abcam

ab231172

Anti-Ki67 antibody [SP6] - BSA free

Anti-Ki67 antibody [SP6] - BSA free (ab231172) is a rabbit monoclonal antibody detecting Ki67 in **Western Blot, Flow Cytometry (Intra), IHC-P, ICC/IF, mIHC**. Suitable for **Human, Mouse, Rat**.

- Clone SP6 is the most cited clone to Ki67
- KO validated for confirmed specificity
- BSA free for higher signal-to noise ratio
- Multiplex IHC validated on the Leica BOND® MAX using Opal reagents

Recombinant | RabMAb | Advanced Validation | KO Validated

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.02% Sodium azide Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	SP6
Purification technique	Affinity purification
Epitope	C-terminus
Concentration	1.074 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

mIHC

Tested

Species	Human
Dilution info	-
Notes	Perform heat-mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Common marmoset
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	-
Notes	If fixing cells in 4% PFA (10 min, room temp), it is recommended to permeabilized cells with 0.1% Triton-X for 5 min.

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Common marmoset
Dilution info	-

Notes	-

WB

Tested

Species	Human
Dilution info	-
Notes	-

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Common marmoset
Dilution info	-
Notes	-

IHC-P

Tested

Species	Mouse
Dilution info	-
Notes	Antigen retrieval: Heat mediated antigen retrieval with sodium citrate buffer (pH 6.0).

Species	Rat
Dilution info	-
Notes	Antigen retrieval: Heat mediated antigen retrieval with sodium citrate buffer (pH 6.0).

Species	Human

Dilution info	-
Notes	Antigen retrieval: Heat mediated antigen retrieval with sodium citrate buffer (pH 6.0).

Predicted

Species	Common marmoset
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-
Notes	ab172730, Rabbit monoclonal isotype, is suitable for use as an isotype control with this antibody.

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Common marmoset
Dilution info	-
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 conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

What is this antibody validated in?
Anti-Ki67 antibody [SP6] - BSA free (ab231172) is a rabbit recombinant monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Intra), Flow Cytometry (Flow Cyt), Immunohistochemistry (IHC-P), Immunocytochemistry/immunofluorescence (ICC/IF), Multiplex IHC (mIHC) in Human, Mouse, Rat samples.

What is the molecular weight of Ki67?
Anti-Ki67 [SP6] - BSA free (ab231172) specifically detects a band for Ki67 (UniProt: P46013) at a molecular weight of 358kDa.

Collaboration
Anti-Ki67 [SP6] - BSA free (ab231172) is a clone from the portfolio of Spring Bioscience (Roche) SP clones which have

been optimised for immunohistochemistry (IHC).

Trusted by the scientific community
Anti-Ki67 [SP6] - BSA free (ab231172) was first used in a scientific publication in 2019 and has been cited over 20 times in peer-reviewed journals.

Specificity confirmed
The specificity of Anti-Ki67 antibody [SP6] - BSA free (ab231172) has been confirmed by Flow Cytometry (Intracellular) testing in MKI67 Knockout HAP1 cells.

Other related products
We have a range of other formats of antibody clone [SP6] also available for your convenience: ab16667, ab21700, Carrier free - ab197547, ab231172, ab238624, Oligonucleotide - ab279361, Alexa Fluor® 488 - ab281847, Alexa Fluor® 647 - ab281928, PE - ab282173, APC - ab314285, ab320709, Carrier free - ab320710, ab323292

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.

Compatibility
This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

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

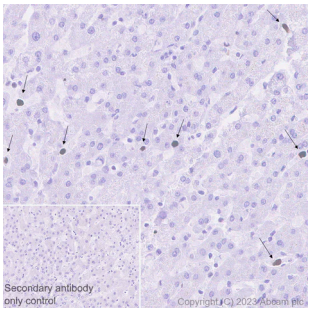
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

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.

10 product images

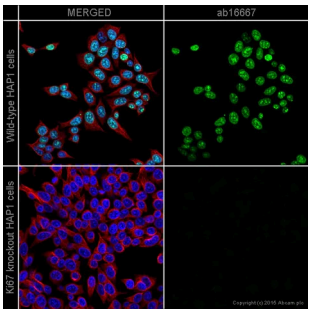


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ki67 antibody [SP6] - BSA free (ab231172)

Ki67 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human liver tissue using rabbit Anti-Ki67 antibody

This data was developed using the same antibody clone in a different buffer formulation (ab16667). Immunohistochemical analysis of paraffin-embedded Human liver tissue labeling Ki67 with ab16667 at 1/200 (0.145 µg/ml) followed by a ready to use LGoat Anti-Rabbit IgG H&L (HRP polymer) secondary antibody (ab214880). Low expression: positive staining only on proliferating cells (arrow) of human liver. The section was incubated with ab16667 at 4°C overnight and

counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP polymer) (ab214880). Heat mediated antigen retrieval was performed using ab93678 (citrate buffer, pH 6.0).

