abcam

ab279657

Anti-Ki67 antibody [B56] - BSA and Azide free

Anti-Ki67 antibody [B56] - BSA and Azide free (ab279657) is a mouse recombinant monoclonal antibody provided in a PBS only buffer for easy conjugation detecting Ki67 in **Flow Cytometry (Intra), IHC-P, ICC/IF**. Suitable for **Human, Mouse, Rat**.

- KO validated for confirmed specificity
- BSA, sodium azide, and glycerol-free for easy conjugation
- Biophysical QC for unrivalled batch-batch consistency

Recombinant | KO Validated

Key facts

Isotype	IgG1
Host species	Mouse
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	B56
Purification technique	Affinity purification Protein A
Light chain type	kappa
Concentration	1.013 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Mouse, Rat, Human
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	-
Notes	-

Expected

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

WB

Not recommended

Species	Mouse, Rat, Human
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-
Notes	-

Expected

Species	Mouse, Rat
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 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 [B56] - BSA and Azide free (ab279657) is a mouse recombinant monoclonal antibody and is validated for use in Flow Cytometry (Intra), Flow Cytometry (Flow Cyt), Immunohistochemistry (IHC-P), Immunocytochemistry/immunofluorescence (ICC/IF) in Human, Mouse, Rat samples.

Specificity confirmed
The specificity of Anti-Ki67 antibody [B56] - BSA and Azide free (ab279657) has been confirmed by Flow Cytometry testing in MKI67 Knockout HeLa cells.

Other related products
We have a range of other formats of antibody clone [B56] also available for your convenience: ab279653, Carrier free - ab279657, Alexa Fluor® 488 - ab283242, Alexa Fluor® 647 - ab283699

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.

Conjugation ready
Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with 1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

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

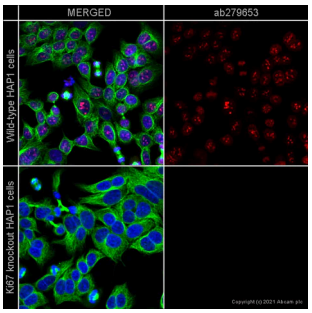
We do not recommend this combination. It is not covered by our product promise.

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.

15 product images



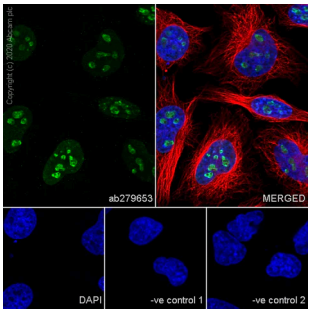
Immunocytochemistry/ Immunofluorescence - Anti-Ki67 antibody [B56] - BSA and Azide free (ab279657)

Ki67 Immunocytochemistry/ Immunofluorescence staining using mouse Anti-Ki67 antibody

This data was developed using ab279653 the same antibody clone in a different buffer formulation.
ab279653 staining Ki67 in wild-type Hap1 cells, with negative expression in Ki67 knockout Hap1 cells. The cells were fixed with 100% methanol (5 min), permeabilised 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 ab279653 at 5 µg/ml and ab6046, Rabbit polyclonal to beta Tubulin at 1 µg/ml. Cells were then incubated with ab150081, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488), pre-adsorbed at 1/1000 dilution (shown in green) and ab150119, Goat polyclonal Secondary Antibody to Mouse IgG - H&L (Alexa Fluor® 647), pre-adsorbed at 1/1000 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).

Image was acquired with a confocal microscope (Leica-Microsystems TCS SP8) and a single confocal section is shown.

This product also work with 4% formaldehyde (10 min) fixation under the same testing conditions.

