

Anti-RFP (RABBIT) Antibody Min X Hu Ms and Rt Serum Proteins - 600-401-379

Code: 600-401-379

Size: 100 µg

Product Description: Anti-RFP (RABBIT) Antibody Min X Hu Ms and Rt Serum Proteins - 600-401-379

Concentration: 1.1mg/ml by UV absorbance at 280 nm

PhysicalState: Liquid (sterile filtered)

| | |
|-------------------------------|--|
| Label | Unconjugated |
| Host | Rabbit |
| Gene Name | DsRed |
| Buffer | 0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2 |
| Stabilizer | None |
| Preservative | 0.01% (w/v) Sodium Azide |
| 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. |
| Synonyms | rabbit anti-RFP antibody, DsRed, rDsRed, Discosoma sp. Red Fluorescent Protein, Red fluorescent protein drFP583 |
| Application Note | Polyclonal anti-RFP is designed to detect RFP and its variants. This antibody can be used to detect RFP by ELISA (sandwich or capture) for the direct binding of antigen. Biotin conjugated polyclonal anti-RFP used in a sandwich ELISA with unconjugated anti-RFP is well suited to titrate RFP in solution. The detection antibody conjugated to biotin is subsequently reacted with streptavidin conjugated HRP (code # S000-03). Fluorochrome conjugated polyclonal anti-RFP can be used to detect RFP by immunofluorescence microscopy in cell expression systems and can detect RFP containing inserts. Significant amplification of signal is achieved using fluorochrome conjugated polyclonal anti-RFP relative to the fluorescence of RFP alone. For immunoblotting use either alkaline phosphatase or peroxidase conjugated polyclonal anti-RFP to detect RFP or RFP containing proteins on western blots. Optimal titers for applications should be determined by the researcher. |
| Background | Fluorescent proteins such as Discosoma Red Fluorescent Protein (DsRed) from sea anemone Discosoma sp. mushroom or green fluorescent protein (GFP) from Aequorea victoria jellyfish are widely used in research practice. Fusion RFP and GFP commonly serve as marker for gene expression and protein localization. As DsRed and GFP share only 19% identity, therefore, in general, anti-GFP antibodies do not recognize DsRed protein and vice versa. Structurally, Discosoma red fluorescent protein is similar to Aequorea green fluorescent protein in terms of its overall fold (a -can) and chromophore-formation chemistry. However, Discosoma red fluorescent protein undergoes an additional step in the chromophore maturation and obligates tetrameric structure. Rockland offers many controls, monoclonal, and polyclonal antibodies for RFP. |
| Purity And Specificity | This product was prepared from monospecific antiserum by immunoaffinity chromatography using Red Fluorescent Protein (Discosoma) coupled to agarose beads followed by solid phase adsorption(s) to remove any unwanted reactivities. Expect reactivity against RFP and its variants: mCherry, tdTomato, mBanana, mOrange, mPlum, mOrange and mStrawberry. Assay by immunoelectrophoresis resulted in a single precipitin arc against anti-Rabbit Serum and purified and partially purified Red Fluorescent Protein (Discosoma). No reaction was observed against Human, Mouse or Rat serum proteins. |
| Assay Dilutions | User Optimized |
| ELISA | 1:20,000 - 1:50,000 |
| Western Blot | 1:1,000 - 1:5,000 |
| Immunohistochemistry | 1:200 - 1:2,000 |
| IF Microscopy | 1:200-1:2,000 |
| Other Assays | User Optimized |
| Expiration | Expiration date is one (1) year from date of opening. |
| Immunogen | The immunogen is a Red Fluorescent Protein (RFP) fusion protein corresponding to the full length amino acid sequence (234aa) derived from the mushroom polyp coral Discosoma. |
| General Reference | Gross LA, Baird GS, Hoffman RC, Baldrige KK, Tsien RY: The structure of the chromophore within DsRed, a red fluorescent protein from coral. Proc. Natl. Acad. Sci. USA 2000, 97: 11990-11995. |

Specific Reference

Takeda N, Jain R, Leboeuf MR, Padmanabhan A, Wang Q, Li L, Lu MM, Millar SE, Epstein JA.. (2013) Hopx expression defines a subset of multipotent hair follicle stem cells and a progenitor population primed to give rise to K6+ niche cells. *Development*. 2013 Apr;140(8):1655-64.

