

Datasheet for 612-143-002-0.5

## Rat IgG (H&L) Antibody DyLight™ 649 Conjugated

### Overview

|                      |                                                                                         |
|----------------------|-----------------------------------------------------------------------------------------|
| <b>Description:</b>  | Goat Anti-Rat IgG (H&L) Antibody DyLight™ 649 Conjugated (5 X 100 µg) - 612-143-002-0.5 |
| <b>Item No.:</b>     | 612-143-002-0.5                                                                         |
| <b>Size:</b>         | 5 x 100 µg                                                                              |
| <b>Applications:</b> | IF, Multiplex                                                                           |
| <b>Reactivity:</b>   | Rat                                                                                     |
| <b>Host Species:</b> | Goat                                                                                    |

### Product Details

|                      |                                                                                                                                                                                                                                                                   |
|----------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Background:</b>   | 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. |
| <b>Synonyms:</b>     | Goat Anti-Rat IgG DyLight 649™ Conjugated Antibody, Goat Anti-Rat IgG Antibody DyLight 649™ Conjugation                                                                                                                                                           |
| <b>Host Species:</b> | Goat                                                                                                                                                                                                                                                              |
| <b>Specificity:</b>  | IgG (H&L)                                                                                                                                                                                                                                                         |
| <b>Conjugate:</b>    | DyLight™ 649                                                                                                                                                                                                                                                      |
| <b>Clonality:</b>    | Polyclonal                                                                                                                                                                                                                                                        |
| <b>Format:</b>       | IgG                                                                                                                                                                                                                                                               |
| <b>F/P Ratio:</b>    | 3.0                                                                                                                                                                                                                                                               |

### Target Details

|                    |                         |
|--------------------|-------------------------|
| <b>Reactivity:</b> | Rat                     |
| <b>Immunogen:</b>  | Rat IgG, whole molecule |

|                            |                                                                                                                                                                                                                                                                                                                                                                                                                                                           |
|----------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Purity/Specificity:</b> | 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. |
|----------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|

## Application Details

|                                |                                                                                                                                                                                                                                                                                                                             |
|--------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Suggested Applications:</b> | IF, Multiplex (Based on references)                                                                                                                                                                                                                                                                                         |
| <b>Application Note:</b>       | 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. |
| <b>Assay Dilutions:</b>        | All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.                                                                                                                                                                                                                             |
| <b>FLISA:</b>                  | >1:20,000                                                                                                                                                                                                                                                                                                                   |
| <b>IF:</b>                     | >1:5,000                                                                                                                                                                                                                                                                                                                    |
| <b>WB:</b>                     | >1:10,000                                                                                                                                                                                                                                                                                                                   |

## Formulation

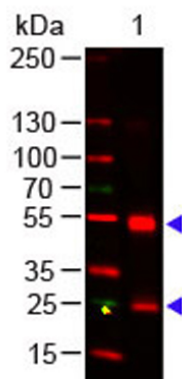
|                               |                                                                        |
|-------------------------------|------------------------------------------------------------------------|
| <b>Physical State:</b>        | Lyophilized                                         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

## Shipping & Handling

|                            |                                                                                                                                                                                                                                                                                                                                                 |
|----------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Shipping Condition:</b> | Ambient                                                                                                                                                                                                                                                                                                                                         |
| <b>Storage Condition:</b>  | 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.

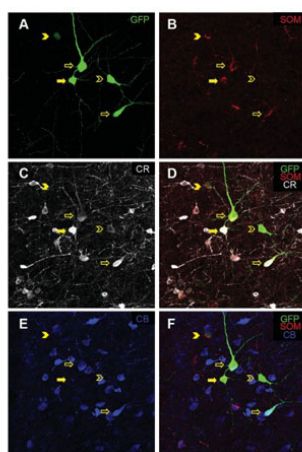
Lane 1: Rat IgG.

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.

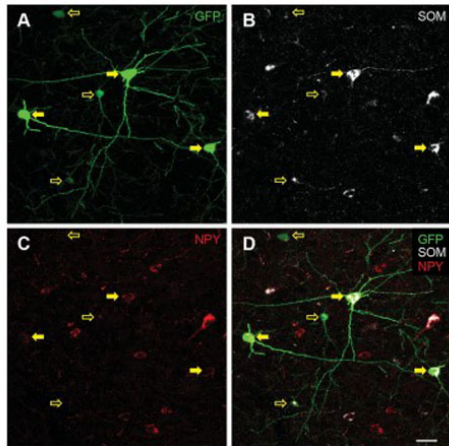


### Immunofluorescence Microscopy

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 GFP1 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/CB1 cells, solid yellow arrows GFP1/SOM1/CR1/CB1 cells, open yellow arrowheads GFP1/SOM1/CR1/CB1 cells, and solid yellow arrowheads GFP1/SOM1/CR1/CB1 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/CB1 cells (d), GFP1/SOM1/CR1/CB2 cells (e), and GFP1/SOM1/CR1/CB1 cells (f) in the cingulate cortex. Scale bar 5 20 μm in F (applies to A–F). Figure 7. PMID: 26669716.

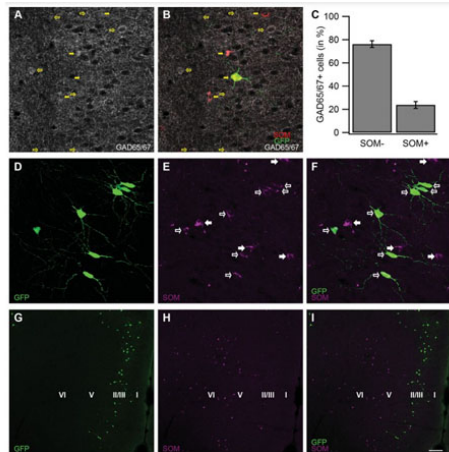


### Immunofluorescence Microscopy

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:

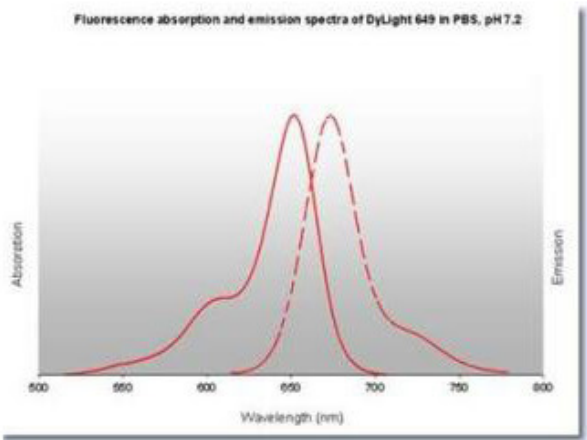
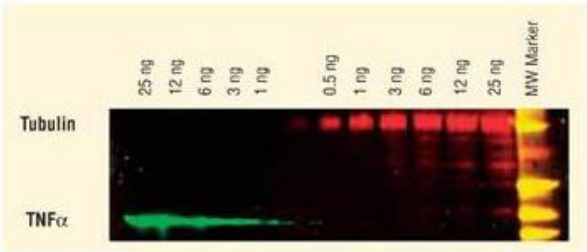
Representative confocal images of cells immunopositive for GAD65/67 (white), SOM (red), and GFP (green). Solid yellow arrows indicate GAD65/ 671/SOM 1 cells and open yellow arrows GAD65/671/SOM2 cells. C: Mean 6 standard deviation of relative numbers of GAD65/671/ SOM2 and GAD65/671/SOM 1 cells in the 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) | $\epsilon$ (M <sup>-1</sup> cm <sup>-1</sup> ) | 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 $\alpha$  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.