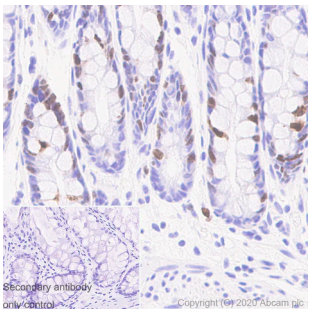


Immunocytochemistry/ Immunofluorescence - Anti-Ki67 antibody [B56] - BSA and Azide free (ab279657)

Ki67 Immunocytochemistry/ Immunofluorescence staining using mouse Anti-Ki67 antibody

This data was developed using ab279653 the same antibody clone in a different buffer formulation.
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized HeLa (Human cervix adenocarcinoma epithelial cell) cells labelling Ki67 with ab279653 at 1/50 dilution (18.72 ug/ml), followed by ab150113 Goat Anti-Mouse IgG H&L (Alexa Fluor® 488) antibody at 1/1000 dilution (Green). Confocal image showing nucleolar staining in HeLa cell line. ab179513 Anti-beta Tubulin rabbit monoclonal antibody at 1/200 dilution followed by ab150080 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 594) at 1/500 dilution was used to counterstain tubulin (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is ab150113 Goat Anti-Mouse IgG H&L (Alexa Fluor® 488) at 1/1000 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ki67 antibody [B56] - BSA and Azide free (ab279657)

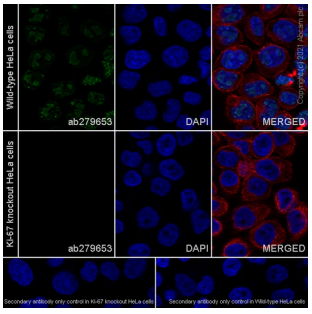
Ki67 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using mouse Anti-Ki67 antibody

This data was developed using ab279653 the same antibody clone in a different buffer formulation.
Immunohistochemical analysis of paraffin-embedded Human colon tissue labeling Ki67 with ab279653 at 1/1000 dilution (0.936 ug/ml) followed by a ready to use LeicaDS9800 (Bond™

Polymer Refine Detection). Nuclear staining on human colon. The section was incubated with ab279653 for 30 mins at room temperature and followed by mouse specific IgG antibody (ab125913) for 8 mins. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins.

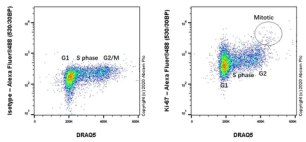


Immunocytochemistry/ Immunofluorescence - Anti-Ki67 antibody [B56] - BSA and Azide free (ab279657)

Ki67 Immunocytochemistry/ Immunofluorescence staining using mouse Anti-Ki67 antibody

This data was developed using ab279653 the same antibody clone in a different buffer formulation.
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized wild-type HeLa (Human cervix adenocarcinoma epithelial cell) cells and KI67 KO HeLa cells labelling Ki67 with ab279653 at 1/50 dilution (18.72 ug/ml), followed by ab150113 Goat Anti-Mouse IgG H&L (Alexa Fluor® 488) antibody at 1/1000 dilution (Green). Confocal image showing nucleolar staining in Parental HeLa cell line and no staining in MKI67 HeLa KO cell line. ab179513 Anti-beta Tubulin rabbit monoclonal antibody at 1/200 dilution followed by ab150080 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 594) at 1/500 dilution was used to counterstain tubulin (Red). The Nuclear counterstain was DAPI (Blue).

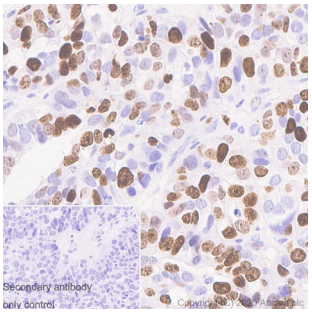
Secondary antibody only control: Secondary antibody is ab150113 Goat Anti-Mouse IgG H&L (Alexa Fluor® 488) at 1/1000 dilution.



Flow Cytometry (Intracellular) - Anti-Ki67 antibody [B56] - BSA and Azide free (ab279657)

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

This data was developed using ab279653 the same antibody clone in a different buffer formulation.
Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized HeLa (Human cervix adenocarcinoma epithelial cell) cells labelling Ki67 with ab279653 at 1/1000 dilution (0.1ug)/ Right compared with a Mouse monoclonal IgG / Left isotype control. A Goat anti mouse IgG (Alexa Fluor® 488, ab150113) at 1/2000 dilution was used as the secondary antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ki67 antibody [B56] - BSA and Azide free (ab279657)

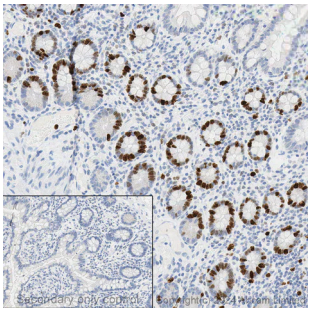
Ki67 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using mouse Anti-Ki67 antibody

