

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

Product Details	
Size	1 mg
Species Reactivity	Rat
Host/Isotype	Rabbit / 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_2535797

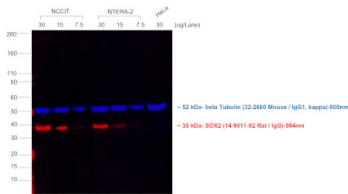
Applications	Tested Dilution	Publications
Western Blot (WB)	1:2,500-1:5,000	-
Immunohistochemistry (IHC)	1-10 µg/mL	0 Publication
Immunohistochemistry (Paraffin) (IHC (P))	-	0 Publication
Immunocytochemistry (ICC/IF)	1-4 µg/mL	-
Flow Cytometry (Flow)	1-10 µg/mL	0 Publication

## Product Specific Information

Product will be shipped at Room Temperature.

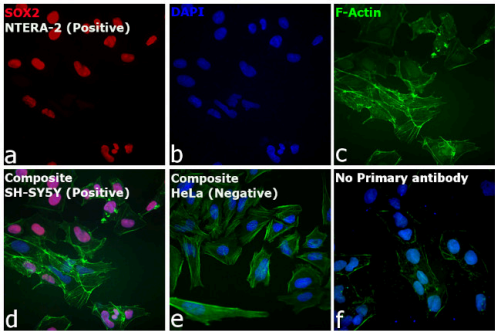
Rat IgG (H+L) Cross-Adsorbed Secondary Antibody (A-21211) in WB

Multiplexed fluorescent western blot was performed using Rabbit anti-Rat IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 594 (Product # A-21211). Whole cell extracts (1% SDS) of NCCIT (Lane 1, 2, 3), NTERA-2 (Lane 4, 5, 6) and HeLa (Lane 7) were electrophoresed using NuPAGE™ 4-12% Bis-Tris Protein Gel (Product # NP0322BOX). Resolved proteins were transferred onto nitrocellulose membrane (Product # IB23001) by iBlot® 2 Dry Blotting System (Product # IB21001). The blot was probed with SOX2 Monoclonal Antibody (Btjce), eBioscience™ (Product # 14-9811-82), and beta Tubulin Monoclonal Antibody (2 28 33) (Product # 32-2600). Secondary antibodies (Product # A-21211, 1:5000 dilution), and (Product # A32789, 1:20,000 dilution) were used for detection of SOX2 and beta tubulin respectively. Fluorescent detection was performed using iBright™ FL1500 (Product # A44115). The anti-rat secondary antibody (Product # A-21211) specifically detects the rat primary antibody.



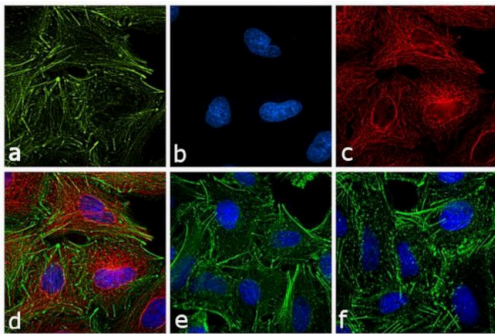
Rat IgG (H+L) Cross-Adsorbed Secondary Antibody (A-21211) in ICC/IF

Immunofluorescence analysis of Rabbit anti-Rat IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 594 (Product # A-21211) was performed using NTERA-2 (positive model) and Hela (negative model) in cells stained with SOX2 Monoclonal Antibody (Btjce), eBioscience (Product # 14-9811-82). 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 overnight at 4C. Rabbit anti-Rat IgG (H+L) Cross-Adsorbed Secondary Antibody, Alexa Fluor™ 594 (Product # A-21211, 1:2000 dilution) in 0.1% BSA in PBS for 1 hour at room temperature, was used for detection of SOX2 in the nucleus (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 HeLa (negative model for SOX2) 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).



Rat IgG (H+L) Cross-Adsorbed Secondary Antibody (A-21211) in ICC/IF

Immunofluorescence analysis of Rabbit anti-Rat IgG (H+L) Secondary Antibody, Alexa Fluor 594 conjugate was performed using A549 cells stained with alpha Tubulin (YL1/2) Rat Monoclonal Antibody (Product # MA1-80017). 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 Rat primary antibody for 3 hours at room temperature. Rabbit anti-Rat IgG (H+L) Secondary Antibody, Alexa Fluor 594 conjugate (Product # A-21211) was used at a concentration of 4 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 (Product # 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.



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## 18 References

Glucagon-Like Peptide-1 (GLP-1) Rescue Diabetic Cardiac Dysfunctions in Human iPSC-Derived Cardiomyocytes. Adv Biol (Weinh) (2023)

E3 ligase MAEA-mediated ubiquitination and degradation of PHD3 promotes glioblastoma progression. Oncogene (2023)

Serotonin distinctly controls behavioral states in restrained and freely moving Drosophila. iScience (2023)

E3 ligase MAEA-mediated ubiquitination and degradation of PHD3 promotes glioblastoma progression Research Square (2022)

Learning-related contraction of grey matter in rodent sensorimotor cortex is associated with adaptive myelination bioRxiv (2022)

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