

MONOCLONAL ANTIBODY

**Anti-GFP (Green Fluorescent Protein) mAb-Alexa Fluor<sup>®</sup> 488**

Code No.	Clone	Subclass	Quantity	Concentration
D153-A48	RQ2	Rat IgG2a $\kappa$	50 $\mu$ L	1 mg/mL

**BACKGROUND:** Since the detection of intracellular Aequorea Victoria Green Fluorescent Protein (GFP) requires only irradiation by UV or blue light, it provides an excellent means for monitoring gene expression and protein localization in living cells. Monoclonal anti-GFP antibody can detect GFP fusion protein on Immunocytochemistry.

**SOURCE:** This antibody was purified from hybridoma (clone RQ2) supernatant using protein G agarose. This hybridoma was established by fusion of mouse myeloma cell PAI with Wistar rat lymph node immunized with GFP purified from GFP expressed 293T cells by affinity chromatographic technique using mouse anti-GFP.

**FORMULATION:** 50  $\mu$ g of IgG in 50  $\mu$ L volume of PBS containing 1% BSA and 0.09% NaN<sub>3</sub>.

\*Azide may react with copper or lead in plumbing system to form explosive metal azides. Therefore, always flush plenty of water when disposing materials containing azide into drain.

**STORAGE:** This antibody solution is stable for one year from the date of purchase when stored at 4°C.

**REACTIVITY:** This antibody reacts with GFP fusion protein on Immunocytochemistry. It reacts with EBFP, ECFP, EGFP, Venus and Sapphire.

**APPLICATION:**

Immunocytochemistry (for PFA fixed cells): 2-5  $\mu$ g/mL

\*This antibody is not suitable for alcohol fixation such as ethanol or methanol.

Please refer to the data sheet (MBL code no. D153-3) for other applications.

Detailed procedure is provided in the following **PROTOCOL**.

**INTENDED USE:**

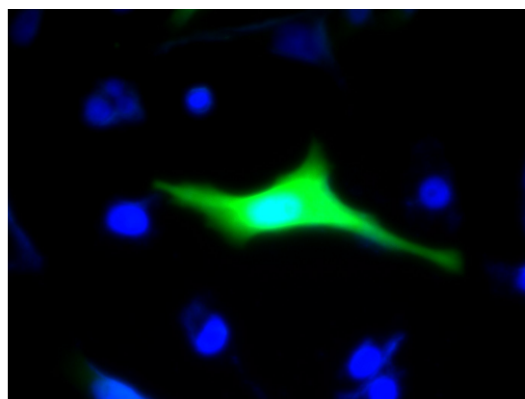
For Research Use Only. Not for use in diagnostic procedures.

**REFERENCES:**

- 1) Kitamura, A., *et al.*, *Genes Cells*. **22**, 521-534 (2017)
- 2) Sato, Y., *et al.*, *J. Biol. Chem.* **284**, 11873-11881 (2009)
- 3) Sakurai, T., *et al.*, *J. Cell Biol.* **183**, 339-352 (2008)
- 4) Kato, A., *et al.*, *J. Virol.* **82**, 6172-6189 (2008)
- 5) Dragone, L. L., *et al.* *PNAS*. **103**, 18202-18207 (2006)

- 6) Darzacq, X., *et al.* *J. Cell Biol.* **173**, 207-218 (2006)
- 7) Hayakawa, T., *et al.* *Plant Cell Physiol.* **47**, 891-904 (2006)
- 8) Obuse, C., *et al.* *Nat. Cell Biol.* **6**, 1135-1141 (2004)

Clone RQ2 is used in these references.