This data was developed using ab279653 the same antibody clone in a different buffer formulation.
Immunohistochemical analysis of paraffin-embedded Human ovarian carcinoma tissue labeling Ki67 with ab279653 at 1/1000 dilution (0.936 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Nuclear staining on human ovarian carcinoma. The section was

incubated with ab279653 for 30 mins at room temperature and followed by mouse specific IgG antibody (ab125913) for 8mins. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ki67 antibody [B56] - BSA and Azide free (ab279657)

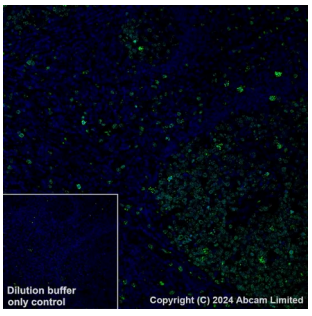
Ki67 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human colon using mouse Anti-Ki67 antibody

This data was developed using ab279653, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of formalin fixed paraffin embedded human colon labelling Ki67 with ab279653 at a concentration of 0.1µg/ml. The immunostaining was performed on a Ventana DISCOVERY ULTRA (Roche Tissue Diagnostics) instrument with a OptiView DAB IHC Detection Kit. Heat mediated antigen retrieval was performed with DISCOVERY cell conditioning solution (CC1) 100°C, pH8.5 for 32mins.

ab279653 Anti-Ki67 antibody [B56] was incubated for 16mins at 37°C. Sections were counterstained with Hematoxylin II. Image inset shows absence of staining in secondary antibody only control.

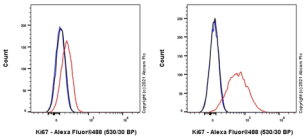
Customers are encouraged to optimise antigen retrieval conditions, antibody concentration, incubation times and temperature for best results in their own IHC assay workflow (automated and manual).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ki67 antibody [B56] - BSA and Azide free (ab279657)

This data was developed using ab279653, the same antibody clone in a different buffer formulation.

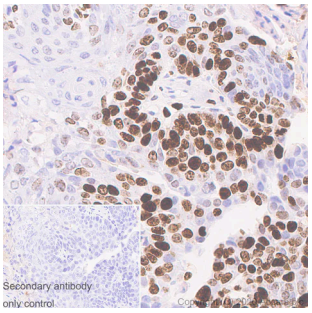
Immunohistochemical analysis of paraffin-embedded Human tonsil tissue labeling Ki67 with ab279653 at 1/1000 dilution. The section was incubated with ab279653 for 60 mins at room temperature. The section was then mounted using Fluoromount®. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8). Counterstained with DAPI. Secondary antibody only control: Goat Anti-Mouse IgG H&L (Alexa Fluor® 488) preadsorbed (ab150117) at 1/1000 dilution. Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution1) for 40 mins.



Flow Cytometry (Intracellular) - Anti-Ki67 antibody [B56] - BSA and Azide free (ab279657)

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

This data was developed using ab279653 the same antibody clone in a different buffer formulation.
Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized Parental HeLa (Human cervix adenocarcinoma epithelial cells) (Right)/ MKI67 KO HeLa cells (Left) labelling Ki67 with ab279653 at 1/1000 dilution (0.1ug) (Red) compared with a Mouse monoclonal IgG (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti mouse IgG (Alexa Fluor® 488, ab150113) at 1/2000 dilution was used as the secondary antibody.



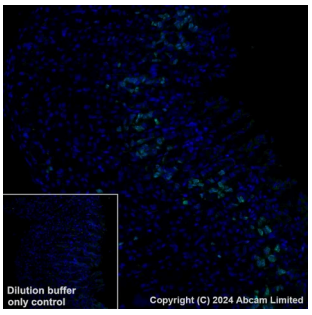
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ki67 antibody [B56] - BSA and Azide free (ab279657)

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

This data was developed using ab279653 the same antibody clone in a different buffer formulation.
Immunohistochemical analysis of paraffin-embedded Mouse breast carcinoma tissue labeling Ki67 with ab279653 at 1/1000 dilution (0.936 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Nuclear staining on mouse breast carcinoma. The section was incubated with ab279653 for 30 mins at room temperature and followed by mouse specific IgG antibody (ab125913) for 8 mins. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

