



Qty: 100µg/400 µL

Rabbit anti-ZO-1 (N-term)

Catalog No. 40-2300

Lot No.

## Rabbit anti-ZO-1 (N-term)

### FORM

This polyclonal antibody is supplied as a 400 µL aliquot at a concentration of 0.25 mg/mL in phosphate buffered saline (pH 7.4) containing 0.1% sodium azide. This antibody is epitope-affinity purified from rabbit antiserum.

PAD: ZMD.437

### IMMUNOGEN

Synthetic peptide derived from the N-terminal region of human, dog, mouse, and rat ZO-1

### SPECIFICITY

This antibody reacts with the ZO-1 protein. Based on sequence homology, reactivity with ZO-2 or ZO-3 is not expected.

### REACTIVITY

Reactivity has been confirmed with MDCK-II, Caco-2, Rat-1, and NRK52E cells by immunofluorescence. Reactivity has also been confirmed with frozen mouse liver, blood vessel (in heart), and mossy fiber terminal (in brain) tissues by immunohistochemistry and immunofluorescence, and with formalin-fixed, paraffin-embedded human pancreas, prostate, and small intestine tissues by immunohistochemistry. This antibody (or Cat# 18-7430) is recommended for Immunohistochemical detection of ZO-1. ZO-1 specific antibodies for other applications can be found at [www.invitrogen.com/antibodies](http://www.invitrogen.com/antibodies).

| Sample | Western Blotting  | Immuno-Fluorescence*** | Immuno-histochemistry (frozen) | Immuno-histochemistry (paraffin)* |
|--------|-------------------|------------------------|--------------------------------|-----------------------------------|
| Human  | Not recommended** | +++                    | ND                             | +++                               |
| Dog    | Not recommended** | +++                    | ND                             | ND                                |
| Rat    | Not recommended** | +++                    | ND                             | ND                                |
| Mouse  | Not recommended** | +++                    | +++                            | ND                                |

(Excellent +++, Good++, Poor +, No reactivity 0, Not applicable N/A, Not Determined ND)

### USAGE

Working concentrations for specific applications should be determined by the investigator. Appropriate concentrations will be affected by several factors, including secondary antibody affinity, antigen concentration, sensitivity of detection method, temperature and length of incubations, etc. The suitability of this antibody for applications other than those listed below has not been determined. The following concentration ranges are recommended starting points for this product.

**Western Blotting:** Not recommended\*\*. See [www.invitrogen.com/antibodies](http://www.invitrogen.com/antibodies) for other applications.  
**Immunofluorescence:** 2.5 µg/mL  
**Immunohistochemistry (frozen):** 2 µg/mL  
**Immunohistochemistry (paraffin)\*:** 1-2 µg/mL

\* For immunohistochemistry in formalin-fixed, paraffin-embedded tissues, tissue pretreatment with pepsin is required prior to staining.

\*\* Some lots of this antibody have successfully been used in Western Blotting using human, dog and rat cell lysates. However, for Western Blotting we recommend the use of monoclonal anti-ZO-1 antibody (Cat# 33-9100), or other polyclonal ZO-1 antibodies (Cat# 61-7300, #40-2200).

\*\*\* For Immunofluorescence methanol fixation is preferred even though paraformaldehyde fixation will also work.

### STORAGE

Store at 2-8°C for up to one month. Store at -20°C for long-term storage. Avoid repeated freezing and thawing.

(cont'd)

[www.invitrogen.com](http://www.invitrogen.com)

Invitrogen Corporation • 542 Flynn Rd • Camarillo • CA 93012 • Tel: 800.955.6288 • E-mail: [techsupport@invitrogen.com](mailto:techsupport@invitrogen.com)

PI402300

(Rev 10/08) DCC-08-1089

**Important Licensing Information** - These products may be covered by one or more Limited Use Label Licenses (see the Invitrogen Catalog or our website, [www.invitrogen.com](http://www.invitrogen.com)). By use of these products you accept the terms and conditions of all applicable Limited Use Label Licenses. Unless otherwise indicated, these products are for research use only and are not intended for human or animal diagnostic, therapeutic or commercial use.