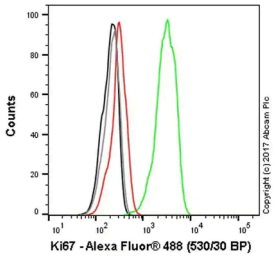


Immunocytochemistry/ Immunofluorescence - Anti-Ki67 antibody [SP6] - BSA free (ab231172)

Ki67 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-Ki67 antibody

This data was developed using the same antibody clone in a different buffer formulation (ab16667).

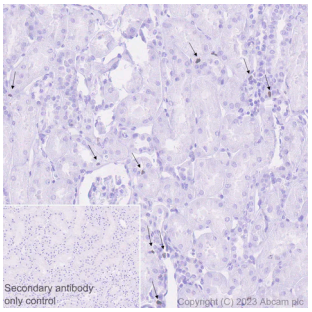
ab16667 staining Ki67 in wild-type HAP1 cells (top panel) and Ki67 knockout HAP1 cells (bottom panel). The cells were fixed with 100% methanol for 5 minutes, permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1 hour. The cells were then incubated with ab16667 at 1/250 dilution and ab195889 at 1/250 dilution (shown in pseudo colour red) overnight at +4°C, followed by a further incubation at room temperature for 1 hour with Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed (ab150081) secondary antibody at 2 µg/ml (shown in green). Nuclear DNA was labelled in blue with DAPI.



Flow Cytometry (Intracellular) - Anti-Ki67 antibody [SP6] - BSA free (ab231172)

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

This data was developed using the same antibody clone in a different buffer formulation (ab16667).
Overlay histogram showing HAP1 wildtype (green line) and HAP1-MKI67 knockout cells (red line) stained with ab16667. 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 (ab16667, 1/1000) for 30 min at 22°C. The secondary antibody used wasGoat Anti-Rabbit IgG H&L (Alexa Fluor®488) preadsorbed (ab150081) secondary antibodyat 1/2000 dilution for 30 min at 22°C. A Rabbit IgG isotype control antibody (ab172730) was used at the same concentration and conditions as the primary antibody (HAP1 wildtype - black line, HAP1-MKI67 knockout - grey line). Unlabelled sample was also used as a control (this line is not shown for the purpose of simplicity). Acquisition of >5,000 events were collected using a 50 mW Blue laser (488nm) and 530/30 bandpass filter. This image was generated from the hybridoma version.

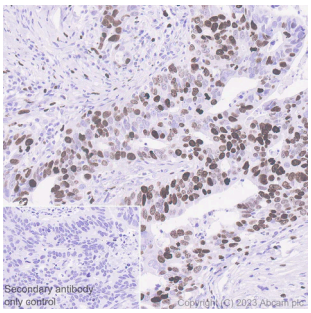


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ki67 antibody [SP6] - BSA free (ab231172)

Ki67 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human kidney tissue using rabbit Anti-Ki67 antibody

This data was developed using the same antibody clone in a different buffer formulation (ab16667). Immunohistochemical analysis of paraffin-embedded Human kidney tissue labeling Ki67 with ab16667 at 1/200 (0.145 µg/ml) followed by a ready to use LGoat Anti-Rabbit IgG H&L (HRP polymer) secondary antibody (ab214880). Low expression: positive staining only on proliferating cells (arrow) of human kidney. The section was incubated with ab16667 at 4°C overnight and counterstained with Hematoxylin. Secondary antibody only control: Secondary

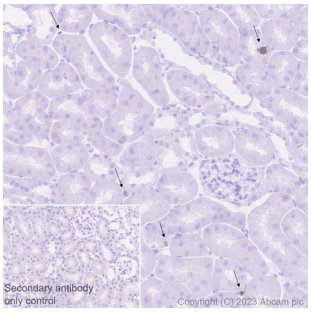
antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP polymer) (ab214880). Heat mediated antigen retrieval was performed using ab93678 (citrate buffer, pH 6.0).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ki67 antibody [SP6] - BSA free (ab231172)

Ki67 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human colon carcinoma tissue using rabbit Anti-Ki67 antibody

This data was developed using the same antibody clone in a different buffer formulation (ab16667). IHC image of ab16667 staining Ki67 in a section of formalin-fixed paraffin-embedded Human colon carcinoma tissue. The section incubated with ab16667 at 1/200 (0.145 µg/ml) dilution and ready to use Goat Anti-Rabbit IgG H&L (HRP polymer) (ab214880) was used as secondary antobody. Positive staining on human colon carcinoma. The section was incubated with ab16667 at 4°C overnight. The section was counterstained with haematoxylin. Heat mediated antigen retrieval was performed using ab93678 (citrate buffer, pH 6.0).



