

Datasheet for 612-143-002**Rat IgG (H&L) Antibody DyLight™ 649 Conjugated****Overview**

Description:	Goat Anti-Rat IgG (H&L) Antibody DyLight™ 649 Conjugated - 612-143-002
Item No.:	612-143-002
Size:	100 µg
Applications:	WB, IF, Multiplex
Reactivity:	Rat
Host Species:	Goat

Product Details

Background:	This product is designed for immunofluorescence microscopy, fluorescence based plate assays (FLISA) and fluorescent western blotting. This product is also suitable for multiplex analysis, including multicolor imaging, utilizing various commercial platforms.
Synonyms:	Goat Anti-Rat IgG DyLight 649™ Conjugated Antibody, Goat Anti-Rat IgG Antibody DyLight 649™ Conjugation
Host Species:	Goat
Specificity:	IgG (H&L)
Conjugate:	DyLight™ 649
Clonality:	Polyclonal
Format:	IgG
F/P Ratio:	2.9

Target Details

Reactivity:	Rat
Immunogen:	Rat IgG, whole molecule

Purity/Specificity:	This product was prepared from monospecific antiserum by immunoaffinity chromatography using Rat IgG coupled to agarose beads followed by conjugation to fluorochrome and extensive dialysis against the buffer stated above. Assay by immunoelectrophoresis resulted in a single precipitin arc against anti-Goat Serum, Rat IgG and Rat Serum. This antibody will react with heavy chains of Rat IgG and with light chains of most Rat immunoglobulins.
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Application Details

Tested Applications:	WB
Suggested Applications:	IF, Multiplex (Based on references)
Application Note:	Anti-Rat IgG DyLight™ 649 has been tested by western blot and is designed for immunofluorescence microscopy, fluorescence based plate assays (FLISA) and fluorescent western blotting. The emission spectra for this DyLight™ conjugate match the principle output wavelengths of most common fluorescence instrumentation.
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
FLISA:	>1:20,000
IF:	>1:5,000
WB:	>1:10,000

Formulation

Physical State:	Lyophilized
Concentration:	1.0 mg/mL by UV absorbance at 280 nm
Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	0.01% (w/v) Sodium Azide
Stabilizer:	10 mg/mL Bovine Serum Albumin (BSA) - Immunoglobulin and Protease free
Reconstitution Volume:	100 µL
Reconstitution Buffer:	Restore with deionized water (or equivalent)

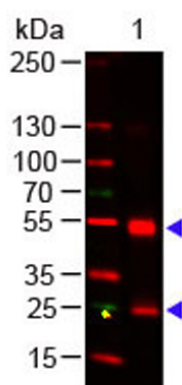
Shipping & Handling

Shipping Condition:	Ambient
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Storage Condition: Store vial at 4° C prior to restoration. For extended storage aliquot contents and freeze at -20° C or below. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use.

Expiration: Expiration date is one (1) year from date of receipt.

Images



Western Blot

Western Blot of Goat anti-Rat IgG (H&L) Antibody DyLight™ 649 Conjugated.

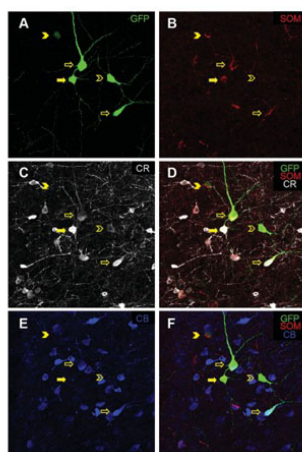
Lane 1: Rat IgG.

Load: 50 ng per lane.

Secondary antibody: Rat IgG (H&L) Antibody DyLight™ 649 Conjugated at 1:1,000 for 60 min at RT.

Block: MB-070 for 30 min at RT.

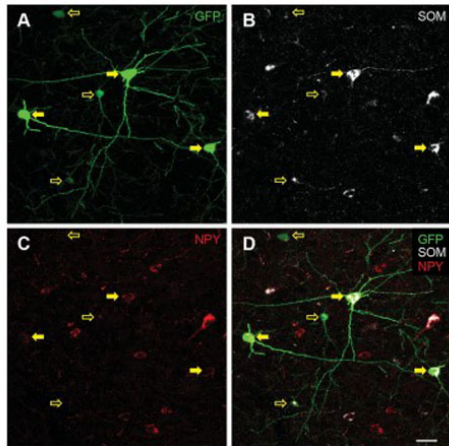
Predicted/Observed size: 55 and 28 kDa.



