

**Datasheet for 610-143-003****Mouse IgG Fc Antibody DyLight™ 649 Conjugated****Overview**

|                      |                                                                       |
|----------------------|-----------------------------------------------------------------------|
| <b>Description:</b>  | Goat Anti-Mouse IgG Fc Antibody DyLight™ 649 Conjugated - 610-143-003 |
| <b>Item No.:</b>     | 610-143-003                                                           |
| <b>Size:</b>         | 100 µg                                                                |
| <b>Applications:</b> | WB                                                                    |
| <b>Reactivity:</b>   | Mouse                                                                 |
| <b>Host Species:</b> | Goat                                                                  |

**Product Details**

|                      |                                                                                                                                                                                                                                                                                                                                                                          |
|----------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Background:</b>   | Anti-Mouse IgG F(c) generated in goat is a proteolytic fragment of immunoglobulin G (IgG) obtained by limited digestion with the enzyme papain under controlled conditions of temperature, time and pH. Receptors bind the Fc portion of mouse IgG and often this fragment is removed from immunoglobulins to minimize receptor binding and lower background reactivity. |
| <b>Synonyms:</b>     | Goat Anti Mouse IgG F(c) Antibody DyLight™ 649 Conjugated, Goat Anti-Mouse IgG Fc Antibody DyLight™ 649 Conjugated, Goat Anti Mouse IgG Fc Fragment Antibody DyLight™ 649 Conjugated                                                                                                                                                                                     |
| <b>Host Species:</b> | Goat                                                                                                                                                                                                                                                                                                                                                                     |
| <b>Specificity:</b>  | IgG Fc                                                                                                                                                                                                                                                                                                                                                                   |
| <b>Conjugate:</b>    | DyLight™ 649                                                                                                                                                                                                                                                                                                                                                             |
| <b>Clonality:</b>    | Polyclonal                                                                                                                                                                                                                                                                                                                                                               |
| <b>Format:</b>       | IgG                                                                                                                                                                                                                                                                                                                                                                      |
| <b>F/P Ratio:</b>    | 3.1                                                                                                                                                                                                                                                                                                                                                                      |

**Target Details**

|                    |                         |
|--------------------|-------------------------|
| <b>Reactivity:</b> | Mouse                   |
| <b>Immunogen:</b>  | Mouse IgG F(c) fragment |

|                            |                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                 |
|----------------------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Purity/Specificity:</b> | This product was prepared from monospecific antiserum by immunoaffinity chromatography using Mouse IgG coupled to agarose beads followed by solid phase adsorption(s) to remove any unwanted reactivities. Assay by immunoelectrophoresis resulted in a single precipitin arc against anti-Goat Serum, Mouse IgG, Mouse IgG F(c) and Mouse Serum. No reaction was observed against Mouse IgG F(ab) This antibody will react with heavy chains of Mouse IgG. Minimal reactivity is expected against other Mouse immunoglobulins. |
|----------------------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|

## Application Details

|                                |                                                                                                                                                                                                                                                                                                                                                                                                        |
|--------------------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Suggested Applications:</b> | WB (Based on references)                                                                                                                                                                                                                                                                                                                                                                               |
| <b>Application Note:</b>       | The emission spectra for this DyLight™ conjugate match the principle output wavelengths of most common fluorescence instrumentation. 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>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

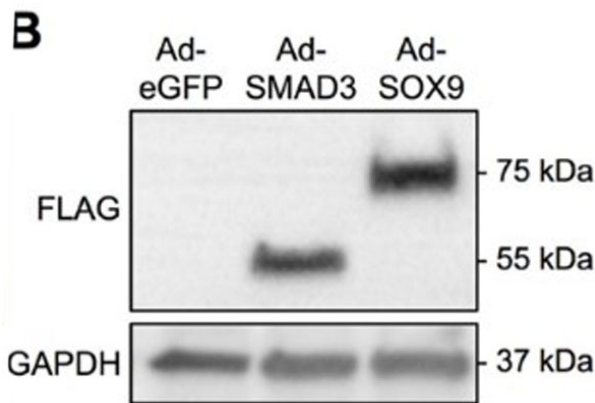
|                               |                                                                        |
|-------------------------------|------------------------------------------------------------------------|
| <b>Physical State:</b>        | Lyophilized                                                            |
| <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

