

Qty: 100 μg/200 μL Mouse anti-Claudin-5 **Catalog No.** 35-2500 **Lot No.** See product label

Mouse anti-Claudin-5

FORM

This monoclonal antibody is supplied as a 200 µL aliquot at a concentration of 0.5 mg/mL in phosphate buffered saline (PBS), pH 7.4, containing 0.1% sodium azide (NaN₃). The antibody is Protein A affinity-purified from mouse ascites.

CLONE: 4C3C2

ISOTYPE: Mouse IgG₁

IMMUNOGEN

Synthetic peptide derived from the mouse Claudin-5 protein

SPECIFICITY

This antibody reacts specifically with the ~ 22-24 kDa endogenous Claudin-5 protein.

REACTIVITY

Reactivity has been confirmed with rat and mouse Claudin-5 by Western blotting using rat lung, kidney, and small intestine homogenates and mouse lung homogenates. Reactivity has also been confirmed with formalin-fixed, paraffin-embedded human colon tissue by immunohistochemistry.*

Sample	ELISA	Immuno- precipitation	Western blotting	Immuno- histochemistry (FFPE)
Rat	nt	nt	+++	nt
Mouse	nt	nt	+++	nt
Human	nt	nt	nt	+++*
Immunogen	+++	nt	nt	nt

nt-not tested

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.

ELISA: 0.1-1.0 μg/mL Western Blotting: 1-3 μg/mL Immunohistochemistry*: 5-10 μg/mL

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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^{*} For best results in immunohistochemistry with formalin-fixed, paraffin-embedded (FFPE) tissues, heat induced epitope retrieval (HIER) with EDTA, pH 8.0, is required prior to staining.

BACKGROUND

Tight junctions are specialized regions of cell-cell contact that are particularly abundant in luminal epithelial cell sheets. In freeze-fracture electron micrographs, tight junctions are visualized as belt-like bands of anastomosing sealing strands (TJ strands) that completely encircle the lateral surfaces of each cell. TJ strands on adjacent cells are presumed to interact with each other to form a sort of "molecular gasket" that prevents ions, water and other molecules from leaking between cells and thus, from one side of the sheet to the other. In addition to this so-called "barrier" function, the "fence" function of tight junctions plays an important role in maintaining epithelial cell-polarity by blocking the diffusion of membrane proteins between apical (luminal) and basolateral cell surfaces. Confinement of the glucose symport to apical surfaces allows glucose to be transported vectorially from the lumen, through the cell, and into the bloodstream.

Several peripheral membrane proteins are associated with tight junctions, including ZO-1, ZO-2, ZO-3, cingulin, the 7H6 antigen, Rab-3b, and symplekin. ¹⁻⁶ While their precise functions are not known, roles for these proteins have been suggested in tight junction assembly and maintenance; signal transduction; and the regulation of tight junction permeability. A growing body of evidence also suggests that actin filaments play a major role in regulating tight junction permeability.

Initially, the only transmembrane protein known to be associated with tight junctions was occludin, a ~65 kDa protein with four transmembrane domains. Despite widespread expectation to the contrary, a critical structural role for occludin in TJ strands was ruled out by the observation of normal tight junctions formed between cells disrupted at both occludin alleles. Closer examination of isolated tight junctions uncovered two related, ~22 kDa, four-transmembrane domain proteins, claudin-1 and claudin-2, with no similarity to occludin. In contrast to occludin, which induces only a small number of short strands at cell-cell contact sites when introduced into fibroblasts lacking tight junctions, claudin-1 and -2 induce networks of strands characteristic of true tight junctions. These findings suggest that claudin-1 and -2 are major structural components of TJ strands and that occludin plays some other accessory role. Excitement in the tight junction field continues to rise following the recent discovery of claudins -3, -4, -5, -6, -7, and -8 and experiments suggesting that tight junctions in different tissues are comprised of different sets of claudin family proteins.

REFERENCES

- 1. Anderson JM. Curr Biol 6:382-384, 1996.
- 2. Anderson JM, Van Itallie CM. Am J Physiol 269:G467-475, 1995.
- 3. Howarth AG, Stevenson BR. Adv Struct Biol 4:25-39, 1996.
- 4. Stevenson BR, Keon BH. Ann Rev Cell Dev Biol 14:89-109, 1998.
- 5. Tsukita S, et al. Cell Struct Funct 21:381-385, 1996.
- 6. Yap AS, et al. Mem Biol 163:159-167, 1998.
- 7. Saitou M, et al. J Cell Biol 141:397-408, 1998.
- 8. Furuse M, et al. J Cell Biol 143(2):391-401, 1998.
- 9. Tsukita S, Furuse M. Genes Cells 3:569-573, 1998.
- 10. Morita K, et al. PNAS 96(2):511-516, 1999.

RELATED PRODUCTS

Primary antibodies	Clone or PAD*	Cat. No.
Rb anti-Claudin-1	JAY.8	51-9000
Rb anti-Claudin-1	MH25	71-7800
Rb anti-Claudin-2	MH44	51-6100
Rb anti-Claudin-3	Z23.JM	34-1700
Rb anti-Claudin-5	Z43.JK	34-1600
Ms anti-ZO-1	ZO1-1A12	33-9100
FITC-Ms anti-ZO-1	ZO1-1A12	33-9111
Rb anti-ZO-1	Z-R1	61-7300
Rb anti-ZO-2		71-1400
Ms anti-Occludin	OC-3F10	33-1500
HRP-Ms anti-Occludin	OC-3F10	33-1520
FITC-Ms anti-Occludin	OC-3F10	33-1511
Rb anti-Occludin	Z-T22	71-1500
Rb anti-Occludin		71-1600
	4DAD 1 1 11 11	

^{*}PAD- polyclonal antibody designation

Conjugate	ZyMAX™ Goat anti- Rabbit IgG (H+L)	ZyMAX™ Goat anti- Mouse IgG (H+L)
Purified	81-6100	81-6500
FITC	81-6111	81-6511
TRITC	81-6114	81-6514
Сутм3	81-6115	81-6515
Сутм5	81-6116	81-6516
HRP	81-6120	81-6520
AP	81-6122	81-6522
Biotin	81-6140	81-6540

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