

Product datasheet

Alexa Fluor® 488 Anti-Ki67 antibody [EPR3610] ab197234

KO VALIDATED Recombinant RabMAb

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Overview

Product name	Alexa Fluor® 488 Anti-Ki67 antibody [EPR3610]
Description	Alexa Fluor® 488 Rabbit monoclonal [EPR3610] to Ki67
Conjugation	Alexa Fluor® 488. Ex: 495nm, Em: 519nm
Tested applications	Suitable for: ICC/IF, Flow Cyt (Intra)
Species reactivity	Reacts with: Human Does not react with: Mouse, Rat
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	ICC/IF: HeLa cells, HAP1 WT and HAP1-Ki67 KO. Flow Cyt (intra): HeLa cells.
General notes	<p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none">- High batch-to-batch consistency and reproducibility- Improved sensitivity and specificity- Long-term security of supply- Animal-free production <p>For more information see here.</p> <p>Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.</p> <p>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: (i) to provide a service, information, or data in return for payment (ii) for therapeutic, diagnostic or prophylactic purposes; or (iii) 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.</p>

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: PBS, 30% Glycerol (glycerin, glycerine), 1% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR3610
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab197234 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		1/100. This product gave a positive signal in HeLa cells fixed with 4% formaldehyde (10 - 20 min) and 100% methanol (5 min). If fixing cells in 4% PFA, it is recommended to permeabilized cells with 0.1% Triton-X for 5 min.
Flow Cyt (Intra)		1/50.

Target

Function	Required to maintain individual mitotic chromosomes dispersed in the cytoplasm following nuclear envelope disassembly (PubMed:27362226). Associates with the surface of the mitotic chromosome, the perichromosomal layer, and covers a substantial fraction of the chromosome surface (PubMed:27362226). Prevents chromosomes from collapsing into a single chromatin mass by forming a steric and electrostatic charge barrier: the protein has a high net electrical charge and acts as a surfactant, dispersing chromosomes and enabling independent chromosome motility (PubMed:27362226). Binds DNA, with a preference for supercoiled DNA and AT-rich DNA (PubMed:10878551). Does not contribute to the internal structure of mitotic chromosomes (By similarity). May play a role in chromatin organization (PubMed:24867636). It is however unclear whether it plays a direct role in chromatin organization or whether it is an indirect consequence of its function in maintaining mitotic chromosomes dispersed.
Sequence similarities	Contains 1 FHA domain. Contains 16 K167R repeats. Contains 1 PP1-binding domain.
Developmental stage	Expression occurs preferentially during late G1, S, G2 and M phases of the cell cycle, while in

cells in G0 phase the antigen cannot be detected (at protein level) (PubMed:6206131). Present at highest level in G2 phase and during mitosis (at protein level). In interphase, forms fiber-like structures in fibrillar-deficient regions surrounding nucleoli (PubMed:2674163, PubMed:8799815).

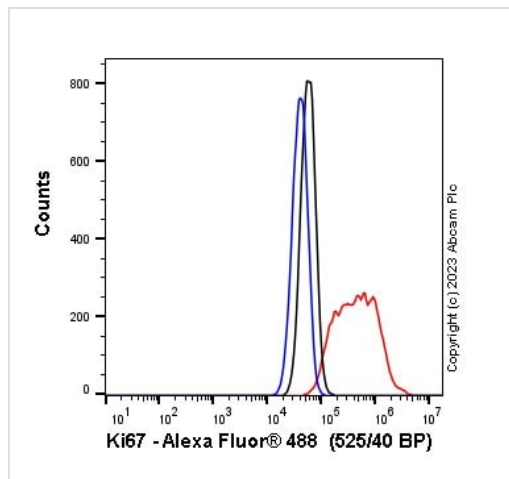
Post-translational modifications

Phosphorylated. Hyperphosphorylated in mitosis (PubMed:10502411, PubMed:10653604). Hyperphosphorylated form does not bind DNA.

