Goat anti-Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor 647

Product Details	
Size	1 mg
Species	Mouse
Published Species	Mouse
Expression System	Goat / IgG
Class	Polyclonal
Type	Secondary Antibody
Conjugate	Alexa Fluor® 647
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_2535805

Applications	Tested Dilution	Publications
Flow Cytometry (Flow)	1-10 µg/mL	3 Publications
Immunocytochemistry (ICC)	2 µg/mL	13 Publications
Immunofluorescence (IF)	2 µg/mL	-
Immunohistochemistry (IHC)	1-10 µg/mL	9 Publications
Immunohistochemistry (Paraffin) (IHC (P))	-	2 Publications
Miscellaneous PubMed (Misc)	-	44 Publications
Western Blot (WB)	-	1 Publication

Product Specific Information

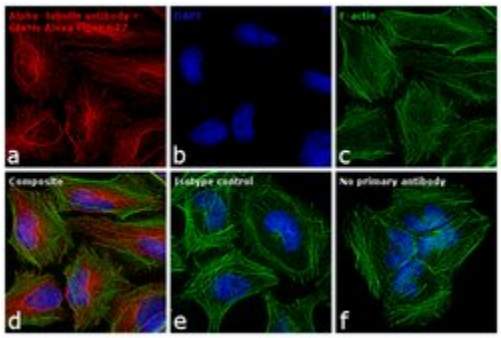
To minimize cross-reactivity, these goat anti-mouse IgG (H+L) whole secondary antibodies have been affinity purified and cross-adsorbed against bovine IgG, goat IgG, rabbit IgG, rat IgG, human IgG, and human serum. Cross-adsorption or pre-adsorption is a purification step to increase specificity of the 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 647 dye is a near-infrared-

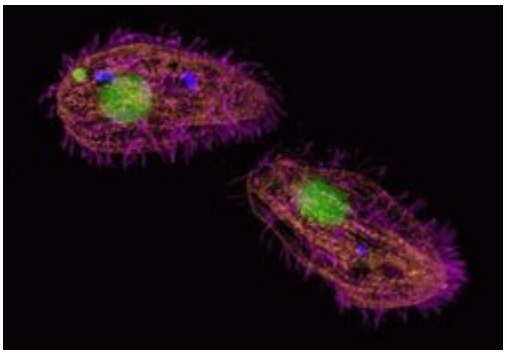
fluorescent dye with excitation ideally suited to the 647 nm laser line. For stable signal generation in imaging and flow cytometry, Alexa Fluor 647 dye is pH-insensitive over a wide molar range. Probes with high fluorescence quantum yield and high photostability allow detection of low-abundance biological structures with great sensitivity. Alexa Fluor 647 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 flow cytometry applications.

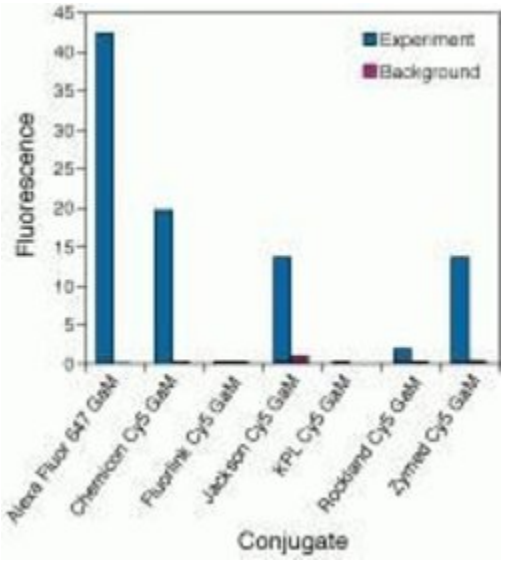
Product Images For Goat anti-Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor 647



Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody (A-21236) in IF
Immunofluorescence analysis of Goat anti-Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor® 647 conjugate 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 2 µg/mL primary antibody for 3 hours at room temperature. Goat anti-Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor® 647 (Product # A-21236) was used at a concentration of 2 µg/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: red). 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: green). 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) Highly Cross-Adsorbed Secondary Antibody (A-21236) in IF
Tetrahymena pyriformis were cultured with EdU (Product # A10044), Click-iT® GalNAz glycoprotein labeling reagent (Product # C33365), and InSpeck™ Blue (350 /440) Intensity Calibration microspheres (I7221). Following fix/perm using Image-iT™ Fixation/Permeabilization kit (Product # R37602), EdU-incorporated DNA was labeled with Alexa Fluor® 488 azide (Product # A10266) and GalNAz-incorporated cellular components with Alexa Fluor® 555 alkyne (Product # A20013). Cilia were labeled with anti-beta-tubulin Ab (Product # 32-2600) and Alexa Fluor® 647 secondary Ab (Product # A-21236). Imaging followed mounting in SlowFade® Gold (S36937).



Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody (A-21236)
Brightness of Alexa Fluor® 647 Goat Anti-Mouse IgG antibody compared with Cy5 goat anti-mouse IgG antibodies commercially available from other companies.

72 References

Immunocytochemistry (13)

Frontiers in cellular neuroscience

Loss of Neurofascin-186 Disrupts Alignment of AnkyrinG Relative to Its Binding Partners in the Axon Initial Segment.

"A-21236 was used in Immunocytochemistry-immunoflourescence to establish that the regulation of AnkG and sodium channel accumulation within the axon initial segment can happen independently to each other so may be mediated by other binding partners."

Authors: Alpizar SA,Baker AL,Gulledge AT,Hoppa MB

Species
Mouse
Not Applicable

Dilution
1:1000
1:1000

Year
2020

PLoS pathogens

HCV and flaviviruses hijack cellular mechanisms for nuclear STAT2 degradation: Up-regulation of PDLIM2 suppresses the innate immune response.

"A-21236 was used in Immunocytochemistry-immunoflourescence to investigate the paradoxical induction of an innate immune response by HCV despite a multitude of mechanisms combating the host response."

Authors: Joyce MA,Berry-Wynne KM,Dos Santos T,Addison WR,McFarlane N,Hobman T,Tyrrell DL

Species
Mouse
Not Applicable

Dilution
Not Cited
Not Cited

Year
2019

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Miscellaneous PubMed (44)

Frontiers in cellular neuroscience

Loss of Neurofascin-186 Disrupts Alignment of AnkyrinG Relative to Its Binding Partners in the Axon Initial Segment.

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Authors: Alpizar SA,Baker AL,Gulledge AT,Hoppa MB

Species
Mouse
Not Applicable

Dilution
1:1000
1:1000

Year
2020

Frontiers in neuroscience

A 3D Printed Device for Low Cost Neural Stimulation in Mice.

"A-21236 was used in Immunohistochemistry to createa reliable, low cost, 3D printed device for Neutral Stimulation in Mice."

Authors: Morrison TJ,Sefton E,Marquez-Chin M,Popovic MR,Morshead CM,Naguib HE

Species
Not Applicable

Dilution
1:500

Year
2020

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IHC (9) IHC (P) (2) WB (1) Flow (3)