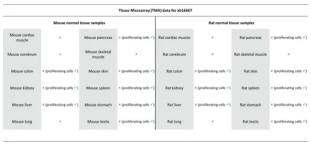
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ki67 antibody [SP6] - BSA free (ab231172)

Ki67 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Mouse kidney tissue using rabbit Anti-Ki67 antibody

This data was developed using the same antibody clone in a different buffer formulation (ab16667).
Immunohistochemical analysis of paraffin-embedded Mouse kidney tissue labeling Ki67 with ab16667 at 1/200 (0.145 µg/ml) followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP polymer) secondary antibody (ab214880). Low expression: positive staining only on proliferating cells (arrow) of mouse kidney. The section was incubated with ab16667 at 4°C overnight and counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP polymer) (ab214880).

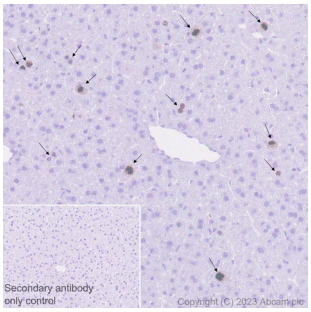
Heat mediated antigen retrieval was performed using ab93678 (citrate buffer, pH 6.0).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ki67 antibody [SP6] - BSA free (ab231172)

Ki67 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Ki67 antibody

Tissue Microarrays stained for Ki67 using ab16667 at 1/200 dilution (0.145 µg/ml) in immunohistochemical analysis. This table provides a detailed overview of positive (tick mark) and negative (cross mark) staining per sample type tested. The sections were pre-treated using Heat mediated antigen retrieval using ab93678 (citrate buffer, pH 6.0). The sections were incubated with ab16667 at +4°C overnight followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP polymer) (ab214880). Hematoxylin was used as the counterstain.
This data was developed using the same antibody clone in a different buffer formulation (ab16667).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ki67 antibody [SP6] - BSA free (ab231172)

Ki67 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Mouse liver tissue using rabbit Anti-Ki67 antibody

This data was developed using the same antibody clone in a different buffer formulation (ab16667).
Immunohistochemical analysis of paraffin-embedded Mouse liver tissue labeling Ki67 with ab16667 at 1/200 (0.145 µg/ml) followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP polymer) secondary antibody (ab214880). Low expression: positive staining only on proliferating cells (arrow) of mouse liver. The section was incubated with ab16667 at 4°C overnight and counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP polymer) (ab214880).

Heat mediated antigen retrieval was performed using ab93678 (citrate buffer, pH 6.0).

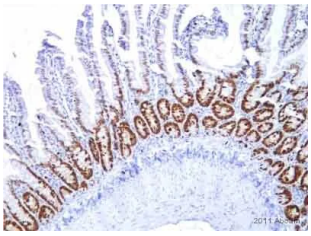


Image courtesy of Jing Ma by customer review.

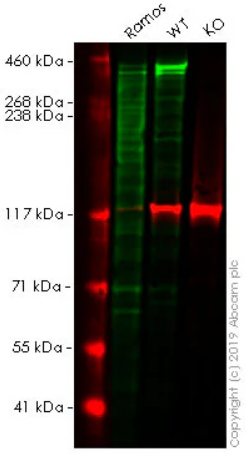
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ki67 antibody [SP6] - BSA free (ab231172)

Ki67 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Ki67 antibody

This data was developed using the same antibody clone in a different buffer formulation (ab16667).

ab16667 staining Ki67 in rat small intestine tissue by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections). Tissue was fixed with formaldehyde and a heat mediated antigen retrieval step was performed using 10 mM citrate buffer pH 6.0. Samples were then blocked with 10% serum for 20 minutes at room temperature followed by incubation with the undiluted primary antibody for 30 minutes. A biotin-conjugated goat anti-rabbit polyclonal was used as secondary antibody at a 1/2000 dilution.

This image was generated from the hybridoma version.



Western blot - Anti-Ki67 antibody [SP6] - BSA free (ab231172)

Ki67 Western blot staining using rabbit Anti-Ki67 antibody

This data was developed using the same antibody clone in a different buffer formulation (ab16667).
Lanes 1-3: Merged signal (red and green). Green - ab16667 observed at 359 kDa. Red - loading control, ab130007 observed at 125 kDa.

ab16667 was shown to react with Ki67 in wild-type HeLa. Loss of signal was observed when knockout sample ab263762 was used. Wild-type and Ki67 knockout samples were subjected to SDS-PAGE. ab16667 and Anti-Vinculin antibody VIN-54] (ab130007) were incubated overnight at 4°C at 1 in 100 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-Ki67 antibody [SP6] (ab16667) at 1/100 dilution

Lane 1:
Ramos cell lysate at 20 µg

Lane 2:
Wild-type HeLa cell lysate at 20 µg

Lane 3:
Western blot - Human MKI67 (Ki67) knockout HeLa cell lysate (ab263762) at 20 µg

Secondary

All lanes:
Western blot - Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/20000 dilution

Performed under reducing conditions.

Predicted band size: 358 kDa

Observed band size: 124 kDa, 359 kDa

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