abcam

Product datasheet

Anti-Calreticulin antibody [EPR3924] - BSA and Azide free ab271865

KO VALIDATED Recombinant RabMAb

15 Images

Overview

Product name	Anti-Calreticulin antibody [EPR3924] - BSA and Azide free
Description	Rabbit monoclonal [EPR3924] to Calreticulin - BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** Flow Cyt, IHC-P, WB, ICC/IF
Species reactivity	**Reacts with:** Mouse, Rat, Human, Monkey
Immunogen	Synthetic peptide within Human Calreticulin aa 50-150. The exact sequence is proprietary.
Positive control	ICC/IF: HAP1 cells (HAP1-CALR as negative cell line) IHC-P: Human colon, kidney, liver, placenta, stomach, breast carcinoma and Papillary carcinoma of thyroid gland tissues; Mouse liver and Rat lung tissues.
General notes	ab271865 is the carrier-free version of ab92516. This format is designed for use in antibody labeling, including fluorochromes, metal isotopes, oligonucleotides, enzymes. Our carrier-free formats are supplied in a buffer free of BSA, sodium azide and glycerol for higher conjugation efficiency. Use our conjugation kits 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® Antibody Labeling Kit from Fluidigm. *Maxpar® 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 monoclonal antibodies. For details on our patents, please refer to RabMAb® patents. Reproducibility is key to advancing scientific discovery and accelerating scientists' next

breakthrough.

Abcam is leading the way with our range of recombinant antibodies, knockout-validated antibodies and knockout cell lines, all of which support improved reproducibility.

We are also planning to innovate the way in which we present recommended applications and species on our product datasheets, so that only applications & species that have been tested in our own labs, our suppliers or by selected trusted collaborators are covered by our Abpromise™ guarantee.

In preparation for this, we have started to update the applications & species that this product is Abpromise guaranteed for.

We are also updating the applications & species that this product has been "predicted to work with," however this information is not covered by our Abpromise guarantee.

Applications & species from publications and Abreviews that have not been tested in our own labs or in those of our suppliers are not covered by the Abpromise guarantee.

Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, as well as customer reviews and Q&As.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	pH: 7.20 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR3924
Isotype	IgG

Applications

Our Abpromise guarantee covers the use of **ab271865** 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		Use at an assay dependent concentration. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
IHC-P		Use at an assay dependent concentration. The use of a HRP/AP polymerized secondary antibody is recommended for enhanced staining.

Application	Abreviews	Notes
WB		Use at an assay dependent concentration. Predicted molecular weight: 48 kDa.
ICC/IF		Use at an assay dependent concentration.

Target

Function

Molecular calcium-binding chaperone promoting folding, oligomeric assembly and quality control in the ER via the calreticulin/calnexin cycle. This lectin interacts transiently with almost all of the monoglucosylated glycoproteins that are synthesized in the ER. Interacts with the DNA-binding domain of NR3C1 and mediates its nuclear export.

Sequence similarities

Belongs to the calreticulin family.

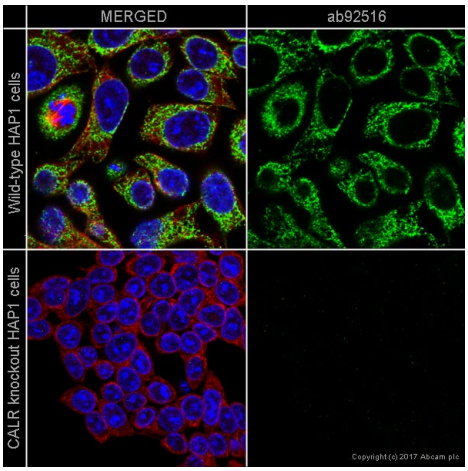
Domain

Can be divided into a N-terminal globular domain, a proline-rich P-domain forming an elongated arm-like structure and a C-terminal acidic domain. The P-domain binds one molecule of calcium with high affinity, whereas the acidic C-domain binds multiple calcium ions with low affinity.
The interaction with glycans occurs through a binding site in the globular lectin domain.
The zinc binding sites are localized to the N-domain.
Associates with PDIA3 through the tip of the extended arm formed by the P-domain.

Cellular localization

Endoplasmic reticulum lumen. Cytoplasm > cytosol. Secreted > extracellular space > extracellular matrix. Cell surface. Also found in cell surface (T cells), cytosol and extracellular matrix. Associated with the lytic granules in the cytolytic T-lymphocytes.

