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Anti-Calnexin/CANX Antibody Picoband™ (monoclonal, 8D10B3)

Catalog # ABO16598

Specification

Anti-Calnexin/CANX Antibody Picoband™ (monoclonal, 8D10B3) - Product Information

Application	**WB, IHC, IF, ICC, FC**
Primary Accession	P27824
Host	**Mouse**
Isotype	**Mouse IgG2b**
Reactivity	**Rat, Human, Mouse**
Clonality	**Monoclonal**
Format	**Lyophilized**

Description
Anti-Calnexin/CANX Antibody Picoband™ (monoclonal, 8D10B3) . Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.

Reconstitution
Adding 0.2 ml of distilled water will yield a concentration of 500 µg/ml.

Anti-Calnexin/CANX Antibody Picoband™ (monoclonal, 8D10B3) - Additional Information

Gene ID 821

Other Names
Calnexin, IP90, Major histocompatibility complex class I antigen-binding protein p88, p90, CANX

Calculated MW
95 kDa KDa

Application Details
Western blot, 0.25-0.5 µg/ml, Human, Mouse, Rat
 Immunohistochemistry(Paraffin-embedded Section), 2-5 µg/ml, Human, Mouse, Rat
 Immunocytochemistry/Immunofluorescence, 5 µg/ml, Human
 Flow Cytometry, 1-3 µg/1x10^6 cells, Human

Contents
Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na2HPO4.

Immunogen
E.coli-derived human Calnexin/CANX recombinant protein (Position: E68-R582).

Purification
Immunogen affinity purified.

Storage	**At -20°C for one year from date of receipt. After reconstitution, at 4°C for one month. It can also be aliquotted and stored frozen at -20°C for six months. Avoid repeated freezing and thawing.**

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Anti-Calnexin/CANX Antibody Picoband™ (monoclonal, 8D10B3) - Protein Information

Name CANX

Function
Calcium-binding protein that interacts with newly synthesized monoglucosylated 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. Associated with partial T-cell antigen receptor complexes that escape the ER of immature thymocytes, it may function as a signaling complex regulating thymocyte maturation. Additionally it may play a role in receptor-mediated endocytosis at the synapse.

Cellular Location
Endoplasmic reticulum membrane; Single-pass type I membrane protein. Mitochondrion membrane {ECO:0000250|UniProtKB:P24643}; Single-pass type I membrane protein. Melanosome membrane; Single-pass type I membrane protein. Note=Identified by mass spectrometry in melanosome fractions from stage I to stage IV (PubMed:12643545, PubMed:17081065). The palmitoylated form preferentially localizes to the perinuclear rough ER (PubMed:22314232) Localizes to endoplasmic reticulum mitochondria-associated membrane (MAMs) that connect the endoplasmic reticulum and the mitochondria (By similarity). {ECO:0000250|UniProtKB:P24643, ECO:0000269|PubMed:12643545, ECO:0000269|PubMed:17081065, ECO:0000269|PubMed:22314232}

Anti-Calnexin/CANX Antibody Picoband™ (monoclonal, 8D10B3) - Protocols

Provided below are standard protocols that you may find useful for product applications.

- Western Blot
- Blocking Peptides
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- Cell Culture

Anti-Calnexin/CANX Antibody Picoband™ (monoclonal, 8D10B3) - Images

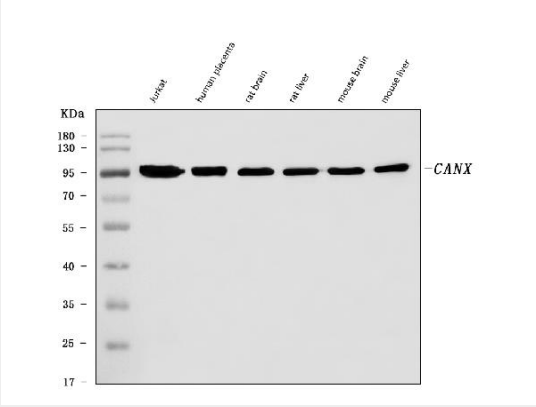

Figure 1. Western blot analysis of Calnexin/CANX using anti-Calnexin/CANX antibody (M03372-2).

Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.
Lane 1: human Jurkat whole cell lysates,
Lane 2: human placenta tissue lysates,
Lane 3: rat brain tissue lysates,
Lane 4: rat liver tissue lysates,
Lane 5: mouse brain tissue lysates,
Lane 6: mouse liver tissue lysates.
After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with mouse anti-Calnexin/CANX antigen affinity purified monoclonal antibody (Catalog # M03372-2) at 0.5 µg/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-mouse IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1001) with Tanon 5200 system. A specific band was detected for Calnexin/CANX at approximately 95 kDa. The expected band size for Calnexin/CANX is at 67 kDa.

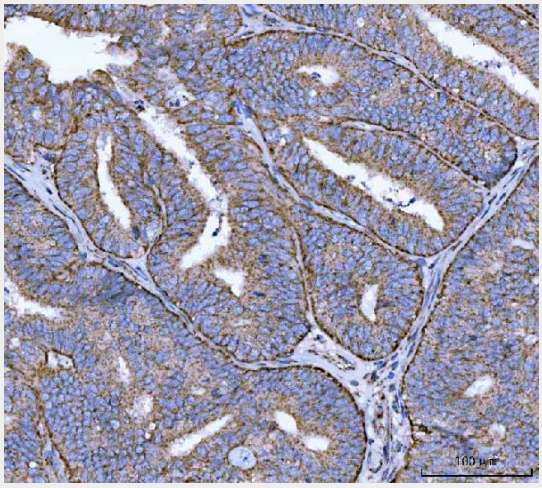

Figure 2. IHC analysis of Calnexin/CANX using anti-Calnexin/CANX antibody (M03372-2).
Calnexin/CANX was detected in a paraffin-embedded section of human endometrial adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-Calnexin/CANX Antibody (M03372-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.

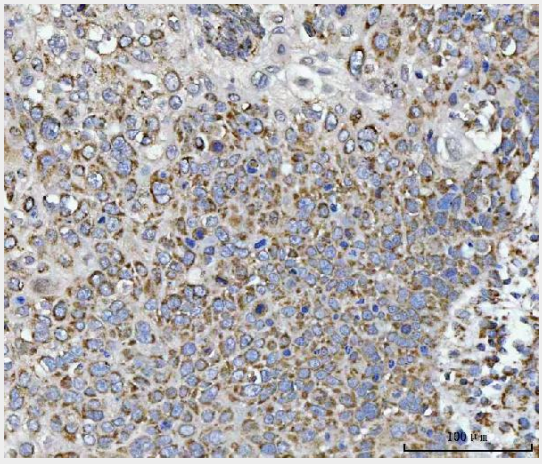

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Figure 3. IHC analysis of Calnexin/CANX using anti-Calnexin/CANX antibody (M03372-2). Calnexin/CANX was detected in a paraffin-embedded section of human lung cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-Calnexin/CANX Antibody (M03372-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.

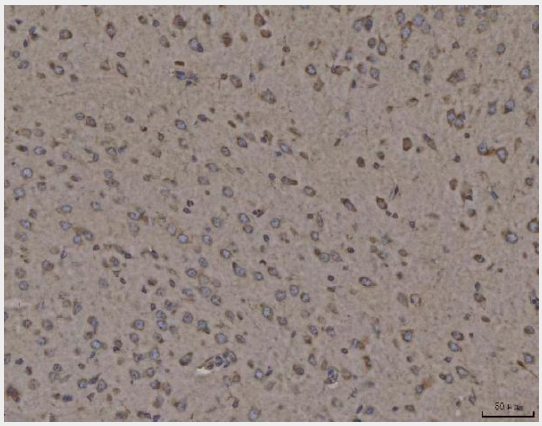

Figure 4. IHC analysis of Calnexin/CANX using anti-Calnexin/CANX antibody (M03372-2). Calnexin/CANX was detected in a paraffin-embedded section of mouse brain tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-Calnexin/CANX Antibody (M03372-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.

