

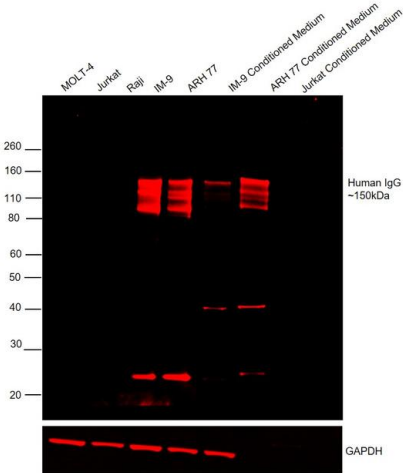
Rabbit anti-Human IgG (H+L) Cross-Adsorbed Secondary Antibody, DyLight™ 650

Product Details	
Size	500 µg
Species Reactivity	Human
Host/Isotype	Rabbit / IgG
Class	Polyclonal
Type	Secondary Antibody
Conjugate	DyLight™ 650
Excitation/Emission Max	651/673 nm
Immunogen	Human IgG-heavy and light chain
Form	Liquid
Concentration	0.5 mg/mL
Purification	Antigen affinity chromatography
Storage buffer	PBS, pH 6.8 to 7.4, with 0.2% BSA
Contains	0.09% sodium azide
Storage conditions	4° C
RRID	AB_2556693

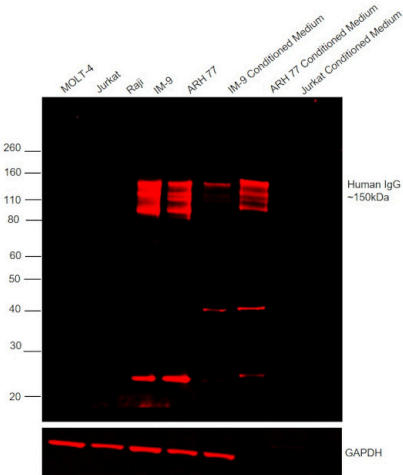
Applications	Tested Dilution	Publications
Western Blot (WB)	1:2,000-1:20,000	-
Immunohistochemistry (IHC)	1:50-1:500	-
Immunocytochemistry (ICC/IF)	1:50-1:500	-
Flow Cytometry (Flow)	1:50 - 1:200	-
Immunoprecipitation (IP)	Assay-dependent	-

Product Specific Information

This antibody is cross-adsorbed and exhibits minimum reactivity to mouse and rat.



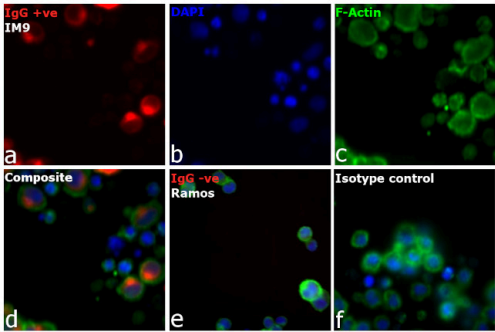
Human IgG (H+L) Cross-Adsorbed Secondary Antibody (SA5-10113) in WB
Western blot (non-reducing) was performed Rabbit anti-Human IgG (H+L) Cross-Adsorbed Secondary Antibody, DyLight 650 (Product # SA5-10113) and a 150 kDa band corresponding to Human IgG was observed in IM-9, ARH-77 and IM-9, ARH-77 conditioned medium8203 (CM) but not in Raji, Jurkat, Molt-4 and Jurkat CM which are known to have low expression. Whole cell lysate (30 µg) of MOLT-4 (Lane 1), Jurkat (Lane 2), Raji (Lane 3), IM-9 (Lane 4), ARH-77 (Lane 5), IM-9 CM (Lane 6), ARH-77 CM (Lane 7) and Jurkat CM (Lane 8) were electrophoresed using NuPAGE™ 4-12% Bis-Tris Protein Gel (Product # NP0322BOX). Resolved proteins were transferred onto a nitrocellulose membrane (Product # IB23001) by iBlot® 2 Dry Blotting System (Product # IB21001). The blot was probed with Rabbit anti-Human IgG (H+L) Cross-Adsorbed Secondary Antibody, DyLight 650 (Product # SA510113) (1:2000 dilution) and detected by fluorescence using iBright FL1500 (Product # A44115). IM-9 and ARH-77 express and secrete IgG whereas Raji is known to express IgM. MOLT-4 and Jurkat (T-cell lines) do not express immunoglobulins (DOI: 10.1002/eji.1830100305;10.3791/3573;10.1016/0022-1759(94)00286-6;PMID: 566614).



Human IgG (H+L) Cross-Adsorbed Secondary Antibody (SA5-10113)
Antibody specificity was demonstrated by detection of differential basal expression of IgG across cell lines owing to their inherent genetic constitution. Relative expression of Human IgG was observed in IM-9, ARH-77 and IM-9, ARH-77 conditioned medium (CM) but not in Raji, MOLT-4, Jurkat and Jurkat CM using Rabbit anti-Human IgG (H+L) Cross-Adsorbed Secondary Antibody, DyLight 650 (Product # SA5-10113) in Western Blot. IM-9 and ARH-77 express and secrete IgG whereas Raji is known to express IgM. MOLT-4 and Jurkat (T-cell lines) do not express immunoglobulins. (DOI:10.1002/eji.1830100305; 10.3791/3573;10.1016/0022-1759(94)00286-6;PMID: 566614). {RE}

Human IgG (H+L) Cross-Adsorbed Secondary Antibody (SA5-10113) in ICC /IF

Immunofluorescence analysis of Rabbit anti-Human IgG (H+L) Cross-Adsorbed Secondary Antibody, DyLight 650 was performed using log phase IM9 cells (IgG producing B-cell line). The cells were fixed with 4% paraformaldehyde for 10 minutes, permeabilized with 0.1% Triton™ X-100 for 15 minutes and blocked with 2% BSA for 1 hour at room temperature. The cells were labeled with Rabbit anti-Human IgG (H+L) Cross-Adsorbed Secondary Antibody, DyLight 650 (Product # SA5-10113) at 1:250 dilution in 0.1% BSA, incubated at 4 degrees Celsius overnight (Panel a: red). Nuclei (Panel b: blue) were stained with Hoechst33342 (Product # H1399). F-actin (Panel c: green) was stained with Alexa Fluor™ 488 Phalloidin (Product # A12379, 1:300 dilution). Panel d represents the merged image showing cytoplasmic (plasma membrane and golgi-body like) localization. Panel e represents Ramos (IgG non-producing B-cell line) which is a negative model for IgG expression. Panel f represents control cells with isotype control antibody to assess background. 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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