Images



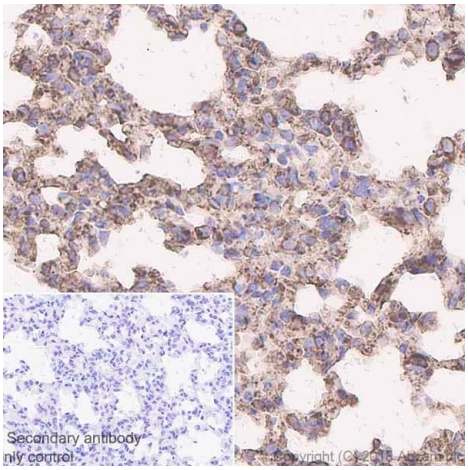
Immunocytochemistry/ Immunofluorescence - Anti-Calreticulin antibody [EPR3924] - BSA and Azide free (ab271865)

ab92516 staining Calreticulin in wild-type HAP1 cells (top panel) and CALR knockout HAP1 cells (bottom panel). The cells were fixed with 100% methanol (5min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab92516 at 1/500 and ab195889 at 1/250 dilution (shown in pseudocolour red) overnight at +4°C, followed by a further incubation at room temperature for 1h with a goat secondary antibody to Rabbit IgG (Alexa Fluor® 488) (ab150081) at 2 µg/ml (shown in green). Nuclear DNA was labelled in blue with DAPI.

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

Alexa Fluor® 488 (ab196158) and Alexa Fluor® 647 (ab196159) conjugated versions are available for this clone.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab92516).

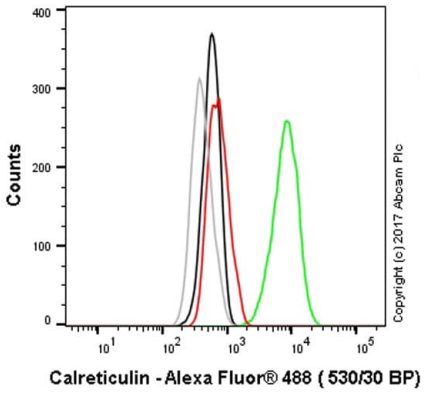


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Calreticulin antibody [EPR3924] - BSA and Azide free (ab271865)

Immunohistochemical analysis of paraffin-embedded Rat lung tissue labeling Calreticulin with ab92516, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Cytoplasmic staining on rat lung.The section was incubated with ab229902 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab92516).



Flow Cytometry - Anti-Calreticulin antibody [EPR3924] - BSA and Azide free (ab271865)

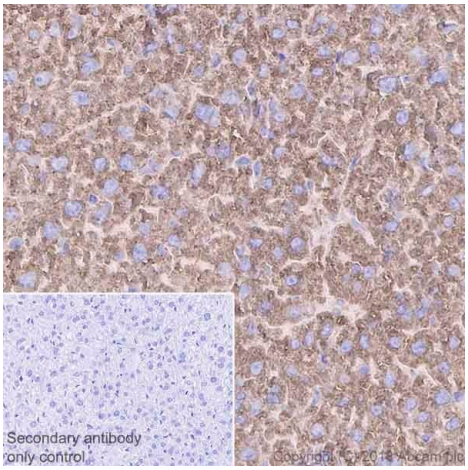
Overlay histogram showing HAP1 wildtype (green line) and HAP1-CALR knockout cells (red line) stained with ab92516. The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS / 10% normal goat serum to block non-specific protein-protein interactions followed by the antibody (ab92516, 1µg/ml) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit IgG (H&L) preadsorbed (ab150081) at 1/2000 dilution for 30 min at 22°C.

A rabbit IgG isotype control antibody (ab172730) was used at the same concentration and conditions as the primary antibody (HAP1 wildtype - black line, HAP1-CALR knockout - grey line). Unlabelled sample was also used as a control (this line is not shown for the purpose of simplicity).

Acquisition of >5,000 events were collected using a 50 mW Blue laser (488nm) and 530/30 bandpass filter.

Alexa Fluor® 488 (ab196158) and Alexa Fluor® 647 (ab196159) conjugated versions are available for this clone.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab92516).

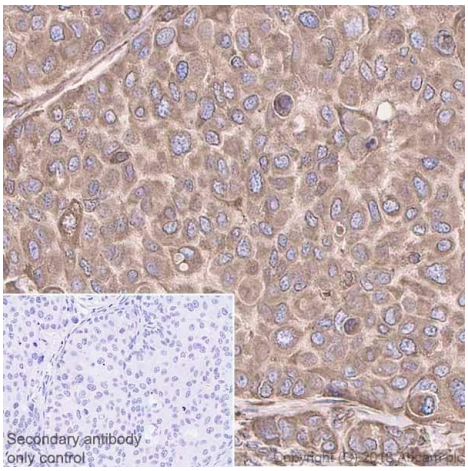


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Calreticulin antibody [EPR3924] - BSA and Azide free (ab271865)

Immunohistochemical analysis of paraffin-embedded Mouse liver tissue labeling Calreticulin with ab92516, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Cytoplasmic staining on mouse liver.The section was incubated with ab229902 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab92516).

