abcam

Product datasheet

Anti-CDT1/DUP antibody [EPR17891] ab202067

Recombinant RabMAb

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Overview

Product name	Anti-CDT1/DUP antibody [EPR17891]
Description	Rabbit monoclonal [EPR17891] to CDT1/DUP
Host species	Rabbit
Tested applications	**Suitable for:** WB, IHC-P, ICC/IF, IP
Species reactivity	**Reacts with:** Human
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: HeLa and HEK-293 cell lysates; Human fetal lung lysate. IHC-P: Human colonic carcinoma and cervix carcinoma tissues. ICC/IF: HeLa and HT-29 cells. IP: HEK-293 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**.

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: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR17891
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab202067 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
WB	★★★★★ (1)	1/1000. Detects a band of approximately 60 kDa (predicted molecular weight: 60 kDa).
IHC-P		1/100. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF	★★★★★ (2)	1/100.
IP		1/30.

Target

Function Cooperates with CDC6 to promote the loading of the mini-chromosome maintenance complex onto chromatin to form the pre-replication complex necessary to initiate DNA replication. Binds DNA in a sequence-, strand-, and conformation-independent manner. Potential oncogene.

Sequence similarities Belongs to the Cdt1 family.

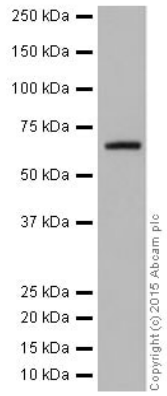
Developmental stage Present during G1 and early S phase of the cell cycle. Degraded during the late S, G2, and M phases.

Domain The PIP-box K+4 motif mediates both the interaction with PCNA and the recuitment of the DCX(DTL) complex: while the PIP-box interacts with PCNA, the presence of the K+4 submotif, recruits the DCX(DTL) complex, leading to its ubiquitination.

Post-translational modifications Ubiquitinated by the DCX(DTL) complex, also named CRL4(CDT2) complex, in response to DNA damage, leading to its degradation. Ubiquitination by the DCX(DTL) complex is necessary to ensure proper cell cycle regulation and is PCNA-dependent: interacts with PCNA via its PIP-box, while the presence of the containing the 'K+4' motif in the PIP box, recruit the DCX(DTL) complex, leading to its degradation. The interaction with GMNN protects it against ubiquitination. Phosphorylated by cyclin A-dependent kinases which results in the binding of CDT1 to the F-box protein SKP2 and subsequent degradation. Binding to GMNN is not affected by phosphorylation.

Cellular localization Nucleus.

Images



Western blot - Anti-CDT1/DUP antibody [EPR17891] (ab202067)

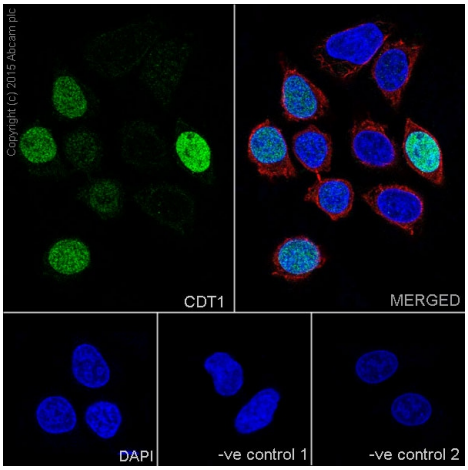
Anti-CDT1/DUP antibody [EPR17891] (ab202067) at 1/10000 dilution + HeLa (Human epithelial cells from cervix adenocarcinoma) cell lysate at 20 µg

Secondary
Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 60 kDa
Observed band size: 60 kDa

Exposure time: 1 minute

Blocking/Dilution buffer: 5% NFDM/TBST.



Immunocytochemistry/ Immunofluorescence - Anti-CDT1/DUP antibody [EPR17891] (ab202067)

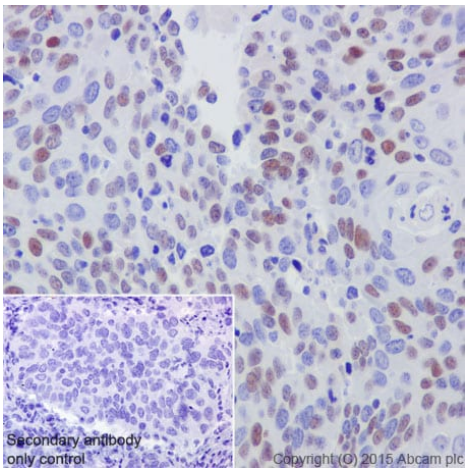
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cells from cervix adenocarcinoma) cells labeling CDT1/DUP with ab202067 at 1/100 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/500 dilution (green).

Confocal image showing nuclear staining on HeLa cell line.

The nuclear counter stain is DAPI (blue).

Tubulin is detected with **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution and **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).

