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

Product Details	
Size	1 mg
Species Reactivity	Rabbit
Host/Isotype	Donkey / 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_141637

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))	1:500	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
in situ PLA (PLA)	-	0 Publication
Miscellaneous PubMed (Misc)	-	0 Publication

Product Specific Information

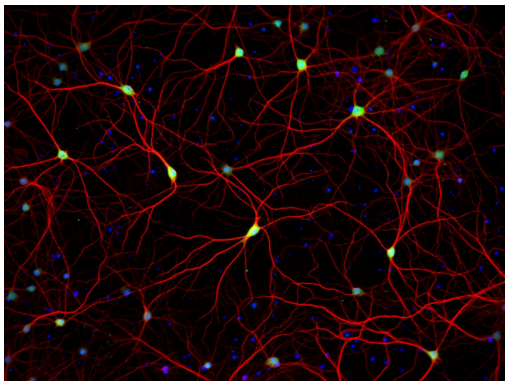
To minimize cross-reactivity, these donkey anti-rabbit IgG whole antibodies have been affinity-purified and show minimum cross-reactivity to bovine, chicken, goat, guinea pig, hamster, horse, human, mouse, rat, and sheep serum proteins. Cross-adsorption or pre-adsorption is a purification step to increase 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 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.

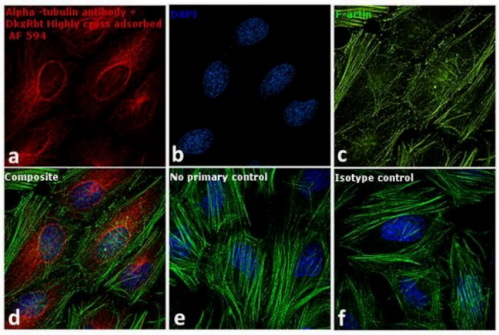
Product will be shipped at Room Temperature.

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



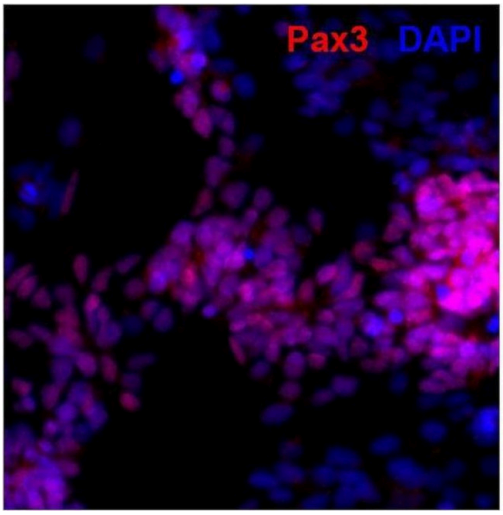
Rabbit IgG (H+L) Highly Cross-Adsorbed Secondary Antibody (A-21207) in ICC/IF

Immunofluorescent analysis of HuC/D (green) and MAP2 (red) on rat primary cortical neurons cultured for 28 days in the B-27 Plus Neuronal Culture System (Product # A3653401). At day 28 the cells were fixed with 4% paraformaldehyde for 15 min, permeabilized with 0.1% triton x-100 for 30min, and blocked with 1% BSA for 30 min at room temperature. Cells were stained with anti-HuC/D antibody (Product # A-21271) at a dilution of 1:250, and anti-MAP2 (Product # PA5-17646) at a dilution of 1:250, in 1% BSA staining buffer, overnight at 4C, and then incubated with Alexa Fluor 488 conjugated donkey anti-mouse (Product # A-21202) and Alexa Fluor 594 donkey anti-rabbit (Product # A-21207) antibodies at a dilution of 1:1000 for 30 min. at room temp. Wash 3 times with DPBS. Stain with DAPI for nucleus. Images were taken on a Thermo Fisher Scientific EVOS M5000 Cell Imaging System at 10x magnification.



Rabbit IgG (H+L) Highly Cross-Adsorbed Secondary Antibody (A-21207) in ICC/IF

Immunofluorescence analysis of Donkey anti-Rabbit IgG (H+L) Highly Cross-Adsorbed Secondary antibody Alexa Fluor® 594 (Product # A-21207)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. Donkey anti-Rabbit IgG (H+L) Highly Cross-Adsorbed Secondary antibody Alexa Fluor® 594 (Product # A-21207) 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 (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) Highly Cross-Adsorbed Secondary Antibody (A-21207) in ICC/IF

Immunofluorescent analysis of Pax3 (red) in human neural stem cells derived from PD-3 iPSCs using Gibco® PSC Neural Induction Medium (Product # A1647801). The cells were fixed and permeabilized using Image-IT® Fixation /Permeabilization kit (Product # R37602), and blocked with blocking buffer included the kit for one hour at room temperature. Cells were stained with a Pax3 polyclonal antibody (Product # PA1-107) at a dilution of 1:200 in blocking buffer for 3 hours at room temperature, and then incubated with an AlexaFluor 594-conjugated donkey anti-rabbit IgG secondary antibody (Product # A-21207) at a dilution of 1:250 for 1 hour at room temperature. Nuclei (blue) were stained with DAPI (Product # D1306). Images were taken on an EVOS® FLoid® Cell Imaging Station at 10X magnification.

View more figures on thermofisher.cn

2832 References

Silencing hepatic PCSK9 via novel chimeric AAV8 mitigates the progression of atherosclerosis by inhibiting inflammation in ApoE-/- mice. Mol Ther Methods Clin Dev (2025)

Extracellular vesicles efficiently deliver survival motor neuron protein to cells in culture. Sci Rep (2025)

Saussurea costus alleviates ulcerative colitis by regulating the gut microbiota and improving intestinal barrier integrity. Front Cell Infect Microbiol (2025)

Cellular senescence induced by cholesterol accumulation is mediated by lysosomal ABCA1 in APOE4 and AD. Mol Neurodegener (2025)

Genome-wide gene expression profiles throughout human malaria parasite liver stage development in humanized mice. Nat Microbiol (2025)