abcam

Product datasheet

Anti-CPT2 antibody [EPR13626] - C-terminal ab181114





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Overview

Product name Anti-CPT2 antibody [EPR13626] - C-terminal

Description Rabbit monoclonal [EPR13626] to CPT2 - C-terminal

Host species Rabbit

Tested applications Suitable for: IHC-P, WB, ICC/IF Species reactivity Reacts with: Mouse, Rat, Human

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

Positive control WB: HAP1, HeLa, MCF7 whole cell lysates; Mouse heart and kidney tissue lysates; kidney and

liver tissue lysates; Human fetal kidney and liver tissue lysates ICC/IF: MCF7 cells IHC-P: Rat

colon; Mouse kidney; Human liver carcinoma and skeletal muscle.

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

Liquid **Form**

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 Preservative: 0.01% Sodium azide

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

Purity Protein A purified

Clonality Monoclonal Clone number EPR13626

Isotype lgG

Applications

The Abpromise guarantee

Our Abpromise quarantee covers the use of ab181114 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
IHC-P		1/50 - 1/100. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See IHC antigen retrieval protocols .
WB		1/1000 - 1/10000. Detects a band of approximately 67 kDa (predicted molecular weight: 74 kDa).
ICC/IF		1/50 - 1/100.

Target

Pathway

Involvement in disease

Lipid metabolism; fatty acid beta-oxidation.

Defects in CPT2 are the cause of carnitine palmitoyltransferase 2 deficiency (CPT2D) [MIM:255110, 600649]; also known as CPT-II deficiency or CPT2 deficiency. CPT2D is an autosomal recessive disorder characterized by recurrent myoglobinuria, episodes of muscle pain, stiffness, and rhabdomyolysis. These symptoms are triggered by prolonged exercise, fasting or viral infection and patients are usually young adults. In addition to this classical, late-onset, muscular type, a hepatic or hepatocardiomuscular form has been reported in infants. Clinical pictures in these children or neonates include hypoketotic hypoglycemia, liver dysfunction, cardiomyopathy and sudden death.

Defects in CPT2 are the cause of carnitine palmitoyltransferase 2 deficiency, lethal neonatal (CPT2D-LN) [MIM:608836]; also known as lethal neonatal CPT-II deficiency. It is a lethal neonatal form of CPT2D. This rarely presentation is antenatal with cerebral periventricular cysts and cystic dysplastic kidneys. The clinical variability of the disease is likely attributed to the variable residual enzymatic activity.

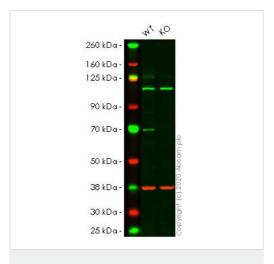
Sequence similarities

Belongs to the carnitine/choline acetyltransferase family.

Cellular localization

Mitochondrion inner membrane.

Images



Western blot - Anti-CPT2 antibody [EPR13626] - C-terminal (ab181114)

All lanes : Anti-CPT2 antibody [EPR13626] - C-terminal (ab181114) at 1/1000 dilution

Lane 1: Wild-type HeLa cell lysate

Lane 2: CPT2 knockout HeLa cell lysate

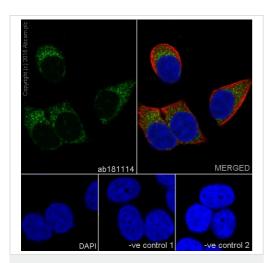
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

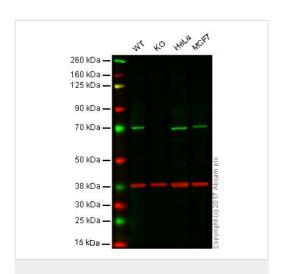
Predicted band size: 74 kDa Observed band size: 74 kDa

Lanes 1-2: Merged signal (red and green). Green - ab181114 observed at 74 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (ab8245) observed at 37 kDa.

