Rabbit anti-Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 488

Product Details	
Size	1 mg
Species Reactivity	Mouse
Host/Isotype	Rabbit / IgG
Class	Polyclonal
Type	Secondary Antibody
Conjugate	Alexa Fluor™ 488
Excitation/Emission Max	499/520 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_2534106

Applications	Tested Dilution	Publications
Western Blot (WB)	1:2,500-1:5,000	-
Immunohistochemistry (IHC)	1-10 µg/mL	0 Publication
Immunohistochemistry (Paraffin) (IHC (P))	-	0 Publication
Immunohistochemistry (Frozen) (IHC (F))	-	0 Publication
Immunocytochemistry (ICC/IF)	1:2,000	0 Publication
Flow Cytometry (Flow)	1:100-1:500	0 Publication
Miscellaneous PubMed (Misc)	-	0 Publication

Product Specific Information

To minimize cross-reactivity, these rabbit anti-mouse IgG (H+L) whole secondary antibodies have been affinity purified and cross-adsorbed against human serum prior to conjugation. Cross-adsorption or pre-adsorption is a purification step to increase specificity of the antibody resulting in higher sensitivity and less background staining. The secondary antibody solution is passed through a column matrix containing immobilized serum proteins from potentially cross-reactive species. Only the nonspecific-binding secondary antibodies are captured in the column, and the highly specific secondaries flow through. The benefits of this extra step are apparent in multiplexing/multicolor-staining experiments (e.g., flow cytometry) where there is potential cross-reactivity with other primary antibodies or in tissue/cell fluorescent staining experiments where there are may be the presence of endogenous immunoglobulins.

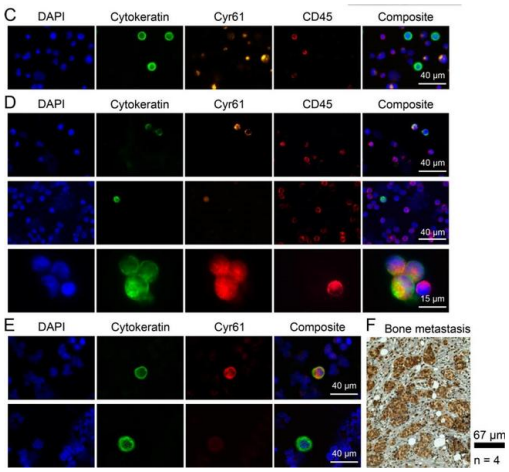
Alexa Fluor dyes are among the most trusted fluorescent dyes available today. Invitrogen™ Alexa Fluor 488 dye is a bright, green-fluorescent dye with excitation ideally suited to the 488 nm laser line. For stable signal generation in imaging and flow cytometry, Alexa Fluor 488 dye is pH-insensitive over a wide molar range. Probes with high fluorescence quantum yield and high photostability allow detection of low-abundance biological structures with great sensitivity. Alexa Fluor 488 dye molecules can be attached to proteins at high molar ratios without significant self-quenching, enabling brighter conjugates and more

sensitive detection. The degree of labeling for each conjugate is typically 2-8 fluorophore molecules per IgG molecule; the exact degree of labeling is indicated on the certificate of analysis for each product lot.

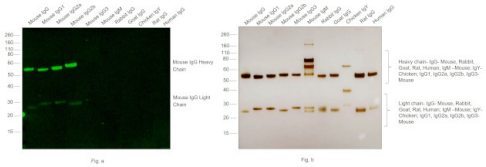
Using conjugate solutions: Centrifuge the protein conjugate solution briefly in a microcentrifuge before use; add only the supernatant to the experiment. This step will help eliminate any protein aggregates that may have formed during storage, thereby reducing nonspecific background staining. Because staining protocols vary with application, the appropriate dilution of antibody should be determined empirically. For the fluorophore-labeled antibodies a final concentration of 1-10 µg/mL should be satisfactory for most immunohistochemistry and 1:100-1:500 for flow cytometry applications.