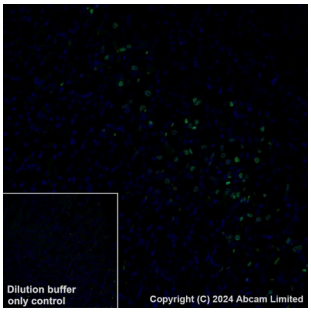
Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ki67 antibody [B56] - BSA and Azide free (ab279657)

This data was developed using ab279653, the same antibody clone in a different buffer formulation.

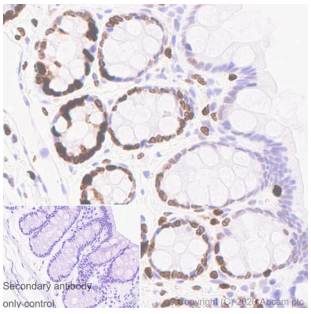
Immunohistochemical analysis of paraffin-embedded Mouse stomach tissue labeling Ki67 with ab279653 at 1/1000 dilution. The section was incubated with ab279653 for 60 mins at room temperature. The section was then mounted using Fluoromount®. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8). Counterstained with DAPI. Secondary antibody only control: Goat Anti-Mouse IgG H&L (Alexa Fluor® 488) preadsorbed (ab150117) at 1/1000 dilution. Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution1) for 40 mins.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ki67 antibody [B56] - BSA and Azide free (ab279657)

This data was developed using ab279653, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Rat stomach tissue labeling Ki67 with ab279653 at 1/1000 dilution. The section was incubated with ab279653 for 60 mins at room temperature. The section was then mounted using Fluoromount®. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8). Counterstained with DAPI. Secondary antibody only control: Goat Anti-Mouse IgG H&L (Alexa Fluor® 488) preadsorbed (ab150117) at 1/1000 dilution. Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution1) for 40 mins.



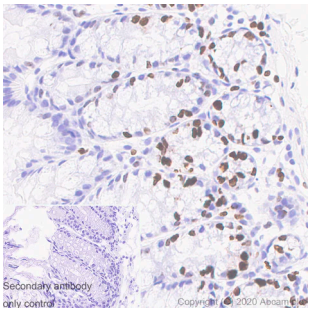
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ki67 antibody [B56] - BSA and Azide free (ab279657)

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

This data was developed using ab279653 the same antibody clone in a different buffer formulation.
Immunohistochemical analysis of paraffin-embedded Rat colon tissue labeling Ki67 with ab279653 at 1/1000 dilution (0.936 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Nuclear staining on rat colon. The section was incubated with ab279653 for 30 mins at room temperature and followed by mouse specific IgG antibody (ab125913) for 8 mins. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins.



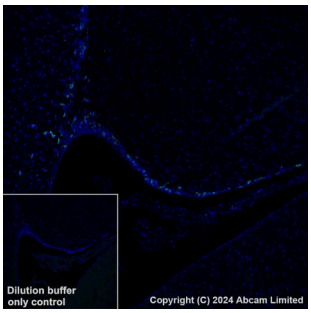
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ki67 antibody [B56] - BSA and Azide free (ab279657)

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

This data was developed using ab279653 the same antibody clone in a different buffer formulation.
Immunohistochemical analysis of paraffin-embedded Mouse colon tissue labeling Ki67 with ab279653 at 1/1000 dilution (0.936 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Nuclear staining on mouse colon. The section was incubated with ab279653 for 30 mins at room temperature and followed by mouse specific IgG antibody (ab125913) for 8 mins. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ki67 antibody [B56] - BSA and Azide free (ab279657)

This data was developed using ab279653, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Mouse cerebrum tissue labeling Ki67 with ab279653 at 1/1000 dilution. The section was incubated with ab279653 for 60 mins at room temperature. The section was then mounted using Fluoromount®. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8). Counterstained with DAPI. Secondary antibody only control: Goat Anti-Mouse IgG H&L (Alexa Fluor® 488) preadsorbed (ab150117) at 1/1000 dilution. Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution1) for 40 mins.

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