**Immunocytochemical detection of GFP expressed in HeLa using D153-A48.**

Green: Alexa Fluor<sup>®</sup> 488 and GFP own fluorescence  
Blue: DAPI

**PROTOCOL:**

**Immunocytochemistry**

- 1) Culture the cells in the appropriate condition on a glass slide. (for example, spread  $1 \times 10^4$  cells of transfectant cells for one slide, then incubate in a CO<sub>2</sub> incubator for one night.)
- 2) Wash the cells 3 times with PBS.
- 3) Fix the cells by immersing the slide in PBS containing 4% paraformaldehyde for 10 minutes at room temperature.
- 4) Wash the glass slide 3 times with PBS.
- 5) Immerse the slide in PBS containing 0.2% Triton X-100 for 10 minutes at room temperature.
- 6) Wash the glass slide 3 times with PBS.
- 7) Add Clear Back (MBL; code no. MTG-001) onto the cells and incubate for 10 minutes at room temperature.
- 8) Tip off Clear Back, add the primary antibody diluted with PBS containing 2% FCS as suggested in the **APPLICATION** onto the cells and incubate for 30 minutes at room temperature (Optimization of antibody concentration or incubation condition is recommended if necessary.)
- 9) Wash the glass slide 2 times with PBS.
- 10) Counter stain with DAPI for 2 minutes at room temperature.
- 11) Wash the glass slide 2 times with PBS.

- 12) Wipe excess buffer off the slide but take care not to touch the cells. Never leave the cells to dry.
- 13) Promptly add mounting medium onto the slide, then put a cover slip on it.

## RELATED PRODUCTS:

### Antibodies

D153-3	Anti-GFP mAb (RQ2)
D153-6	Anti-GFP mAb-Biotin (RQ2)
D153-8	Anti-GFP mAb-Agarose (RQ2)
M047-A48	Anti-GFP mAb-Alexa Fluor <sup>®</sup> 488 (RQ2)
M047-A59	Anti-GFP mAb-Alexa Fluor <sup>®</sup> 594 (RQ2)
M047-A64	Anti-GFP mAb-Alexa Fluor <sup>®</sup> 647 (RQ2)
M048-3	Anti-GFP mAb (1E4)
598	Anti-GFP pAb (polyclonal)
598-7	Anti-GFP pAb-HRP-Direct (polyclonal)
PM073	Anti-Renilla GFP pAb (polyclonal)
M208-3	Anti-RFP mAb Cocktail (1G9, 3G5)
M155-3	Anti-RFP mAb (8D6)
M165-3	Anti-RFP mAb (3G5)
M165-8	Anti-RFP mAb-Agarose (3G5)
M204-3	Anti-RFP mAb (1G9)
M204-7	Anti-RFP mAb-HRP-Direct (1G9)
PM005	Anti-RFP pAb (polyclonal)
PM005-7	Anti-RFP pAb-HRP-Direct (polyclonal)
M180-3	Anti-HA-tag mAb (TANA2) (200 µL)
M180-A48	Anti-HA-tag mAb-Alexa Fluor <sup>®</sup> 488 (TANA2)
M180-A59	Anti-HA-tag mAb-Alexa Fluor <sup>®</sup> 594 (TANA2)
M180-A64	Anti-HA-tag mAb-Alexa Fluor <sup>®</sup> 647 (TANA2)
561	Anti-HA-tag pAb (polyclonal) (0.1 mL)
M132-3	Anti-HA-tag mAb (5D8)
M185-3L	Anti-DDDDK-tag mAb (FLA-1) (1 mL)
M185-A48	Anti-DDDDK-tag mAb-Alexa Fluor <sup>®</sup> 488 (FLA-1)
M185-A59	Anti-DDDDK-tag mAb-Alexa Fluor <sup>®</sup> 594 (FLA-1)
M185-A64	Anti-DDDDK-tag mAb-Alexa Fluor <sup>®</sup> 647 (FLA-1)
PM020	Anti-DDDDK-tag pAb (polyclonal)
M047-3	Anti-Myc-tag mAb (PL14)
M047-A48	Anti-Myc-tag mAb-Alexa Fluor <sup>®</sup> 488 (PL14)
M047-A59	Anti-Myc-tag mAb-Alexa Fluor <sup>®</sup> 594 (PL14)
M047-A64	Anti-Myc-tag mAb-Alexa Fluor <sup>®</sup> 647 (PL14)
M192-3	Anti-Myc-tag mAb (My3) (200 µL)
562	Anti-Myc-tag pAb (polyclonal) (0.1 mL)
D291-3	Anti-His-tag mAb (OGHis) (200 µL)
D291-A48	Anti-His-tag mAb-Alexa Fluor <sup>®</sup> 488 (OGHis)
D291-A59	Anti-His-tag mAb-Alexa Fluor <sup>®</sup> 594 (OGHis)
D291-A64	Anti-His-tag mAb-Alexa Fluor <sup>®</sup> 647 (OGHis)
M089-3	Anti-His-tag mAb (6C4)
M136-3	Anti-His-tag mAb (2D8)
PM032	Anti-His-tag pAb (polyclonal)
M167-3	Anti-V5-tag mAb (1H6)
M215-3	Anti-V5-tag mAb (OZA3)
PM003	Anti-V5-tag pAb (polyclonal)
PM021	Anti-S-tag pAb (polyclonal)
PM070	Anti-E-tag pAb (polyclonal)
PM022	Anti-T7-tag pAb (polyclonal)
563	Anti-VSV-G-tag pAb (polyclonal)