Ortega F, Gascón S, Masserdotti G, Deshpande A, Simon C, Fischer J, Dimou L, Chichung Lie D, Schroeder T, Berninger B. (2013) Oligodendroglial and neurogenic adult subependymal zone neural stem cells constitute distinct lineages and exhibit differential responsiveness to Wnt signalling. *Nat Cell Biol*. 2013 Jun;15(6):602-13. doi: 10.1038/ncb2736. Epub 2013 May 5.

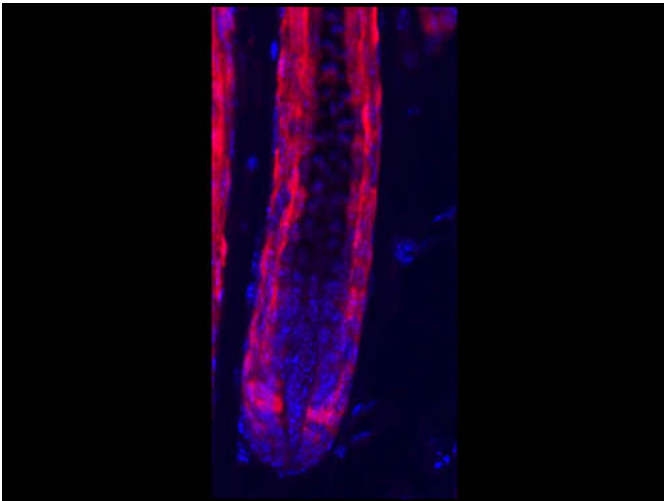
Tian X, Hu T, He L, Zhang H, Huang X, Poelmann RE, Liu W, Yang Z, Yan Y, Pu WT, Zhou B. (2013) Peritruncal coronary endothelial cells contribute to proximal coronary artery stems and their aortic orifices in the mouse heart. *PLoS One*. 2013 Nov 21;8(11):e80857. doi: 10.1371/journal.pone.0080857. eCollection 2013.

Related Products

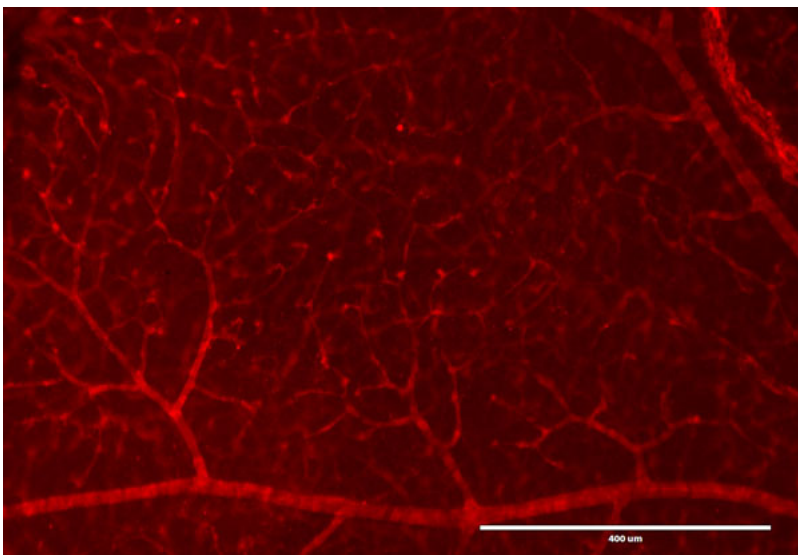
| | |
|-------------|---|
| 000-001-379 | Recombinant Red Fluorescent Protein (RFP) Control - 000-001-379 |
| 200-301-268 | Anti-AKT pS473 (MOUSE) Monoclonal Antibody - 200-301-268 |
| 600-301-215 | Anti-GFP Antibody (Mouse) Monoclonal Antibody - 600-301-215 |
| 600-401-215 | Anti-GFP (RABBIT) Antibody - 600-401-215 |

Images

- 1 Immunofluorescence Microscopy of Rabbit Anti-RFP antibody. Tissue: HopERCre/+; R26Tom/+ mice. Fixation: 0.5% PFA. Antigen retrieval: Tamoxifen. Primary antibody: RFP antibody at 10 µg/mL for 1 h at RT. Secondary antibody: Fluorescein rabbit secondary antibody at 1:10,000 for 45 min at RT. Localization: RFP is nuclear and occasionally cytoplasmic. Staining: Hop-derived cells in the hair follicle, labeled in red.

