# abcam

# Product datasheet

# Anti-CPT1A antibody [EPR21843-71-1C] - BSA and Azide free ab234906



リコンピナント

RabMAb

# 画像数 11

#### 製品の概要

製品名 Anti-CPT1A antibody [EPR21843-71-1C] - BSA and Azide free

製品の詳細 Rabbit monoclonal [EPR21843-71-1C] to CPT1A - BSA and Azide free

由来種 Rabbit

特異性 Our WB images were generated by testing un-bolied samples.

アプリケーション 適用あり: Flow Cyt (Intra), WB, IHC-P, ICC/IF, IP

種交差性 交差種: Human

免疫原 Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

ポジティブ・コントロール WB: Wild-type HAP1 whole cell lysate; HEK-293T, HeLa, SK-OV-3, MCF7 and HepG2 whole cell

lysates; Human kidney lysate; His-tagged human CPT1A recombinant protein (aa406-755). IHC-P: Human kidney and ovarian carcinoma tissues. ICC/IF: HeLa and SK-OV-3 cells. Flow Cyt

(intra): HeLa cells. IP: SK-OV-3 whole cell lysate.

特記事項 ab234906 is the carrier-free version of ab220789.

Our <u>carrier-free</u> antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our **conjugation kits** for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

This product is compatible with the Maxpar<sup>®</sup> Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar<sup>®</sup> is a trademark of Fluidigm Canada Inc.

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

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For more information see here.

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

#### 製品の特性

製品の状態 Liquid

保存方法 Shipped at 4°C. Store at +4°C. Do Not Freeze.

**バッファー** pH: 7.2

Constituent: PBS

キャリア・フリー はい

精製度 Protein A purified

**ポリ/モノ** モノクローナル

**クローン名** EPR21843-71-1C

アイソタイプ lgG

#### アプリケーション

**The Abpromise guarantee** <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab234906の使用に適用されます アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
Flow Cyt (Intra)		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 88 kDa (predicted molecular weight: 88 kDa). Our WB images were generated by testing un-bolied samples.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.

# ターゲット情報

組織特異性 Strong expression in kidney and heart, and lower in liver and skeletal muscle.

גליבל Lipid metabolism; fatty acid beta-oxidation.

**関連疾患**Defects in CPT1A are the cause of carnitine palmitoyltransferase 1A deficiency (CPT1AD)

[MIM:255120]; also known as CPT-I deficiency or CPT1A deficiency. CPT1AD is a rare autosomal recessive metabolic disorder of long-chain fatty acid oxidation characterized by severe

episodes of hypoketotic hypoglycemia usually occurring after fasting or illness. Onset is in infancy

or early childhood.

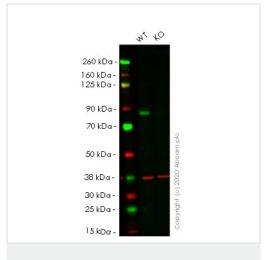
配列類似性

Belongs to the carnitine/choline acetyltransferase family.

細胞内局在

Mitochondrion outer membrane.

#### 画像



Western blot - Anti-CPT1A antibody [EPR21843-71-1C] - BSA and Azide free (ab234906)

**All lanes :** Anti-CPT1A antibody [EPR21843-71-1C] (<u>ab220789</u>) at 1/1000 dilution

Lane 1: Wild-type HEK-293T cell lysate

Lane 2: CPT1A knockout HEK-293T cell lysate

Lysates/proteins at 20 µg per lane.

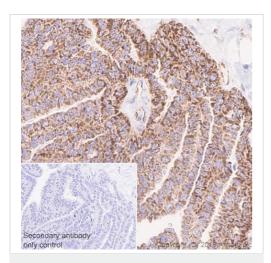
Performed under reducing conditions.

Predicted band size: 88 kDa Observed band size: 88 kDa

This data was developed using the same antibody clone in a different buffer formulation (ab220789).

Lanes 1-2: Merged signal (red and green). Green - <u>ab220789</u> observed at 88 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (<u>ab8245</u>) observed at 37 kDa.

ab220789 was shown to react with CPT1A in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line ab266319 (knockout cell lysate ab256880) was used. Wild-type HEK-293T and CPT1A knockout HEK-293T 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. ab220789 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.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CPT1A antibody

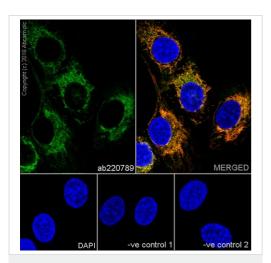
[EPR21843-71-1C] - BSA and Azide free (ab234906)

Immunohistochemical analysis of paraffin-embedded human ovarian carcinoma tissue labeling CPT1A with <u>ab220789</u> at 1/1000 dilution, followed by Goat Anti-Rabbit lgG H&L (HRP) Ready to use. Granular cytoplasmic staining in human ovarian carcinoma (PMID: 26716645). Counter stained with hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab220789).

