <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution followed by <u>ab150077</u> (Alexa Fluor®488 Goat Anti-Rabbit lgG H&L) at 1/500 dilution.



Western blot - Anti-COX IV antibody

[EPR9442(ABC)] - Mitochondrial Loading Control
(ab202554)

All lanes : Anti-COX IV antibody [EPR9442(ABC)] - Mitochondrial Loading Control (ab202554) at 1/10000 dilution

Lane 1 : Mouse heart lysate

Lane 2 : Rat heart lysate

Lysates/proteins at 10 µg per lane.

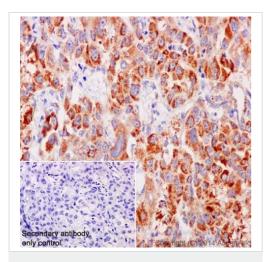
Secondary

All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 20 kDa **Observed band size:** 17 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-COX IV antibody

[EPR9442(ABC)] - Mitochondrial Loading Control (ab202554)

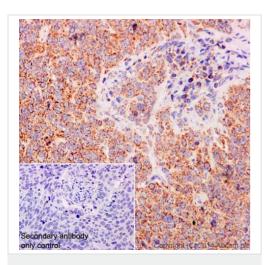
Immunohistochemical analysis of paraffin-embedded Human hepatocellular carcinoma tissue labeling COX IV with ab202554 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution.

Cytoplasmic staining on Human hepatocellular carcinoma tissue is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-COX IV antibody

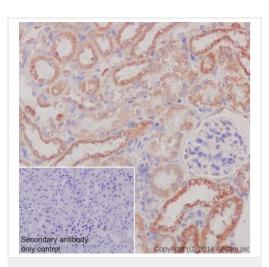
[EPR9442(ABC)] - Mitochondrial Loading Control (ab202554)

Immunohistochemical analysis of paraffin-embedded Human cervix carcinoma tissue labeling COX IV with ab202554 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution.

Cytoplasmic staining on Human cervix carcinoma tissue is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-COX IV antibody

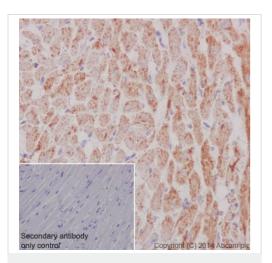
[EPR9442(ABC)] - Mitochondrial Loading Control (ab202554)

Immunohistochemical analysis of paraffin-embedded Mouse kidney tissue labeling COX IV with ab202554 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution.

Cytoplasmic staining on mouse kidney tissue is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-COX IV antibody

[EPR9442(ABC)] - Mitochondrial Loading Control (ab202554)

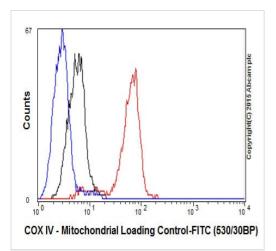
Immunohistochemical analysis of paraffin-embedded Rat cardiac muscle tissue labeling COX IV with ab202554 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution.

Cytoplasmic staining on Human cervix carcinoma tissue is observed.

Counter stained with Hematoxylin.

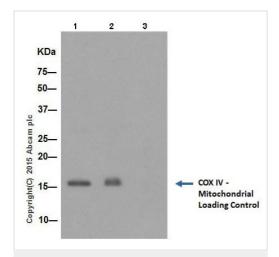
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-COX IV antibody [EPR9442(ABC)] - Mitochondrial Loading Control (ab202554)

Intracellular flow cytometric analysis of 2% paraformaldehyde-fixed MCF7 (Human breast adenocarcinoma cell line) cells labeling COX IVwith ab202554 at 1/20 dilution (red) compared with a rabbit monoclonal IgG isotype control (ab172730;black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti-rabbit IgG (FITC) at 1/150 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-COX IV antibody

[EPR9442(ABC)] - Mitochondrial Loading Control
(ab202554)

COX IV was immunoprecipitated from 1mg of Human fetal heart whole cell lysate with ab202554 at 1/20 dilution.

Western blot was performed from the immunoprecipitate using ab202554 at 1/1000 dilution.

VeriBlot for IP Detection Reagent (HRP) (<u>ab131366</u>) was used for detection at 1/1500 dilution.

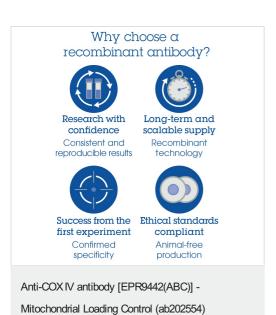
Lane 1: Human fetal heart whole cell lysate 10 µg (Input).

Lane 2: ab202554 IP in Human fetal heart whole cell lysate.

Lane 3: Rabbit monoclonal $\lg G (\underline{ab172730})$ instead of ab202554 in Human fetal heart whole cell lysate.

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

Exposure time: 3 seconds.



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