invitrogen



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

<b>Product Details</b>		
Size	1 mg	
Species Reactivity	Mouse	
Host/Isotype	Goat / IgG	
Class	Polyclonal	
Туре	Secondary Antibody	
Conjugate	Alexa Fluor™ Plus 647	
Excitation/Emission Max	658/675 nm	
Immunogen	Gamma Immunoglobins Heavy and Light chains	
Form	Liquid	
Concentration	2 mg/mL	
Purification	Affinity chromatography	
Storage buffer	proprietary buffer, pH 6.5	
Contains	0.016% Methylisothiazolone, 0.016% Bromonitrodioxane	
Storage conditions	4° C, store in dark	
RRID	AB_2633277	

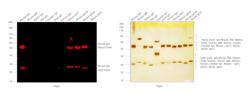
Applications	Tested Dilution	Publications
Western Blot (WB)	0.05-0.2 μg/mL	-
Immunohistochemistry (IHC)	-	0 Publication
Immunohistochemistry (Frozen) (IHC (F))	-	0 Publication
Immunocytochemistry (ICC/IF)	1-10 μg/mL	0 Publication
Miscellaneous PubMed (Misc)	-	0 Publication

### **Product Specific Information**

To minimize cross-reactivity, the goat anti-mouse IgG whole antibodies have been pre cross-adsorbed against bovine IgG, goat 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 less background staining and cross-reactivity. 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. Further passages through additional columns result in highly cross-adsorbed preparations of secondary antibody. The benefits of these extra steps are apparent in multiplexing/multicolor-staining experiments where there is potential cross-reactivity with other primary antibodies or in tissue/cell fluorescent staining experiments where there may be the presence of endogenous immunoglobulins.

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.

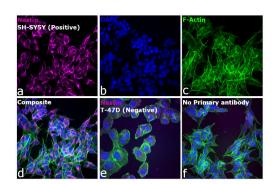
## Product Images For Goat anti-Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor™ Plus 647



Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody (A32728) Specificity of secondary antibody was demonstrated by specific detection of the target immunoglobulin. Antibody specificity was demonstrated by specific detection of Mouse IgG. A band at ~50 and 25 kDa corresponding to Mouse IgG Heavy and Light Chain was observed in Mouse IgG, IgG1, IgG2a, IgG2b but not in other species using Goat anti-Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor Plus 647 (Product # A32728) in Western Blot. {RE}

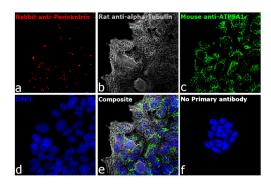
## Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody (A32728) in ICC/IF

Immunofluorescence analysis of Goat anti-Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor Plus 647 (Product # A32728) was performed using SH-SY5Y (positive model) and T-47D (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 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 Plus 647 (Product # A32728, 1:2000 dilution) in 0.1% BSA in PBS for 45 minutes at room temperature, was used for detection of Nestin in the cytoskeleton (Panel a: Pink). Nuclei (Panel b: blue) were stained with Hoechst33342 (Product # H1399). F-actin was stained with Alexa Fluor® 488 Phalloidin (Product # A12379, 1:300) (Panel c: green). Panel d represents the composite image. The specificity of the secondary antibody was proved by the absence of signal in T-47D (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) and externally deconvoluted (D.Sage et al./Methods 115 (2017) 28-41).



# Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody (A32728) in ICC/IF

Immunofluorescence analysis of A32731, A-11081 and A32728 was performed using primary antibodies against Pericentrin (Product # PA5-53498), alpha Tubulin (Product # MA1-80017) and ATP5A1 (Product # 43-9800) in 70% confluent log phase HEK 293 cells. The cells were fixed with 4% Paraformaldehyde, permeabilized with 0.1% Triton X-100 and blocked with 2% BSA, then incubated with the primary antibodies at 1:100 dilution each at 4 degree celsius overnight. The cells were then incubated with Goat anti-Rabbit IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor™ Plus 488 (Product # A32731), Goat anti-Rat IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 546 (Product # A-11081) and Goat anti-Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor™ Plus 647 (Product # A32728) at 1:2000 dilution each in 0.1% BSA at room temperature for 45 minutes. The images were captured at 40X magnification in CellInsight CX7 LZR High-Content Screening (HCS) Platform (Product # CX7A1110LZR) and externally deconvoluted (D.Sage et al./Methods 115 (2017) 28-41). The specific centrosomal, cytoskeletal and mitochondrial localization of Pericentrin (Panel a), Tubulin (Panel b) and ATP5A1 (Panel c) in the respective channels alone shows the specificity of all the 3 secondary antibodies used. Nuclei (Panel d) were stained with Hoechst33342 (Product # H1399). Panel e is the composite of Panels a-d, showing co-localisation. Panel f is control cells with



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### **□ 511 References**

The Seneca Valley virus 3C protease cleaves DCP1A to attenuate its antiviral effects. Vet Res (2025)

Structural insights into how DEK nucleosome binding facilitates H3K27 trimethylation in chromatin. Nat Struct Mol Biol (2025)

Blebbisomes are large, organelle-rich extracellular vesicles with cell-like properties. Nat Cell Biol (2025)

The integrated stress response engages a cell-autonomous, ligand-independent, DR5-driven apoptosis switch. Cell Death Dis (2025)

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