abcam

Product datasheet

Anti-CNOT7 antibody [EPR18722] ab195587

KO VALIDATED Recombinant RabMAb

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Overview

Product name	Anti-CNOT7 antibody [EPR18722]
Description	Rabbit monoclonal [EPR18722] to CNOT7
Host species	Rabbit
Tested applications	**Suitable for:** Flow Cyt (Intra), ICC/IF, IP, WB
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: Mouse ovary, embryo, brain and heart lysates; F9, SP2/0, C6, PC-12, HeLa, HepG2, NTERA-2/D1, MOLT-4 and NCCIT whole cell lysates; rat kidney, spleen and heart lysates; human fetal brain, fetal heart, fetal kidney and fetal spleen lysates. IP: HeLa whole cell lysate.
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

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, 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR18722

Isotype IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab195587 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)		1/1000.
ICC/IF		1/1000.
IP	★★★★★ (2)	1/40.
WB	★★★★☆ (5)	1/1000. Detects a band of approximately 33 kDa (predicted molecular weight: 33 kDa).

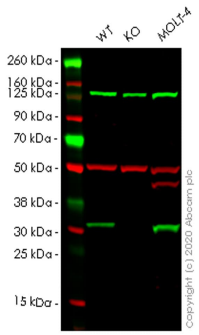
Target

Function Ubiquitous transcription factor required for a diverse set of processes. It is a component of the CCR4 complex involved in the control of gene expression.

Sequence similarities Belongs to the CAF1 family.

Cellular localization Nucleus.

Images



Western blot - Anti-CNOT7 antibody [EPR18722] (ab195587)

All lanes : Anti-CNOT7 antibody [EPR18722] (ab195587) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate

Lane 2 : CNOT7 knockout HeLa cell lysate

Lane 3 : MOLT-4 cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

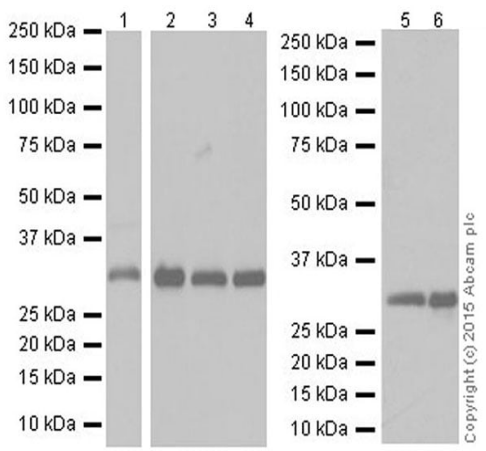
All lanes : Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) at 1/10000 dilution

Predicted band size: 33 kDa

Observed band size: 33 kDa

Lanes 1-3: Merged signal (red and green). Green - ab195587 observed at 33 kDa. Red - loading control **ab7291** observed at 50 kDa.

ab195587 Anti-CNOT7 antibody [EPR18722] was shown to specifically react with CNOT7 in wild-type HeLa cells. Loss of signal was observed when knockout cell line **ab265811** (knockout cell lysate **ab258370**) was used. Wild-type and CNOT7 knockout samples were subjected to SDS-PAGE. ab195587 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (**ab7291**) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-CNOT7 antibody [EPR18722] (ab195587)

All lanes : Anti-CNOT7 antibody [EPR18722] (ab195587) at 1/1000 dilution

Lane 1 : Mouse ovary lysate
Lane 2 : Mouse embryo lysate
Lane 3 : F9 (Mouse embryonic testicular cancer cell line) whole cell lysate
Lane 4 : SP2/0 (Mouse spleen cell line) whole cell lysate
Lane 5 : C6 (Rat glial tumor cell line) whole cell lysate
Lane 6 : PC-12 (Rat adrenal gland pheochromocytoma cell line) whole cell lysate

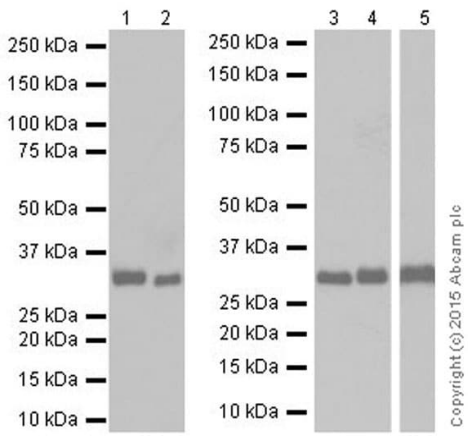
Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

Predicted band size: 33 kDa
Observed band size: 33 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: Lane 1: 2 seconds; Lane 2-6: 5 seconds.



