# abcam

# Product datasheet

# Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) ab150077

★★★★★ 20 Abreviews 2871 References 画像数 16

### 製品の概要

製品名 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488)

由来種Goatターゲット生物種Rabbit

特異性 This antibody is specific to Rabbit IgG.

アプリケーション 適用あり: ICC/IF, Flow Cyt, IHC-P, ELISA, IHC-Fr

免疫原 The details of the immunogen for this antibody are not available.

標識 Alexa Fluor® 488. Ex: 495nm, Em: 519nm

#### 製品の特性

製品の状態 Liquid

保存方法 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.

パッファー Preservative: 0.02% Sodium azide

Constituents: 23% Glycerol (glycerin, glycerine), PBS, 1% BSA

精製度 Immunogen affinity purified

特記事項(精製) This antibody was isolated by affinity chromatography using antigen coupled to agarose beads.

**ポリ/モノ** ポリクローナル

アイソタイプ IgG

特記事項 Fluorochrome chart – a complete quide:

A quick and easy guide to help you select the most appropriate fluorochromes for your next

experiment.

Please see here.

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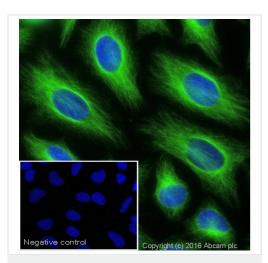
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#### アプリケーション

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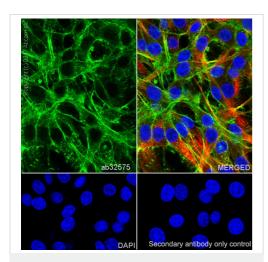
アプリケーション	Abreviews	特記事項
ICC/IF	<b>★★★★★ (7)</b>	1/200 - 1/1000.
Flow Cyt		1/2000 - 1/4000.
IHC-P	<b>★★★★★</b> (3)	Use at an assay dependent concentration.
ELISA		Use at an assay dependent concentration.
IHC-Fr	<b>★★★★★</b> (5)	Use at an assay dependent concentration.

#### 画像



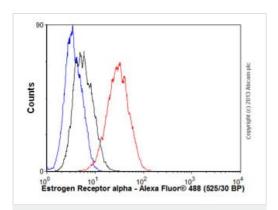
Immunocytochemistry/ Immunofluorescence - Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) ICC/IF image of beta Tubulin staining in HeLa cells. The cells were 100% methanol fixed (5 min), permeabilized with 0.1% Triton X-100 for 5 minutes and then incubated in 1% BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to block nonspecific protein-protein interactions. The cells were then incubated with the primary antibody (ab6046, 5µg/ml) overnight at +4°C. The secondary antibody (green) was ab150077 Alexa Fluor 488 goat anti-rabbit lgG (H+L) used at 2µg/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.



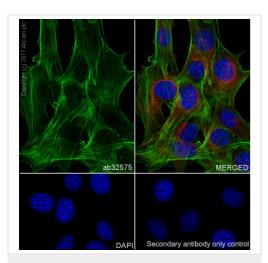
Immunocytochemistry/ Immunofluorescence - Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077)

Immunocytochemistry/ Immunofluorescence analysis of C6(Rat glial tumor glial cell) cells labeling alpha smooth muscle Actin with purified  $\underline{ab32575}$  at 1/100 dilution (0.71  $\mu$ g/ml). Cells were fixed in 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1/200 (2.5  $\mu$ g/ml). Goat anti rabbit lgG (Alexa Fluor® 488, ab150077) was used as the secondary antibody at 1/1000 dilution (2  $\mu$ g/ml) dilution. DAPI was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



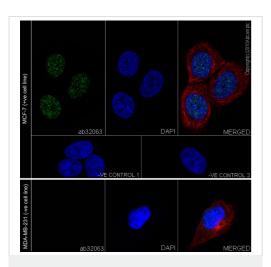
Flow Cytometry (Intracellular) - Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077)

Overlay histogram showing MCF7 cells stained with unpurified ab32063 (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 goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab32063, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit lgG (H+L) (ab150077) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (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 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This antibody gave a positive signal in MCF7 cells fixed with 80% methanol (5 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.



Immunocytochemistry/ Immunofluorescence - Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077)

Immunocytochemistry/ Immunofluorescence analysis of NIH/3T3 (Mouse embryonic fibroblast) cells labeling alpha smooth muscle Actin with purified <u>ab32575</u> at 1/500 dilution (5.2  $\mu$ g/ml). Cells were fixed in 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. Cells were counterstained with Ab195889 Antialpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1/200 (2.5  $\mu$ g/ml). Goat anti rabbit lgG (Alexa Fluor® 488, ab150077) was used as the secondary antibody at 1/1000 dilution (2  $\mu$ g/ml) dilution. DAPI was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.

