

 Rudolf-Wissell-Str. 28

 37079 Göttingen, Germany

 Phone:
 +49 551-50556-0

 Fax:
 +49 551-50556-384

 E-mail:
 sales@sysy.com

 Web:
 www.sysy.com

MLC-2A

Cat.No. 311 011AT1; Monoclonal mouse antibody, 100 µg purified IgG (lyophilized)

Data Sheet

Reconstitution/ Storage	100 μ g purified IgG, lyophilized, fluorescence-labeled with ATTO [®] 647N. For reconstitution add 100 μ l H ₂ O to get a 1mg/ml solution in PBS. Either add 1:1 (v/v) glycerol, then aliquot and store at -20°C until use, or store aliquots at -80°C without additives. Reconstitute immediately upon receipt! Avoid bright light when working with the antibody to minimize photo bleeching of the fluorescent dye.
Applications	WB: N/A IP: N/A ICC: yes (see remarks) IHC: yes (see remarks) IHC-P/FFPE: 1 : 200
Label	ATTO 647N
Clone	56F5
Subtype	IgG2b (κ light chain)
Immunogen	Recombinant protein corresponding to AA 1 to 175 from human MLC-2A (UniProt Id: Q01449)
Epitop	Epitop: AA 1 to 175 from human MLC-2A (UniProt Id: Q01449)
Reactivity	Reacts with: human (Q01449), rat, mouse (Q9QVP4). No signal: chicken. Other species not tested yet.
Specificity	Specific for MLC-2A, no cross-reactivity to MLC-2V.
matching control	311-0P
Remarks	ICC: Only methanol fixation, recommended protocol.
	IHC : Protocol for immunohistochemistry of this antibody will be provided with the goods.

TO BE USED IN VITRO / FOR RESEARCH ONLY NOT TOXIC, NOT HAZARDOUS, NOT INFECTIOUS, NOT CONTAGIOUS

During cardiogenesis two major isoforms of **m**yosin **l**ight **c**hain **2** are co-expressed in a tightly regulated manner. **MLC-2A** is only present in the atrium while MLC-2V is exclusively expressed in the ventricle. Knock out studies revealed that the 2A isoform cannot substitute for the 2V variant in the ventricular chamber.

Recently it has been demonstrated that embryonic and adult stem cells can be differentiated into

cardiomyocytes which may generate suitable replacements for damaged heart tissue in the future. This monoclonal antibody is a useful tool to distinguish between ventricle and atrium specific cardiomyocytes.

Selected General References

Mechanism of spontaneous excitability in human embryonic stem cell derived cardiomyocytes. Satin J, Kehat I, Caspi O, Huber I, Arbel G, Itzhaki I, Magyar J, Schroder EA, Perlman I, Gepstein L The Journal of physiology (2004) 559(Pt 2): 479-96.

Selection of ventricular-like cardiomyocytes from ES cells in vitro.

Müller M, Fleischmann BK, Selbert S, Ji GJ, Endl E, Middeler G, Müller OJ, Schlenke P, Frese S, Wobus AM, Hescheler J, et al. FASEB journal : official publication of the Federation of American Societies for Experimental Biology (2000) 14(15): 2540-8.

Transgenic remodeling of the contractile apparatus in the mammalian heart. Palermo J, Gulick J, Colbert M, Fewell J, Robbins J Circulation research (1996) 78(3): 504-9.



Protocol for immunohistochemistry for MLC-2A antibodies

mouse monoclonal antibody (Cl. 56F5); purified IgG; cat. no. 311 011AT1

Deparaffinization and rehydration:

- Incubate sections with xylene twice for 5-10min.
- Incubate section with acetone once for 10min.
- Incubate section with acetone diluted 1:2 with TBS pH 7.6 once for 10min.
- Incubate section with TBS once for 10min.
- Rinse slides in dest. H2O.

Antigen retrieval: is necessary

- Microwave sections for 15 min in freshly mixed 10 mM citrate buffer, pH 6.0 to retrieve the antigen.
- Cool slides slowly to RT (20-30min).
- Rinse slides in PBS:
 - Citrate buffer: A: 21.01g citric acid in 1000ml dest. H₂O

B: 29.41g sodium citrate in 1000ml dest. H₂O

Mix 9ml A with 41ml B and 450ml dest. H_2O . Adjust the pH to 6.0.

Immunofluorescence:

- Block sections in 10% normal goat serum in PBS (30min at RT).
- Remove blocking solution and incubate sections overnight at 4°C with the primary antibody (anti-MLC-2V) in PBS / 2% BSA at a dilution of 1:200 (5µg/ml).
- Remove primary antibody and wash thoroughly with PBS +BSA 3 times for 5min.
- Mount slices and examine microscopically.