## BACKGROUND

The tight junction is a cell-cell adhesion structure that constitutes the epithelial junctional complex along with adherens junctions and desmosomes. Tight junctions create a primary barrier to the diffusion of solutes across the cellular sheet, and also function as a boundary between the apical and basolateral membrane domains to produce their polarization.<sup>1</sup> Tight junction strands are mainly composed of claudins, occludin, and JAM.<sup>2</sup> Various scaffold proteins, ZO-1, ZO-2, and ZO-3, are concentrated at the cytoplasmic surfaces of the junctional complex regions to determine the specialization and localization of junctions. The zona occludens (ZO) proteins constitute the plaque structures underlying plasma membranes together with various proteins including cingulin, symplekin, the Par-3/Par-6/atypical protein kinase C complex, ZONAB, and guanine nucleotide exchange factor-H1/Lfc.<sup>2</sup> All three ZO proteins have three PDZ domains, one Src homology 3 domain, and one guanylate kinase-like homologue domain in this order from their NH<sub>2</sub> termini, indicating that ZO-1, ZO-2, and ZO-3 are membrane-associated guanylate kinase-like homologues (MAGUKs).<sup>3</sup>

A recent study showed that low levels of ZO-1 expression correlated with poor patient prognosis in breast cancer.<sup>4</sup> Another study observed aberrant ZO-1 expression in synovial sarcoma samples.<sup>5</sup>

ZO-1 antibodies are useful as tight junction markers because ZO-1 is exclusively concentrated at tight junctions, and directly binds to claudin, occludin, and JAM proteins.

## REFERENCES

1. Anderson JM and Van Itallie CM. *Am J Physiol* 269:G467-G475, 1995.
2. Umeda K, et al. *J Biol Chem* 279(43):44785-44794, 2004.
3. Itoh M, et al. *J Cell Biol* 121:491-502, 1993.
4. Martin TA, et al. *Eur J Cancer* 40(18):2717-2725, 2004.
5. Billings SD, et al. *Mod Pathol* 17(2):141-149, 2004.

## RELATED PRODUCTS

| <b>Product</b>              | <b>Conjugate</b> | <b>Cat. No.</b> |
|-----------------------------|------------------|-----------------|
| Protein A                   | Sepharose 4B     | 10-1041         |
| rec-Protein G               | Sepharose 4B     | 10-1241         |
| ZyMAX™ Goat anti-rabbit IgG | Unconjugated     | 81-6100         |
| ZyMAX™ Goat anti-mouse IgG  | Unconjugated     | 81-6500         |

Secondary antibody conjugates.

| <b>Conjugate</b> | <b>Goat anti-rabbit IgG (H+L)</b> | <b>Goat anti-mouse IgG (H+L)</b> | <b>Ex/Em*</b> | <b>Fluorescence similar to--</b> |
|------------------|-----------------------------------|----------------------------------|---------------|----------------------------------|
| Alexa Fluor® 488 | A11008                            | A11001                           | 495/519       | FITC                             |
| Alexa Fluor® 555 | A21428                            | A21422                           | 555/565       | Cy3                              |
| Alexa Fluor® 594 | A11012                            | A11005                           | 590/617       | Texas Red                        |
| Alexa Fluor® 647 | A21244                            | A21235                           | 650/668       | Cy5                              |
| HRP              | 81-6120                           | 81-6520                          | NA**          | NA                               |
| AP               | 81-6122                           | 81-6522                          | NA            | NA                               |
| Biotin           | B2770                             | B2763                            | NA            | NA                               |

\*Excitation/emission (nm); \*\*Not applicable

For additional secondary antibody conjugates, visit [www.invitrogen.com/antibodies](http://www.invitrogen.com/antibodies)

## For Research Use Only

[www.invitrogen.com](http://www.invitrogen.com)

Invitrogen Corporation • 542 Flynn Rd • Camarillo • CA 93012 • Tel: 800.955.6288 • E-mail: [techsupport@invitrogen.com](mailto:techsupport@invitrogen.com)

PI402300

(Rev 10/08) DCC-08-1089

**Important Licensing Information** - These products may be covered by one or more Limited Use Label Licenses (see the Invitrogen Catalog or our website, [www.invitrogen.com](http://www.invitrogen.com)). By use of these products you accept the terms and conditions of all applicable Limited Use Label Licenses. Unless otherwise indicated, these products are for research use only and are not intended for human or animal diagnostic, therapeutic or commercial use.