

Chicken anti-Goat IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 647

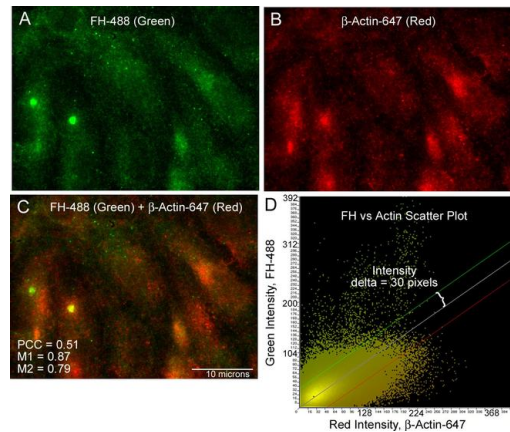
Product Details	
Size	1 mg
Species Reactivity	Goat
Host/Isotype	Chicken / IgY
Class	Polyclonal
Type	Secondary Antibody
Conjugate	Alexa Fluor™ 647
Excitation/Emission Max	650/671 nm
Immunogen	Gamma Immunoglobins Heavy and Light chains
Form	liquid
Concentration	2 mg/mL
Purification	purified
Storage buffer	PBS, pH 7.5
Contains	5mM sodium azide
Storage conditions	4° C, store in dark
RRID	AB_2535872

Applications	Tested Dilution	Publications
Western Blot (WB)	1:5000-1:10,000	-
Immunohistochemistry (IHC)	1-10 µg/mL	-
Immunohistochemistry (Paraffin) (IHC (P))	-	0 Publication
Immunocytochemistry (ICC/IF)	0.5-10 µg/mL	0 Publication
Flow Cytometry (Flow)	-	0 Publication
Miscellaneous PubMed (Misc)	-	0 Publication

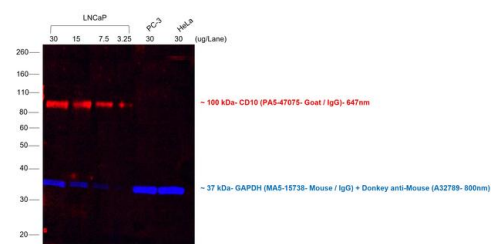
Product Specific Information

This Chicken anti-goat antibody reacts with IgG heavy chains and all classes of immunoglobulin light chains from goat. Chicken secondary antibodies have gained popularity because they demonstrate a lower level of nonspecific binding. Chicken antibodies lack a classic “Fc” domain and will not bind to protein A or protein G, nor will they bind to mammalian IgG Fc receptors. Fluorescence of this long-wavelength Alexa Fluor dye is not visible by looking through a conventional fluorescence microscope.

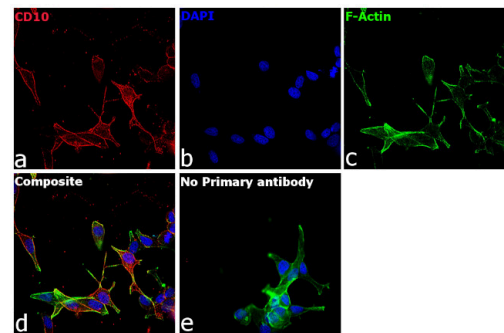
Product will be shipped at Room Temperature.



Goat IgG (H+L) Cross-Adsorbed Secondary Antibody (A-21469) in ICC/IF
Intensity scatter plots of FH with β -actin shows a colocalization in the HUVEC cytoplasm. HUVECs were formaldehyde-fixed, treated with Triton-X and stained concurrently for FH and β -actin. (A) FH was detected using 2 mouse monoclonal antibodies to human FH plus donkey anti-mouse AF IgG-488 (green). (B) β -actin was detected using polyclonal goat anti- β -actin plus chicken anti-goat antibody AF IgG-647 (red). (C) Shows the merged image detecting both FH (green) and β -actin (red) and the calculated values for the Pearson's (PCC) and Manders' (M1 and M2) correlation coefficients. (D) The intensity scatter plot of the merged image in (C) shows a single linear correlation indicative of a signal overlap. Image collected and cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/25803806>), licensed under a CC BY license.



Goat IgG (H+L) Cross-Adsorbed Secondary Antibody (A-21469) in WB
Multiplexed fluorescent western blot was performed using Chicken anti-Goat IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 647 (Product # A-21469). Membrane enriched extracts of LNCap (Lane 1, 2, 3, 4), PC-3 (Lane 5) and HeLa (Lane 6) were electrophoresed using NuPAGE™ 4-12% Bis-Tris Protein Gel (Product # NP0321BOX). Resolved proteins were transferred onto nitrocellulose membrane (Product # IB23001) by iBlot® 2 Dry Blotting System (Product # IB21001). The blot was probed with CD10 Polyclonal Antibody (Product # PA5-47075) and GAPDH Loading Control Monoclonal Antibody (GA1R) (Product # MA5-15738). Secondary antibodies (Product # A-21469, 1:10,000 dilution) and (Product # A32789, 1:20,000 dilution) were used for detection of CD10 and GAPDH respectively. Fluorescent detection was performed using iBright™ FL1500 (Product # A44115). The anti-goat secondary antibody (Product # A-21469) specifically detects the goat primary antibody.



Goat IgG (H+L) Cross-Adsorbed Secondary Antibody (A-21469) in ICC/IF
Immunofluorescence analysis of Chicken anti-Goat IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 647 (Product # A-21469) was performed using LNCaP cells stained with CD10 Polyclonal Antibody (Product # PA5-47075). 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 Chicken anti-Goat IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 647 (Product # A-21469, 1:3000 dilution) in 0.1% BSA in PBS for 45 minutes at room temperature. Membrane localization of CD10 was seen in LNCaP (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).

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