

# Goat anti-Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 568

Product Details	
Size	1 mg
Species Reactivity	Mouse
Host/Isotype	Goat / IgG
Class	Polyclonal
Type	Secondary Antibody
Conjugate	Alexa Fluor™ 568
Excitation/Emission Max	579/603 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_144696

Applications	Tested Dilution	Publications
Western Blot (WB)	-	0 Publication
Immunohistochemistry (IHC)	-	0 Publication
Immunohistochemistry (Paraffin) (IHC (P))	-	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-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 may be the presence of endogenous immunoglobulins.

Alexa Fluor dyes are among the most trusted fluorescent dyes available today. Invitrogen™ Alexa Fluor 568 dye is a bright, orange/red-fluorescent dye with excitation ideally suited to the 568 nm laser line. For stable signal generation in imaging and flow cytometry, Alexa Fluor 568 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 568 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.

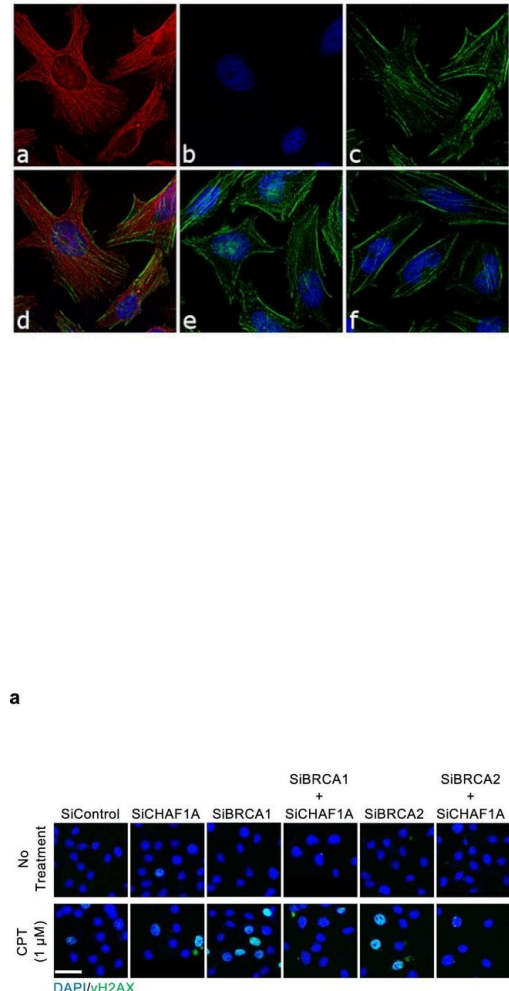
**Product Images For Goat anti-Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 568**

**Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody (A-11031) in ICC/IF**

Immunofluorescence analysis of Goat anti-Mouse IgG (H+L) Secondary Antibody, Alexa Fluor 568 conjugate (Product # A11031) 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 mouse primary antibody (1:250 dilution) for 3 hours at room temperature. Goat anti-Mouse IgG (H+L) Secondary Antibody, Alexa Fluor 568 conjugate was used at 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.

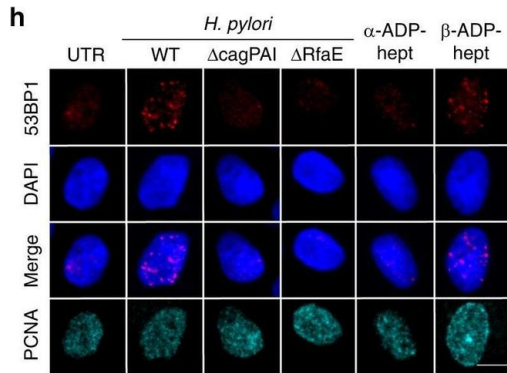
**Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody (A-11031) in ICC/IF**

Loss of CAF-1 suppresses genomic instability in BRCA-deficient cells. a, b H2AX immunofluorescence experiment showing that CHAF1A depletion suppresses CPT-induced DNA damage accumulation in BRCA1 or BRCA2-depleted cells. HeLa cells were treated with 1 µM CPT for 1 h followed by media removal and chase in fresh media for 3 h. Representative micrographs (scale bar represents 50 µm) (a) and quantifications (b) are shown. At least 50 cells were quantified for each condition. The mean values are represented on the graph, and the p-values (t-test, two-tailed, unpaired) are listed at the top. Western blots confirming the co-depletions are shown in Supplementary Fig. 2a. c-e Neutral comet assay showing that CHAF1A depletion suppresses CPT-induced (c, d) and cisplatin-induced (e) DSB formation in BRCA1 or BRCA2-depleted cells. HeLa cells were treated with 100 nM CPT for 4 h or 2.5 µM cisplatin for 24 h. Representative micrographs (c) and quantifications (d, e) are shown. At least 60 nuclei were quantified for each condition. The mean values are represented on the graph, and the p-values (t-test, two-tailed, unpaired) are listed at the top. f, g Clonogenic survival experiments showing that CHAF1A co-depletion in BRCA2-knockdown (f) or BRCA1-knockdown (g) HeLa cells promotes cisplatin resistance. The average of three experiments, with standard deviations indicated as error bars, is shown. Asterisks indicate sta... Image collected and cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/36085347>), licensed under a CC BY license.



## Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody (A-11031) in ICC/IF

The bifunctional RfaE enzyme is required, and its product -ADP-heptose is sufficient for the induction of DNA DSBs. a–c AGS cells were infected for 6 h with *H. pylori* P12 or its isogenic RfaE and Cag-PAI mutants at an MOI of 50 and subjected to immunofluorescence staining for 53BP1 and PCNA as well as DAPI. Representative images are shown in a alongside scatter dot plots of >730 and up to 1655 cells per condition in b. IL-8 secretion as assessed by ELISA is shown in c. Data in b and c are pooled from four independent experiments. Red lines indicate medians. Data in c are represented as mean  $\pm$  SEM of four independent experiments. Scale bar in a, 10  $\mu$ m. d, e AGS cells were infected for 6 h as described above and subjected to PFGE. A representative gel is shown in d alongside the quantification of three gels (independent experiments) in e. Fragmented DNA was normalized to the intact DNA retained in the slot, and to the uninfected control condition, which was set as 100%. Data are presented as mean values  $\pm$  SEM. f, g Wild-type (CTRL), ALPK1-deficient (ALPK1), and TIFA-deficient (TIFA) AGS cells were exposed to - or -ADP-heptose at 0.5 M final concentration for 6 h and subjected to immunofluorescence staining for 53BP1 and PCNA as well as DAPI. Scatter dot plots of >579 and up to 706 cells per condition are shown in f, with red lines indicating medians. IL-8 secretion as assessed ... Image collected and cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/33037203>), licensed under a CC BY license.



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## 1525 References

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