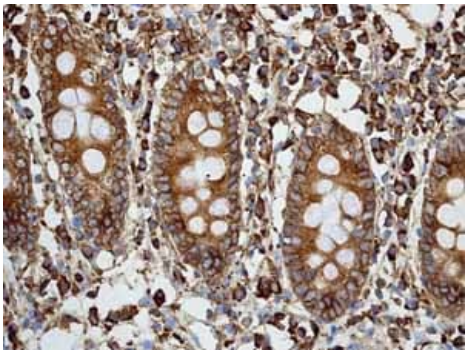


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Calreticulin antibody [EPR3924] - BSA and Azide free (ab271865)

Immunohistochemical analysis of paraffin-embedded Human breast carcinoma tissue labeling Calreticulin with ab92516, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Cytoplasmic staining human breast carcinoma.The section was incubated with ab229902 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab92516).

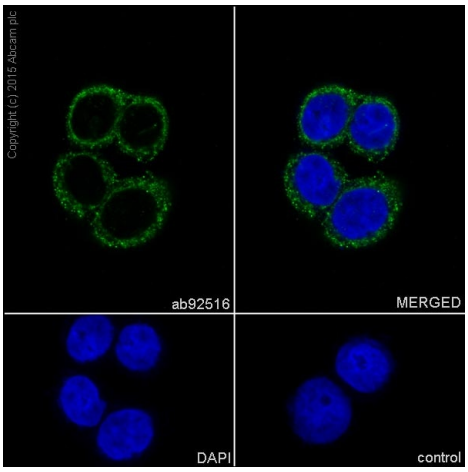


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Calreticulin antibody [EPR3924] - BSA and Azide free (ab271865)

Formalin-fixed, paraffin-embedded normal human colon tissue stained for Calreticulin using ab92516 at 1/250 dilution in immunohistochemical analysis.

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

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab92516).

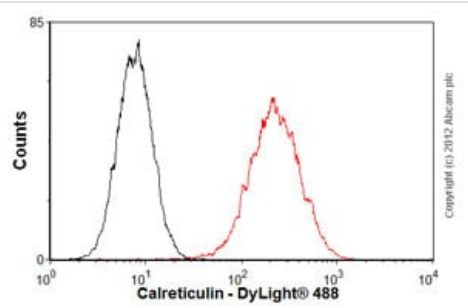


Immunocytochemistry/ Immunofluorescence - Anti-Calreticulin antibody [EPR3924] - BSA and Azide free (ab271865)

Immunocytochemistry/Immunofluorescence analysis of HT-29 (human colorectal adenocarcinoma) labelling Calreticulin with purified ab92516 at 1/500. Cells were fixed with 100% methanol. An Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000) was used as the secondary antibody (Ab150077). Nuclei counterstained with DAPI (blue).

Control: PBS only

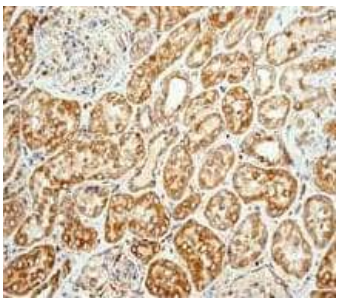
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab92516).



Flow Cytometry - Anti-Calreticulin antibody [EPR3924] - BSA and Azide free (ab271865)

Overlay histogram showing HeLa cells stained with ab92516 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab92516, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit IgG (H+L) (ab96899) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab92516).

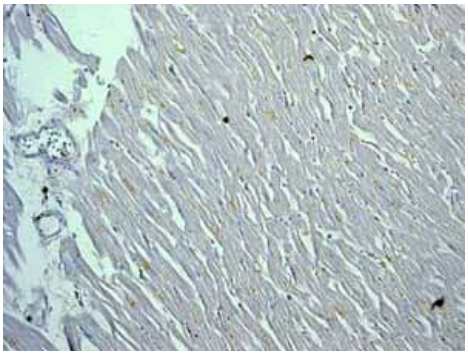


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Calreticulin antibody [EPR3924] - BSA and Azide free (ab271865)

ab92516, at 1/250 dilution, staining Calreticulin in paraffin embedded Human kidney tissue by Immunohistochemistry.

