

# Donkey anti-Sheep IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 647

Product Details	
Size	1 mg
Species Reactivity	Sheep
Host/Isotype	Donkey / IgG
Class	Polyclonal
Type	Secondary Antibody
Conjugate	Alexa Fluor™ 647
Excitation/Emission Max	650/671 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_2535865

Applications	Tested Dilution	Publications
Western Blot (WB)	1:5000	-
Immunohistochemistry (IHC)	1-10 µg/mL	0 Publication
Immunocytochemistry (ICC/IF)	1-10 µg/mL	0 Publication
Miscellaneous PubMed (Misc)	-	0 Publication

## Product Specific Information

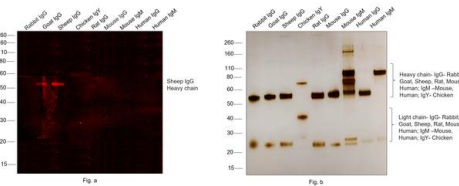
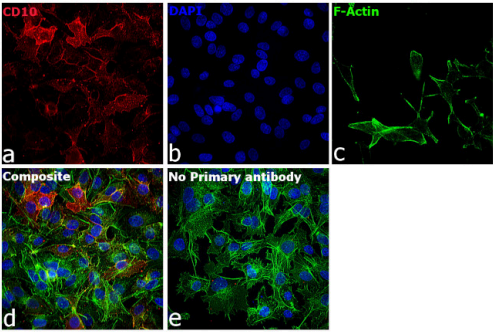
Fluorescence of this long-wavelength Alexa Fluor dye is not visible by looking through a conventional fluorescence microscope.

Product will be shipped at Room Temperature.

**Sheep IgG (H+L) Cross-Adsorbed Secondary Antibody (A-21448) in ICC/IF**  
Immunofluorescence analysis of Donkey anti-Sheep IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 647 (Product # A-21448) was performed using Hep G2 cells stained with CD49a Polyclonal Antibody (Product # PA5-47763). The cells were fixed with 4% paraformaldehyde for 10 minutes, permeabilized with 0.1% Triton™ X-100 for 10 minutes, blocked with 2% BSA for 1 hour and labeled with 1:100 dilution of primary antibody for 3 hours at room temperature. The cells were probed with Donkey anti-Sheep IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 647 (Product # A-21448, 1:2000 dilution) in 0.1% BSA in PBS for 45 minutes at room temperature and detected with Donkey anti-Sheep IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 647 (Product # A-21448). Membrane localization of CD49a was seen in Hep G2 (Panel a: Red). 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. Nonspecific staining was not observed with secondary antibody alone (panel e). 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).

**Sheep IgG (H+L) Cross-Adsorbed Secondary Antibody (A-21448) in WB**  
Fluorescent western blot was performed using Donkey anti-Sheep IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 647 (Product # A-21448). Membrane enriched extracts of HeLa (Lane 1), Hep G2 (Lane 2) and K-562 (Lane 3) were electrophoresed using NuPAGE™ 3 to 8%, Tris-Acetate, 1.0 mm, Mini Protein Gel, 10-well (Product # EC6695BOX). Resolved proteins were transferred onto nitrocellulose membrane (Product # IB23001) by iBlot® 2 Dry BlottingSystem (Product # IB21001). The blot was probed with CD49a Sheep Polyclonal Antibody (Product # PA5-47763) and GAPDH Loading Control Monoclonal Antibody (GA1R) (Product # MA5-15738). Secondary antibodies (Product # A-21448, 1:5000 dilution) and (Product # SA5-35521B, 1:30,000 dilution) were used for detection of CD49a and GAPDH respectively. Fluorescent detection was performed using iBright™ FL1500 (Product # A44115). The anti-sheep secondary antibody (Product # A-21448) specifically detects the sheep primary antibody.

**Sheep IgG (H+L) Cross-Adsorbed Secondary Antibody (A-21448) in WB**  
Western blot was performed using Donkey anti-Sheep IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 647 (Product # A-21448) and ~50 kDa band corresponding to Sheep IgG Heavy Chain was observed in Sheep IgG but not in Rabbit IgG, Rat IgG, Chicken IgY, Mouse IgG, Mouse IgM, Human IgG and Human IgM. Purified protein (100 ng) of Rabbit IgG (Lane 1), Goat IgG (Lane 2), Sheep IgG (Lane 3), Chicken IgY (Lane 4), Rat IgG (Lane 5), Mouse IgG (Lane 6), Mouse IgM (Lane 7), Human IgG (Lane 8), Human IgM (Lane 9) (Fig. a) were electrophoresed using NuPAGE™ 4-12% Bis-Tris Protein Gel (Product # NP0321BOX). Resolved proteins were then transferred onto a nitrocellulose membrane (Product # IB23001) by iBlot® 2 Dry Blotting System (Product # IB21001). The blot was probed with Donkey anti-Sheep IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 647 (Product # A-21448, 1:2000 dilution) and detected using the iBright™ FL1500 (Product # A44115). Silver staining was performed to establish equivalent loading of purified proteins using the Pierce™ Silver Stain Kit (Product # 24612) (Fig. b). The secondary antibody showed cross reactivity with Goat IgG.



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Early intervention anti-A immunotherapy attenuates microglial activation without inducing exhaustion at residual plaques. *Mol Neurodegener* (2025)

Efficient generation of germline chimeras in a non-rodent species using rabbit induced pluripotent stem cells. *Nat Commun* (2025)

Harmonizing TUNEL with multiplexed iterative immunofluorescence enriches spatial contextualization of cell death. *Cell Rep Methods* (2025)

CRISPR-Cas9 genetic screens reveal regulation of TMPRSS2 by the Elongin BC-VHL complex. *Sci Rep* (2025)

Capture, mutual inhibition and release mechanism for aPKC-Par6 and its multisite polarity substrate Lgl. *Nat Struct Mol Biol* (2025)

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