

Rabbit anti-Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 633

Product Details	
Size	1 mg
Species Reactivity	Mouse
Host/Isotype	Rabbit / IgG
Class	Polyclonal
Type	Secondary Antibody
Conjugate	Alexa Fluor™ 633
Excitation/Emission Max	631/650 nm
Immunogen	Gamma Immunoglobins Heavy and Light chains
Form	Liquid
Concentration	2 mg/mL
Purification	purified
Storage buffer	PBS, pH 7.5
Contains	5mM sodium azide
Storage conditions	4°C, store in dark
RRID	AB_2535727

Applications	Tested Dilution	Publications
Western Blot (WB)	1:10,000	-
Immunohistochemistry (IHC)	1-10 µg/mL	-
Immunohistochemistry (Paraffin) (IHC (P))	-	0 Publication
Immunohistochemistry (PFA fixed) (IHC (PFA))	-	0 Publication
Immunocytochemistry (ICC/IF)	4 µg/mL	0 Publication
Flow Cytometry (Flow)	1-10 µg/mL	-

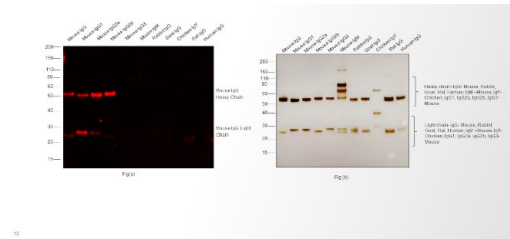
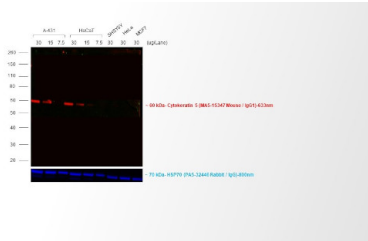
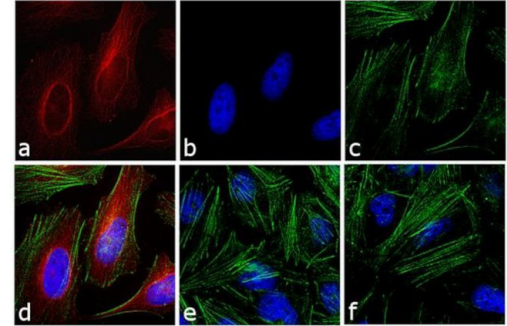
Product Specific Information

Product will be shipped at Room Temperature.

Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody (A-21063) in ICC/IF
Immunofluorescence analysis of Rabbit anti-Mouse IgG (H+L) Secondary Antibody, Alexa Fluor 633 (Product # A-21063) was performed using HeLa cells stained with alpha Tubulin (236-10501) Mouse Monoclonal Antibody (Product # A11126). The cells were fixed with 4% paraformaldehyde for 10 minutes, permeabilized with 0.1% Triton™ X-100 for 10 minutes, blocked with 1% BSA for 1 hour and labeled with mouse primary antibody (1:250 dilution) for 3 hours at room temperature. Rabbit anti-Mouse IgG (H+L) Secondary Antibody, Alexa Fluor 633 was used at concentration of 4µg/mL in phosphate buffered saline containing 0.2 % BSA for 45 minutes at room temperature, for detection of alpha Tubulin in the cytoplasm (Panel a: green). Nuclei (Panel b: blue) were stained with DAPI in SlowFade® Gold Antifade Mountant (Product # S36938). F-actin was stained with Alexa Fluor® 488 Phalloidin (Product # A12379, 1:300) (Panel c: red). Panel d represents the composite image. No nonspecific staining was observed with the secondary antibody alone (panel f), or with an isotype control (panel e). The images were captured at 60X magnification.

Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody (A-21063) in WB
Multiplexed fluorescent western blot was performed using Rabbit anti-Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 633 (Product # A-21063). Whole cell extracts of A-431 (Lane 1, 2, 3), HaCaT (Lane 4, 5,6), SH-SY5Y (Lane 7), HeLa (Lane 8) and MCF7 (Lane 9) were electrophoresed usingNuPAGE™ 4-12% Bis-Tris Protein Gel (Product # NP03221BOX). Resolved proteins were transferred onto anitrocellulose membrane (Product # IB23001) byiBlot® 2 Dry BlottingSystem (Product # IB21001). The blot was probed with Cytokeratin 5 Monoclonal Antibody (3E2F1) (Product # MA5-15347), and HSP70 Polyclonal Antibody (Product # PA5-32446). Secondary antibodies (Product # A-21063, 1:10,000 dilution), and (Product # SA5-35571, 1:10000 dilution) were used for detection of Cytokeratin 5, and HSP70 respectively. Fluorescent detection was performed usingiBrightFL1500 (Product # A44115). The anti-mouse secondary antibody (Product # A-21063) specifically detects the mouse primary antibody.

Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody (A-21063) in WB
Western blot was performed using Rabbit anti-Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 633 (Product # A-21063) and ~55, 25 kDa bands corresponding to Mouse IgG Heavy Chain and Light chain respectively were observed in Mouse IgG, Mouse IgG1, Mouse IgG2a, Mouse IgG2b but not in Mouse IgG3, Mouse IgM, Rabbit IgG, Goat IgG, Chicken IgY, Rat IgG and Human IgG. Purified protein (100 ng) of Mouse IgG (Lane 1), Mouse IgG1 (Lane 2), Mouse IgG2a (Lane 3), Mouse IgG2b (Lane 4), Mouse IgG3 (Lane 5), Mouse IgM (Lane 6), Rabbit IgG (Lane 7), Goat IgG (Lane 8), Chicken IgY (Lane 9), Rat IgG (Lane 10), Human IgG (Lane 11) were electrophoresed usingNuPAGE™ 4-12% Bis-Tris Protein Gel (Product # NP0322BOX). Resolved proteins were then transferred onto a nitrocellulose membrane(Product # IB23001) byiBlot® 2 Dry Blotting System (Product # IB21001). The blot was probed with Rabbit anti-Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 633 (Product # A-21063, 1:5000 dilution) and detected using theiBrightFL1500 (Product # A44115). Silver staining was performed to establish equivalent loading of purified proteins using the Pierce™ Silver Stain Kit (Product # 24612) (Fig b).



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27 References

Single-cell sequencing of tumor-associated macrophages in a *Drosophila* model. *Front Immunol* (2023)

Multi-level Fgf4- and apoptosis-dependent regulatory mechanism ensures the plasticity of ESC-chimaeric mouse embryo. *Development* (2023)

TIM-3 increases the abundance of type-2 dendritic cells during *Leishmania donovani* infection by enhancing IL-10 production via STAT3. *Cell Death Dis* (2023)

Stress-inducible phosphoprotein 1 (HOP/STI1/STIP1) regulates the accumulation and toxicity of - synuclein in vivo. *Acta Neuropathol* (2022)

Paracrine interactions through FGFR1 and FGFR2 receptors regulate the development of preimplantation mouse chimaeric embryo. *Open Biol* (2022)

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