

Datasheet for 600-401-938**Myosin Antibody****Overview**

Description:	Anti-Myosin (RABBIT) Antibody - 600-401-938
Item No.:	600-401-938
Size:	100 µg
Applications:	ELISA, WB, FC, Other
Reactivity:	Mouse, Rat
Host Species:	Rabbit

Product Details

Background:	Myosin is the major component of thick muscle filaments, and is a long asymmetric molecule containing a globular head and a long tail. The molecule consists of two heavy chains each ~200,000 daltons, and four light chains each ~16,000 - 21,000 daltons. Activation of smooth and cardiac muscle primarily involves pathways that increase calcium and myosin phosphorylation resulting in contraction. Myosin light chain phosphatase acts to regulate muscle contraction by dephosphorylating activated myosin light chain. The selected peptide sequence used to generate the polyclonal antibody is located near the amino terminal end of the polypeptide corresponding to the smooth/non-muscle form of myosin regulatory light chain found in cardiac myocytes in addition to smooth and non-muscle cells.
Synonyms:	rabbit anti-Myosin antibody, Myosin regulatory light polypeptide 12A, Myosin regulatory light chain 12A, Myosin regulatory light chain MRLC3
Host Species:	Rabbit
Clonality:	Polyclonal
Format:	IgG

Target Details

Gene Name:	MYL12A
Reactivity:	Mouse, Rat
Immunogen Type:	Conjugated Peptide

Immunogen:	This affinity purified antibody was prepared from whole rabbit serum produced by repeated immunizations with a synthetic peptide corresponding to amino acids aa 10-35 of human myosin light chain protein.
Purity/Specificity:	This affinity purified antibody is directed against the regulatory light chain of smooth and non-muscle myosin. The antibody detects both unphosphorylated and monophosphorylated forms of the protein. The product was affinity purified from monospecific antiserum by immunoaffinity purification. Cross reactivity is expected with myosin light chain from human, mouse and rat sources. Reactivity with the protein from other species has not been determined; however, the sequence of the immunogen is nearly identical in mammalian and avian species. BLAST search analysis was used to determine that the smooth and non-muscle forms of myosin regulatory light chain have identical sequences. Cross reactivity is expected.
Relevant Links:	• NCBI - NP_001289978.1 • UniProtKB - J3QRS3 • GeneID - 10627

Application Details

Tested Applications:	ELISA, WB
Suggested Applications:	FC, Other (Based on references)
Application Note:	This affinity-purified antibody was tested by ELISA and immunoblotting and was found to be reactive with both the unphosphorylated and mono-phosphorylated forms of the protein. Although not tested, this antibody is likely functional in immunohistochemistry and immunoprecipitation.
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
ELISA:	1:10,000 - 1:70,000
IHC:	User Optimized
IP:	1:100
WB:	1:500 - 1:2,000

Formulation

Physical State:	Liquid (sterile filtered)
Concentration:	0.98mg/mL by UV absorbance at 280 nm
Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	0.01% (w/v) Sodium Azide

Stabilizer: None

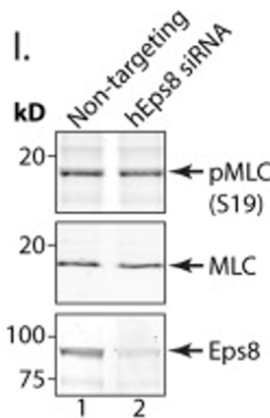
Shipping & Handling

Shipping Condition: Dry Ice

Storage Condition: Store vial at -20° C prior to opening. Aliquot contents and freeze at -20° C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use.

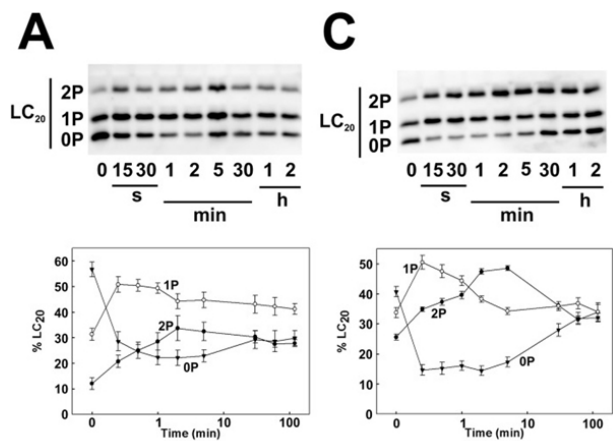
Expiration: Expiration date is one (1) year from date of receipt.

Images



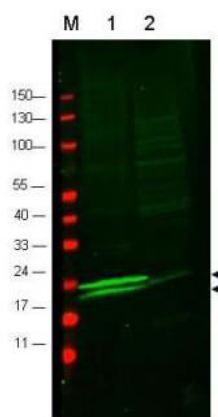
Western Blot

I) Western blot analyses of Eps8, myosin II regulatory light chain (MLC p/n 600-401-938), and myosin II regulatory light chain phosphorylated on serine 19 (pMLC (S19) p/n 600-401-416) in lysates of A375 cells that were treated with non-targeting siRNAs (non-targeting) or siRNAs targeting human Eps8 to deplete Eps8 (hEps8 siRNA). Fig 2. PMID: 26163656



Western Blot

Human coronary (A, B) and umbilical arterial smooth muscle cells (C, D) were seeded in 4-well plates, serum starved overnight and stimulated with 5% FBS at time zero. Cells were lysed in Laemmli sample buffer at the indicated times of serum stimulation and subjected to Phos-tag SDS-PAGE and western blotting with anti-LC20 (Myosin p/n 600-401-938) to detect unphosphorylated (0P), mono- (1P) and diphosphorylated LC20 (2P). Representative western blots are shown in panels A and C above cumulative quantitative data for each of the LC20 bands. Fig 1. PMID: 31834925



Western Blot

Western blot using Rockland's anti-RLC of Smooth and Non-muscle Myosin antibody to detect vascular myosin (rat aorta, lane 1) but not cardiac myosin (mouse heart, lane2). Each lane was loaded with 35 µg of lysate. Arrowheads indicate the detection of both mono-phosphorylated (upper) and unphosphorylated (lower) forms of the protein. After blocking with 5% Normal goat serum and 0.5% BLOTTO in PBS, the membrane was probed with the primary antibody diluted in blocking buffer to 1:600 for 2 h at room temperature. The membrane was washed and reacted with a 1:10,000 dilution of IRDye800™ conjugated Gt-a-Rabbit IgG [H&L] MX (611-132-122) for 45 min at room temperature (800 nm channel, green). Molecular weight estimation was made by comparison to prestained MW markers in lane M (700 nm channel, red). IRDye™800 fluorescence image was captured using the Odyssey® Infrared Imaging System developed by LI-COR. IRDye is a trademark of LI-COR, Inc. Other detection systems will yield similar results.

References

- Aguilar-Cuenca R et al. Tyrosine Phosphorylation of the Myosin Regulatory Light Chain Controls Non-muscle Myosin II Assembly and Function in Migrating Cells. *Curr Biol.* (2020)
- Deng JT et al. Rho-associated kinase and zipper-interacting protein kinase, but not myosin light chain kinase, are involved in the regulation of myosin phosphorylation in serum-stimulated human arterial smooth muscle cells. *PLoS One.* (2019)
- Lahey et al. Signaling pathways induced by serine proteases to increase intestinal epithelial barrier function. *PLOS One* (2017)
- Logue et al. Erk regulation of actin capping and bundling by Eps8 promotes cortex tension and leader bleb-based migration. *Elife* (2015)

Disclaimer

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