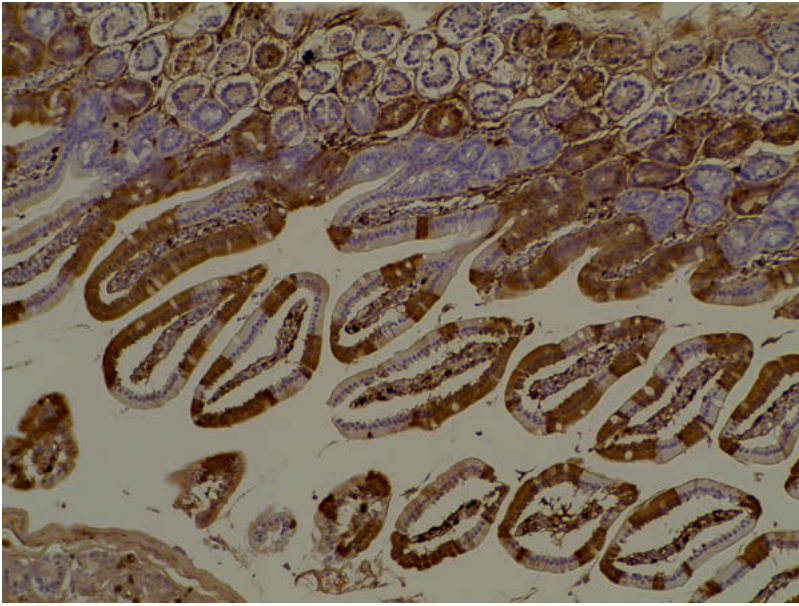


- 2 Immunofluorescence Microscopy of Rabbit Anti-RFP antibody. Tissue: DsRed transgenic mouse retina. Fixation: 4% PFA. Blocking: 3% BSA, 0.3% Triton. Primary antibody: RFP antibody at 1:100 for 12 h at 4°C. Secondary antibody: Alexa488 secondary antibody at 1:10,000 for 4 hours at RT. Localization: RFP is nuclear and occasionally cytoplasmic. Staining: labeled in red.



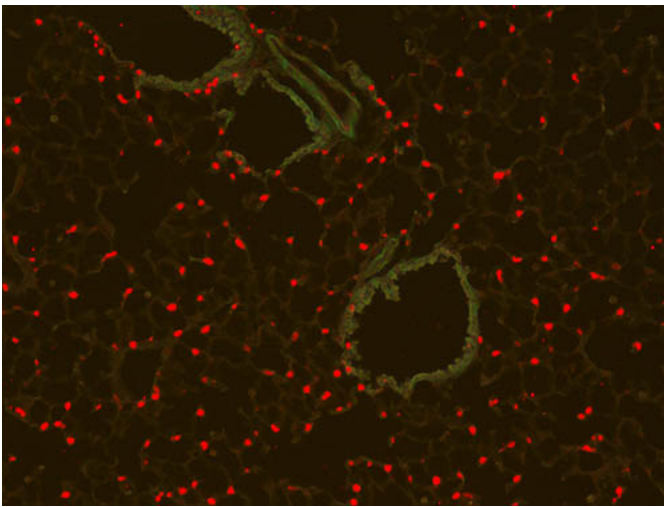
3

Immunohistochemistry of Anti-RFP Antibody. Tissue: Mouse gut tissue in tomato transgenic mice. Fixation: formalin fixed paraffin embedded. Antigen retrieval: heat 5 min at high temp in 1X rodent decloaker. Primary antibody: RFP antibody at 1:200 for 1 h 30 min at RT. Secondary Antibody: HRP anti-rabbit (p/n 611-103-122). Image courtesy: Kwan Hyun Kim.



