



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.

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 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)

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PI402300

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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.¹ Tight junction strands are mainly composed of claudins, occludin, and JAM.² 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.² 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 NH₂ termini, indicating that ZO-1, ZO-2, and ZO-3 are membrane-associated guanylate kinase-like homologues (MAGUKs).³

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

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

Product	Conjugate	Cat. No.
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.

Conjugate	Goat anti-rabbit IgG (H+L)	Goat anti-mouse IgG (H+L)	Ex/Em*	Fluorescence similar to--
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

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