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

MBL
A JSR Life Sciences Company

Smart-IP Series

Anti-GFP (Green Fluorescent Protein) mAb -Magnetic Beads

CODE No. D153-11

CLONALITY Monoclonal
CLONE RQ2
ISOTYPE Rat IgG2a κ
QUANTITY 20 tests (Slurry: 1 mL)

SOURCE Purified IgG from hybridoma supernatant
IMMUNOGEN GFP purified from GFP expressed 293T cells
REACTIVITY This antibody reacts with EBFP, ECFP, EGFP, Venus and Sapphire as well as GFP.
FORMULATION 10 mg magnetic beads in 1 mL PBS/0.1% BSA/0.09% NaN_3
*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 beads suspension is stable for one year from the date of purchase when stored at 4°C.

APPLICATION-CONFIRMED
Immunoprecipitation 50 µL of beads slurry/sample
*The purification capacity of Anti-GFP (Green Fluorescent Protein) mAb-Magnetic Beads varies depending upon the characteristics of a GFP fusion protein. For example, 50 µL of beads slurry bounds 4.2 µg of a GFP fusion protein (32 kDa).

APPLICATION-REPORTED
RNP Immunoprecipitation (RIP) Reference 6)

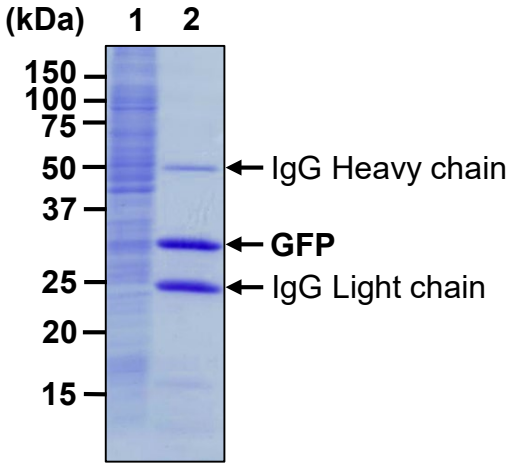
REFERENCES
1) Zhang, D., *et al*., *Rice*. **11**, 45 (2018) [IP]
2) Jessen, T. N. and Jessen, J. R., *Exp. Cell Res*. **361**, 265-276 (2017) [IP]
3) Aoyama, S., *et al*., *Biochem. Biophys. Res. Commun.* **491**, 33-39 (2017) [IP]
4) Chen, C. C., *et al*., *J. Biol. Chem.* **292**, 12560-12576 (2017) [Co-IP]
5) Amara, C. S., *et al*., *Front. Cell Dev. Biol.* **5**, 20 (2017) [IP]
6) Sommer, G., *et al., PLoS One* **12**, e0173246 (2017) [RIP]
7) Wang, Y., *et al*., *Plant Physiol.* **173**, 1235-1246 (2016) [IP]

For more information, please visit our web site **https://ruo.mbl.co.jp/**

The descriptions of the following protocols are examples. Each user should determine the appropriate condition.

Immunoprecipitation

1) Wash 1 x 10^7 cells 3 times with PBS and suspend with 1 mL of cold Lysis buffer [50 mM Tris-HCl (pH 7.5), 150 mM NaCl, 1% NP-40] containing appropriate protease inhibitors.
2) Centrifuge the tube at 12,000 x g for 5 min. at 4°C and transfer the supernatant to another tube.
3) Add magnetic beads as suggested in the **APPLICATION** and purified GFP protein into 300 µL of the supernatant prepared in step 2). Mix well and incubate with gentle agitation for 1 hr. at 4°C.
4) Place the tube on the magnetic rack (MBL; code no. 3190) for a few seconds.
5) Remove the supernatant.
6) Add 1 mL of cold Wash buffer [50 mM Tris-HCl (pH 7.5), 150 mM NaCl, 0.05% NP-40] and resuspend the magnetic beads.
7) Place the tube on the magnetic rack for a few seconds.
8) Remove the supernatant.
9) Repeat Steps 6)-8) 3 times.
10) Resuspend the magnetic beads in 20 µL of Laemmli's sample buffer, boil for 2 min., and place the tube on the magnetic rack for a few seconds.
11) Load 20 µL of the sample per lane in a 1-mm-thick SDS-polyacrylamide gel and carry out electrophoresis.
12) Visualize the protein bands by CBB staining.



Immunoprecipitation of GFP protein

Sample: 293T cell lysate from 3x10^6 cells + GFP protein 10 µg
Lane 1: Input (5 µL/lane)
Lane 2: Post-IP beads of Anti-GFP mAb (M153-11)