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

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab92516).

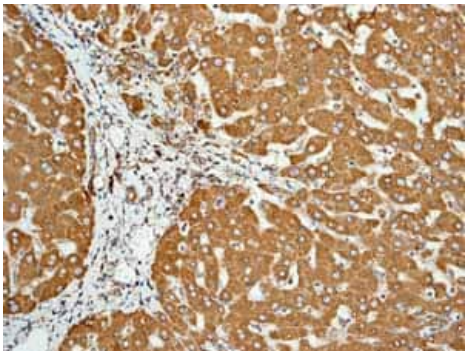


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Calreticulin antibody [EPR3924] - BSA and Azide free (ab271865)

ab92516 showing negative staining in Normal human heart tissue.

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

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab92516).

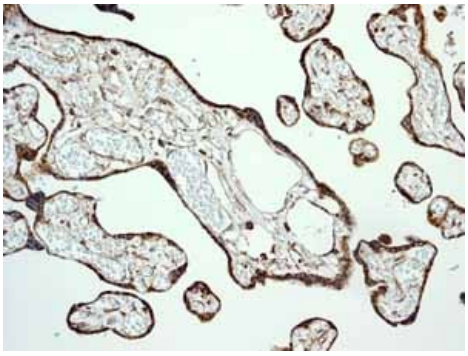


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Calreticulin antibody [EPR3924] - BSA and Azide free (ab271865)

Formalin-fixed, paraffin-embedded normal human liver tissue stained for Calreticulin using ab92516 at 1/250 dilution in immunohistochemical analysis.

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

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab92516).

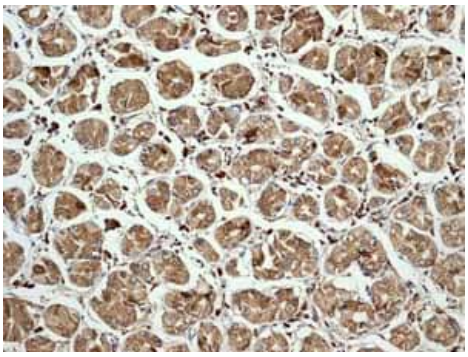


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Calreticulin antibody [EPR3924] - BSA and Azide free (ab271865)

Formalin-fixed, paraffin-embedded normal human placenta tissue stained for Calreticulin using ab92516 at 1/250 dilution in immunohistochemical analysis.

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

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab92516).

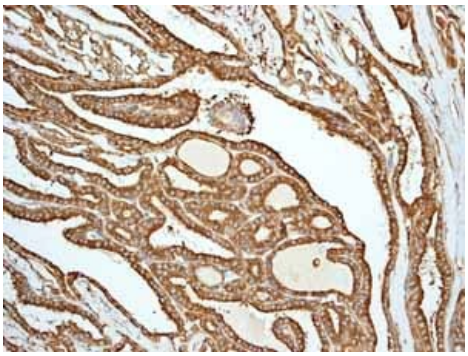


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Calreticulin antibody [EPR3924] - BSA and Azide free (ab271865)

Formalin-fixed, paraffin-embedded normal human stomach tissue stained for Calreticulin using ab92516 at 1/250 dilution in immunohistochemical analysis.

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

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab92516).

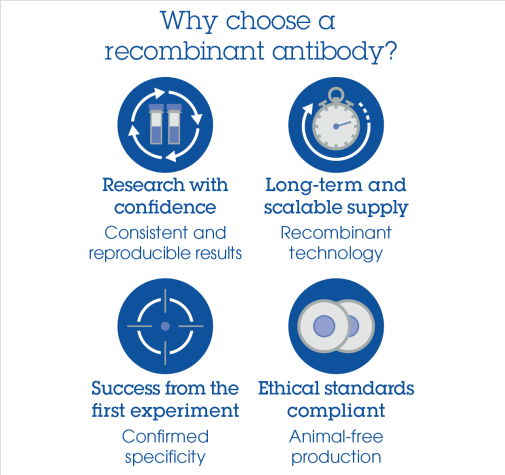


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Calreticulin antibody [EPR3924] - BSA and Azide free (ab271865)

Formalin-fixed, paraffin-embedded Papillary carcinoma of human thyroid gland tissue.stained for Calreticulin using ab92516 at 1/250 dilution in immunohistochemical analysis.

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

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab92516).



Anti-Calreticulin antibody [EPR3924] - BSA and Azide free (ab271865)

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