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



Immunocytochemistry/ Immunofluorescence - Anti-CPT1A antibody [EPR21843-71-1C] - BSA and Azide free (ab234906)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized SK-OV-3 (human ovarian cancer cell line) cells labeling CPT1A with <u>ab220789</u> at 1/100 dilution followed by Goat Anti-Rabbit lgG H&L (Alexa Fluor<sup>®</sup> 488) (<u>ab150077</u>) secondary antibody at 1/1000 dilution (green). Confocal image showing mitochondrial staining in SK-OV-3 cell line.

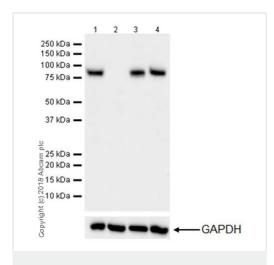
The nuclear counter stain is DAPI (blue). COX IV is detected with Anti-COX IV antibody [mAbcam33985] - Mitochondrial Marker (<u>ab33985</u>) at 1/1000 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor<sup>®</sup> 594) preadsorbed (<u>ab150120</u>) (red).

The negative controls are as follows:

-ve control 1: <u>ab220789</u> at 1/100 dilution followed by Goat Anti-Mouse IgG H&L (Alexa Fluor<sup>®</sup> 594) preadsorbed (<u>ab150120</u>) secondary antibody at 1/1000 dilution.

-ve control 2: Anti-COX IV antibody [mAbcam33985] - Mitochondrial Marker (ab33985) at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab220789).



Western blot - Anti-CPT1A antibody [EPR21843-71-1C] - BSA and Azide free (ab234906)

**All lanes :** Anti-CPT1A antibody [EPR21843-71-1C] (<u>ab220789</u>) at 1/5000 dilution

Lane 1: Wild-type HAP1 whole cell lysate

Lane 2: CPT1A knockout HAP1 whole cell lysate

Lane 3: HeLa (human epithelial cell line from cervix

adenocarcinoma) whole cell lysate

Lane 4: SK-OV-3 (human ovarian cancer cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

#### Secondary

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

Developed using the ECL technique.

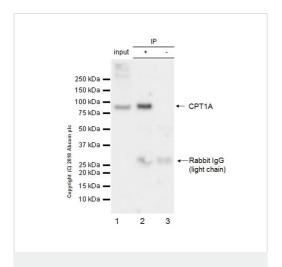
**Predicted band size:** 88 kDa **Observed band size:** 88 kDa

Exposure time: 92 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

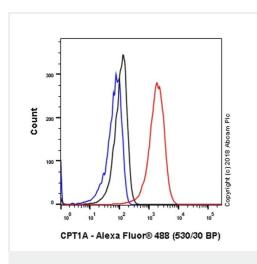
ab220789 was shown to specifically react with CPT1A in wild-type HAP1 cells as signal was lost in CPT1A knockout cells. Wild-type and CPT1A knockout samples were subjected to SDS-PAGE.
ab220789 and ab181602 (Rabbit anti-GAPDH loading control) were incubated 1 hour at room temperature at 1/5000 dilution and 1/200,000 dilution respectively. Blots were developed with Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (ab97051) secondary antibody at 1/100,000 dilution for 1 hour at room temperature before imaging. The blot was developed on a BIO-RAD® ChemiDoc™ MP instrument using the ECL technique.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab220789).



Immunoprecipitation - Anti-CPT1A antibody

[EPR21843-71-1C] - BSA and Azide free (ab234906)



Flow Cytometry (Intracellular) - Anti-CPT1A antibody [EPR21843-71-1C] - BSA and Azide free (ab234906)

CPT1A was immunoprecipitated from 0.35 mg of SK-OV-3 (human ovarian cancer cell line) whole cell lysate with <u>ab220789</u> at 1/30 dilution. Western blot was performed from the immunoprecipitate using <u>ab220789</u> at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (<u>ab131366</u>), was used for detection at 1/5000 dilution.

Lane 1: SK-OV-3 whole cell lysate 10 µg (Input).

Lane 2: ab220789 IP in SK-OV-3 whole cell lysate.

Lane 3: Rabbit monoclonal  $\lg G$  ( $\underline{ab172730}$ ) instead of  $\underline{ab220789}$  in SK-OV-3 whole cell lysate.

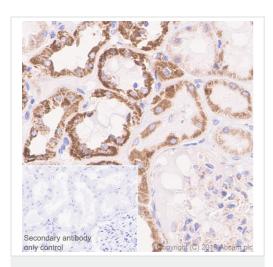
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 30 seconds.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab220789).