Western blot - Anti-CNOT7 antibody [EPR18722] (ab195587)

All lanes : Anti-CNOT7 antibody [EPR18722] (ab195587) at 1/1000 dilution

Lane 1 : Mouse brain lysate
Lane 2 : Mouse heart lysate
Lane 3 : Rat kidney lysate
Lane 4 : Rat spleen lysate
Lane 5 : Rat heart lysate

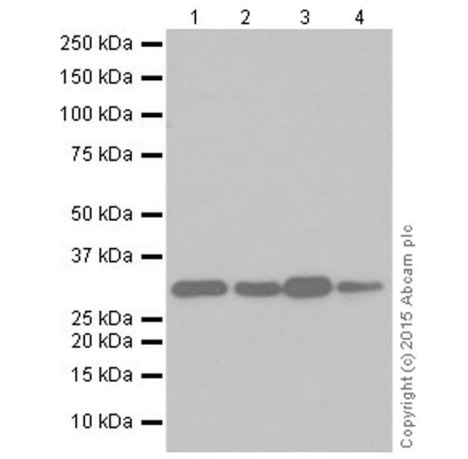
Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

Predicted band size: 33 kDa
Observed band size: 33 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: Lane 1/2: 15 seconds; Lane 3/4: 5 seconds; Lane 5: 30 seconds.



Western blot - Anti-CNOT7 antibody [EPR18722] (ab195587)

All lanes : Anti-CNOT7 antibody [EPR18722] (ab195587) at 1/1000 dilution

Lane 1 : Human fetal brain lysate
Lane 2 : Human fetal heart lysate
Lane 3 : Human fetal kidney lysate
Lane 4 : Human fetal spleen lysate

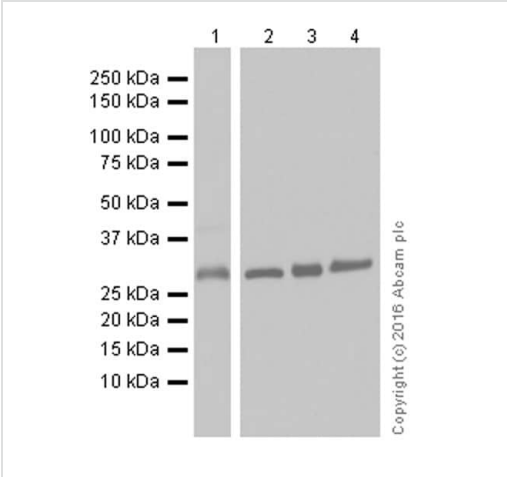
Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/10000 dilution

Predicted band size: 33 kDa
Observed band size: 33 kDa

Exposure time: 30 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-CNOT7 antibody [EPR18722] (ab195587)

All lanes : Anti-CNOT7 antibody [EPR18722] (ab195587) at 1/1000 dilution

Lane 1 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 2 : HepG2 (Human liver hepatocellular carcinoma cell line) whole cell lysate
Lane 3 : NTERA-2/D1 (Human malignant pluripotent embryonic carcinoma cell line) whole cell lysate
Lane 4 : NCCIT (Human pluripotent embryonic carcinoma cell line) whole cell lysate

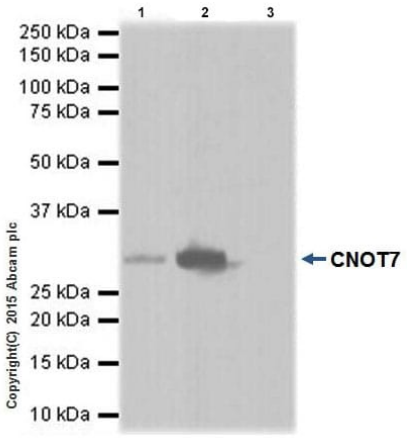
Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

Predicted band size: 33 kDa
Observed band size: 33 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: Lane 1: 1 seconds; Lane 2-4: 2 seconds.



Immunoprecipitation - Anti-CNOT7 antibody [EPR18722] (ab195587)

CNOT7 was immunoprecipitated from 1mg of HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate with ab195587 at 1/40 dilution. Western blot was performed from the immunoprecipitate using ab195587 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/10000 dilution.

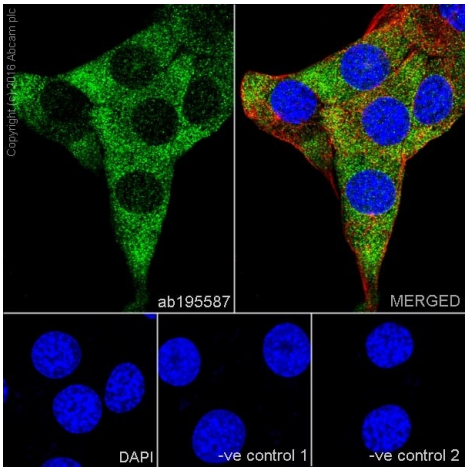
Lane 1: HeLa whole cell lysate 10µg (Input).

Lane 2: ab195587 IP in HeLa whole cell lysate.

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of ab195587 in HeLa whole cell lysate.

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

Exposure time: 1 second.



