abcam

Product datasheet

Alexa Fluor® 647 Rabbit IgG, monoclonal [EPR25A] - Isotype Control ab199093

リコンビナント RabMAb

14 References 画像数 5

製品の概要

製品名	Alexa Fluor® 647 Rabbit IgG, monoclonal [EPR25A] - Isotype Control
標識	Alexa Fluor® 647. Ex: 652nm, Em: 668nm
特異性	Please note Abcam have optimised the validation of this product. In our hands, we observe an increase in background signal intensity with the use of Triton X-100 and would recommend using an alternative permeabilisation method such as methanol or saponin.
アプリケーション	**適用あり:** ICC/IF, Flow Cyt
免疫原	Chemical/ Small Molecule conjugated to keyhole limpet haemocyanin. KLH is a copper containing oxygen carrier occurring freely dissolved in the hemolymph of many molluscs and arthropods.KLH forms a large complex composed of ~50 kDa subunits.
特記事項	KLH is often used in molecular immunology as a carrier protein conjugated to low molecular weight molecules such as peptides, amino acids, nucleic acids, drugs or toxins to render them more immunogenic due to the size of the conjugate complex and the immunogenicity of KLH. This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information **see here**. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**. 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

prophylactic purposes; or (iv) for resale, regardless of whether they are sold for use in research. For information on purchasing a license to this product for purposes other than research, contact Life Technologies Corporation, 5781 Van Allen Way, Carlsbad, CA 92008 USA or **outlicensing@thermofisher.com**.

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 long term. Avoid freeze / thaw cycle. Store In the Dark.
バッファー	pH: 7.40 Preservative: 0.02% Sodium azide Constituents: PBS, 30% Glycerol (glycerin, glycerine), 1% BSA
精製度	Protein A purified
ポリ/モノ	モノクローナル
クローン名	EPR25A
アイソタイプ	IgG

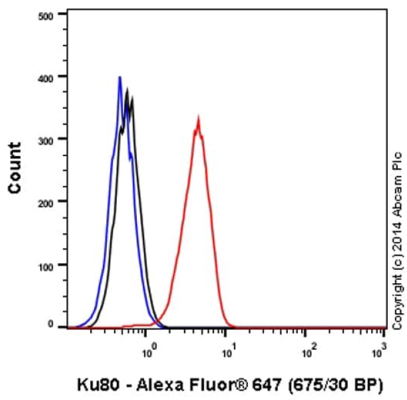
アプリケーション

The Abpromise guarantee **Abpromise保証は、次のテスト済みアプリケーションにおけるab199093の使用に適用されます**

アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
ICC/IF		Use at an assay dependent concentration.
Flow Cyt		Use at an assay dependent concentration. **Please note**: This product should be diluted to the same concentration (not dilution) of the primary antibody to be used.

画像

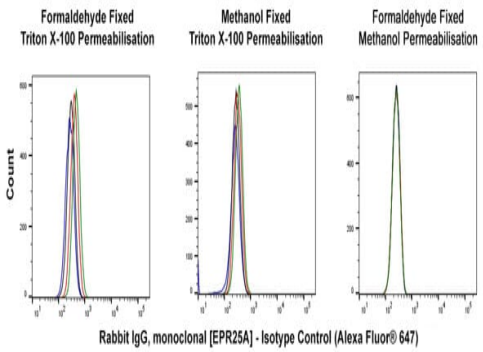


Flow Cytometry - Alexa Fluor® 647 Rabbit IgG, monoclonal [EPR25A] - Isotype Control (ab199093)

Overlay histogram showing HeLa (human epithelial cell line from cervix adenocarcinoma) cells stained with **ab198587** (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 (**ab198587**, 1/50 dilution) for 30 min at 22ºC.

Isotype control antibody (black line) was rabbit IgG (monoclonal) Alexa Fluor® 647 (ab199093) used at the same concentration and conditions as the primary antibody. Unlabeled sample (blue line) was also used as a control.

Acquisition of >5,000 events were collected using a solid-state 25mW red diode laser (635 nm) and 675/30 bandpass filter.



