

## Product datasheet

# Anti-Calnexin antibody [EPR3632] ab92573

**KO VALIDATED** Recombinant RabMAb<sup>®</sup>

[1 Abreviews](#) [38 References](#) [8 Images](#)

### Overview

<b>Product name</b>	Anti-Calnexin antibody [EPR3632]
<b>Description</b>	Rabbit monoclonal [EPR3632] to Calnexin
<b>Host species</b>	Rabbit
<b>Specificity</b>	Recognizes ER membrane, mitochondria and cis-Golgi
<b>Tested applications</b>	<b>Suitable for:</b> ICC/IF, WB, IP, IHC-P <b>Unsuitable for:</b> Flow Cyt
<b>Species reactivity</b>	<b>Reacts with:</b> Human
<b>Immunogen</b>	Synthetic peptide within Human Calnexin aa 1-100. The exact sequence is proprietary. Database link: <a href="#">P27824</a>
<b>Positive control</b>	WB: HeLa, A431, SH-SY5Y, HEK-293T, MCF7, U-2 OS and HepG2 whole cell lysate ( <a href="#">ab7900</a> ). IHC-P: Human tonsil tissue. ICC/IF: Wild-type HAP1 cells. IP: HeLa lysate.
<b>General notes</b>	References regarding specificity:  Horner SM <i>et al.</i> Mitochondrial-associated endoplasmic reticulum membranes (MAM) form innate immune synapses and are targeted by hepatitis C virus. <i>Proc Natl Acad Sci U S A</i> 108:14590-5 (2011). <a href="#">PubMed: 21844353</a>  Myhill N <i>et al.</i> The subcellular distribution of calnexin is mediated by PACS-2. <i>Mol Biol Cell</i> 19:2777-88 (2008). <a href="#">PubMed: 18417615</a>  Yoshimura SI <i>et al.</i> Direct targeting of cis-Golgi matrix proteins to the Golgi apparatus. <i>J Cell Sci</i> 114:4105-15 (2001). <a href="#">PubMed: 11739642</a>  This product is a recombinant monoclonal antibody, which offers several advantages including: <ul style="list-style-type: none"><li>- High batch-to-batch consistency and reproducibility</li><li>- Improved sensitivity and specificity</li><li>- Long-term security of supply</li><li>- Animal-free production</li></ul> For more information <a href="#">see here</a> .  Our RabMAb <sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to <a href="#">RabMAb<sup>®</sup> patents</a> .  Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

## Properties

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<b>Form</b>	Liquid
<b>Storage instructions</b>	Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid repeated freeze / thaw cycles.
<b>Storage buffer</b>	pH: 7.20 Preservative: 0.05% Sodium azide Constituents: 0.1% BSA, 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue culture supernatant
<b>Purity</b>	Protein A purified
<b>Clonality</b>	Monoclonal
<b>Clone number</b>	EPR3632
<b>Isotype</b>	IgG

## Applications

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**The Abpromise guarantee** Our **Abpromise guarantee** covers the use of ab92573 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
ICC/IF		1/1000.
WB		1/20000 - 1/100000. Predicted molecular weight: 90 kDa.
IP		1/50.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

**Application notes** Is unsuitable for Flow Cyt.

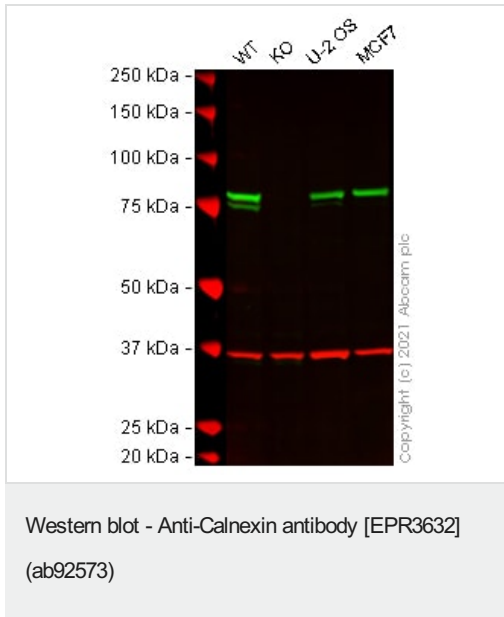
## Target

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<b>Function</b>	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.
<b>Sequence similarities</b>	Belongs to the calreticulin family.
<b>Cellular localization</b>	Endoplasmic reticulum membrane. Melanosome. Identified by mass spectrometry in melanosome fractions from stage I to stage IV.