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol-permeabilized HeLa (human epithelial cell line from cervix adenocarcinoma) cell line labeling CPT1A with <a href="mailto:ab220789">ab220789</a> at 1/600 dilution (red) compared with a Rabbit lgG, monoclonal [EPR25A] - Isotype Control (<a href="mailto:ab172730">ab172730</a>) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) (<a href="mailto:ab150077">ab150077</a>) at 1/2000 dilution was used as the secondary antibody.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab220789).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CPT1A antibody

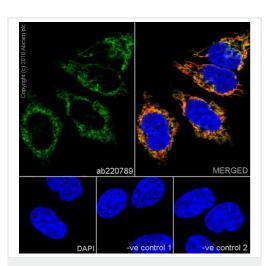
[EPR21843-71-1C] - BSA and Azide free (ab234906)

Immunohistochemical analysis of paraffin-embedded human kidney tissue labeling CPT1A with <u>ab220789</u> at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Granular cytoplasmic staining in human kidney (PMID: 18192268; PMID: 28956034). Counter stained with hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab220789).

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



Immunocytochemistry/ Immunofluorescence - Anti-CPT1A antibody [EPR21843-71-1C] - BSA and Azide free (ab234906)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (human epithelial cell line from cervix adenocarcinoma) cells labeling CPT1A with <u>ab220789</u> at 1/100 dilution followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor<sup>®</sup> 488) (<u>ab150077</u>) secondary antibody at 1/1000 dilution (green). Confocal image showing mitochondrial staining in HeLa cell line.

The nuclear counter stain is DAPI (blue). COX IV is detected with Anti-COX IV antibody [mAbcam33985] - Mitochondrial Marker (ab33985) at 1/1000 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor<sup>®</sup> 594) preadsorbed (ab150120) (red).

The negative controls are as follows:

-ve control 1: <u>ab220789</u> at 1/100 dilution followed by Goat Anti-Mouse IgG H&L (Alexa Fluor<sup>®</sup> 594) preadsorbed (<u>ab150120</u>) secondary antibody at 1/1000 dilution.

-ve control 2: Anti-COX IV antibody [mAbcam33985] - Mitochondrial Marker (<u>ab33985</u>) at 1/1000 dilution, followed by Goat Anti-Rabbit lgG H&L (Alexa Fluor<sup>®</sup> 488) (<u>ab150077</u>).

This data was developed using the same antibody clone in a

1 2 3

250 kDa —

150 kDa —

75 kDa —

50 kDa —

37 kDa —

25 kDa —

20 kDa —

15 kDa —

10 kDa —

Western blot - Anti-CPT1A antibody [EPR21843-71-1C] - BSA and Azide free (ab234906)

different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab220789).

**All lanes :** Anti-CPT1A antibody [EPR21843-71-1C] (<u>ab220789</u>) at 1/1000 dilution

**Lane 1 :** MCF7 (Human breast adenocarcinoma epithelial cell) cell lysate

Lane 2 : HeLa (Human cervix adenocarcinoma epithelial cell) cell lvsate

Lane 3 : HepG2 (Human hepatocellular carcinoma epithelial cell) cell lysate

Lysates/proteins at 20 µg per lane.

#### Secondary

**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/20000 dilution

**Predicted band size:** 88 kDa **Observed band size:** 88 kDa

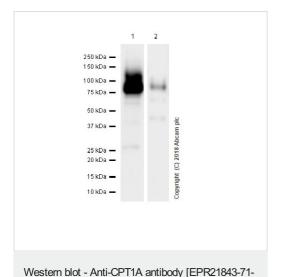
Exposure time: 180 seconds

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab220789).

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

CPT1A is strongly expressed in kidney and heart, and lower in liver and skeletal muscle.

We recommend loading higher amount of lysate or using lower antibody dilution to detect signal in HepG2 lysate.



1C] - BSA and Azide free (ab234906)

**All lanes :** Anti-CPT1A antibody [EPR21843-71-1C] (<u>ab220789</u>) at 1/1000 dilution

Lane 1: Human kidney tissue lysate

Lane 2: Human liver tissue lysate

Lysates/proteins at 20 µg per lane.

# Secondary

**All lanes :** VeriBlot for IP Detection Reagent (HRP) (<u>ab131366</u>) at 1/1000 dilution

Predicted band size: 88 kDa Observed band size: 88 kDa

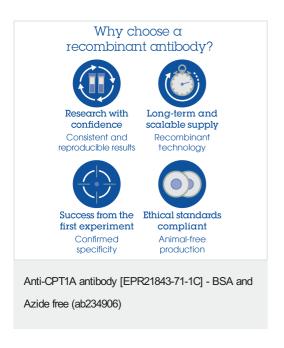
Exposure time: 92 seconds

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab220789).

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

CPT1A is strongly expressed in kidney and heart, and lower in liver and skeletal muscle.

We recommend loading higher amount of lysate or using lower antibody dilution to detect signal in liver lysate.



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