Cellular localization

Chromosome. Nucleus. Nucleus, nucleolus. Associates with the surface of the mitotic chromosome, the perichromosomal layer, and covers a substantial fraction of the mitotic chromosome surface (PubMed:27362226). Associates with satellite DNA in G1 phase (PubMed:9510506). Binds tightly to chromatin in interphase, chromatin-binding decreases in mitosis when it associates with the surface of the condensed chromosomes (PubMed:15896774, PubMed:22002106). Predominantly localized in the G1 phase in the perinucleolar region, in the later phases it is also detected throughout the nuclear interior, being predominantly localized in the nuclear matrix (PubMed:22002106).

Images

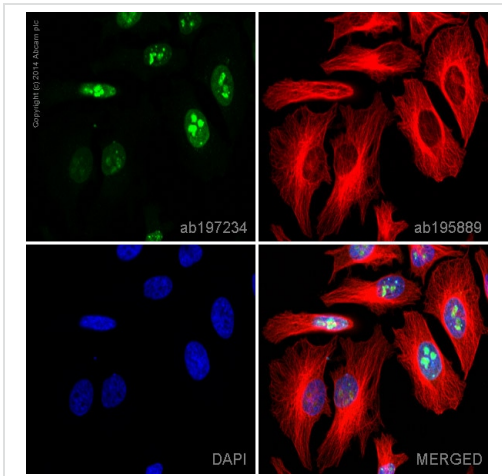


Flow Cytometry (Intracellular) - Alexa Fluor® 488
Anti-Ki67 antibody [EPR3610] (ab197234)

Flow cytometry overlay histogram showing HeLa cells stained with ab197234 (red line). The cells were fixed with 4% formaldehyde (10 min) and then permeabilised with 0.1% PBS-Triton X-100 for 15 min. The cells were incubated in 1x PBS containing 10% normal goat serum to block non-specific protein-protein interaction followed by the antibody (ab197234) (1×10^6 in 100µl at 1µg/ml (1/500)) for 30min at 22°C.

Isotype control antibody (black line) was Alexa Fluor® 488 Rabbit IgG, monoclonal [EPR25A] - Isotype Control used at the same concentration and conditions as the primary antibody. Unlabelled sample (blue line) was also used as a control.

Acquisition of >5000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter.

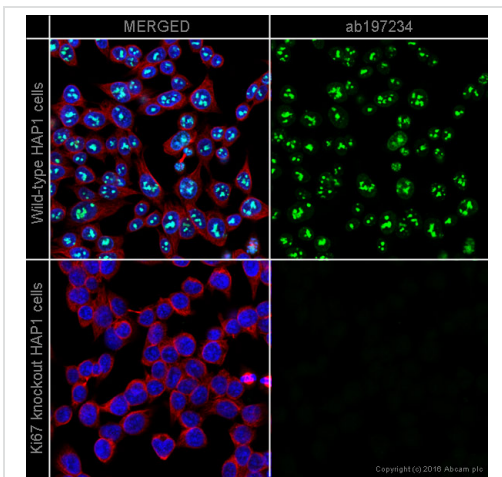


Immunocytochemistry/ Immunofluorescence - Alexa Fluor® 488 Anti-Ki67 antibody [EPR3610] (ab197234)

ab197234 staining Ki67 in HeLa cells. The cells were fixed with 4% formaldehyde (10 min), 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 ab197234 at a 1/100 dilution (shown in green) and **ab195889**, Mouse monoclonal to alpha Tubulin (Alexa Fluor® 594), at a 1/250 dilution (shown in red). Nuclear DNA was labelled 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 (5 min).



Immunocytochemistry/ Immunofluorescence - Alexa Fluor® 488 Anti-Ki67 antibody [EPR3610] (ab197234)

ab197234 staining Ki67 in wild-type HAP1 cells (top panel) and Ki67 knockout HAP1 cells (bottom panel). The cells were fixed with 100% methanol (5min), 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 with ab197234 at 1/100 dilution (shown in green) and **ab195889** at 1/250 dilution (shown in pseudo colour red) overnight at +4°C. Nuclear DNA was labelled in blue with DAPI.

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

Why choose a recombinant antibody?



Research with confidence
Consistent and reproducible results



Long-term and scalable supply
Recombinant technology



Success from the first experiment
Confirmed specificity



Ethical standards compliant
Animal-free production

Alexa Fluor® 488 Anti-Ki67 antibody [EPR3610]
(ab197234)

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