abcam

Product datasheet

Anti-Calnexin antibody [EPR3633(2)] - ER Membrane Marker (HRP) ab195198

KO VALIDATED Recombinant RabMAb

1 References 3 Images

Overview

Product name	Anti-Calnexin antibody [EPR3633(2)] - ER Membrane Marker (HRP)
Description	Rabbit monoclonal [EPR3633(2)] to Calnexin - ER Membrane Marker (HRP)
Host species	Rabbit
Conjugation	HRP
Tested applications	**Suitable for:** WB, IHC-P
Species reactivity	**Reacts with:** Human **Does not react with:** Mouse
Immunogen	Synthetic peptide within Human Calnexin aa 550 to the C-terminus (C terminal). The exact sequence is proprietary.
Positive control	WB: HepG2, A431 and HeLa whole cell lysates. IHC-P: normal human colon tissue.
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. Stable for 12 months at -20°C. Store In the Dark.
Storage buffer	pH: 7.40 Preservative: 0.1% Proclin Constituents: 1% BSA, 30% Glycerol, PBS
Purity	Protein A purified

Clonality	Monoclonal
Clone number	EPR3633(2)
Isotype	IgG

Applications

Our Abpromise guarantee covers the use of **ab195198** 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		1/5000. Detects a band of approximately 82 kDa (predicted molecular weight: 68 kDa).
IHC-P		Use at an assay dependent concentration.

Target

Function	Calcium-binding protein that interacts with newly synthesized glycoproteins in the endoplasmic reticulum. It may act in assisting protein assembly and/or in the retention within the ER of unassembled protein subunits. It seems to play a major role in the quality control apparatus of the ER by the retention of incorrectly folded proteins.
Sequence similarities	Belongs to the calreticulin family.
Cellular localization	Endoplasmic reticulum membrane. Melanosome. Identified by mass spectrometry in melanosome fractions from stage I to stage IV.

Images

Western blot - Anti-Calnexin antibody [EPR3633(2)] - ER Membrane Marker (HRP) (ab195198)

All lanes : Anti-Calnexin antibody [EPR3633(2)] - ER Membrane Marker (HRP) (ab195198) at 1/5000 dilution

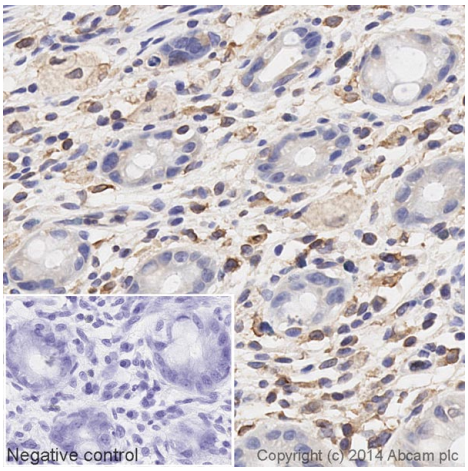
Lane 1 : Wild-type HAP1 whole cell lysate
Lane 2 : CANX (Calnexin) knockout HAP1 whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 68 kDa

ab195198 was shown to recognize Calnexin in wild-type HAP1 cells as signal was lost at the expected MW in CANX (Calnexin) knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and CANX (Calnexin) knockout samples were subjected to SDS-PAGE. Ab195198 and

ab184095 (Mouse monoclonal [mAbcam 9484] to GAPDH - Loading Control (Alexa Fluor® 680) loading control) were incubated overnight at 4°C at 1/5000 dilution and 1/1000 dilution respectively. The loading control was imaged using the Licor Odyssey CLx prior to blots being developed with ECL technique.

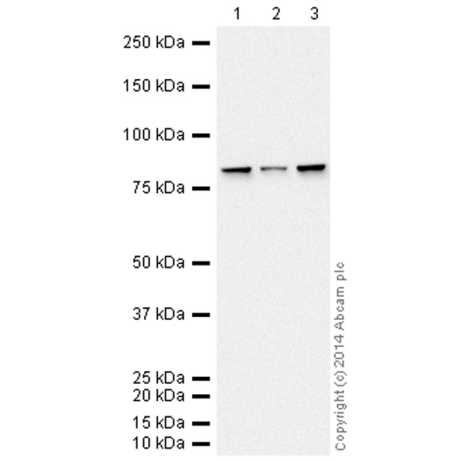


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Calnexin antibody [EPR3633(2)] - ER Membrane Marker (HRP) (ab195198)

IHC image of Calnexin staining in a section of formalin-fixed paraffin-embedded normal human colon tissue*, performed on a Leica BOND. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20mins. The section was then incubated with ab195198 at 1/500 dilution, for 15 mins at room temperature. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. The inset negative control image is taken from an identical assay without primary antibody.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre



Western blot - Anti-Calnexin antibody [EPR3633(2)] - ER Membrane Marker (HRP) (ab195198)

All lanes : Anti-Calnexin antibody [EPR3633(2)] - ER Membrane Marker (HRP) (ab195198) at 1/5000 dilution

Lane 1 : HepG2 (Human hepatocellular liver carcinoma cell line) Whole Cell Lysate
Lane 2 : A431 (Human epithelial carcinoma cell line) Whole Cell Lysate
Lane 3 : HeLa whole cell lysate (ab150035)

Lysates/proteins at 10 µg per lane.

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 68 kDa
Observed band size: 82 kDa
why is the actual band size different from the predicted?

Exposure time: 20 minutes

This blot was produced using a 4-12% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 50 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 3% milk before being incubated with ab195198 overnight at 4°C. Antibody binding was visualised using ECL development solution ab133406.

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