M071-3	Anti-GST-tag mAb (3B2)
M209-3	Anti-GST-tag mAb (GT5)
PM022	Anti-GST-tag pAb (polyclonal)
M013-3	anti-Thioredoxin (2C9)
M095-3	Anti-Luciferase mAb (2D4)
PM016	Anti-Luciferase pAb (polyclonal)
PM047	Anti-Renilla Luciferase pAb (polyclonal)
M094-3	Anti-β-galactosidase mAb (5A3)
PM049	Anti-β-galactosidase pAb (polyclonal)
M091-3	Anti-MBP (Maltose Binding Protein) mAb (1G12)
M013-3	Anti-Thioredoxin (Trx-tag) mAb (2C9)
PM015	Anti-Chitin Binding Domain pAb (polyclonal)
PM071	Anti-Calmodulin Binding Protein-tag pAb (polyclonal)
M211-3	Anti-Strep-tag II mAb (4F1)
M214-3	Anti-mini-AID-tag mAb (1E4)

### Smart-IP series

3190	Magnetic Rack
D153-11	Anti-GFP mAb-Magnetic Beads (RQ2)
M165-11	Anti-RFP mAb-Magnetic Beads (3G5)
M180-11	Anti-HA-tag mAb-Magnetic Beads (TANA2)
M132-11	Anti-HA-tag mAb-Magnetic Beads (5D8)
M185-11	Anti-DDDDK-tag mAb-Magnetic Beads (FLA-1)
M047-11	Anti-Myc-tag mAb-Magnetic Beads (PL14)
D291-11	Anti-His-tag mAb-Magnetic Beads (OGHis)
M167-11	Anti-V5-tag mAb-Magnetic Beads (1H6)
M198-9	Anti-E-tag mAb-Magnetic beads (21D11)
D058-9	Anti-Multi Ubiquitin mAb-Magnetic beads (FK2)
M075-11	Mouse IgG1 (isotype control)-Magnetic Beads
M076-11	Mouse IgG2a (isotype control)-Magnetic Beads
M077-11	Mouse IgG2b (isotype control)-Magnetic Beads
M081-11	Rat IgG2a (isotype control)-Magnetic Beads
M180-10	Anti-HA-tag mAb-Magnetic Agarose (TANA2)
M132-10	Anti-HA-tag mAb-Magnetic Agarose (5D8)
M185-10	Anti-DDDDK-tag mAb-Magnetic Agarose (FLA-1)
M047-10	Anti-Myc-tag mAb-Magnetic Agarose (PL14)
D291-10	Anti-His-tag mAb-Magnetic Agarose (OGHis)
D153-10	Anti-GFP mAb-Magnetic Agarose (RQ2)
M165-10	Anti-RFP mAb-Magnetic Agarose (3G5)
M167-10	Anti-V5-tag mAb-Magnetic Agarose (1H6)
M198-10	Anti-E-tag mAb-Magnetic Agarose (21D11)

### Isotype Controls

M081-3	Rat IgG2a isotype control (2H3)
M081-A48	Rat IgG2a isotype control-Alexa Fluor <sup>®</sup> 488 (2H3)

### Blocking Reagent

MTG-001	Clear Back
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Other related antibodies and kits are also available.  
Please visit our website at <http://ruo.mbl.co.jp/>

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## LABEL LICENSES:

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