Immunofluorescence Microscopy

Analysis of somatostatin (SOM)-, calretinin (CR)-, and calbindin (CB)-expressing interneurons in the cingulate cortex of FVB-Tg(GadGFP)45704Swn/J mice. A–F:

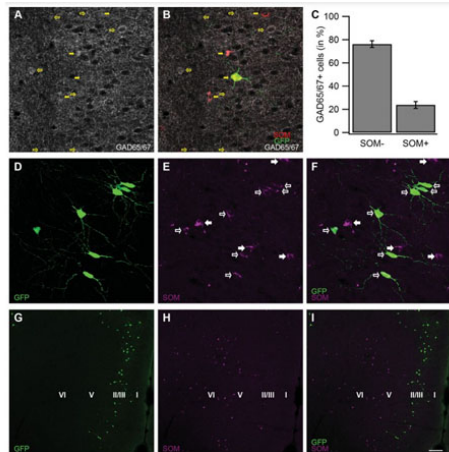
Representative confocal images of GFP 1 cells (green) immunopositive for SOM (red), CR (white), and CB (blue) in layers II–III of the cingulate cortex. Open yellow arrows indicate GFP1/SOM1/CR1/CB 1 cells, solid yellow arrows GFP1/SOM1/CR 1 cells, open yellow arrowheads GFP1/SOM2/CR 1 cells, and solid yellow arrowheads GFP1/SOM 1 cells. G: Mean 6 standard deviation of relative numbers of GFP1/SOM2/CR2/CB2 cells (a), GFP1/SOM2/CR1/CB2 cells (b), GFP1/SOM1/CR2/CB2 cells (c), GFP1/SOM1/ CR2/CB 1 cells (d), GFP1/SOM1/CR1/CB2cells (e), and GFP1/SOM1/CR1/CB 1 cells (f) in the cingulate cortex. Scale bar 5 20 lm in F (applies to A–F). Figure 7. PMID: 26669716.



Immunofluorescence Microscopy

Analysis of neuropeptide Y (NPY) expressing interneurons in the cingulate cortex of FVB-Tg(GadGFP)45704Swn/J mice.

A–D: Representative confocal images of cells immunopositive for GFP (green), somatostatin (SOM, white), and NPY (red), and merged image of all channels. Solid yellow arrows indicate GFP1/SOM1/NPY 1 cells and open yellow arrows GFP1/ SOM1/NPY2 cells. E: Mean 6 standard deviation of relative numbers of GFP1/SOM2/NPY2 cells, GFP1/SOM1/NPY2 cells, and GFP1/SOM1/NPY 1 cells in the cingulate cortex. Scale bar 5 20 lm in D (applies to A–D). Figure 10. PMID: 26669716.



Immunofluorescence Microscopy

Analysis of somatostatin-expressing interneurons in the cingulate cortex of FVB-Tg(GadGFP)45704Swn/J mice. A,B: Representative confocal images of cells immunopositive for GAD65/67 (white), SOM (red), and GFP (green). Solid yellow arrows indicate GAD65/ 671/SOM 1 cells and open yellow arrows GAD65/671/SOM2 cells. C: Mean 6 standard deviation of relative numbers of GAD65/671/ SOM2 and GAD65/671/SOM 1 cells in the cingulate cortex. D–F: Representative confocal images of GFP 1 cells (green) in the cingulate cortex immunopositive for SOM (magenta). The combined expression of GFP and SOM in the same neuron is shown in the merged image (F). Open white arrows represent GFP1/SOM 1 cells and solid white arrows SOM1/GFP- cells. G–I: Overview of the laminar distribution of SOM 1 cells in the cingulate cortex shown in representative confocal images. The laminar distribution of GFP 1 cells in the cingulate cortex is shown in G. Layer-specific distribution of SOM 1 neurons is shown in H. The merged image of both channels is shown in I. Whereas GFP 1 cells were clearly confined to layers II–III, SOM 1 cells were also found in the deeper cortical layers. Roman numbers I–VI indicate cortical layers I–VI. J: Mean 6 standard deviation of relative numbers of GFP1/SOM 1 and GFP1/SOM2 cells in the cingulate cortex. K: Mean 6 standard deviation of relative numbers of GFP-/SOM 1 and GFP1/SOM 1 cells in the cingulate cortex. Scale bar in I 5 20 lm for A,B,D–F; 100 lm for G–I. Figure 4. PMID: 26669716.

