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

My select sampler set

Anti-RFP mAb

CODE No.	M204-3MS
CLONALITY	Monoclonal
CLONE	1G9
ISOTYPE	Mouse IgG2b κ
QUANTITY	20 µL, 1 mg/mL
SOURCE	Purified IgG from hybridoma supernatant
IMMUNOGEN	RFP recombinant protein
REACTIVITY	This clone reacts with mRFP1, DsRed, mCherry, mOrange and mPlumn. It does not cross-react with GFP.
FORMULATION	PBS containing 50% glycerol (pH 7.2). No preservative is contained.
STORAGE	This antibody solution is stable for one year from the date of purchase when stored at -20°C.

APPLICATION-CONFIRMED

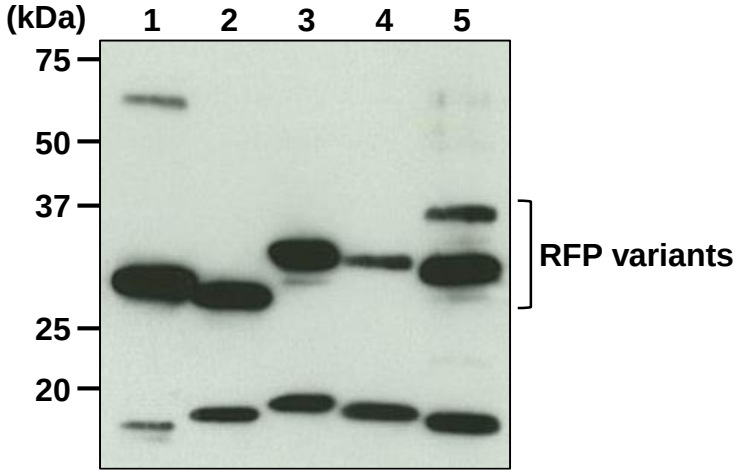
Western blotting	1 µg/mL
Immunoprecipitation	Not recommended
Immunocytochemistry	Not recommended

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.

SDS-PAGE & Western blotting

1) Mix the sample with equal volume of Laemmili's sample buffer.
2) Boil the sample for 3 min. and centrifuge. Load 10 μL of the sample per lane in a 1-mm-thick SDS-polyacrylamide gel (12.5% acrylamide) for electrophoresis.
3) Blot the protein to a polyvinylidene difluoride (PVDF) membrane at 1 mA/cm^2 for 1 hr. in a semi-dry transfer system (Transfer Buffer: 25 mM Tris, 190 mM glycine, 20% methanol). See the manufacturer's manual for precise transfer procedure.
4) To reduce nonspecific binding, soak the membrane in 10% skimmed milk (in PBS, pH 7.2) overnight at 4°C.
5) Wash the membrane with PBS-T [0.05% Tween-20 in PBS] (5 min. x 3).
6) Incubate the membrane with primary antibody diluted with 1% skimmed milk (in PBS, pH 7.2) as suggested in the **APPLICATIONS** for 1 hr. at room temperature. (The concentration of antibody will depend on the conditions.)
7) Wash the membrane with PBS-T (5 min. x 3).
8) Incubate the membrane with the 1:10,000 of anti-IgG (Mouse) pAb-HRP (MBL; code no. 330) diluted with 1% skimmed milk (in PBS, pH 7.2) for 1 hr. at room temperature.
9) Wash the membrane with PBS-T (5 min. x 3).
10) Wipe excess buffer on the membrane, and then incubate it with appropriate chemiluminescence reagent for 1 min. Remove extra reagent from the membrane by dabbing with paper towel, and seal it in plastic wrap.
11) Expose to an X-ray film in a dark room for 3 min. Develop the film as usual. The condition for exposure and development may vary.

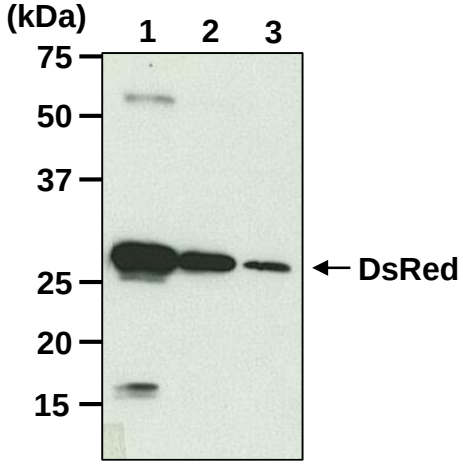


Western blotting analysis of RFP variants

Lane 1: DsRed
Lane 2: mRFP1*
Lane 3: mCherry*
Lane 4: mOrange*
Lane 5: mPlumn*

Immunoblotted with Anti-RFP mAb (M204-3)

*Samples were provided by RIKEN.



Western blotting analysis of DsRed recombinant protein

Lane 1: 10 ng/lane
Lane 2: 2 ng/lane
Lane 3: 0.4 ng/lane

Immunoblotted with Anti-RFP mAb (M204-3)