Goat anti-Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 555

Product Details	
Size	1 mg
Species Reactivity	Mouse
Host/Isotype	Goat / IgG
Class	Polyclonal
Type	Secondary Antibody
Conjugate	Alexa Fluor™ 555
Excitation/Emission Max	553/568 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_2535844

Applications	Tested Dilution	Publications
Western Blot (WB)	-	0 Publication
Immunohistochemistry (IHC)	1-10 µg/mL	0 Publication
Immunohistochemistry (Paraffin) (IHC (P))	-	0 Publication
Immunohistochemistry (Frozen) (IHC (F))	-	0 Publication
Immunohistochemistry - Free Floating (IHC (Free))	-	0 Publication
Immunocytochemistry (ICC/IF)	2 µg/mL	0 Publication
Flow Cytometry (Flow)	1-10 µg/mL	0 Publication
Miscellaneous PubMed (Misc)	-	0 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 human IgG and 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.

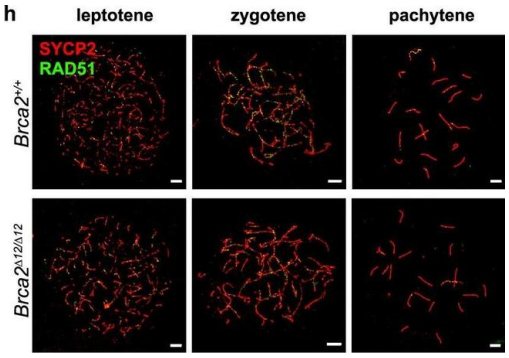
Alexa Fluor dyes are among the most trusted fluorescent dyes available today. Invitrogen™ Alexa Fluor 555 dye is a bright, orange-fluorescent dye with excitation ideally suited to the 555 nm laser line. For stable signal generation in imaging and flow

cytometry, Alexa Fluor 555 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 555 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.

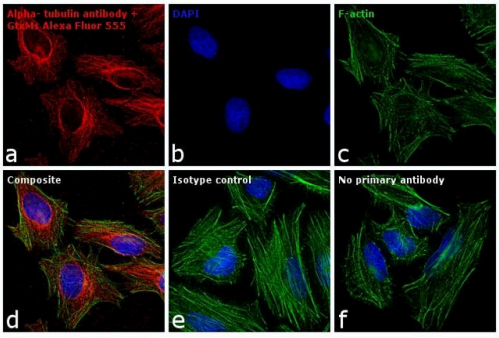
Using conjugate solutions: Centrifuge the protein conjugate solution briefly in a microcentrifuge before use; add only the supernatant to the experiment. This step will help eliminate any protein aggregates that may have formed during storage, thereby reducing nonspecific background staining. Because 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 will be shipped at Room Temperature.

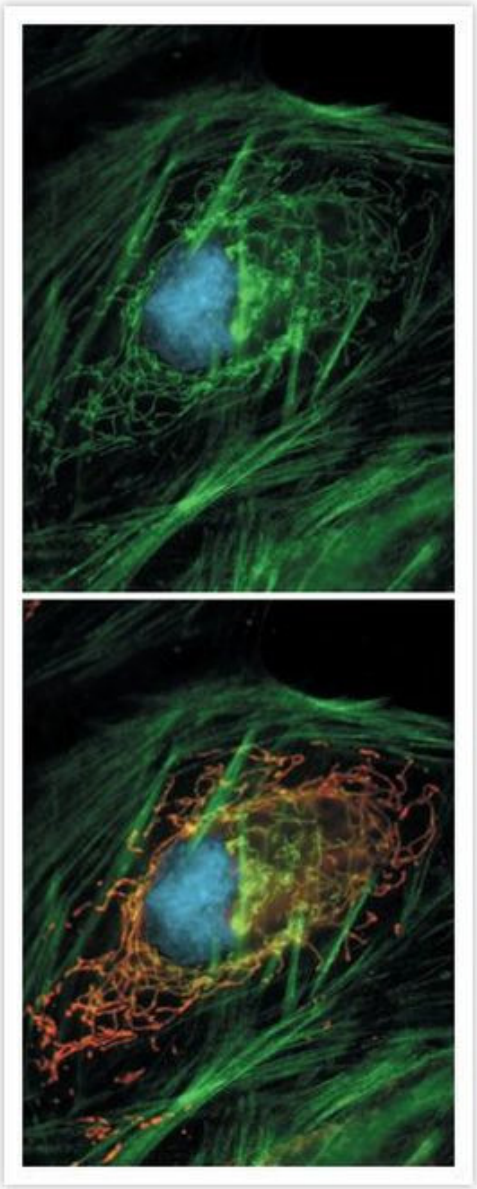
Product Images For Goat anti-Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 555



Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody (A-21422) in ICC/IF
Meiotic phenotype of Brca2 exon 12 deletion mouse model.a Schematic depiction of the domain composition of the BRCA2 protein and the exons 11-15 encoding HSF2BP-binding and DMC1-binding domains. Introns are not drawn to scale; different exon phases are indicated by the shape of the boundary. Location of Cas9 cut sites for exon 12 and exons 12-14 excision is shown. b RT-PCR on cDNA from mouse testis with indicated genotypes confirming the loss of exon 12; primer locations are shown on the exon scheme in a. The PCR was performed twice with the same results. c Immunoblot analysis of proteins precipitated from Brca2+/+ and Brca212/12 mouse testes using anti-HSF2BP antibody and, as control, with anti-RAD51 antibodies; and the input samples; performed as described in “Methods” section. The experiment was performed four times with similar results. d testis weight, e bodyweight, f sperm count, and g epididymis weight in Brca212/12 and control mice. n = 5 animals for Brca2+/+, n = 6 for Brca212/12, n = 3 (testis weight and bodyweight), and n = 2 (epididymis weight and sperm count) for Brca212/+. Mean, s.e.m, and p values from one-way ANOVA with Tukey test are indicated. h-n Immunofluorescent analysis of meiotic protein localization on spermatocyte spreads from Brca212/12 and control mice. Representative images (h, l; scale bars = 5 µm) and quantificatio… Image collected and cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/34326328), licensed under a CC BY license.



Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody (A-21422) in ICC/IF
Immunofluorescence analysis of Goat anti-Mouse IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor® 555 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) Cross-Adsorbed Secondary Antibody, Alexa Fluor® 555 (Product # A-21422) 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) Cross-Adsorbed Secondary Antibody (A-21422) in ICC/IF
HeLa cell transfected with pShooter pCMV/myc/mito/GFP, then fixed and permeabilized. Green-fluorescent protein (GFP) localized in the mitochondria was labeled with anti-GFP mouse IgG_{2a} (Product # A-11120) and detected with orange-fluorescent Alexa Fluor® 555 goat anti-mouse IgG (Product # A-21422), which colocalized with the dim GFP fluorescence. F-actin was labeled with green-fluorescent Alexa Fluor® 488 phalloidin (Product # A12379), and the nucleus was stained with blue-fluorescent DAPI (Product # D1306, D3571, D21490). The sample was mounted using ProLong® Gold antifade reagent (Product # P36930). Some GFP fluorescence is retained in the mitochondria after fixation (top), but immunolabeling and detection greatly improve visualization (bottom).

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1077 References

Astrocyte-derived lactoferrin reduces -amyloid burden by promoting the interaction between p38 kinase and PP2A phosphatase in male APP/PS1 transgenic mice. Br J Pharmacol (2024)

Sox9 regulates alternative splicing and pancreatic beta cell function. Nat Commun (2024)

Disease-associated nonsense and frame-shift variants resulting in the truncation of the GluN2A or GluN2B C-terminal domain decrease NMDAR surface expression and reduce potentiating effects of neurosteroids. Cell Mol Life Sci (2024)

TRPP2 is located in the primary cilia of human non-pigmented ciliary epithelial cells. Graefes Arch Clin Exp Ophthalmol (2024)

PNLDC1 catalysis and postnatal germline function are required for piRNA trimming, LINE1 silencing, and spermatogenesis in mice bioRxiv (2023)