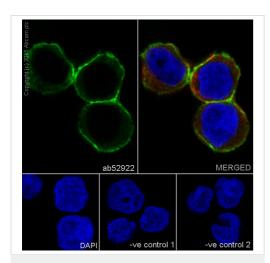


Immunocytochemistry/ Immunofluorescence - Goat
Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077)

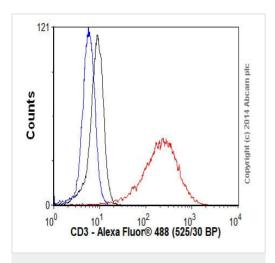
Immunocytochemistry/Immunofluorescence analysis of MCF-7 cells labelling Estrogen Receptor alpha with purified <u>ab32063</u> at 1/1000. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit lgG (1/1000) was used as the secondary antibody. The cells were co-stained with <u>ab7291</u>, a mouse anti-tubulin (1/1000) using <u>ab150120</u>, an Alexa Fluor® 594-conjugated goat anti-mouse lgG (1/1000) as the secondary antibody. Nuclei counterstained with DAPI (blue).

Control 1: primary antibody (1/1000) and secondary antibody, **ab150120**, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/1000).

Control 2: <u>ab7291</u> (1/1000) and secondary antibody, ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit lgG (1/1000).



Immunocytochemistry/ Immunofluorescence - Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077)



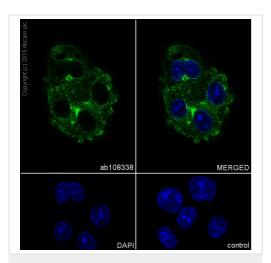
Flow Cytometry - Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077)

Immunocytochemistry/Immunofluorescence analysis of Raji (human Burkitt's lymphoma) cells labelling HLA A with purified <a href="mailto:ab52922">ab52922</a> at 1/100. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit lgG (1/1000) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. <a href="mailto:ab7291">ab7291</a>, a mouse anti-tubulin (1/1000) and <a href="mailto:ab7291">ab150120</a>, an Alexa Fluor® 594-conjugated goat anti-mouse lgG (1/1000) were also used.

Control 1: primary antibody (1/100) and secondary antibody, **ab150120**, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/500).

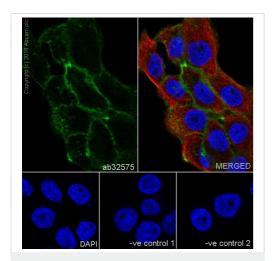
Control 2: <u>ab7291</u> (1/1000) and secondary antibody, ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit lgG (1/500).

Overlay histogram showing Jurkat cells stained with <u>ab16669</u> (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 goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (<u>ab16669</u>, 1/1000 dilution) for 30 min at 22°C. The secondary antibody Goat anti-rabbit IgG H&L (Alexa Fluor® 488) (ab150077) was used at 1/4000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monclonal) (0.1µg/1x10<sup>6</sup> 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 20mW Argon ion laser (488nm) and 525/30 bandpass filter.



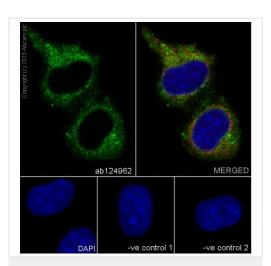
Immunocytochemistry/ Immunofluorescence - Goat
Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077)

Immunocytochemistry/ Immunofluorescence analysis of HepG2 (Human hepatocellular carcinoma epithelial cell) cells labeling ATG9A with Purified <a href="mailto:ab108338">ab108338</a> at 1/100 dilution. Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. ab150077 Goat anti rabbit lgG (Alexa Fluor® 488) was used as the secondary antibody at 1/1000 dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Immunocytochemistry/ Immunofluorescence - Goat
Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077)

Immunocytochemistry/Immunofluorescence analysis of A431 (human epidermoid carcinoma) cells labeling alpha smooth muscle Actin (green) with purified <a href="mailto:ab32575">ab32575</a> at 1/500. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000) was used as the secondary antibody. Cells were counterstained with <a href="mailto:ab7291">ab7291</a>, anti-Tubulin (mouse mAb) at 1/1000 followed by <a href="mailto:ab150120">ab150120</a> Alexa Fluor®594 goat anti-mouse secondary (1/1000). Nuclei were counterstained with DAPI (blue). For negative control 1, rabbit primary antibody and anti-mouse secondary antibody (<a href="mailto:ab150120">ab150120</a>) were used. For negative control 2, <a href="mailto:ab150120">ab7291</a> (mouse primary antibody) was used followed by anti-rabbit secondary antibody (<a href="mailto:ab150077">ab150077</a>).