## Images

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**All lanes :** Anti-Calnexin antibody [EPR3632] (ab92573) at 1/20000 dilution

**Lane 1 :** Wild-type HEK-293T cell lysate

**Lane 2 :** CANX knockout HEK-293T cell lysate

**Lane 3 :** U-2 OS cell lysate

**Lane 4 :** MCF7 cell lysate

Lysates/proteins at 20 µg per lane.

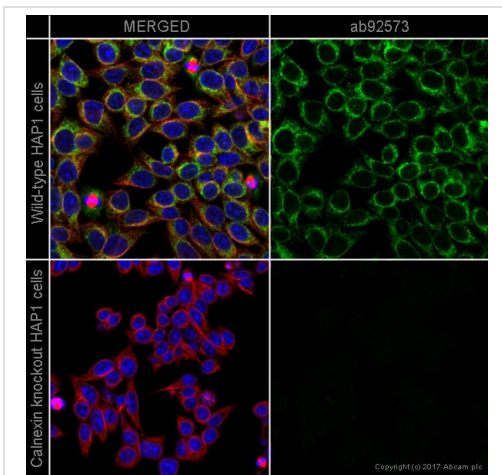
Performed under reducing conditions.

**Predicted band size:** 90 kDa

**Observed band size:** 80 kDa

**Lanes 1 - 4:** Merged signal (red and green). Green - ab92573 observed at 80 kDa. Red - loading control **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

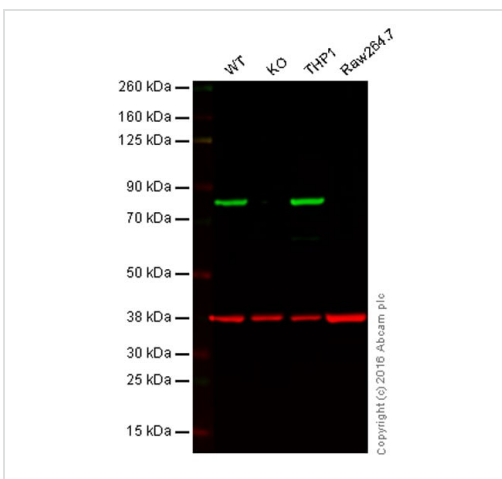
ab92573 was shown to react with Calnexin in wild-type HEK-293T cells in Western blot with loss of signal observed in CANX knockout cell line **ab255368** (CANX knockout cell lysate **ab263805**). Wild-type HEK-293T and CANX knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween<sup>®</sup>) before incubation with ab92573 and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4 °C at a 1 in 20000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye<sup>®</sup> 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye<sup>®</sup> 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-Calnexin antibody [EPR3632] (ab92573)

ab92573 staining Calnexin in wild-type HAP1 cells (top panel) and CANX 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 ab92573 at 1/1000 dilution 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).



Western blot - Anti-Calnexin antibody [EPR3632] (ab92573)

**All lanes** : Anti-Calnexin antibody [EPR3632] (ab92573) at 1/20000 dilution

**Lane 1** : Wild-type HAP1 cell lysate

**Lane 2** : Calnexin knockout HAP1 cell lysate

**Lane 3** : THP-1 cell lysate

**Lane 4** : RAW 264.7 cell lysate

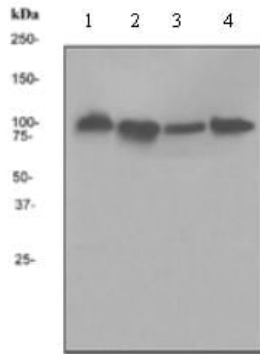
Lysates/proteins at 20 µg per lane.