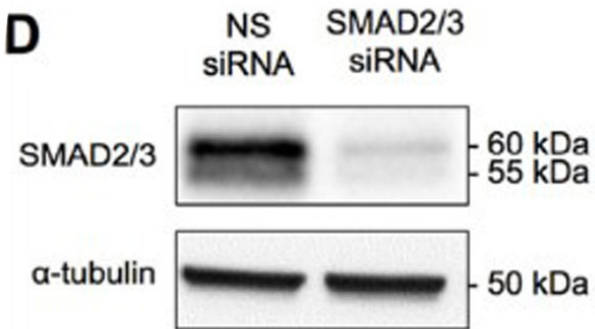
|                            |         |
|----------------------------|---------|
| <b>Shipping Condition:</b> | Ambient |
|----------------------------|---------|

|                    |                                                                                                                                                                                                                                                                                                                                                 |
|--------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| 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.                                                                                                                                                                                                                                                                                           |

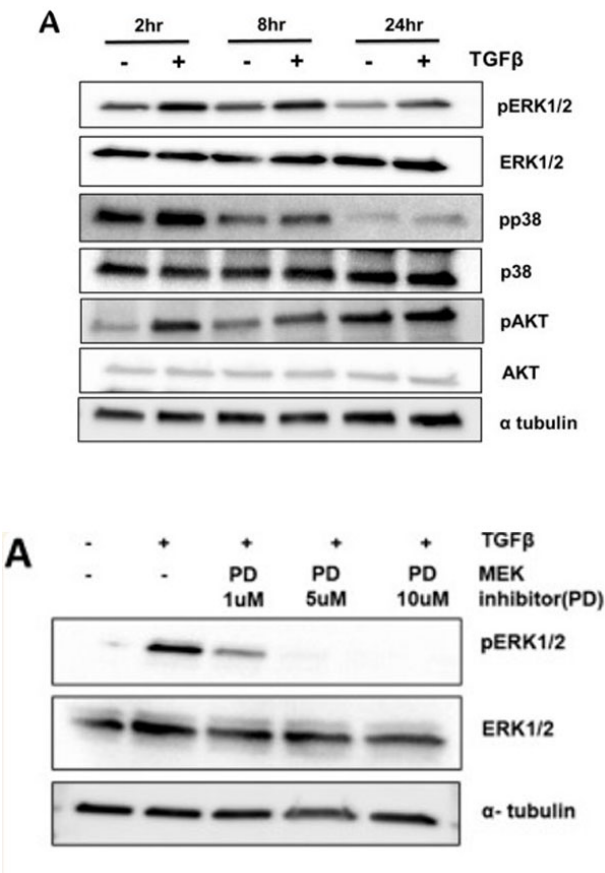
Images



**Western Blot**  
SMAD3 and SOX9 adenoviral vectors infect cells, induce expression of FLAG-tagged proteins, and up-regulate SMAD3 and SOX9 function respectively. Bovine chondrocytes that were infected with either Ad-eGFP, Ad-SMAD3, or Ad-SOX9. Western blots of extracts from bovine chondrocytes showed expression of FLAG-tagged proteins at approximately 52 kDa and 75 kDa, corresponding to SMAD3 and SOX9 molecular weights respectively (n = 4) (B). Primary antibodies: anti-GAPDH antibody (1:1000) or anti-FLAG antibody (1:500), with secondary antibodies: HRP-conjugated anti-rabbit (1:2000) and anti-mouse [1:2500, p/n 610-143-003] Figure 2. PMID: 27746378.



