abcam

Product datasheet

Anti-CMT2 antibody [EPR9584] ab150363

KO VALIDATED Recombinant RabMAb

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Overview

Product name	Anti-CMT2 antibody [EPR9584]
Description	Rabbit monoclonal [EPR9584] to CMT2
Host species	Rabbit
Tested applications	**Suitable for:** WB, IHC-P, ICC/IF **Unsuitable for:** Flow Cyt or IP
Species reactivity	**Reacts with:** Human **Predicted to work with:** Mouse, Rat
Immunogen	Synthetic peptide within Human CMT2 aa 250 to the C-terminus. The exact sequence is proprietary. Database link: **Q15013**
Positive control	Recombinant Human CMT2 (**ab123183**) can be used as a positive control in WB. HepG2, A431, and HeLa cell lysates, Human brain and Human cervical carcinoma tissues This antibody gave a positive result when used in the following formaldehyde fixed cell lines: HeLa. WB: Wild-type HeLa cell lysate. THP-1, HepG2 and MDA-MB-231 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. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.
Storage buffer	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: 9% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA, 50% Tissue culture

	supernatant
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR9584
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab150363 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

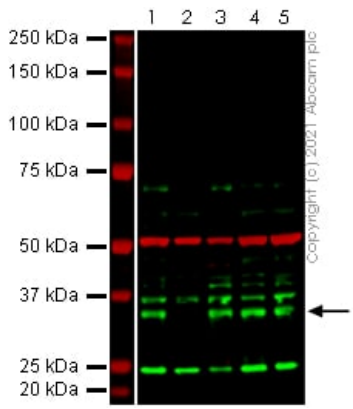
Application	Abreviews	Notes
WB	★★☆☆☆ (1)	1/1000 - 1/10000. Detects a band of approximately 34 kDa (predicted molecular weight: 31 kDa).
IHC-P		1/100 - 1/250. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
ICC/IF		1/100 - 1/250.

Application notes Is unsuitable for Flow Cyt or IP.

Target

Function	May function to silence the spindle checkpoint and allow mitosis to proceed through anaphase by binding MAD2L1 after it has become dissociated from the MAD2L1-CDC20 complex.
Sequence similarities	Belongs to the MAD2L1BP family.
Developmental stage	During the cell cycle, levels increase and then remain constant until late mitosis after which they drop.
Post-translational modifications	Phosphorylated upon DNA damage, probably by ATM or ATR.
Cellular localization	Nucleus. Cytoplasm > cytoskeleton > spindle. During early mitosis, unevenly distributed throughout the nucleoplasm. From metaphase to anaphase, concentrated on the spindle.

Images



Western blot - Anti-CMT2 antibody [EPR9584] (ab150363)

All lanes : Anti-CMT2 antibody [EPR9584] (ab150363) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : MAD2L1BP knockout cell line **ab265854** (knockout cell lysate **ab257510**)
Lane 3 : THP-1 cell lysate
Lane 4 : HepG2 cell lysate
Lane 5 : MDA-MB-231 cell lysate

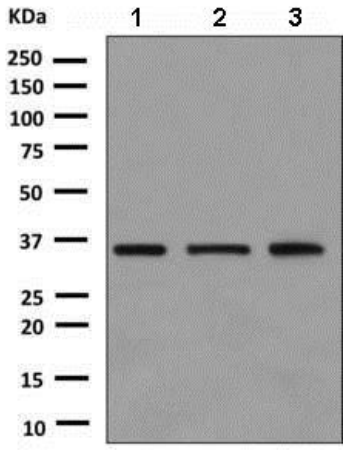
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 31 kDa
Observed band size: 35 kDa

False colour image of Western blot: Anti-CMT2 antibody [EPR9584] staining at 1/1000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] (**ab7291**) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab150363 was shown to bind specifically to CMT2. A band was observed at 35 kDa in wild-type HeLa cell lysates with no signal observed at this size in MAD2L1BP knockout cell line **ab265854** (knockout cell lysate **ab257510**). To generate this image, wild-type and MAD2L1BP knockout HeLa cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with primary antibodies overnight at 4°C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) at

1/20000 dilution.