**Predicted band size:** 90 kDa

**Lanes 1 - 4:** Merged signal (red and green). Green - ab92573 observed at 80 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab92573 was shown to specifically react with Calnexin when Calnexin knockout samples were used. Wild-type and Calnexin

knockout samples were subjected to SDS-PAGE. ab92573 and **ab8245** (loading control to GAPDH) were diluted at 1/20,000 and 1/10,000 respectively and incubated overnight at 4°C. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed **ab216773** and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed **ab216776** secondary antibodies at 1/10,000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-Calnexin antibody [EPR3632] (ab92573)

**All lanes** : Anti-Calnexin antibody [EPR3632] (ab92573) at 1/20000 dilution

**Lane 1** : HeLa cell lysate

**Lane 2** : A431 cell lysate

**Lane 3** : SH-SY5Y cell lysate

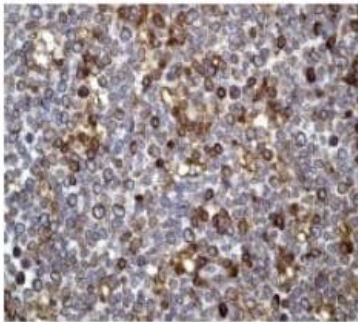
**Lane 4** : HepG2 cell lysates

Lysates/proteins at 10 µg per lane.

### Secondary

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

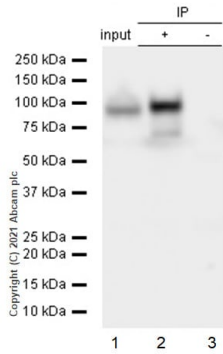
**Predicted band size:** 90 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Calnexin antibody [EPR3632] (ab92573)

Immunohistochemical analysis of paraffin embedded Human tonsil tissue using ab92573 at a 1/100 dilution.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunoprecipitation - Anti-Calnexin antibody  
[EPR3632] (ab92573)

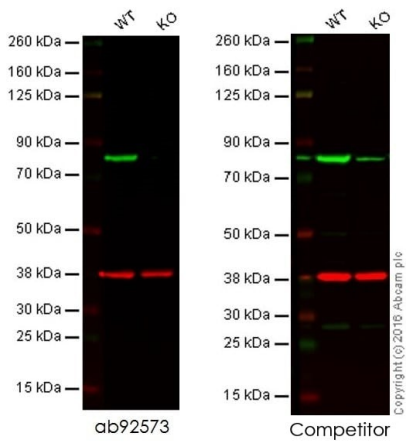
Calnexin was immunoprecipitated from 0.35 mg HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate 10 µg with 92573 at 1/100 dilution (2µg). VeriBlot for IP Detection Reagent (HRP)([ab131366](#)) was used at 1/5000 dilution.

**Lane 1:** HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate 10 µg

**Lane 2:** ab92573 IP in HeLa whole cell lysate

**Lane 3:** Rabbit monoclonal IgG ([ab172730](#)) instead of ab92573 in HeLa whole cell lysate

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



Western blot - Anti-Calnexin antibody [EPR3632]  
(ab92573)

**All lanes :** Anti-Calnexin antibody [EPR3632] (ab92573)

**Lane 1 :** Wild-type HAP1 cell lysate

**Lane 2 :** Calnexin knockout HAP1 cell lysate

Lysates/proteins at 20 µg per lane.

**Predicted band size:** 90 kDa

**Lanes 1 - 2:** Merged signal (red and green). Green - ab92573 observed at 80 kDa. Red - loading control, [ab8245](#), observed at 37 kDa.

This western blot image is a comparison between ab92573 and a competitor's rabbit polyclonal antibody.

### Why choose a recombinant antibody?



**Research with confidence**  
Consistent and reproducible results



**Long-term and scalable supply**  
Recombinant technology



**Success from the first experiment**  
Confirmed specificity



**Ethical standards compliant**  
Animal-free production

Anti-Calnexin antibody [EPR3632] (ab92573)

**Please note:** All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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- Extensive multi-media technical resources to help you
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