**Western Blot**  
Knock-down of Sox9 attenuates TGF-β-mediated regulation of Paps2. ATDC5 cells were transfected with Sox9 siRNA, Smad2/3 siRNA, or control non-specific (NS) siRNA. Western blots showed Smad2/3 protein levels were reduced in the presence of Smad2/3 siRNA. α-Tubulin was used as a loading control (n = 6) (D). Primary antibodies: anti-SMAD2/3 antibody (1:2000) or anti-α-Tubulin antibody [1:2500, p/n 200-301-880], with secondary antibodies anti-mouse [1:2500, p/n 610-143-003]. Figure 4. PMID: 27746378.



**Western Blot**

TGF-β signaling regulates noncanonical pathways in the sclerotome. Sclerotome was treated with vehicle control or TGFβ1 for 2, 8 or 24 h. Immunoblot was used to determine activity of ERK, p38 and AKT. α tubulin was used as a general loading control. Immunoblots were cropped for clarity. Primary antibodies: pERK1/2 , ERK1/2, pp38, p38, pAKT, AKT, and Alpha tubulin [p/n 200-301-880] with secondary antibodies: anti-Rabbit-HRP and anti-mouse DyLight™649 [p/n 610-143-003]. Figure 3. PMID: 33288795.

**Western Blot**

ERK is required for fibrous tissue marker regulation but not required to inhibit chondrogenesis. (A) Sclerotome was treated with a MEK inhibitor PD184352, PD, to inhibit ERK activity, for 24 h and then cells were treated with TGFβ1 for 8 h. Immunoblot was used to determine relative levels of pERK1/2, ERK1/2, and α-tubulin. Primary antibodies: pERK1/2 , ERK1/2, and Alpha tubulin [p/n 200-301-880] with secondary antibodies: anti-Rabbit-HRP and anti-mouse DyLight™649 [p/n 610-143-003]. Figure 5. PMID: 33288795.

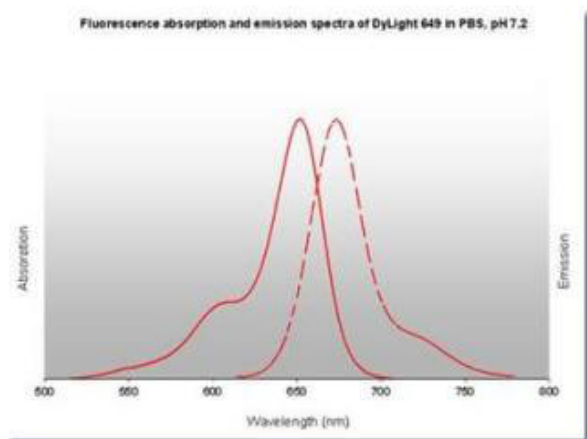
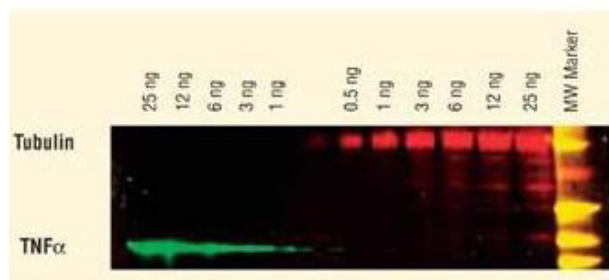
**Diagram**

Properties of DyLight™ Conjugates.

| Emission      | Color                                                                               | DyLight™ Dye | Ex/Em (nm) | ε (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

## References

- Clayton SW et al. Canonical and noncanonical TGF- $\beta$  signaling regulate fibrous tissue differentiation in the axial skeleton. *Sci Rep.* (2020)
- Chavez et al. SOX9 protein is stabilized by TGF- $\beta$  and regulates PAPSS2 mRNA expression in chondrocytes. *Osteoarthritis Cartilage* (2017)

## 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.