abcam

Product datasheet

Anti-Cyclophilin B antibody ab16045



★★★★★ 14 Abreviews 101 References 画像数 9

製品の概要

製品名 Anti-Cyclophilin B antibody

製品の詳細 Rabbit polyclonal to Cyclophilin B

由来種 Rabbit

特異性 Replenishment batches of our polyclonal antibody, ab16045 are tested in WB. Previous batches

were additionally validated in ICC/IF and IP. These applications are still expected to work and are covered by our Abpromise guarantee. You may also be interested in our alternative recombinant

antibody, ab178397.

アプリケーション 適用あり: WB, ICC/IF, IP

種交差性 交差種: Mouse, Rat, Horse, Chicken, Dog, Human

交差が予測される動物種: Cow, Pig, Xenopus laevis 🔷

免疫原 Synthetic peptide corresponding to Human Cyclophilin B aa 150 to the C-terminus (C terminal)

conjugated to keyhole limpet haemocyanin. (Peptide available as <u>ab16277</u>, <u>ab5016</u>)

特記事項

The Life Science industry has been in the grips of a reproducibility crisis for a number of years.

Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

製品の特性

製品の状態 Liquid

保存方法 Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -

80°C. Avoid freeze / thaw cycle.

バッファー pH: 7.40

Preservative: 0.02% Sodium azide

Constituent: PBS

Batches of this product that have a concentration < 1mg/ml may have BSA added as a stabilising agent. If you would like information about the formulation of a specific lot, please contact our scientific support team who will be happy to help.

精製度 Immunogen affinity purified

ポリ/モノ ポリクローナル

アイソタイプ IgG

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ab16045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
WB	****(7)	Use a concentration of 0.5 µg/ml. Detects a band of approximately 21 kDa (predicted molecular weight: 21 kDa).
ICC/IF	**** <u>(3)</u>	Use a concentration of 1 µg/ml.
IP		Use at an assay dependent concentration.

ターゲット情報

機能 PPlases accelerate the folding of proteins. It catalyzes the cis-trans isomerization of proline imidic

peptide bonds in oligopeptides.

関連疾患 Defects in PPIB are the cause of osteogenesis imperfecta type 9 (OI9) [MIM:259440]. OI9 is a

connective tissue disorder characterized by bone fragility, low bone mass and bowing of limbs due to multiple fractures. Short limb dwarfism and blue sclerae are observed in some but not all

patients.

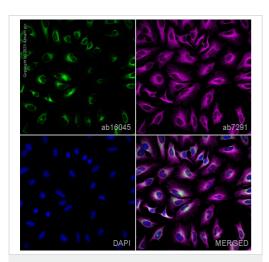
配列類似性 Belongs to the cyclophilin-type PPlase family. PPlase B subfamily.

Contains 1 PPlase cyclophilin-type domain.

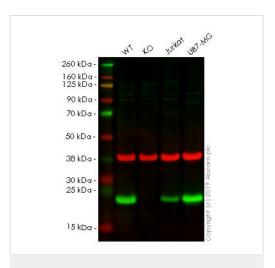
細胞内局在 Endoplasmic reticulum lumen. Melanosome. Identified by mass spectrometry in melanosome

fractions from stage I to stage IV.

画像



Immunocytochemistry/ Immunofluorescence - Anti-Cyclophilin B antibody (ab16045)



Western blot - Anti-Cyclophilin B antibody (ab16045)

ab16045 staining Cyclophilin B in HeLa cells. The cells were fixed with 100% methanol (5 min), permeabilized with 0.1% PBS-Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1%PBS-Tween for 1h. The cells were then incubated overnight at 4°C with ab16045 at 1µg/ml and ab7291, Mouse monoclonal [DM1A] to alpha Tubulin - Loading Control. Cells were then incubated with ab150081, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488), preadsorbed at 1/1000 dilution (shown in green) and ab150120, Goat polyclonal Secondary Antibody to Mouse IgG - H&L (Alexa Fluor® 594), pre-adsorbed at 1/1000 dilution (shown in pseudocolour magenta). Nuclear DNA was labelled with DAPI (shown in blue). Image was acquired with a high-content analyser (Operetta CLS, Perkin Elmer) and a maximum intensity projection of confocal sections is shown.

All lanes: Anti-Cyclophilin B antibody (ab16045) at 1/5000 dilution

Lane 1: Wild-type HAP1 whole cell lysate

Lane 2: PPIB knockout HAP1 whole cell lysate

Lane 3: Jurkat whole cell lysate

Lane 4: U87-MG whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 21 kDa **Observed band size:** 24 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab16045 observed at 24 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab16045 was shown to specifically react with PPIB in wild-type HAP1 cells as signal was lost in PPIB knockout cells. Wild-type and

PPIB knockout samples were subjected to SDS-PAGE. Ab16045 and <u>ab8245</u> (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/5000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed <u>ab216773</u> and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed <u>ab216776</u> secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.

1 2 3 4
250kDa__
150kDa__
150kDa__
100kDa__
75kDa__
50kDa__
37kDa__
25kDa__
20kDa__
15kDa__

Western blot - Anti-Cyclophilin B antibody (ab16045)

All lanes: Anti-Cyclophilin B antibody (ab16045) at 1 µg/ml

Lane 1: Rat Liver
Lane 2: Mouse 3T3
Lane 3: Dog

Lane 4: Chicken

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (ab6721) at 1/5000 dilution

Performed under reducing conditions.