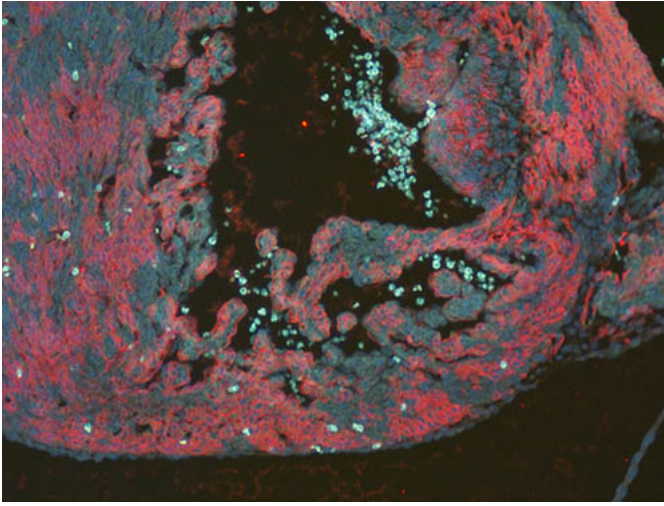
4

Immunofluorescence Microscopy of Rabbit Anti-RFP antibody. Tissue: (10X) Mouse lung tissue. Fixation: 4% PFA. Antigen retrieval: Heat. Primary antibody: Anti-RFP antibody at 1:50 for 1 h at RT. Secondary antibody: Fluorescein rabbit secondary antibody at 1:250 for 1 hr at RT. Staining: SPC+ cells are RFP positive in red.



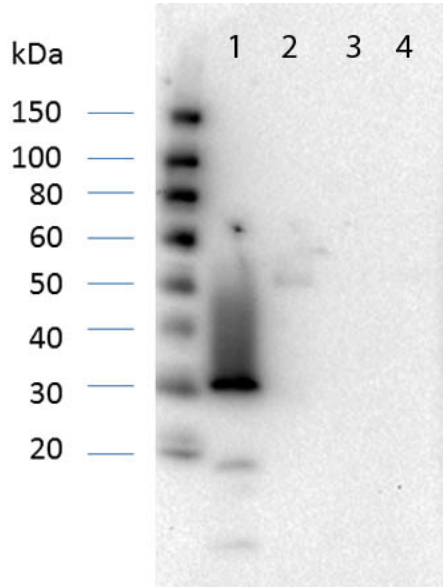
5

Immunofluorescence Microscopy of Rabbit Anti-RFP antibody. Tissue: (10X) Mouse E14.5 embryo heart tissue. Fixation: 4% PFA. Antigen retrieval: Heat. Primary antibody: Anti-RFP antibody at 1:50 for 1 h at RT. Secondary antibody: Fluorescein rabbit secondary antibody at 1:250 for 1 hr at RT. Staining: cardiac cells are RFP positive in red in tomato transgenic mice.



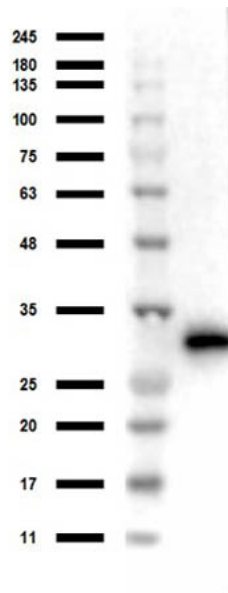
6

Western Blot of RFP Antibody Pre-Absorbed. Lane 1: RFP. Lane 2: Human IgG. Lane 3: Goat IgG. Lane 4: Mouse IgG. Load: 50ng per lane. Primary antibody: RFP Antibody Pre-Absorbed at 1:1,000 overnight at 4°C. Secondary antibody: Peroxidase conjugated rabbit secondary antibody at 1:40,000 for 30 min at RT. Block: MB-070 Blocking Buffer for 30 min at RT. Predicted/Observed size: 27 kDa, 30 kDa.