The negative controls are as follows:
-ve control 1: ab202067 at 1/100 dilution followed by **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
-ve control 2: **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution followed by **ab150077** (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/500 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CDT1/DUP antibody [EPR17891] (ab202067)

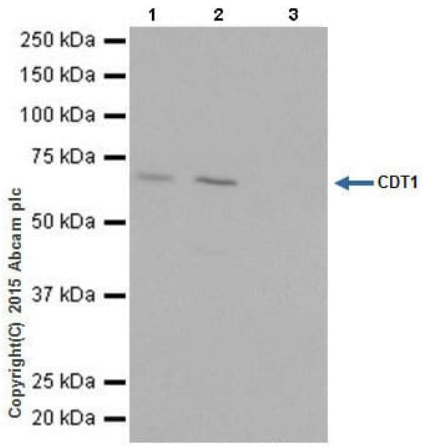
Immunohistochemical analysis of paraffin-embedded human cervix carcinoma tissue labeling CDT1/DUP with ab202067 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) secondary antibody at 1/500 dilution.

Nuclear staining on human cervix carcinoma tissue is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

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



Immunoprecipitation - Anti-CDT1/DUP antibody [EPR17891] (ab202067)

CDT1/DUP was immunoprecipitated from 1mg of HEK-293 (Human epithelial cells from embryonic kidney) whole cell lysate with ab202067 at 1/30 dilution.

Western blot was performed from the immunoprecipitate using ab202067 at 1/1000 dilution.

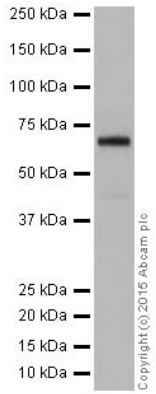
Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG was used as secondary antibody at 1/1500 dilution.

Lane 1: HEK-293 whole cell lysate 10 µg (Input).

Lane 2: ab202067 IP in HEK-293 whole cell lysate.

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of ab202067 in HEK-293 whole cell lysate.
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 5 seconds.



Western blot - Anti-CDT1/DUP antibody [EPR17891] (ab202067)

Anti-CDT1/DUP antibody [EPR17891] (ab202067) at 1/2000 dilution + HEK-293 (Human epithelial cells from embryonic kidney) cell lysate at 20 µg

Secondary

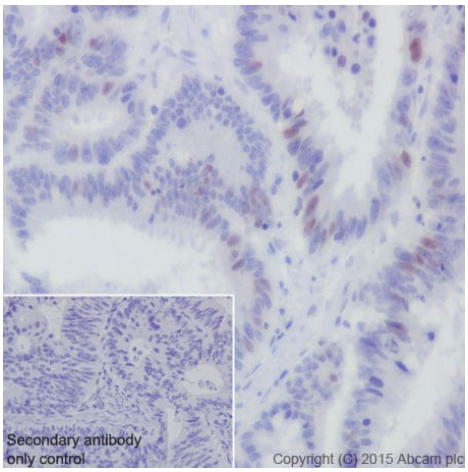
Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 60 kDa

Observed band size: 60 kDa

Exposure time: 1 minute

Blocking/Dilution buffer: 5% NFDM/TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CDT1/DUP antibody [EPR17891] (ab202067)

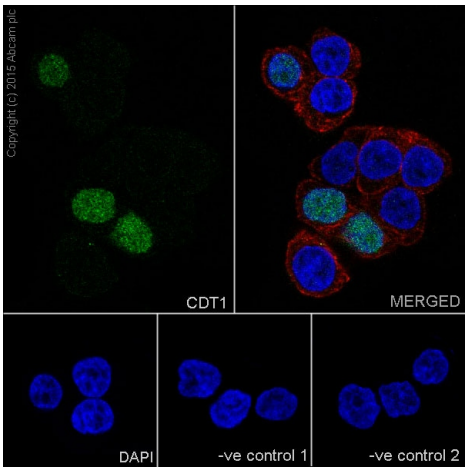
Immunohistochemical analysis of paraffin-embedded Human colonic carcinoma tissue labeling CDT1/DUP with ab202067 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) secondary antibody at 1/500 dilution.

Nuclear staining on Human colonic carcinoma tissue is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

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



Immunocytochemistry/ Immunofluorescence - Anti-CDT1/DUP antibody [EPR17891] (ab202067)

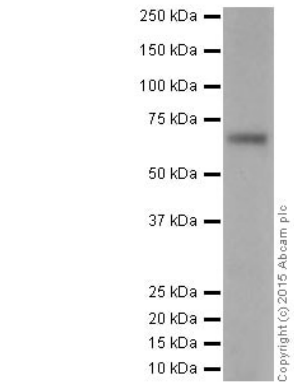
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HT-29 (Human colorectal adenocarcinoma cells) cells labeling CDT1/DUP with ab202067 at 1/100 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/500 dilution (green).

Confocal image showing nuclear staining on T-29 cell line.

The nuclear counter stain is DAPI (blue).

Tubulin is detected with **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution and **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).

The negative controls are as follows:
-ve control 1: ab202067 at 1/100 dilution followed by **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
-ve control 2: **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution followed by **ab150077** (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/500 dilution.



Western blot - Anti-CDT1/DUP antibody [EPR17891] (ab202067)

Anti-CDT1/DUP antibody [EPR17891] (ab202067) at 1/1000 dilution + Human fetal lung lysate at 10 µg

Secondary
Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/1000 dilution

Predicted band size: 60 kDa
Observed band size: 60 kDa

Exposure time: 1 minute

Blocking/Dilution buffer: 5% NFDM/TBST.

Anti-CDT1/DUP antibody [EPR17891] (ab202067)

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