# abcam

# Product datasheet

# Anti-CNOT7 antibody [EPR18722] - BSA and Azide free ab240994



Recombinant

RabMAb

## 7 Images

#### Overview

Product name Anti-CNOT7 antibody [EPR18722] - BSA and Azide free

**Description** Rabbit monoclonal [EPR18722] to CNOT7 - BSA and Azide free

Host species Rabbit

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

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; MOLT-4, F9, SP2/0, C6, PC-12, HeLa,

HepG2, NTERA-2/D1 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. ICC:

NIH/3T3 and F9 cells. Flow Cyt (intra): NIH/3T3 and F9 cells.

**General notes** ab240994 is the carrier-free version of <u>ab195587</u>.

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 <u>conjugation kits</u> 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

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit

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### 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. Do Not Freeze.

Storage buffer pH: 7.2

Constituent: PBS

Carrier free Yes

**Purity** Protein A purified

ClonalityMonoclonalClone numberEPR18722

**Isotype** IgG

#### **Applications**

The Abpromise guarantee Our Abpromise guarantee covers the use of ab240994 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)		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 33 kDa (predicted molecular weight: 33 kDa).
ICC/IF		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.

#### **Target**

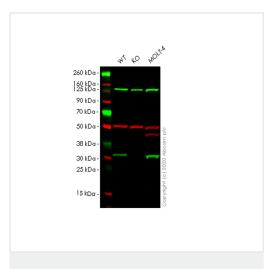
**Function** Ubiquitous transcription factor required for a diverse set of processes. It is a component of the

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] - BSA and Azide free (ab240994)

**All lanes :** Anti-CNOT7 antibody [EPR18722] (<u>ab195587</u>) 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**

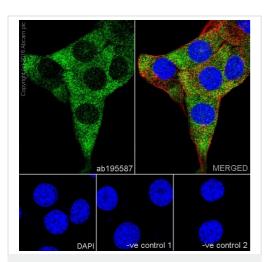
**All lanes :** Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/10000 dilution

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

This data was developed using <u>ab195587</u>, the same antibody clone in a different buffer formulation.

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

<u>ab195587</u> 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 <u>ab265811</u> (knockout cell lysate <u>ab258370</u>) was used. Wild-type and CNOT7 knockout samples were subjected to SDS-PAGE. <u>ab195587</u> and Anti-alpha Tubulin antibody [DM1A] - Loading Control (<u>ab7291</u>) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (<u>ab216773</u>) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (<u>ab216776</u>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-CNOT7 antibody [EPR18722] - BSA and Azide free (ab240994)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized NIH/3T3 (Mouse embryonic fibroblast cell line) cells labeling CNOT7 with <a href="mailto:ab195587">ab195587</a> at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor<sup>®</sup> 488) (<a href="mailto:ab150077">ab150077</a>) 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 (<a href="mailto:ab7291">ab7291</a>) at 1/1000 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor<sup>®</sup> 594) (<a href="mailto:ab150120">ab150120</a>) secondary antibody at 1/1000 dilution (red).

The negative controls are as follows:

-ve control 1: <u>ab195587</u> at 1/1000 dilution, followed by Anti-Mouse lgG H&L (Alexa Fluor® 594) (<u>ab150120</u>) secondary at 1/XXXX dilution.

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

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab195587</u>).

ab 195587 MERGED

DAPI -ve control 1 -ve control 2

Immunocytochemistry/ Immunofluorescence - Anti-CNOT7 antibody [EPR18722] - BSA and Azide free (ab240994)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized F9 (Mouse embryonic testicular cancer cell line) cells labeling CNOT7 with <u>ab195587</u> at 1/1000 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 cytoplasmic staining on NIH/3T3 cell line. The nuclear counter stain is DAPI (blue).

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

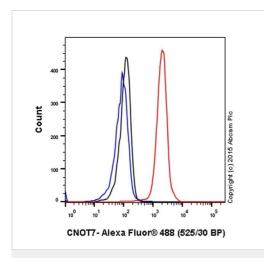
The negative controls are as follows:

-ve control 1: <u>ab195587</u> at 1/1000 dilution, followed by Anti-Mouse lgG H&L (Alexa Fluor<sup>®</sup> 594) (<u>ab150120</u>) secondary at 1/XXXX dilution.

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

3 250 kDa -150 kDa -100 kDa -75 kDa 🕳 50 kDa -37 kDa -Copyright(C) 2015 Abcam CNOT7 25 kDa -20 kDa -15 kDa -10 kDa

Immunoprecipitation - Anti-CNOT7 antibody [EPR18722] - BSA and Azide free (ab240994)



Flow Cytometry (Intracellular) - Anti-CNOT7 antibody [EPR18722] - BSA and Azide free (ab240994)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab195587)

CNOT7 was immunoprecipitated from 1mg of HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate with <u>ab195587</u> 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.

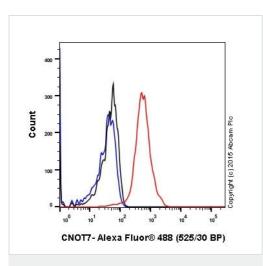
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 1 second.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab195587)

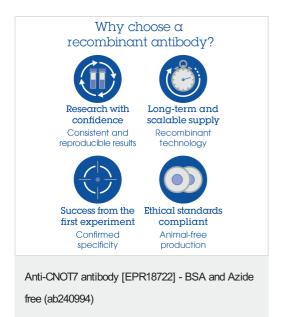
Intracellular Flow Cytometry analysis of 4% paraformaldehyde fixed NIH/3T3 (Mouse embryonic fibroblast cell line) cell line labelling CNOT7 with ab195587 at 1/1000 dilution (red), isotype control with ab172730 (black) and unlabelled control cells without incubation with primary antibody and secondary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

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab195587)



Flow Cytometry (Intracellular) - Anti-CNOT7 antibody [EPR18722] - BSA and Azide free (ab240994) Intracellular Flow Cytometry analysis of 4% paraformaldehyde fixed F9 (Mouse embryonic testicular cancer cell line) cells labeling CNOT7 with <u>ab195587</u> at 1/1000 dilution (red), isotype control with <u>ab172730</u> (black) and unlabeled control cells without incubation with primary antibody and secondary antibody (blue).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab195587</u>)



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