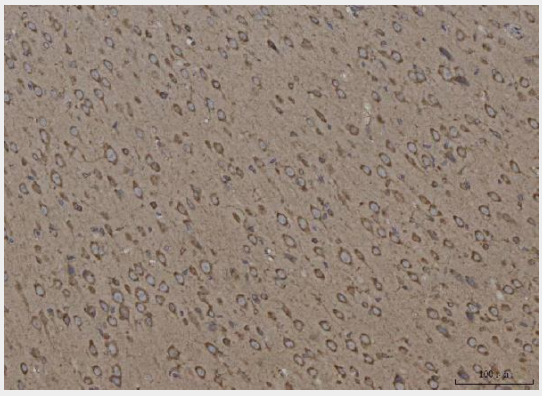

Figure 5. IHC analysis of Calnexin/CANX using anti-Calnexin/CANX antibody (M03372-2). Calnexin/CANX was detected in a paraffin-embedded section of rat brain tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-Calnexin/CANX Antibody (M03372-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.

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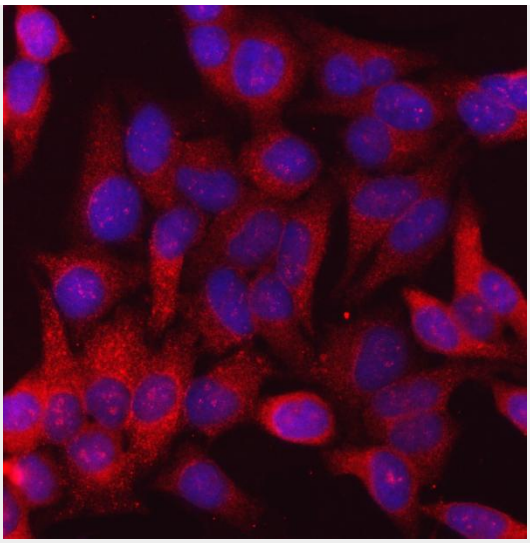

Figure 6. IF analysis of Calnexin/CANX using anti-Calnexin/CANX antibody (M03372-2). Calnexin/CANX was detected in an immunocytochemical section of Hela cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 µg/mL mouse anti-Calnexin/CANX Antibody (M03372-2) overnight at 4°C. Cy3 Conjugated Goat Anti-Mouse IgG (BA1031) was used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

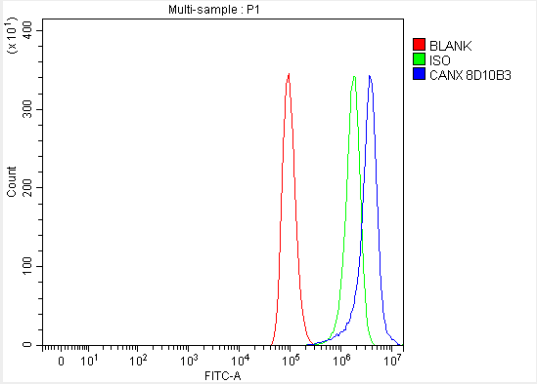

Figure 7. Flow Cytometry analysis of A549 cells using anti-Calnexin/CANX antibody (M03372-2). Overlay histogram showing A549 cells stained with M03372-2 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-Calnexin/CANX Antibody (M03372-2, 1 µg/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

Anti-Calnexin/CANX Antibody Picoband™ (monoclonal, 8D10B3) - Background

Calnexin (CNX) is a 67 kDa integral protein of the endoplasmic reticulum. This gene encodes a member of the calnexin family of molecular chaperones. The encoded protein is a calcium-binding, endoplasmic reticulum (ER)-associated protein that interacts transiently with newly synthesized N-linked glycoproteins, facilitating protein folding and assembly. It may also play a central role in the quality control of protein folding by retaining incorrectly folded protein subunits within the ER for degradation. Alternatively spliced transcript variants encoding different isoforms have been described.