Flow Cytometry - Alexa Fluor® 647 Rabbit IgG, monoclonal [EPR25A] - Isotype Control (ab199093)

Overlay histogram showing HeLa (human epithelial cell line from cervix adenocarcinoma) cells stained with ab199093 using various fixation and permeablisation. The cells were fixed, washed and permeablised as indicated below;

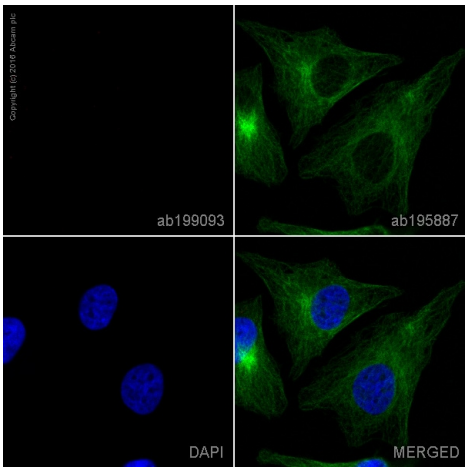
4% formaldehyde (10 min, room temperature)/0.1% PBS-Triton X-100 (15 min, room temperature)

80% methanol (5 min, -20°C)/0.1% PBS-Triton X-100 (15 min, room temperature)

4% formaldehyde (10 min, room temperature)/90% methanol (30 min, -20°C)

The cells were then incubated in 1x PBS / 10% normal goat serum to block non-specific protein-protein interactions followed by the antibody (**ab199091**) for 30 min at 22ºC at the following concentrations - Blue line (Unlabelled), Black line (0.01µg/ml), Red line (0.1µg/ml) and Green line (1µg/ml) .

Acquisition of >5,000 events were collected using a 40mW Red laser (640nm) and 670/14 bandpass filter.

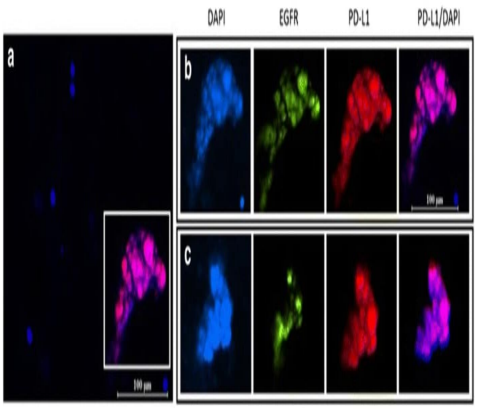


Immunocytochemistry/ Immunofluorescence - Alexa Fluor® 647 Rabbit IgG, monoclonal [EPR25A] - Isotype Control (ab199093)

Immunofluorescent analysis of HeLa (human epithelial cell line from cervix adenocarcinoma) cells, fixed with 4% formaldehyde (10 min). The cells were permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with ab199093 (Rabbit IgG, monoclonal [EPR25A] - Isotype Control) at 1/500 dilution (showing no signal) and **ab195887**, Mouse monoclonal to alpha Tubulin (Alexa Fluor® 488), at 1/250 dilution (shown in green). Nuclear DNA was labeled with DAPI (shown in blue).

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

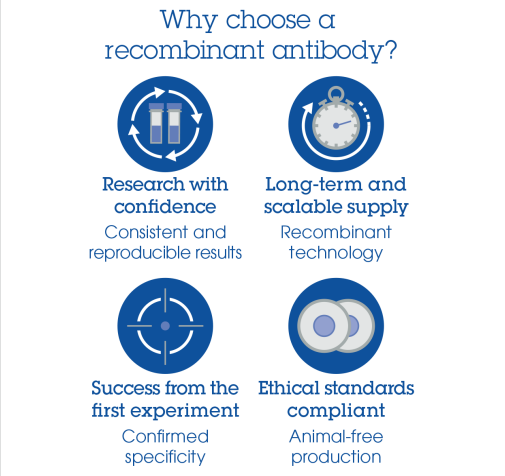
This product also gave a positive signal under the same testing conditions in HeLa cells fixed with 100% methanol (5min).



Immunocytochemistry/ Immunofluorescence - Alexa Fluor® 647 Rabbit IgG, monoclonal [EPR25A] - Isotype Control (ab199093)

Kulasinghe et al BMC Cancer. 2017 May 16;17(1):333. doi: 10.1186/s12885-017-3316-3. Fig 4.

The initial sample was stained using an antibody cocktail and anti-EGFR antibody. A further slide was fixed with 4% formaldehyde for 10 mins, permeabilized with 0.2% Triton X-100 for 5 mins and blocked with 10% fetal-bovine serum in 0.1% PBS-Tween for 1 h at room temperature. The cells were incubated overnight at 4 °C with anti-EGFR antibody and anti-PD-L1 antibody [28-8] (Alexa Fluor® 647) (Abcam **ab209960**) 1/100 dilution. Nuclear DNA was visualized with DAPI. Rabbit IgG monoclonal isotype control (Alexa Fluor®647) (ab199093) was used to identify nonspecific binding. Cells were imaged on the Olympus IX3 inverted microscope.



Alexa Fluor® 647 Rabbit IgG, monoclonal [EPR25A] - Isotype Control (ab199093)

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