Diagram
Properties of DyLight™ Fluorescent Dyes.

Emission	Color	DyLight™ Dye	Ex/Em (nm)	ϵ (M ⁻¹ cm ⁻¹)	Similar Dyes
Blue		405	400/420	30,000	Alexa™ 405, Cascade Blue
Green		488	493/518	70,000	Alexa™ 488, Cy2®, FITC
Yellow		549	550/568	150,000	Alexa™ 546, Alexa 555, Cy3®, TRITC
Red		649	646/674	250,000	Alexa™ 647, Cy5®
Near Infrared		680	682/715	140,000	Alexa™ 680, Cy5.5®, IRDye™ 700
Infrared		800	770/794	270,000	IRDye™ 800

Western Blot
DyLight™ dyes can be used for two-color Western Blot detection with low background and high signal. Anti-tubulin was detected using a DyLight™ 549 conjugate. Anti-TNF α was detected using a DyLight™ 649 conjugate. The image was captured using the Typhoon™ 9410 Imaging System.

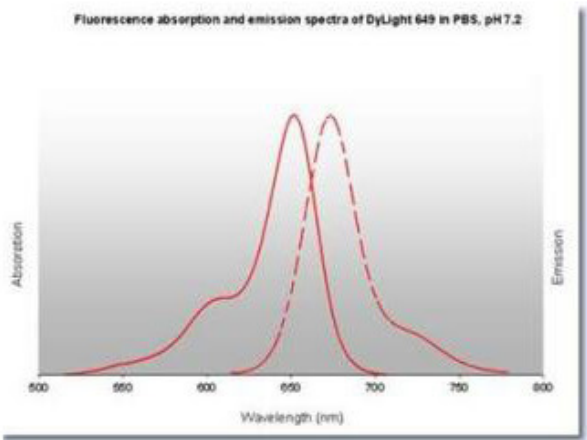
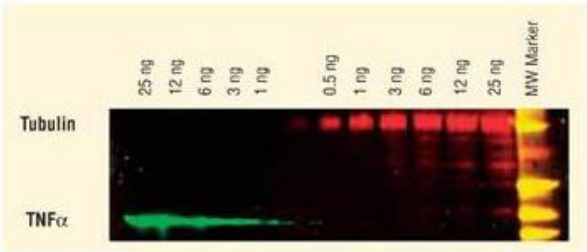


Diagram
DyLight™ 649 Fluorescence Spectra.

References

- Riedemann S et al. Gad1-promotor-driven GFP expression in non-GABAergic neurons of the nucleus endopiriformis in a transgenic mouse line. *J Comp Neurol.* (2019)
- Riedemann et al. Two types of somatostatin-expressing GABAergic interneurons in the superficial layers of the mouse cingulate cortex. *PLOS One* (2018)
- Riedemann T et al. Immunocytochemical heterogeneity of somatostatin-expressing GABAergic interneurons in layers II and III of the mouse cingulate cortex: A combined immunofluorescence/design-based stereologic study. *J Comp Neurol.* (2016)

Disclaimer

This product is for research use only and is not intended for therapeutic or diagnostic applications. Please contact a technical service representative for more information. All products of animal origin manufactured by Rockland Immunochemicals are derived from starting materials of North American origin. Collection was performed in United States Department of Agriculture (USDA) inspected facilities and all materials have been inspected and certified to be free of disease and suitable for exportation. All properties listed are typical characteristics and are not specifications. All suggestions and data are offered in good faith but without guarantee as conditions and methods of use of our products are beyond our control. All claims must be made within 30 days following the date of delivery. The prospective user must determine the suitability of our materials before adopting them on a commercial scale. Suggested uses of our products are not recommendations to use our products in violation of any patent or as a license under any patent of Rockland Immunochemicals, Inc. If you require a commercial license to use this material and do not have one, then return this material, unopened to: Rockland Inc., P.O. BOX 5199, Limerick, Pennsylvania, USA.