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Datasheet for 611-151-122**Rabbit IgG (H&L) Antibody ATTO 425 Conjugated Pre-Adsorbed****Overview**

Description:	Goat Anti-Rabbit IgG (H&L) Antibody ATTO 425 Conjugated (Min X Bv Ch Gt GP Ham Hs Hu Ms Rt & Sh Serum Proteins) - 611-151-122
Item No.:	611-151-122
Size:	500 µg
Applications:	Dot Blot, WB, IF, IHC, Multiplex
Reactivity:	Rabbit
Host Species:	Goat

Product Details

Background:	Anti-Rabbit IgG (H&L) ATTO 425 Antibody generated in goat detects reactivity to Rabbit IgG. Secreted as part of the adaptive immune response by plasma B cells, immunoglobulin G constitutes 75% of serum immunoglobulins. Immunoglobulin G binds to viruses, bacteria, as well as fungi and facilitates their destruction or neutralization via agglutination (and thereby immobilizing them), activation of the complement cascade, and opsonization for phagocytosis. The whole IgG molecule possesses both the F(c) region, recognized by high-affinity Fc receptor proteins, as well as the F(ab) region possessing the epitope-recognition site. Both the Heavy and Light chains of the antibody molecule are present. Secondary Antibodies are available in a variety of formats and conjugate types. When choosing a secondary antibody product, consideration must be given to species and immunoglobulin specificity, conjugate type, fragment and chain specificity, level of cross-reactivity, and host-species source and fragment composition.
Synonyms:	Goat anti-Rabbit IgG Antibody ATTO425 Conjugation, Goat anti-Rabbit IgG ATTO 425 Conjugated Antibody
Host Species:	Goat
Specificity:	IgG (H&L)
Conjugate:	ATTO 425
Clonality:	Polyclonal
Format:	IgG
F/P Ratio:	2.3

Target Details

Reactivity:	Rabbit
Immunogen:	Rabbit IgG whole molecule
Purity/Specificity:	Rabbit IgG (H&L) Antibody ATTO 425 was prepared from monospecific antiserum by immunoaffinity chromatography using Rabbit 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, Rabbit IgG and Rabbit Serum. No reaction was observed against Bovine, Chicken, Goat, Guinea Pig, Hamster, Horse, Human, Mouse, Rat and Sheep Serum Proteins. This antibody will react with heavy chains of rabbit IgG and with light chains of most rabbit immunoglobulins.

Application Details

Tested Applications:	Dot Blot, WB
Suggested Applications:	IF, IHC, Multiplex (Based on references)
Application Note:	Anti-Rabbit IgG (H&L) conjugated to ATTO 425 has been tested by dot blot and western blot and is designed for STED microscopy, FRET, 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. The emission spectra for this ATTO conjugate matches 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

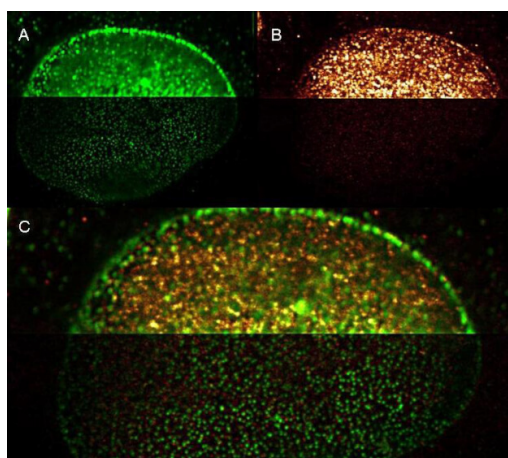
Physical State:	Lyophilized
Concentration:	1.0 mg/mL by UV absorbance at 280 nm
Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	0.01% (w/v) Sodium Azide
Stabilizer:	10 mg/mL Bovine Serum Albumin (BSA) - Immunoglobulin and Protease free

Reconstitution Volume:	500 μ L
Reconstitution Buffer:	Restore with deionized water (or equivalent)

Shipping & Handling

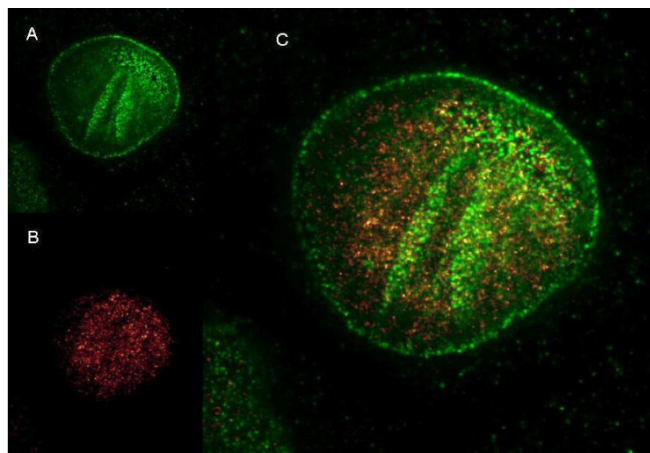
Shipping Condition:	Ambient
Storage Condition:	Store vial at 4° C prior to restoration. For extended storage aliquot contents and freeze at -20° C or below. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for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