ab181114 was shown to react with CPT2/CPT1 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line ab265931 (knockout cell lysate ab257180) was used. Wild-type HeLa and CPT2 knockout HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab181114 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) overnight at 4°C at a 1 in 1000 Dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye®800CW) preadsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye®680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-CPT2 antibody [EPR13626] - C-terminal (ab181114)



Western blot - Anti-CPT2 antibody [EPR13626] - C-terminal (ab181114)

Immunocytochemistry/Immunofluorescence analysis of MCF-7 (human breast carcinoma) cells labelling CPT2 with purified ab181114 at 1/100. Cells were fixed with 100% methanol and permeabilized with 0.1% Triton X-100. **ab150077**, Alexa Fluor[®] 488-conjugated goat anti-rabbit lgG (1/1000) was used as the secondary antibody. The cells were co-stained with **ab7291**, a mouse anti-tubulin antibody (1/1000) using **ab150120**, an Alexa Fluor[®] 594-conjugated goat anti-mouse lgG (1/1000) as the secondary. Nuclei couterstained with DAPI (blue).

For negative control 1, rabbit primary antibody was used, followed by anti-mouse secondary antibody (<u>ab150120</u>). For negative control 2, mouse primary antibody (<u>ab7291</u>) was used followed by anti-rabbit secondary antibody (<u>ab150077</u>).

All lanes : Anti-CPT2 antibody [EPR13626] - C-terminal (ab181114) at 1/1000 dilution

Lane 1: Wild-type HAP1 whole cell lysate

Lane 2: CPT2 knockout HAP1 whole cell lysate

Lane 3 : HeLa whole cell lysate

Lane 4 : MCF7 whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 74 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab181114 observed at 70 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab181114 was shown to specifically react with CPT2 in wild-type HAP1 cells. No band was observed when CPT2 knockout samples were examined. Wild-type and CPT2 knockout samples were subjected to SDS-PAGE. Ab181114 and ab8245 (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/10,000 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 <u>ab216776</u> secondary antibodies at 1/10,000 dilution for 1 hour at room temperature before imaging.

1 2 3 4

250 kDa —
150 kDa —
100 kDa —
75 kDa —
37 kDa —
25 kDa —
20 kDa —
15 kDa —
10 kDa —
110 kDa —

Western blot - Anti-CPT2 antibody [EPR13626] - C-terminal (ab181114)

All lanes : Anti-CPT2 antibody [EPR13626] - C-terminal (ab181114) at 1/2000 dilution (purified)

Lane 1: Human fetal kidney tissue lysate

Lane 2: MCF-7 (human breast carcinoma) whole cell lysate

Lane 3 : Mouse heart tissue lysate

Lane 4 : Mouse kidney tissue lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Anti-Rabbit lgG (HRP), specific to the non-reduced form of lgG at 1/2000 dilution

Predicted band size: 74 kDa **Observed band size:** 67 kDa

1 2

250 KDa —
150 KDa —
150 KDa —
75 KDa —
37 KDa —
25 KDa —
20 KDa —
15 KDa —
15 KDa —
10 KDa —
11 KDa —

Western blot - Anti-CPT2 antibody [EPR13626] - C-terminal (ab181114)

Blocking and diluting buffer 5% NFDM/TBST

All lanes : Anti-CPT2 antibody [EPR13626] - C-terminal (ab181114) at 1/1000 dilution (purified)

Lane 1 : Rat kidney tissue lysate

Lane 2 : Rat liver tissue lysate

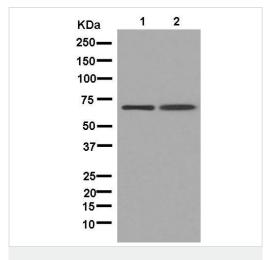
Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 74 kDa **Observed band size:** 67 kDa

Blocking and diluting buffer 5% NFDM/TBST



Western blot - Anti-CPT2 antibody [EPR13626] - C-terminal (ab181114)

All lanes : Anti-CPT2 antibody [EPR13626] - C-terminal (ab181114) at 1/10000 dilution (unpurified)