Western blot - Anti-CMT2 antibody [EPR9584] (ab150363)

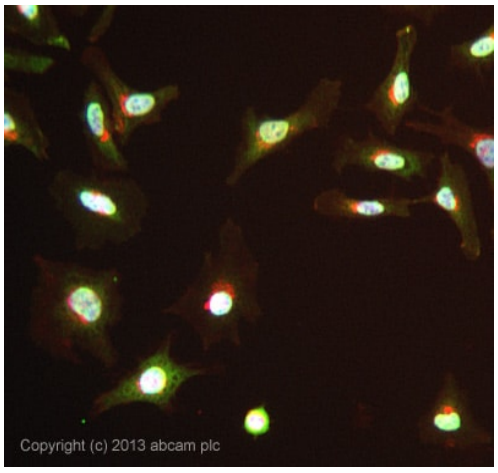
All lanes : Anti-CMT2 antibody [EPR9584] (ab150363) at 1/1000 dilution

Lane 1 : HepG2 cell lysate
Lane 2 : A431 cell lysate
Lane 3 : HeLa cell lysate

Lysates/proteins at 10 µg per lane.

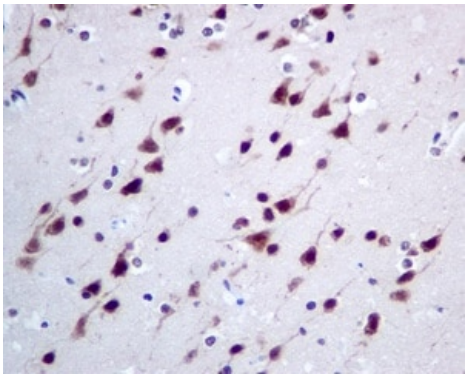
Secondary
All lanes : HRP labelled goat anti-rabbit at 1/2000 dilution

Predicted band size: 31 kDa
Observed band size: 34 kDa



Immunocytochemistry/ Immunofluorescence - Anti-CMT2 antibody [EPR9584] (ab150363)

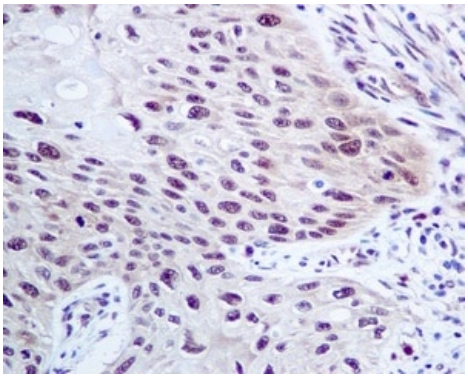
ab150363 stained HeLa cells. The cells were 4% formaldehyde fixed (10 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody ab150363 at 5µg/ml overnight at +4°C. The secondary antibody (green) was DyLight® 488 goat anti- rabbit (**ab96899**) IgG (H+L) used at a 1/250 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CMT2 antibody [EPR9584] (ab150363)

Immunohistochemical analysis of paraffin embedded Human brain tissue labelling CMT2 binding protein with ab150363 antibody at a dilution of 1/100.

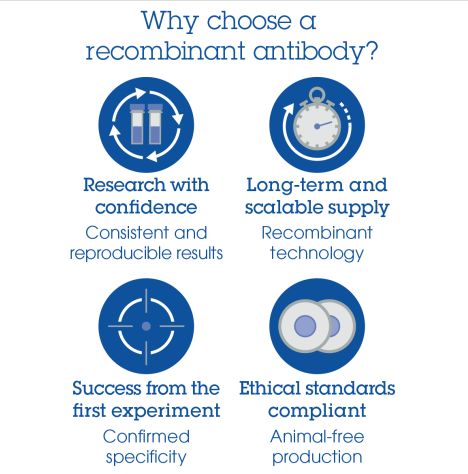
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CMT2 antibody [EPR9584] (ab150363)

Immunohistochemical analysis of paraffin embedded Human cervical carcinoma tissue labelling CMT2 binding protein with ab150363 antibody at a dilution of 1/100.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Anti-CMT2 antibody [EPR9584] (ab150363)

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