Product will be shipped at Room Temperature.

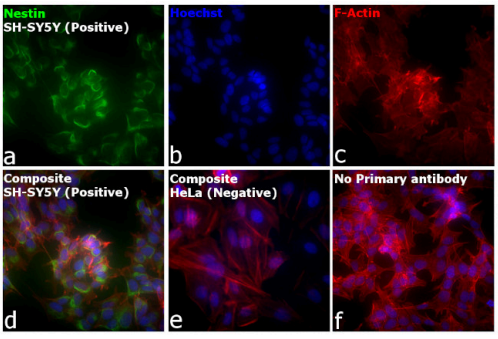
Product Images For Rabbit anti-Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 488



Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody (A-11059) in ICC/IF
Cyr61 detection in breast cancer cells. (A) Comparison of the cytoplasmic Cyr61 levels with cytokeratin levels, as analysed by the pan-cytokeratin antibody cocktails A45/BB3 and AE1/AE3 by Western blot analysis. (B) A comparison of the Cyr61 levels in the peripheral blood mononuclear cells (PBMC) of healthy women with the Cyr61 levels in breast cancer cell lines. (C) Cyr61 detection in BC-M1 and MDA-MB-468 spiked into blood samples from healthy women by immunocytochemical double staining. (D) Cyr61 detection in CTC from the peripheral blood of breast cancer patients (details: Table 1). (E) Detection of Cyr61 in the DTC from the bone marrow of breast cancer patients. The upper row shows a Cyr61-positive DTC, and the bottom row shows a Cyr61-negative DTC. (F) An immunohistochemical Cyr61 detection in the bone metastases of breast cancer patients. (C-E) The composite images are overlays of the Cytokeratin, Cyr61, Dapi and CD45 (if applied) signals, nbiol: 3 (A,C,E), 4 (F). Image collected and cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/33540545), licensed under a CC BY license.



Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody (A-11059)
Specificity of secondary antibody was demonstrated by specific detection of the target immunoglobulin. Antibody specificity was demonstrated by specific detection of Mouse IgG. Band at ~55 and 25 kDa corresponding to Mouse IgG Heavy and Light Chain were observed in Mouse IgG but not in other species using Rabbit anti-Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 488 (Product # A-11059) in Western Blot. {RE}



Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody (A-11059) in ICC/IF
Immunofluorescence analysis of Rabbit anti-Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 488 (Product # A-11059) was performed using SH-SY5Y (positive model) and HeLa (negative model) cells stained with Nestin Monoclonal Antibody (10C2), eBioscience™ (Product # 14-9843-80). The cells were fixed with 4% paraformaldehyde for 10 minutes, permeabilized with 0.1% Triton™ X-100 for 15 minutes, blocked with 2% BSA for 1 hour and labeled with 1:500 dilution of primary antibody overnight at 4C. Rabbit anti-Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 488 (Product # A-11059, 1:2000 dilution) in 0.1% BSA in PBS for 1 hour at room temperature, was used for detection of Nestin in the cytoskeleton (Panel a: green). Nuclei (Panel b: blue) were stained with Hoechst33342 (Product # H1399). F-actin was stained with Rhodamine Phalloidin (Product # R415, 1:500) (Panel c: red). Panel d represents the composite image. The specificity of the secondary antibody was proved by the absence of signal in HeLa (negative model for Nestin) due to no primary antibody binding (Panel e). Non-specific staining was not observed with secondary antibody alone (panel f). The images were captured at 40X magnification in CellInsight CX7 LZR High-Content Screening (HCS) Platform (Product # CX7A1110LZR).

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294 References

Acute and persistent effects of oral glutamine supplementation on growth, cellular proliferation, and tight junction protein transcript abundance in jejunal tissue of low and normal birthweight pre-weaning piglets. PLoS One (2024)

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Britanin inhibits titanium wear particleinduced osteolysis and osteoclastogenesis. Mol Med Rep (2023)