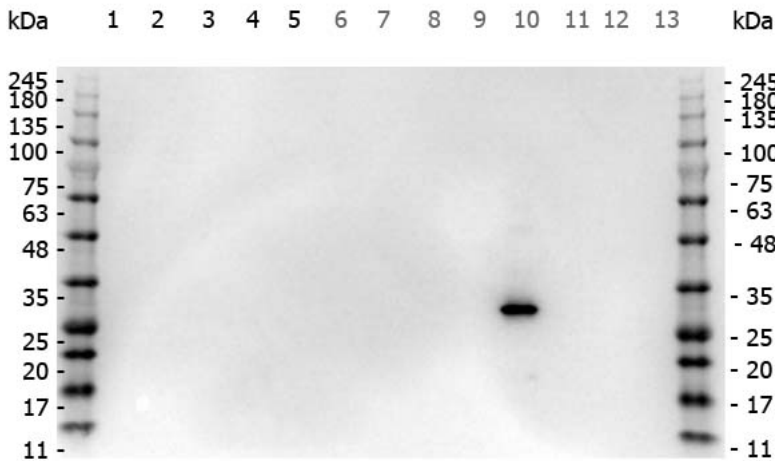
7

Western Blot of Rabbit anti-RFP Antibody. Lane 1: Opal Prestained Marker (p/n MB-210-0500). Lane 2: 50ng of RFP. Primary Antibody: Anti-RFP at 1µg/mL overnight at 2-8°C. Secondary Antibody: Goat anti-Rabbit HRP (p/n 611-103-122) at 1:70,000 for 30mins at RT. Block: BlockOut Universal blocking buffer (p/n MB-073). Expect ~27kDa.



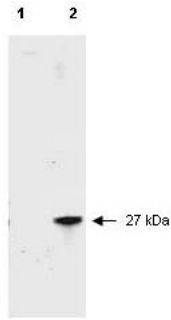
8

Western Blot of Rabbit anti-RFP antibody. Marker: Opal Pre-stained ladder (p/n MB-210-0500). Lane 1: HEK293 lysate (p/n W09-000-365). Lane 2: HeLa Lysate (p/n W09-000-363). Lane 3: CHO/K1 Lysate (p/n W07-000-357). Lane 4: MDA-MB-231 (p/n W09-001-GK6). Lane 5: A431 Lysate (p/n W09-000-361). Lane 6: Jurkat Lysate (p/n W09-001-370). Lane 7: NIH/3T3 Lysate (p/n W10-000-358). Lane 8: E-coli HCP Control (p/n 000-001-J08). Lane 9: FLAG Positive Control Lysate (p/n W00-001-383). Lane 10: Red Fluorescent Protein (p/n 000-001-379). Lane 11: Green Fluorescent Protein (p/n 000-001-215). Lane 12: Glutathione-S-Transferase Protein. Lane 13: Maltose Binding Protein (p/n 000-001-385). Load: 10 µg of lysate or 50ng of purified protein per lane. Primary antibody: RFP antibody at 1µg/mL overnight at 4C. Secondary antibody: Peroxidase rabbit secondary antibody (p/n 611-103-122) at 1:30,000 for 60 min at RT. Blocking Buffer: 1% Casein-TTBS for 30 min at RT. Predicted/Observed size: 30 kDa for RFP.



9

Western blot of RFP recombinant protein detected with Rockland's polyclonal anti-RFP antibody. Lane 1 shows no reaction against a GFP recombinant protein present in 10 µg of HeLa cell extract. Lane 2 shows a single band detected in 10 µg of a HeLa lysate containing RFP recombinant protein as a 27 kDa band. A 4-12% Bis-Tris gradient gel (Invitrogen) was used for SDS-PAGE. The membrane was blocked and then probed with Anti-RFP diluted 1:2,500 for 1 h at RT followed by washes and reaction with a 1:5,000 dilution of IRDye™800 conjugated Goat-a-Rabbit IgG [H&L] MX (611-132-122). 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.



Disclaimer

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