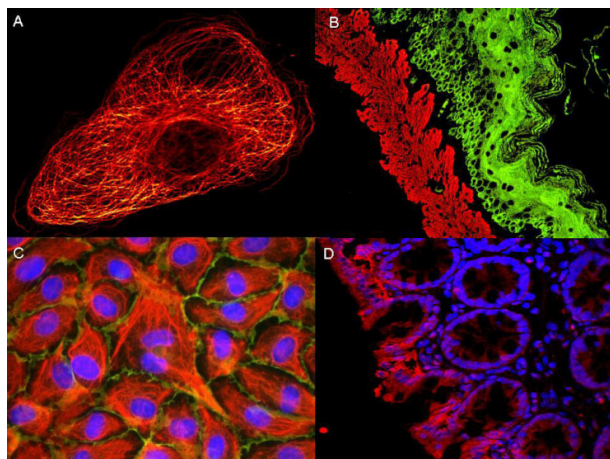
Immunofluorescence Microscopy

Rockland Dylight and ATTO®dye conjugated antibodies provide high signal and low background for confocal microscopy (upper images) and high resolution Stimulated Emission Depletion (STED) Microscopy (lower images). Both Dylight and Atto conjugated secondary antibodies maintained robust, intense signal during repeated laser excitation and de-excitation used during STED microscopy. Shown here are: A. (Green) Mouse anti NuP (NuP=Nuclear Pore Protein) detected with Dylight 488 Goat anti mouse (610-141-121) B. (Red) Rabbit Anti Ezh1/2 Pab (Ezh=enhancer of zeste homology) with detection by Rockland ATTO®425 conjugated Goat anti Rabbit (611-151-122) (Red and Green) Images combined. Data was collected on a STED-CW TCS-SP5 Confocal system (Leica Microsystems) equipped with a DFC 350FX Camera allowing sequential acquisition in widefield, confocal and STED CW imaging modes and provided courtesy of: Myriam Gastard, PhD, personal communication, Leica Microsystems, Inc. USA



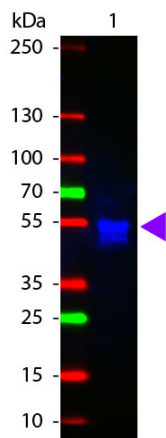
Immunofluorescence Microscopy

Rockland DyLight and ATTO[®] dye conjugated antibodies provide high signal and low background for confocal microscopy and high resolution Stimulated Emission Depletion (STED) Microscopy. Both Dylight and Atto conjugated secondary antibodies maintained robust, intense signal during repeated laser excitation and de-excitation used during STED microscopy. Shown here are: A. (Green) Mouse anti NuP (NuP=Nuclear Pore Protein) detected with Dylight 488 Goat anti mouse (610-141-121) B. (Red) Rabbit Anti Ezh1/2 Pab (Ezh=enhancer of zeste homology) with detection by Rockland ATTO[®]425 conjugated Goat anti Rabbit (611-151-122) C. (Red and Green) Images combined. Data was collected on a STED-CW TCS-SP5 Confocal system (Leica Microsystems) equipped with a DFC 350FX Camera allowing sequential acquisition in wide-field, confocal and STED CW imaging modes and provided courtesy of: Myriam Gastard, PhD, personal communication, Leica Microsystems, Inc. USA



Immunofluorescence Microscopy

ATTO[®] dyes can be used for multicolor immunofluorescent detection with low background and high signal. Examples shown are: A. Tubulin in PtK2- male Rat Kangaroo Kidney Epithelial Cells was detected using ATTO 532 labeled secondary antibody. B. Muscle alpha-actin was stained with a mouse primary antibody and ATTO 488 anti-mouse IgG (green) while Cytokeratin was stained with polyclonal rabbit anti-cytokeratin and ATTO 647N anti-rabbit IgG (red). C. HUVEC (Human umbilical vein endothelial cells) were stained with anti- Vimentin-ATTO 532 (green), anti-E-Cadherin-ATTO 655 (red) and DAPI (blue). D. Rat colon sections were stained with Anti-Aquaporin 3-ATTO 594 antibody. Hoechst 33342 (blue) is used as counterstain. Images provided courtesy of Dr. Jörg Reichwein, ATTO-TEC GmbH



Western Blot

Western Blot of ATTO 425 conjugated Goat anti-Rabbit IgG antibody. Lane 1: Rabbit IgG. Lane 2: none. Load: 50 ng per lane. Primary antibody: none. Secondary antibody: ATTO 425 rabbit secondary antibody at 1:1,000 for 60 min at RT. Block: MB-070 for 30 min RT. Predicted/Observed size: 55 kDa, 28 kDa/55 kDa for Rabbit IgG. Other band(s): none.

References

- Hruska M et al. Nanoscale rules governing the organization of glutamate receptors in spine synapses are subunit specific. *Nat Commun.* (2022)
- Henderson NT, Le Marchand SJ, Hruska M, Hippenmeyer S, Luo L, Dalva MB. Ephrin-B3 controls excitatory synapse density through cell-cell competition for EphBs. *Elife.* (2019)
- Hruska M et al. Anchoring and synaptic stability of PSD-95 is driven by ephrin-B3. *Nat Neurosci.* (2015)

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.