Donkey anti-Rabbit IgG (H+L) Secondary Antibody, Qdot™ 655

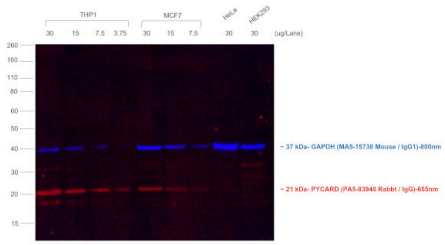
Product Details	
Size	100 µL
Species Reactivity	Rabbit
Host/Isotype	Donkey / IgG
Class	Polyclonal
Type	Secondary Antibody
Conjugate	Qdot™ 655
Excitation/Emission Max	300/654 nm
Immunogen	Gamma Immunoglobins Heavy and Light chains
Form	Liquid
Concentration	1 µM
Purification	purified
Storage buffer	0.05M borate, pH 8.3, with 1M betaine
Contains	0.05% sodium azide
Storage conditions	4° C, store in dark
RRID	AB_2556496

Applications	Tested Dilution	Publications
Western Blot (WB)	1:50-1:500	-
Immunohistochemistry (IHC)	1:50	-
Immunocytochemistry (ICC/IF)	1:50-1:500	-
Flow Cytometry (Flow)	1:50	-

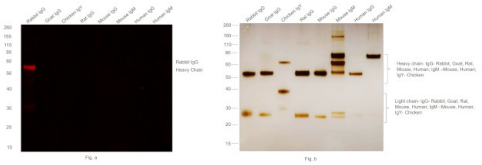
Product Specific Information

Qdot nanocrystals are composed of semi-conductor material to generate a fluorescent particle which is exceptionally bright and does not photobleach. Qdot nanocrystals paired with the correct optical filters are as much as 50 times brighter than traditional organic dyes.

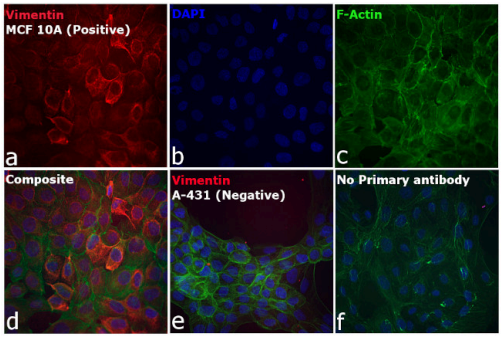
Product Images For Donkey anti-Rabbit IgG (H+L) Secondary Antibody, Qdot™ 655



Rabbit IgG (H+L) Secondary Antibody (Q22089) in WB
Multiplexed fluorescent western blot was performed using Donkey anti-Rabbit IgG (H+L) Secondary Antibody, Qdot™ 655 (Product # Q22089). Membrane enriched extracts of THP1 (Lane 1, 2, 3, 4), MCF7 (Lane 5, 6, 7), HeLa (Lane 8) and HEK293 (Lane 9) were electrophoresed usingNuPAGE™ 4-12% Bis-Tris Protein Gel (Product # NP0322BOX). Resolved proteins were transferred onto anitrocellulose membrane (Product # IB23001) byiBlot® 2 Dry BlottingSystem (Product # IB21001). The blot was probed with PYCARD Polyclonal Antibody (Product # PA5-83948), and GAPDH Loading Control Monoclonal Antibody (GA1R) (Product # MA5-15738). Secondary antibodies (Product # Q22089, 1: 500 dilution), and (Product # A32789, 1:20000 dilution) were used for detection of PYCARD and GAPDH respectively. Fluorescent detection was performed usingiBrightFL1500 (Product # A44115). The anti-rabbit secondary antibody (Product # Q22089) specifically detects the rabbit primary antibody.



Rabbit IgG (H+L) Secondary Antibody (Q22089)
Specificity of secondary antibody was demonstrated by specific detection of the target immunoglobulin. Antibody specificity was demonstrated by specific detection of Rabbit IgG. A band at ~55 kDa corresponding to Rabbit IgG Heavy Chain was observed in Rabbit IgG but not in other species using Donkey anti-Rabbit IgG (H+L) Secondary Antibody, Qdot 655 (Product # Q22089) in Western Blot. {RE}



Rabbit IgG (H+L) Secondary Antibody (Q22089) in ICC/IF
Immunofluorescence analysis of Donkey anti-Rabbit IgG (H+L) Secondary Antibody, Qdot™ 655 was performed using MCF 10A (positive model) and A-431 (negative model) cells stained with Vimentin Polyclonal antibody (Product # PA5-27231). 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 1:500 dilution of primary antibody at 4 degree celsius. Donkey anti-Rabbit IgG (H+L) Secondary Antibody, Qdot™ 655, 1:500 dilution) in 0.1% BSA in PBS for 45 minutes at room temperature, was used for detection of Vimentin in the cytoskeleton (Panel a: Red). Nuclei (Panel b: blue) were stained with Hoechst33342 (Product # H1399). F-actin was stained with Alexa Fluor® 488 Phalloidin (Product # A12379, 1:500) (Panel c: green). Panel d represents the composite image. The specificity of the secondary antibody was proved by the absence of signal in A-431 (negative model for Vimentin) due to no primary antibody binding (Panel e). Non-specific staining was not observed with secondary antibody alone (panel f). 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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1 Reference

Polygalasaponin F protects hippocampal neurons against glutamate-induced cytotoxicity. Neural Regen Res (2022)