Immunocytochemistry/ Immunofluorescence - Anti-CNOT7 antibody [EPR18722] (ab195587)

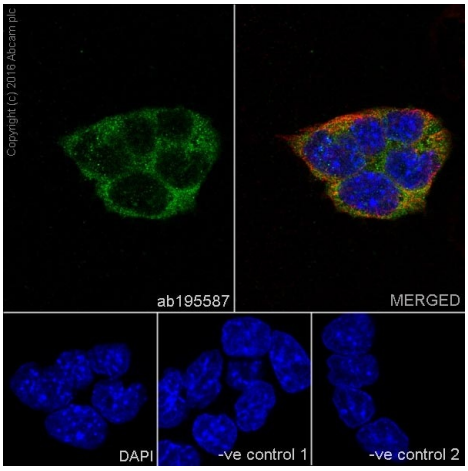
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized NIH/3T3 (Mouse embryonic fibroblast cell line) cells labeling CNOT7 with ab195587 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution (green). Confocal image showing cytoplasmic staining on NIH/3T3 cell line. The nuclear counter stain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin mouse MAb (**ab7291**) at 1/1000 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) (**ab150120**) secondary antibody at 1/1000 dilution (red).

The negative controls are as follows:

-ve control 1: ab195587at 1/1000 dilution, followed by Anti-Mouse IgG H&L (Alexa Fluor® 594) (**ab150120**) secondary.

-ve control 2: Anti-alpha Tubulin mouse MAb (**ab7291**) at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150077**) secondary at 1/1000 dilution.



Immunocytochemistry/ Immunofluorescence - Anti-CNOT7 antibody [EPR18722] (ab195587)

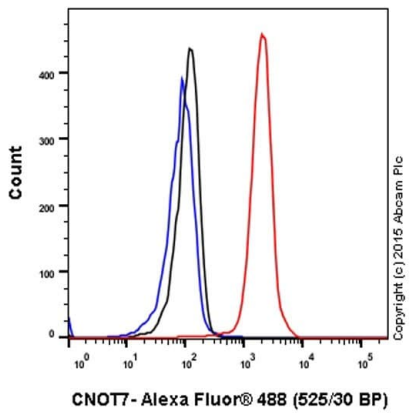
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized F9 (Mouse embryonic testicular cancer cell line) cells labeling CNOT7 with ab195587 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution (green). Confocal image showing cytoplasmic staining on NIH/3T3 cell line. The nuclear counter stain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin mouse MAb (**ab7291**) at 1/1000 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) (**ab150120**) secondary antibody at 1/1000 dilution (red).

The negative controls are as follows:

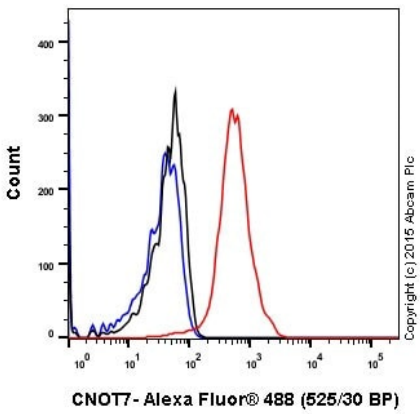
-ve control 1: ab195587at 1/1000 dilution, followed by Anti-Mouse IgG H&L (Alexa Fluor® 594) (**ab150120**) secondary at 1/XXXX dilution.

-ve control 2: Anti-alpha Tubulin mouse MAb (**ab7291**) at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150077**) secondary at 1/1000 dilution.



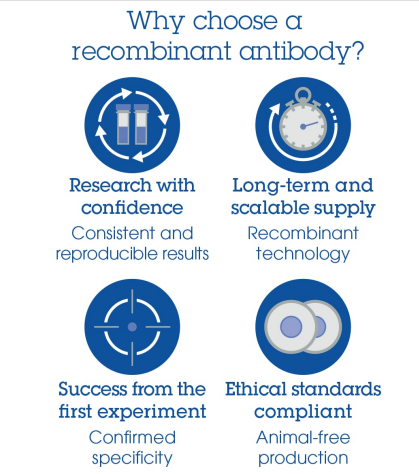
Flow Cytometry (Intracellular) - Anti-CNOT7 antibody [EPR18722] (ab195587)

Intracellular Flow Cytometry analysis of 4% paraformaldehyde fixed NIH/3T3 (Mouse embryonic fibroblast cell line) cell linelabelling CNOT7 with ab195587 at 1/1000 dilution (red), isotype control with **ab172730** (black) and unlabelled control cells without incubation with primary antibody and secondary antibody (blue).



Flow Cytometry (Intracellular) - Anti-CNOT7 antibody [EPR18722] (ab195587)

Intracellular Flow Cytometry analysis of 4% paraformaldehyde fixed F9 (Mouse embryonic testicular cancer cell line) cells labeling CNOT7 with ab195587 at 1/1000 dilution (red), isotype control with **ab172730** (black) and unlabeled control cells without incubation with primary antibody and secondary antibody (blue).



Anti-CNOT7 antibody [EPR18722] (ab195587)

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