abcam

ab215226

Alexa Fluor® 555 Anti-Ki67 antibody [EPR3610]

Recombinant | RabMAb | KO Validated

Key facts

Isotype	IgG
Host species	Rabbit
Conjugation	Alexa Fluor® 555
Excitation/Emission	Ex: 555nm, Em: 565nm
Storage buffer	pH: 7.4 Preservative: 0.02% Sodium azide Constituents: PBS, 30% Glycerol (glycerin, glycerine), 1% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EPR3610
Purification technique	Affinity purification Protein A
Concentration	0.5 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

ICC

Tested

Species	Human

Dilution info	1/100 - 1/500
Notes	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)

Tested

Species	Human
Dilution info	1/500
Notes	-

ICC/IF

Expected

Species	Human
Dilution info	Use at an assay dependent concentration.
Notes	-

IHC-P

Expected

Species	Human
Dilution info	Use at an assay dependent concentration.
Notes	-

Target data

See full target information MKI67

Function	Protein that associates with the surface of mitotic chromosomes and acts both as a chromosome repellent during early mitosis and chromosome attractant during late mitosis (PubMed:27362226, PubMed:32879492, PubMed:35513709, PubMed:39153474). Required to maintain individual mitotic chromosomes dispersed in the cytoplasm following nuclear envelope disassembly (PubMed:27362226). During early mitosis, relocalizes from nucleoli to the chromosome surface where it forms extended brush structures that cover a

substantial fraction of the chromosome surface (PubMed:27362226). The MKI67 brush structure 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). During mitotic anaphase, the MKI67 brush structure collapses and MKI67 switches from a chromosome repellent to a chromosome attractant to promote chromosome clustering and facilitate the exclusion of large cytoplasmic particles from the future nuclear space (PubMed:32879492, PubMed:39153474). Mechanistically, dephosphorylation during mitotic exit and simultaneous exposure of a conserved basic patch induce the RNA-dependent formation of a liquid-like condensed phase on the chromosome surface, promoting coalescence of neighboring chromosome surfaces and clustering of chromosomes (PubMed:39153474). Binds premature ribosomal RNAs during anaphase; promoting liquid-liquid phase separation (PubMed:28935370, PubMed:39153474). 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; it is however unclear whether it plays a direct role in chromatin organization or whether it is an indirect consequence of its function in mitotic chromosome (PubMed:24867636).

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle, Store in the dark

Notes

Patented technology
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?
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 batch production

For more information, read more on recombinant antibodies.

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Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	Ki67 is a nuclear protein often referred to as MKI67 with a molecular weight of approximately 345 kDa. This protein is strongly associated with cell proliferation. Scientists commonly use Ki67 staining techniques to identify and quantify proliferating cells in tissues. Ki67 is expressed in the nucleus during active cell cycle phases—G1 S G2 and M—but not in resting cells in G0 phase. It is detectable through methods such as Ki67 immunofluorescence and Ki67 flow cytometry which are important for analyzing cell division.
Biological function summary	The Ki67 protein plays an essential role in cellular proliferation processes. It does not form part of a stable complex but interacts transiently with other cell cycle-related proteins. Research indicates that Ki67 maintains the structure of the perichromosomal layer during mitosis impacting chromosome separation. It is particularly active in tissues with high cell turnover such as bone marrow and lymphoid organs.
Pathways	Ki67 influences several key cellular pathways that control cell proliferation and differentiation. The protein acts within the regulation of the cell cycle and the PI3K/AKT signaling pathway important for cell growth and survival. Within these pathways Ki67 interacts with proteins like cyclin-dependent kinases (CDKs) which regulate transitions between different phases of the cell cycle.
Associated diseases and disorders	Ki67 has significant implications in oncology and can be used as a biomarker for cancer prognosis. Its expression levels help determine the aggressiveness of tumors especially in breast cancer and prostate cancer. High Ki67 levels correlate with poor prognosis as it indicates rapid cell division. In cancer Ki67 associates with p53 a protein that regulates the cell cycle and function as a tumor suppressor.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

We do not recommend this combination. It is not covered by our product promise.

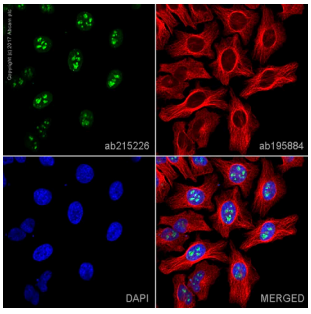
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In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

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3 product images

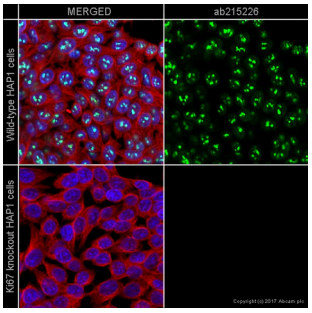


Immunocytochemistry - Alexa Fluor® 555 Anti-Ki67 antibody [EPR3610] (ab215226)

Ki67 Immunocytochemistry staining using rabbit Anti-Ki67 antibody

ab215226 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 ab215226 at 1/100 dilution (**pseudocolored in green**) and ab195884, Rat monoclonal to Tubulin (Alexa Fluor® 647), at 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).

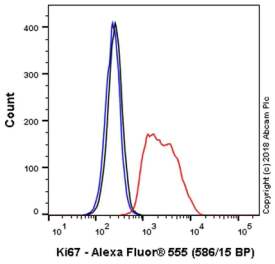
This product also gave a positive signal under the same testing conditions in HeLa cells fixed with 100% methanol (5 min).



Immunocytochemistry - Alexa Fluor® 555 Anti-Ki67 antibody [EPR3610] (ab215226)

Ki67 Immunocytochemistry staining using rabbit Anti-Ki67 antibody

ab215226 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 overnight at +4°C with ab215225 at a 1/500 dilution (**pseudocolored in green**) and ab195884, Rat monoclonal [YOL1/34] to Tubulin (Alexa Fluor® 647), at a 1/250 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue). This product also gave a positive signal under the same testing conditions in HAP1 cells fixed with 4% formaldehyde (10 min).
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



Flow Cytometry (Intracellular) - Alexa Fluor® 555 Anti-Ki67 antibody [EPR3610] (ab215226)

Ki67 Flow Cytometry (Intracellular) staining using rabbit Anti-Ki67 antibody

Overlay histogram showing HeLa cells stained with ab215226 (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 (ab215226, 1/500 dilution) for 30 min at 22°C.
Isotype control antibody (black line) was Rabbit IgG (monoclonal) Alexa Fluor® 555 (ab208569) 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 50 mW Yellow/Green laser (561nm) and 586/15 bandpass filter.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.