Goat anti-Rabbit IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 594

Product Details	
Size	1 mg
Species Reactivity	Rabbit
Host/Isotype	Goat / IgG
Class	Polyclonal
Type	Secondary Antibody
Conjugate	Alexa Fluor™ 594
Excitation/Emission Max	590/618 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_2534079

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

Product Specific Information

To minimize cross-reactivity, these goat anti-rabbit IgG whole antibodies have been cross-adsorbed against human IgG, human serum, mouse IgG, mouse serum, and bovine 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.

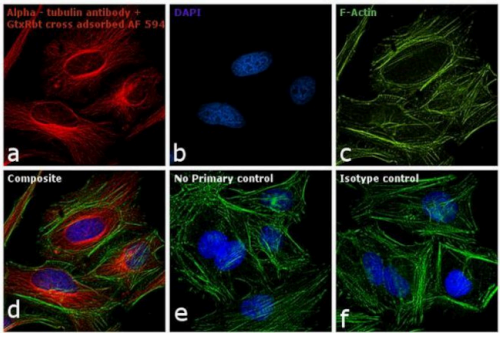
Alexa Fluor dyes are among the most trusted fluorescent dyes available today. Invitrogen™ Alexa Fluor 594 dye is a bright, red-fluorescent dye with excitation ideally suited to the 594 nm laser line. For stable signal generation in imaging and flow cytometry, Alexa Fluor 594 dye is pH-insensitive over 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 594 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.

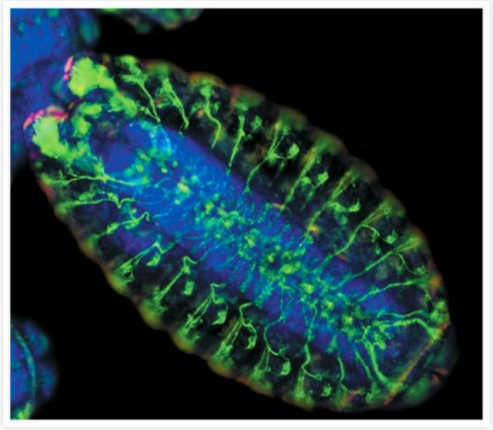
Using conjugate solutions: Centrifuge the protein conjugate solution briefly in a microcentrifuge before use; add only the supernatant to the experiment. This step will help eliminate any protein aggregates that may have formed during storage, thereby reducing nonspecific background staining. Because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

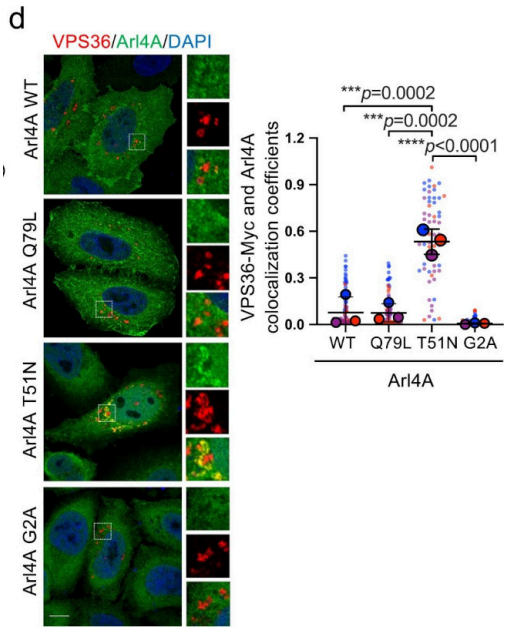
Product Images For Goat anti-Rabbit IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 594



Rabbit IgG (H+L) Cross-Adsorbed Secondary Antibody (A-11012) in ICC/IF
Immunofluorescence analysis of Goat anti-Rabbit IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor 594 (Product # A-11012) was performed using HeLa cells stained with alpha Tubulin Rabbit Polyclonal Antibody (Product # PA5-16891). 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 of rabbit primary antibody for 3 hours at room temperature. Goat anti-Rabbit IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor 594 (A-11012) 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.



Rabbit IgG (H+L) Cross-Adsorbed Secondary Antibody (A-11012) in ICC/IF
The peripheral nervous system of a wild-type (Canton-S) Drosophila melanogaster embryo labeled with the monoclonal 22c10 antibody (which detects a microtubule-associated protein) and subsequently visualized using green-fluorescent Alexa Fluor® 488 Rabbit Anti-Mouse IgG antibody (Product # A-11059). The actively dividing cells of the developing denticle bands were labeled with a rabbit anti-histone-H3 antibody and visualized using red-fluorescent Alexa Fluor® 594 Goat Anti-Rabbit IgG antibody (Product # A-11012). Finally, the nuclei, which are concentrated in the central nervous system, were counterstained with blue-fluorescent DAPI (Product # D1306, D3571, D21490). Image contributed by Neville Cobbe, University of Edinburgh.



Rabbit IgG (H+L) Cross-Adsorbed Secondary Antibody (A-11012) in ICC/IF
Arl4A interacts with VPS36.a The yeast reporter strain L40 was transformed with a construct encoding Gal4 transcriptional activation domain-fused VPS36 (330-386) and the indicated wild-type and mutant constructs of Arl4A, Arl4C, and Arl4D fused to the LexA DNA-binding domain. Colonies were patched on selective plates and assayed for -galactosidase activity. Lamin was included as a negative control. b Equal amounts of His-tagged Arl4A WT, Q79L, T51N, and Arl1 (negative control) were pulled down by either GST or GST-VPS36 (201-386). Inputs and pulled-downed Arl4 proteins were probed using an anti-His antibody. Equal inputs of GST fusion proteins used in the assay were visualized by Coomassie Blue staining as shown at the bottom of each figure. c VPS36 interacted with Arl4AWT, Arl4AQ79L, and Arl4AT51N in vivo. Lysates of HeLa cells transfected with the indicated plasmids were immunoprecipitated with HA beads, and the bound proteins were analyzed by Western blotting with anti-HA, anti-Arl4A, or anti--tubulin antibodies. The amounts of coimmunoprecipitated Arl4A were determined by densitometric quantification. The results represent the mean ± SD of six independent experiments and the p-values were assessed by one-way ANOVA with Tukey's test. d HeLa cells were cotransfected with VPS36 WT-Myc and Arl4A WT, Q79L, T51N, or G2A (myristoylation-defective) - Image collected and cropped by CiteAb under a CC-BY … Image collected and cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/38030597), licensed under a CC BY license.

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3285 References

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