

#### Anti-TJP1 Antibody Picoband™ (monoclonal, 3E12) Catalog # ABO14891

Specification

# Anti-TJP1 Antibody Picoband<sup>™</sup> (monoclonal, 3E12) - Product Information

ApplicationWB, IIPrimary Accession00715HostMouseIsotypeMouseReactivityHumaClonalityMonoeFormatLyophDescriptionAnti-TIP1 Antibody Picoband™ (monoclonal, 3E12), Teste

WB, IHC, IF, ICC, FC <u>007157</u> Mouse Mouse IgG1 Human Monoclonal Lyophilized

Anti-TJP1 Antibody Picoband<sup>™</sup> (monoclonal, 3E12) . Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human.

Reconstitution

Add 0.2ml of distilled water will yield a concentration of 500  $\mu$ g/ml.

### Anti-TJP1 Antibody Picoband<sup>™</sup> (monoclonal, 3E12) - Additional Information

Gene ID 7082

**Other Names** Tight junction protein ZO-1, Tight junction protein 1, Zona occludens protein 1, Zonula occludens protein 1, TJP1, ZO1

Calculated MW 220 kDa KDa

**Application Details** 

Western blot, 0.1-0.5 µg/ml, Human<br> Immunohistochemistry (Paraffin-embedded Section), 0.5-1 µg/ml, Human, By Heat<br> Immunocytochemistry/Immunofluorescence, 2 µg/ml, Human<br> Flow Cytometry, 1-3 µg/1x10^6 cells, Human<br> M

Subcellular Localization

Cell membrane; Peripheral membrane protein; Cytoplasmic side; tight junction; Cell junction; gap junction; podosome

**Tissue Specificity** 

The alpha-containing isoform is found in most epithelial cell junctions. The short isoform is found both in endothelial cells and the highly specialized epithelial junctions of renal glomeruli and Sertoli cells of the seminiferous tubules.

Contents

Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na<sub>2</sub>HPO<sub>4</sub>, 0.05mg NaN<sub>3</sub>.

Immunogen



E.coli-derived human TJP1 recombinant protein (Position: H1178-F1527). Human TJP1 shares 82% amino acid (aa) sequence identity with mouse TJP1.

# Cross Reactivity

No cross-reactivity with other proteins.

Storage

Store 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 freeze-thaw cycles.

### Anti-TJP1 Antibody Picoband<sup>™</sup> (monoclonal, 3E12) - Protein Information

Name TJP1 (HGNC:11827)

#### **Function**

TIP1, TIP2, and TIP3 are closely related scaffolding proteins that link tight junction (TJ) transmembrane proteins such as claudins, junctional adhesion molecules, and occludin to the actin cytoskeleton (PubMed: <a href="http://www.uniprot.org/citations/7798316" target=" blank">7798316</a>, PubMed:<a href="http://www.uniprot.org/citations/9792688" target=" blank">9792688</a>). Forms a multistranded TJP1/ZO1 condensate which elongates to form a tight junction belt, the belt is anchored at the apical cell membrane via interaction with PATJ (By similarity). The tight junction acts to limit movement of substances through the paracellular space and as a boundary between the compositionally distinct apical and basolateral plasma membrane domains of epithelial and endothelial cells. Necessary for lumenogenesis, and particularly efficient epithelial polarization and barrier formation (By similarity). Plays a role in the regulation of cell migration by targeting CDC42BPB to the leading edge of migrating cells (PubMed:<a href="http://www.uniprot.org/citations/21240187" target="\_blank">21240187</a>). Plays an important role in podosome formation and associated function, thus regulating cell adhesion and matrix remodeling (PubMed: <a href="http://www.uniprot.org/citations/20930113" target=" blank">20930113</a>). With TIP2 and TIP3, participates in the junctional retention and stability of the transcription factor DBPA, but is not involved in its shuttling to the nucleus (By similarity). May play a role in mediating cell morphology changes during ameloblast differentiation via its role in tight junctions (By similarity).

### **Cellular Location**

Cell membrane; Peripheral membrane protein; Cytoplasmic side. Cell junction, tight junction. Cell junction. Cell junction, gap junction. Cell projection, podosome. Note=Moves from the cytoplasm to the cell membrane concurrently with cell-cell contact (PubMed:7798316). Forms a condensed tight junction-linked belt of protein during junction formation which becomes anchored to the apical cell membrane via interaction with PATJ (By similarity). At podosomal sites, is predominantly localized in the ring structure surrounding the actin core (PubMed:20930113). Colocalizes with SPEF1 at sites of cell- cell contact in intestinal epithelial cells (PubMed:31473225) {ECO:0000250|UniProtKB:O97758, ECO:0000269|PubMed:20930113, ECO:0000269|PubMed:31473225, ECO:0000269|PubMed:7798316}

#### **Tissue Location**

The alpha-containing isoform is found in most epithelial cell junctions. The short isoform is found both in endothelial cells and the highly specialized epithelial junctions of renal glomeruli and Sertoli cells of the seminiferous tubules

### Anti-TJP1 Antibody Picoband<sup>™</sup> (monoclonal, 3E12) - Protocols



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

- <u>Western Blot</u>
- Blocking Peptides
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- <u>Cell Culture</u>

# Anti-TJP1 Antibody Picoband™ (monoclonal, 3E12) - Images



Figure 1. Western blot analysis of TJP1 using anti-TJP1 antibody (M00860).

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 CACO-2 whole cell lysates,

Lane 2: human U20S whole cell lysates,

Lane 3: human K562 whole cell lysates,

Lane 4: human PC-3 whole cell lysates,

Lane 5: human T47D whole 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-TJP1 antigen affinity purified monoclonal antibody (Catalog # M00860) at 0.5  $\mu$ g/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 TJP1 at approximately 220 kDa. The expected band size for TJP1 is at 195 kDa.





Figure 2. IHC analysis of TJP1 using anti-TJP1 antibody (M00860).

TJP1 was detected in paraffin-embedded section of human placenta tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1  $\mu$ g/ml mouse anti-TJP1 Antibody (M00860) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen



Figure 3. IHC analysis of TJP1 using anti-TJP1 antibody (M00860).

TJP1 was detected in paraffin-embedded section of human kidney cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-TJP1 Antibody (M00860) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.





Figure 4. Flow Cytometry analysis of hela cells using anti-TJP1 antibody (M00860). Overlay histogram showing hela cells stained with M00860 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-TJP1 Antibody (M00860, 1  $\mu$ g/1x10<sup>6</sup> cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10  $\mu$ g/1x10<sup>6</sup> cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1  $\mu$ g/1x10<sup>6</sup>) used under the same conditions. Unlabelled sample (Red line) was also used as a control.



Figure 5. IF analysis of TJP1 using anti-TJP1 antibody (M00860).

TJP1 was detected in immunocytochemical section of MCF7 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 2  $\mu$ g/mL mouse anti-TJP1 Antibody (M00860) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Mouse IgG (BA1126) 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.