

Product datasheet

Donkey Anti-Rabbit IgG H&L (Alexa Fluor® 647)
ab150075

★★★★★ 4 Abreviews 236 References 5 Images

Overview

Product name	Donkey Anti-Rabbit IgG H&L (Alexa Fluor® 647)
Description	Donkey polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 647)
Host species	Donkey
Target species	Rabbit
Tested applications	Suitable for: ICC/IF, ELISA, IHC-P, Flow Cyt, IHC-Fr
Immunogen	The details of the immunogen for this antibody are not available.
Conjugation	Alexa Fluor® 647. Ex: 652nm, Em: 668nm

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle. Stable for 12 months at -20°C. Store In the Dark.
Storage buffer	Preservative: 0.02% Sodium azide Constituents: 23% Glycerol (glycerin, glycerine), PBS, 1% BSA
Purity	Immunogen affinity purified
Purification notes	This antibody was isolated by affinity chromatography using antigen coupled to agarose beads.
Clonality	Polyclonal
Isotype	IgG
General notes	Alexa Fluor® is a registered trademark of Molecular Probes, Inc, a Thermo Fisher Scientific Company. The Alexa Fluor® dye included in this product is provided under an intellectual property license from Life Technologies Corporation. As this product contains the Alexa Fluor® dye, the purchase of this product conveys to the buyer the non-transferable right to use the purchased product and components of the product only in research conducted by the buyer (whether the buyer is an academic or for-profit entity). As this product contains the Alexa Fluor® dye the sale of this product is expressly conditioned on the buyer not using the product or its components, or any materials made using the product or its components, in any activity to generate revenue, which may include, but is not limited to use of the product or its components: in manufacturing; (ii) to provide a service, information, or data in return for payment (iii) for therapeutic, diagnostic or

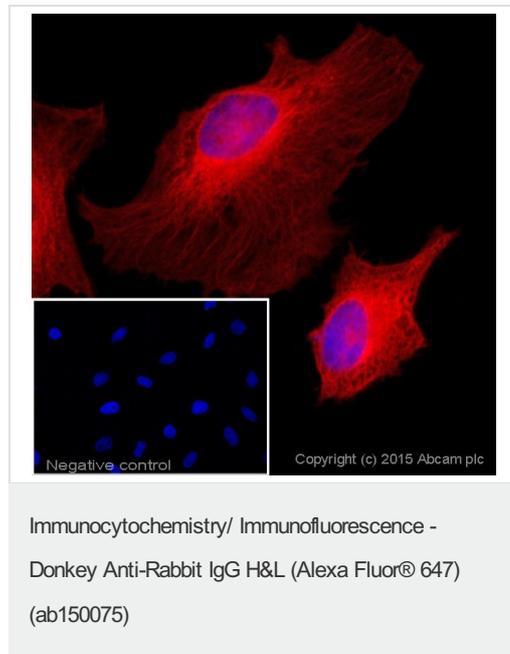
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Applications

The Abpromise guarantee Our [Abpromise guarantee](#) covers the use of ab150075 in the following tested applications. The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

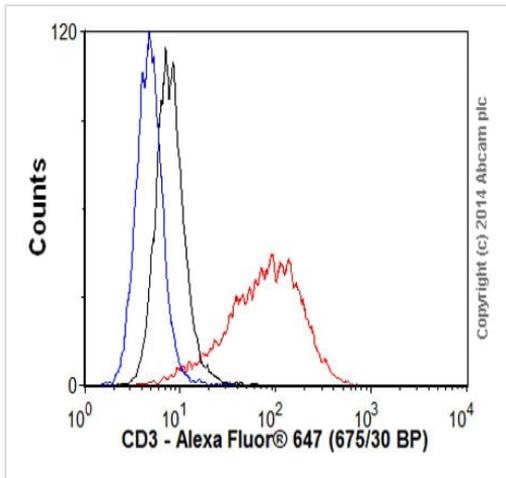
Application	Abreviews	Notes
ICC/IF	★★★★★ (1)	1/200 - 1/1000.
ELISA		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration.
Flow Cyt		1/2000 - 1/4000. ab199093 - Rabbit monoclonal IgG (Alexa Fluor® 647), is suitable for use as an isotype control to complement this secondary antibody.
IHC-Fr	★★★★★ (1)	Use at an assay dependent concentration.

