abcam

Product datasheet

Alexa Fluor® 647 Anti-Hamartin antibody [EP318Y] ab223388

Recombinant RabMAb

3 Images

Overview

Product name	Alexa Fluor® 647 Anti-Hamartin antibody [EP318Y]
Description	Alexa Fluor® 647 Rabbit monoclonal [EP318Y] to Hamartin
Host species	Rabbit
Conjugation	Alexa Fluor® 647. Ex: 652nm, Em: 668nm
Tested applications	**Suitable for:** Flow Cyt (Intra), IHC-P **Unsuitable for:** ICC/IF
Species reactivity	**Reacts with:** Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	IHC-P: normal human kidney tissue sections. Flow Cyt (intra): HeLa cells
General notes	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.

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. Store In the Dark.
Storage buffer	pH: 7.40 Preservative: 0.02% Sodium azide Constituents: 30% Glycerol (glycerin, glycerine), 1% BSA, PBS
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EP318Y
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab223388 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
Flow Cyt (Intra)		1/5000.
IHC-P		1/100. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

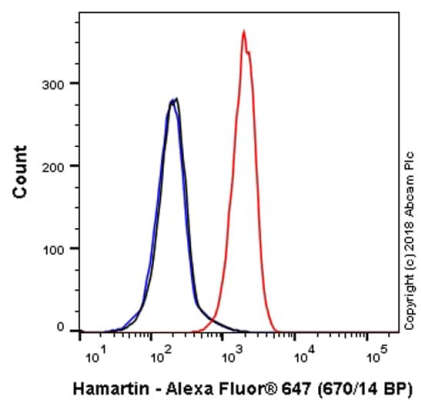
Application notes Is unsuitable for ICC/IF.

Target

Function	In complex with TSC2, inhibits the nutrient-mediated or growth factor-stimulated phosphorylation of S6K1 and EIF4EBP1 by negatively regulating mTORC1 signaling. Seems not to be required for TSC2 GAP activity towards RHEB. Implicated as a tumor suppressor. Involved in microtubule-mediated protein transport, but this seems to be due to unregulated mTOR signaling.
Tissue specificity	Highly expressed in skeletal muscle, followed by heart, brain, placenta, pancreas, lung, liver and kidney. Also expressed in embryonic kidney cells.
Involvement in disease	Defects in TSC1 are the cause of tuberous sclerosis type 1 (TSC1) [MIM:191100]. It is an autosomal dominant multi-system disorder that affects especially the brain, kidneys, heart, and skin. TS1C is characterized by hamartomas (benign overgrowths predominantly of a cell or tissue type that occurs normally in the organ) and hamartias (developmental abnormalities of tissue combination). Clinical symptoms can range from benign hypopigmented macules of the skin to profound mental retardation with intractable seizures to premature death from a variety of disease-associated causes. Defects in TSC1 may be a cause of focal cortical dysplasia of Taylor balloon cell type (FCDBC) [MIM:607341]. FCDBC is a subtype of cortical displasias linked to chronic intractable epilepsy.

	Cortical dysplasias display a broad spectrum of structural changes, which appear to result from changes in proliferation, migration, differentiation, and apoptosis of neuronal precursors and neurons during cortical development.
Domain	The C-terminal putative coiled-coil domain is necessary for interaction with TSC2.
Post-translational modifications	Phosphorylation at Ser-505 does not affect interaction with TSC2. Phosphorylated upon DNA damage, probably by ATM or ATR.
Cellular localization	Cytoplasm. Membrane. At steady state found in association with membranes.

Images



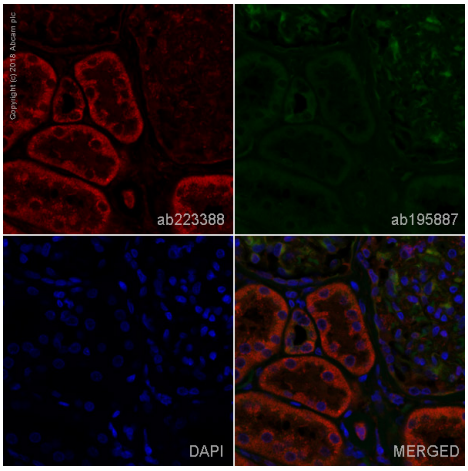
Flow Cytometry (Intracellular) - Alexa Fluor® 647 Anti-Hamartin antibody [EP318Y] (ab223388)

Overlay histogram showing HeLa cells stained with ab223388 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS / 10% normal goat serum to block non-specific protein-protein interactions followed by the antibody (ab223388, 1/5000 dilution) for 30 min at 22°C.

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

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

This antibody gave a positive signal in HeLa cells fixed with 4% formaldehyde (10 min)/permeabilized with 0.1% PBS-Triton X-100 for 15 min used under the same conditions.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Alexa Fluor® 647 Anti-Hamartin antibody [EP318Y] (ab223388)

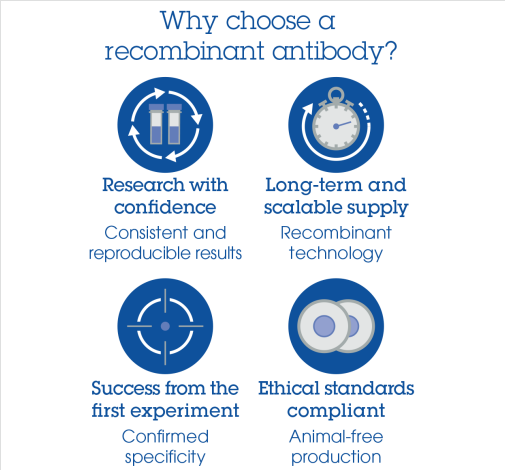
IHC image of Hamartin staining in a section of formalin-fixed paraffin-embedded normal human kidney*.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20mins performed on a Leica BOND™. Non-specific protein-protein interactions were then blocked in TBS containing 0.025% (v/v) Triton X-100, 0.3M (w/v) glycine and 1% (w/v) BSA for 1h at room temperature. The section was then incubated overnight at +4°C in TBS containing 0.025% (v/v) Triton X-100 and 1% (w/v) BSA with ab223388 at 1/100 dilution (shown in red) and counterstained using **ab195887**, Mouse monoclonal to alpha Tubulin (Alexa Fluor® 488), at 1/250 dilution (shown in green). Nuclear DNA was labelled with DAPI (shown in blue). The section was then mounted using Fluoromount®.

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

For other IHC staining systems (automated and non-automated), customers should optimize variable parameters such as antigen retrieval conditions, antibody concentrations and incubation times.

*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre.



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