Lane 1 : Human fetal liver tissue lysate

Lane 2: Human fetal kidney tissue lysate

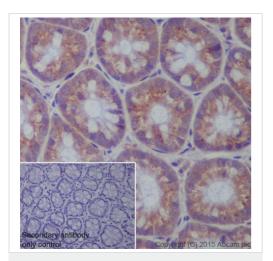
Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab136636) at 1/500

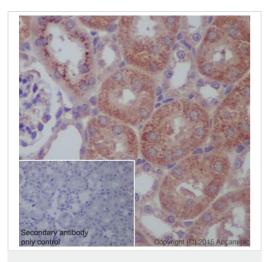
dilution

Predicted band size: 74 kDa **Observed band size:** 67 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CPT2 antibody [EPR13626] - C-terminal (ab181114)

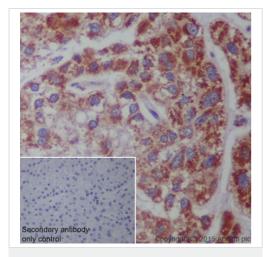
Immunohistochemical analysis of paraffin embedded rat colon tissue section labelling CPT2 with purified ab181114 at dilution of 1/50. The secondary antibody used was HRP-conjugated Goat Anti-Rabbit IgG H&L (ab97051) at dilution of 1/500. The sample was counter-stained with hematoxylin. Antigen retrieval was performed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control and is shown in the inset.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CPT2 antibody

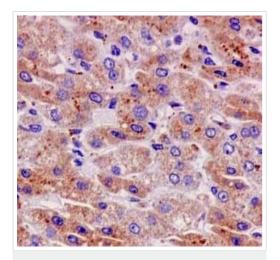
[EPR13626] - C-terminal (ab181114)

Immunohistochemical analysis of paraffin embedded mouse kidney tissue section labelling CPT2 with purified ab181114 at dilution of 1/50. The secondary antibody used was HRP-conjugated Goat Anti-Rabbit IgG H&L (ab97051) at dilution of 1/500. The sample was counter-stained with hematoxylin. Antigen retrieval was performed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control and is shown in the inset.



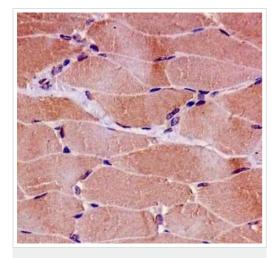
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CPT2 antibody
[EPR13626] - C-terminal (ab181114)

Immunohistochemical analysis of paraffin embedded human liver carcinoma tissue section labelling CPT2 with purified ab181114 at dilution of 1/50. The secondary antibody used was Goat Anti-Rabbit IgG H&L (HRP) (ab97051), at a dilution of 1/500. The sample was counter-stained with hematoxylin. Antigen retrieval was performed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control and is shown in the inset.



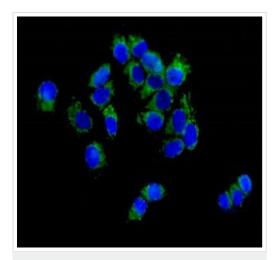
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CPT2 antibody
[EPR13626] - C-terminal (ab181114)

Immunohistochemical analysis of paraffin-embedded Human liver tissue labeling CPT2 with unpurified ab181114 at 1/100 dilution, followed by prediluted HRP Polymer for Rabbit lgG. Counter stained with Hematoxylin.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CPT2 antibody
[EPR13626] - C-terminal (ab181114)

Immunohistochemical analysis of paraffin-embedded Human skeletal muscle tissue labeling CPT2 with unpurified ab181114 at 1/100 dilution, followed by prediluted HRP Polymer for Rabbit lgG. Counter stained with Hematoxylin.



Immunocytochemistry/ Immunofluorescence - Anti-CPT2 antibody [EPR13626] - C-terminal (ab181114)

Immunofluorescent analysis of 4% paraformaldehyde-fixed MCF7 cells labeling CPT2 with unpurified ab181114 at 1/100 dilution, followed by Goat anti rabbit lgG (Alexa Fluor® 488) secondary antibody at 1/200 dilution. Counter stained with Dapi.



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