Predicted band size: 21 kDa Observed band size: 25 kDa

Exposure time: 30 seconds

250kDa 1 2 3 4 5 6 7 8 9 10 150kDa — 100kDa — 75kDa — 50kDa — 37kDa — 25kDa — 20kDa — 15kDa —

Western blot - Anti-Cyclophilin B antibody (ab16045)

All lanes: Anti-Cyclophilin B antibody (ab16045) at 1 µg/ml

Lane 1 : HeLa nuclear lysate

Lane 2: HeLa whole cell lysate

Lane 3: A431 whole cell lysate

Lane 4: Jurkat whole cell lysate

Lane 5: HEK293 whole cell lysate

 $\textbf{Lane 6:} \ \textit{HeLa nuclear lysate with Human Cyclophilin B peptide}$

(ab16277) at 1 µg/ml

Lane 7: HeLa whole cell lysate with Human Cyclophilin B peptide

 $(\underline{ab16277})$ at 1 $\mu g/ml$

 $\textbf{Lane 8:} \ \text{A431 whole cell lysate with Human Cyclophilin B peptide}$

(ab16277) at 1 µg/ml

Lane 9: Jurkat whole cell lysate with Human Cyclophilin B peptide

(ab16277) at 1 µg

Lane 10 : HEK293 whole cell lysate with Human Cyclophilin B peptide (ab16277) at 1 μ g/ml

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab6721</u>) at 1/5000 dilution

Performed under reducing conditions.

Predicted band size: 21 kDa **Observed band size:** 21 kDa

Exposure time: 30 seconds

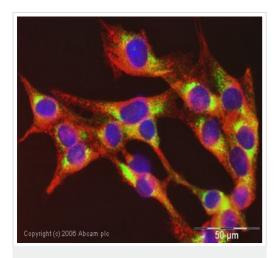
Immunoprecipitation - Anti-Cyclophilin B antibody (ab16045)

Cyclophilin B was immunoprecipitated using 0.5mg Hela whole cell extract, 5µg of Rabbit polyclonal to Cyclophilin B and 50µl of protein G magnetic beads (+). No antibody was added to the control (-). The antibody was incubated under agitation with Protein G beads for 10min, Hela whole cell extract lysate diluted in RIPA buffer was added to each sample and incubated for a further 10min under agitation.

Proteins were eluted by addition of $40\mu l$ SDS loading buffer and incubated for 10min at $70^{o}C$; $10\mu l$ of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab16045.

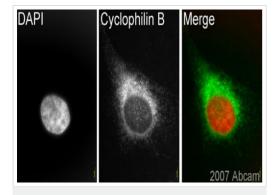
Secondary: Clean blot (HRP conjugate) at 1/1000 dilution.

Band: 21kDa: Cyclophilin B.



Immunocytochemistry/ Immunofluorescence - Anti-Cyclophilin B antibody (ab16045)

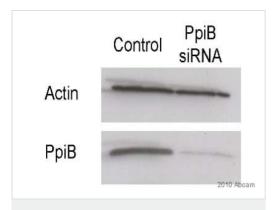
ICC/IF image of ab16045 stained NIH/3T3 cells. The cells were methanol fixed (5 min) and incubated with the antibody (ab16045, 1µg/ml) for 1h at room temperature. The secondary antibody (green) was Alexa Fluor® 488 goat anti-rabbit lgG (H+L) used at a 1/1000 dilution for 1h. Image-iTTMFX Signal Enhancer was used as the primary blocking agent, 5% BSA (in TBS-T) was used for all other blocking steps. DAPI was used to stain the cell nuclei (blue). Alexa Fluor® 594 WGA was used to label plasma membranes (red).



Immunocytochemistry/ Immunofluorescence - Anti-Cyclophilin B antibody (ab16045)

This image is courtesy of an Abreview submitted by Dr Kirk McManus

ab16045 (1/1000) staining Cyclophilin B in assynchronous HeLa cells (green). Cells were fixed with Paraformaldehyde and counterstained with DAPI in order to highlight the nucleus (red). Please refer to abreview for further experimental details.



Western blot - Anti-Cyclophilin B antibody (ab16045) Image courtesy of Dr MSchrader by Abreview.

All lanes: Anti-Cyclophilin B antibody (ab16045) at 1/1000 dilution

Lane 1: Whole cell lysate prepared from rat pancreatic AR42J cells, which were treated with 10nM dexamethasone for 48 hours.

Lane 2: Whole cell lysate for negative control, prepared from rat pancreatic AR42J cells (specific knock down of cyclophilin B/PpiB by siRNA), which were treated with 10nM dexamethasone for 48 hours.

Secondary

All lanes : Goat-anti-Rabbit HRP-conjugated polyclonal at 1/2000 dilution

Developed using the ECL technique.

Predicted band size: 21 kDa

Observed band size: 23 kDa

Exposure time: 1 minute

Primary antibody incubated for 12 hours at 4°C.

Blocking step performed using 5% milk, 1 hour at 20°C.

Anti-Cyclophilin B antibody (ab16045) at 0.5 μ g/ml + Recombinant Human Cyclophilin B protein (ab88801) at 0.01 μ g

Secondary

Goat Anti-Rabbit IgG H&L (HRP) preadsorbed (ab97080) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 21 kDa

Exposure time: 30 seconds

150 kDa —

100 kDa —

75 kDa —

50 kDa —

37 kDa —

25 kDa —

20 kDa —

10 kDa —

10 kDa —

10 kDa —

Western blot - Anti-Cyclophillin B antibody (ab16045)

250 kDa -

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