Images



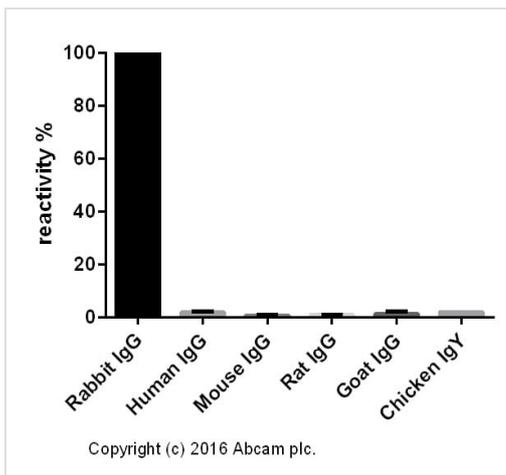
ICC/IF image of [ab6046](#) stained HeLa cells. The cells were 100% formaldehyde fixed (10 min), permeabilized with 0.1% Triton X-100 for 5 minutes and then incubated in 1%BSA / 10% normal donkey serum / 0.3M glycine in 0.1% PBS-Tween for 1h to block non-specific protein-protein interactions. The cells were then incubated with the primary antibody ([ab6046](#), 5µg/ml) overnight at +4°C. The secondary antibody (red) was ab150075 Alexa Fluor® 647 donkey anti-rabbit IgG (H+L) used at 1µg/ml for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.

The negative control (inset) is a secondary-only assay to demonstrate low non-specific binding of the secondary antibody.



Flow Cytometry - Donkey Anti-Rabbit IgG H&L (Alexa Fluor® 647) (ab150075)

Overlay histogram showing Jurkat cells stained with [ab16669](#) (red line). The cells were fixed with 4% paraformaldehyde (10 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal donkey serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody ([ab16669](#), 1/1000 dilution) for 30 min at 22°C. The secondary antibody Donkey anti-rabbit IgG H&L (Alexa Fluor® 647) ([ab150075](#)) was used at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (0.1µg/1x10⁶ cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a solid-state 25mW red diode laser (635nm) and 675/30 bandpass filter.

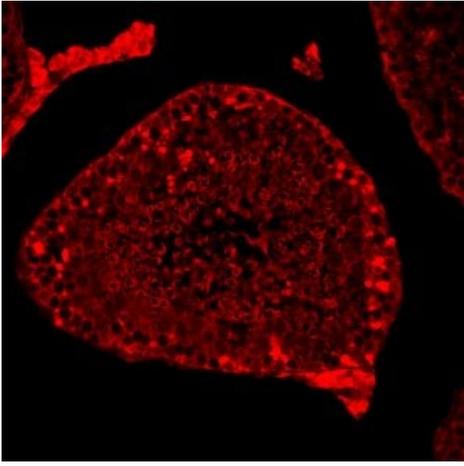


ELISA - Donkey Anti-Rabbit IgG H&L (Alexa Fluor® 647) (ab150075)

Cross-reactivity of the polyclonal secondary antibody [ab182020](#) was tested using a sandwich ELISA approach. The wells were coated with the indicated IgG standards at 1 µg/ml (50 µl/well) and incubated overnight at 4°C, followed by a 5% BSA blocking step for 2h at RT. [ab182020](#) was then added starting at 1 µg/ml and gradually diluted 1/4 (50 µl/well), followed by incubation for 2h. For the detection Goat anti-Donkey IgG H&L (HRP) ([ab6988](#)) was used at 1/20,000 dilution (50 µl/well), followed by incubation for 1h at RT.

For the batch tested, [ab182020](#) showed a cross-reactivity below 2% towards human IgG, mouse IgG, rat IgG, goat IgG and chicken IgY.

This data was developed using the unconjugated antibody ([ab182020](#)).

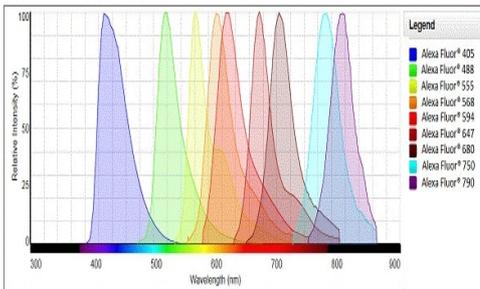


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Donkey Anti-Rabbit IgG H&L (Alexa Fluor® 647) (ab150075)

This image is courtesy of an anonymous Abreview.

ab62050 staining prostaglandin E synthase in mouse testis tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffin-embedded sections). Tissue was fixed with paraformaldehyde, permeabilized with Triton X-100, and blocked with 10% serum for 30 minutes at 24°C; antigen retrieval was by heat mediation in a citrate buffer. Samples were incubated with primary antibody (1/50) for 20 hours at 4°C. An Alexa Fluor® 647-conjugated donkey anti-rabbit IgG H&L (ab150075) (1/200) was used as the secondary antibody.

Emission Spectra of the Alexa Fluor® Conjugated Secondary Antibodies



Alexa Fluor® - Donkey Anti-Rabbit IgG H&L (Alexa Fluor® 647) (ab150075)

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