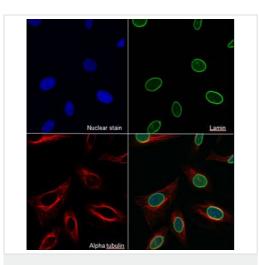


Immunocytochemistry/ Immunofluorescence - Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077)

Immunocytochemistry/Immunofluorescence analysis of HeLa cells labelling IL-1RA with purified <u>ab124962</u> at 1/100. Cells were fixed with 100% methanol and permeabilized with 0.1% Triton X-100. ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. <u>ab7291</u>, a mouse anti-tubulin (1/1000) and <u>ab150120</u>, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/1000) were also used.

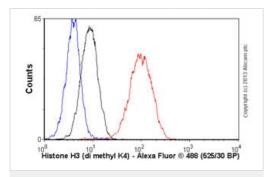
Control 1: primary antibody (1/100) and secondary antibody, **ab150120**, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/1000).

Control 2: <u>ab7291</u> (1/1000) and secondary antibody, ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit lgG (1/1000).

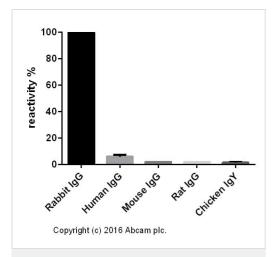


Immunocytochemistry/ Immunofluorescence - Goat
Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077)

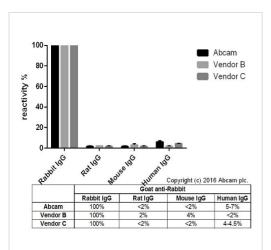
The cells were 100% methanol fixed (5 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab7291, 1µg/ml) and (ab16048, 1µg/ml) overnight at +4°C. The secondary antibodies were ab150115 Alexa Fluor® 647 (red) goat anti-mouse IgG (H+L) used at 2µg/ml for 1h and ab150077 Alexa Fluor® 488 (green) goat anti-rabbit IgG (H+L) used at 2µg/ml for 1h. DAPI was used to stain the cell nuclei.



Flow Cytometry - Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077)



ELISA - Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077)



ELISA - Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077)

Overlay histogram showing HeLa cells stained with <u>ab32356</u> (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (<u>ab32356</u>, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit lgG (H&L) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit lgG (monoclonal) (1µg/1x10^6 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 20mW Argon ion laser (488nm) and 525/30 bandpass filter.

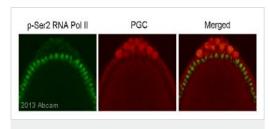
Cross-reactivity of the polyclonal secondary antibody <u>ab182016</u> was tested using a sandwich ELISA approach. The wells were coated with the indicated lgG standards at 1  $\mu$ g/ml (50  $\mu$ l/well) and incubated overnight at 4°C, followed by a 5% BSA blocking step for 2h at RT. <u>ab182016</u> was then added starting at 1  $\mu$ g/ml and gradually diluted 1/4 (50  $\mu$ l/well), followed by incubation for 2h. For the detection Donkey anti-Goat lgG H&L (HRP) (<u>ab6885</u>) was used at 1/10,000 dilution (50  $\mu$ l/well), followed by incubation for 1h at RT.

For the batch tested, <u>ab182016</u> showed a cross-reactivity of 5-7% towards Human IgG and below 2% towards Mouse IgG, Rat IgG and Chicken IgY.

This data was developed using the unconjugated antibody (ab182016).

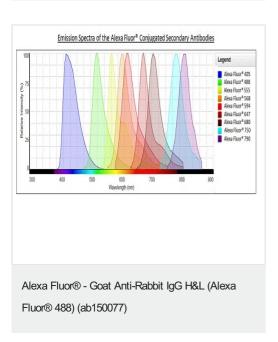
Cross-reactivity of Goat anti-Rabbit IgG H&L (ab182016) and Goat anti-Rabbit IgG H&L obtained from two different vendors was tested using a sandwich ELISA approach. The wells were coated with the indicated IgG standards (Rabbit, Human, Mouse and Rat) at 1  $\mu$ g/ml (50  $\mu$ l/well) and incubated overnight at 4°C, followed by a 5% BSA blocking step for 2h at RT. Secondary antibodies were then added starting at 1  $\mu$ g/ml and gradually diluted 1/4 (50  $\mu$ l/well), followed by incubation for 2h. For the detection Donkey anti-Goat IgG H&L (HRP) (ab6885) was used at 1/10,000 dilution (50  $\mu$ l/well), followed by incubation for 1h at RT. This data is from a representative dilution.

This data was developed using the unconjugated antibody (ab182016).



IHC - Wholemount - Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077)

This image is courtesy of an anonymous Abreview.



IHC - Wholemount of Caenorhabditis elegans larvae labelling RNA polymerase II CTD repeat YSPTSPS (phospho S2) with <u>ab5095</u>. The sample was incubated with primary antibody (1/500 in PBS + 3% BSA + 0.1% Triton X-100) for 12 hours at 4°C. ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG polyclonal (1/1000), was used as the secondary antibody.

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