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Anti-ERp57 Antibody Picoband™ (monoclonal, 7E5)

Catalog # ABO16254

Product Information

Application	WB, IHC, FC
Primary Accession	P30101
Host	Mouse
Isotype	Mouse IgG1
Reactivity	Human
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-ERp57 Antibody Picoband™ (monoclonal, 7E5) . Tested in Flow Cytometry, IHC, WB applications. This antibody reacts with Human.
Reconstitution	Adding 0.2 ml of distilled water will yield a concentration of 500 □g/ml.

Additional Information

Gene ID	2923
Other Names	Protein disulfide-isomerase A3, 5.3.4.1, 58 kDa glucose-regulated protein, 58 kDa microsomal protein, p58, Disulfide isomerase ER-60, Endoplasmic reticulum resident protein 57, ER protein 57, ERp57, Endoplasmic reticulum resident protein 60, ER protein 60, ERp60, PDIA3 (HGNC:4606), ERP57, ERP60, GRP58
Calculated MW	56782
Application Details	Western blot, 0.25-0.5 □g/ml, Human Immunohistochemistry(Paraffin-embedded Section), 2-5 □g/ml, Human Flow Cytometry, 1-3 □g/1x10^6 cells, Human
Source	Eukaryota
Contents	Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na2HPO4.
Clone Names	Clone: 7E5
Immunogen	A synthetic peptide corresponding to a sequence at the C-terminus of human ERp57, different from the related mouse and rat sequences by two amino acids.
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.

Protein Information

Name	PDIA3 (HGNC:4606)
Synonyms	ERP57, ERP60, GRP58
Function	Protein disulfide isomerase that catalyzes the formation, isomerization, and reduction or oxidation of disulfide bonds in client proteins and functions as a protein folding chaperone (PubMed:11825568, PubMed:16193070, PubMed:27897272, PubMed:36104323, PubMed:7487104). Core component of the major histocompatibility complex class I (MHC I) peptide loading complex where it functions as an essential folding chaperone for TAPBP. Through TAPBP, assists the dynamic assembly of the MHC I complex with high affinity antigens in the endoplasmic reticulum. Therefore, plays a crucial role in the presentation of antigens to cytotoxic T cells in adaptive immunity (PubMed:35948544, PubMed:36104323).
Cellular Location	Endoplasmic reticulum. Endoplasmic reticulum lumen {ECO:0000250|UniProtKB:P11598}. Melanosome Note=Identified by mass spectrometry in melanosome fractions from stage I to stage IV (PubMed:12643545).
Tissue Location	Detected in the flagellum and head region of spermatozoa (at protein level) (PubMed:20400973). Expressed in liver, stomach and colon (at protein level). Expressed in gastric parietal cells and chief cells (at protein level) (PubMed:24188822)

Background

PDIA3 (Protein disulfide isomerase family A, member 3), also called GRP58, Erp57 or ER60, is an isomerase enzyme. It is mapped on 15q15.3. PDIA3 is also part of the major histocompatibility complex (MHC) class I peptide-loading complex, which is essential for formation of the final antigen conformation and export from the endoplasmic reticulum to the cell surface. This gene encodes a protein of the endoplasmic reticulum that interacts with lectin chaperones calreticulin and calnexin to modulate folding of newly synthesized glycoproteins. The protein was once thought to be a phospholipase; however, it has been demonstrated that the protein actually has protein disulfide isomerase activity. It is thought that complexes of lectins and this protein mediate protein folding by promoting formation of disulfide bonds in their glycoprotein substrates.

Images

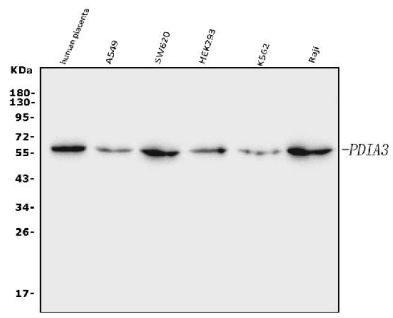


Figure 1. Western blot analysis of ERp57 using anti-ERp57 antibody (M01464-4).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours.
The sample well of each lane was loaded with 30 ug of sample under reducing conditions.
Lane 1: human placenta tissue lysates,
Lane 2: human A549 whole cell lysates,
Lane 3: human SW620 whole cell lysates,
Lane 4: human HEK293 whole cell lysates,
Lane 5: human K562 whole cell lysates,
Lane 6: human Raji whoe cell 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-ERp57 antigen affinity purified monoclonal antibody (Catalog # M01464-4) 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 1:5000 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 ERp57 at approximately 57 kDa. The expected band size for ERp57 is at 57 kDa.

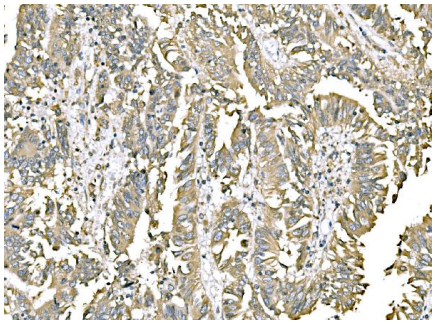


Figure 2. IHC analysis of ERp57 using anti-ERp57 antibody (M01464-4).
ERp57 was detected in a paraffin-embedded section of human rectal cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-ERp57 Antibody (M01464-4) 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.

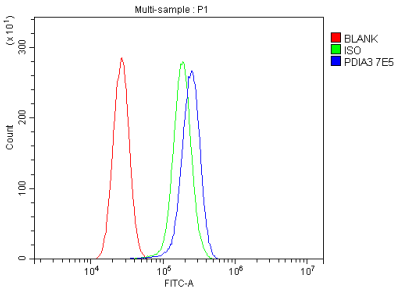


Figure 3. Flow Cytometry analysis of U87 cells using anti-ERp57 antibody (M01464-4).
Overlay histogram showing U87 cells stained with M01464-4 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-ERp57 Antibody (M01464-4, 1 µg/1x10^6 cells) for 30 min at 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.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.