abcam

ab6326

Anti-BrdU antibody [BU1/75 (ICR1)] - Proliferation Marker

Anti-BrdU antibody [BU1/75 (ICR1)] - Proliferation Marker (ab6326) is a rat monoclonal antibody detecting BrdU in **Flow Cytometry (Intra), IHC-P, ICC/IF**.

- Over 1920 publications
- Trusted since 2002

Key facts

Isotype	IgG2a
Host species	Rat
Storage buffer	pH: 7.4 Preservative: 0.02% Sodium azide Constituents: PBS, 6.97% L-Arginine
Form	Liquid
Clonality	Monoclonal
Clone number	BU1/75 (ICR1)
Purification technique	Affinity purification Protein G
Specificity	This antibody reacts with BrdU in single stranded DNA, BrdU attached to a protein carrier or free BrdU. It detects nucleated cells in S-Phase which have had BrdU incorporated into their DNA. Also reacts with chlorodeoxyuridine but with reduced staining. The antibody does not react with thymidine. It has been reported in the literature that this antibody clone cross-reacts with Edu (PMID: 23272138) and some customers reported that it cross reacts with IdU.
Light chain type	kappa
Concentration	1 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Chemical
Dilution info	1-3 µg/mL
Notes	Perform heat-mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Flow Cyt (Intra)

Tested

Species	Chemical
Dilution info	1/25 - 1/200
Notes	Rat IgG2a, kappa monoclonal [RTK2758] (Low endotoxin, Azide Free) (ab18450), is suitable for use as an isotype control with this antibody.

ICC/IF

Tested

Species	Chemical
Dilution info	1/250
Notes	-

Target data

See full target information BrdU

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

Notes

Anti-BrdU antibody [BU1/75 (ICR1)] - Proliferation Marker (ab6326) is a rat monoclonal antibody and is validated for use in Flow Cyt (Intra), ICC/IF and IHC-P.

Anti-BrdU antibody [BU1/75 (ICR1)] - Proliferation Marker (ab6326) was first used in a scientific publication in 1985 and has been cited over 1925 times in peer reviewed journals. It's performance in IHC in mouse samples is trusted by the scientific community.

Abcam's high quality validation processes ensure Anti-BrdU antibody [BU1/75 (ICR1)] - Proliferation Marker (ab6326) has high sensitivity and specificity.

Anti-BrdU antibody [BU1/75 (ICR1)] - Proliferation Marker (ab6326) has 105 independent reviews from customers.

Anti-BrdU antibody [BU1/75 (ICR1)] - Proliferation Marker (ab6326) specifically detects BrdU (UniProt ID: N/A; Molecular weight: 0.3kDa) and is sold in 100 ug and 1 mg selling sizes.

Conjugation-ready, carrier free format available for antibody clone BU1/75 (ICR1) - ab264079.

Antibody clone BU1/75 (ICR1) is also available pre-conjugated to a variety of labels for your convenience - Alexa Fluor® 488, Alexa Fluor® 647, Alexa Fluor® 594, HRP, Alexa Fluor® 555 (ab2274, ab2275, ab2276, ab2257, ab22124).

Top cited antibody on the market for the proliferation marker BrdU with >2400 citations. ab6326 has over 80 5 star reviews. BrdU antibody is essential for accurately tracking cell proliferation and DNA synthesis in various research applications, including cancer and neurogenesis studies

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

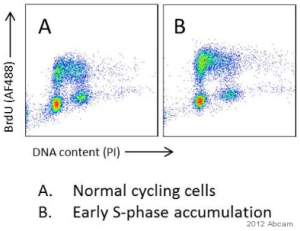
We do not recommend this combination. It is not covered by our product promise.

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.

9 product images

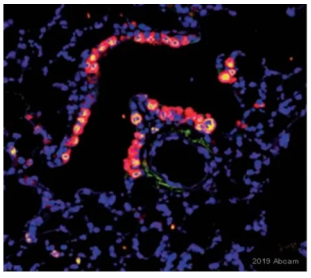


This image is courtesy of an anonymous customer review

Flow Cytometry (Intracellular) - Anti-BrdU antibody [BU1/75 (ICR1)] - Proliferation Marker (ab6326)

BrdU Flow Cytometry (Intracellular) staining of HeLa cells using rat Anti-BrdU antibody

ab6326 staining BrdU in HeLa cells by Flow Cytometry. Cells were incubated with 10 µM BrdU for 30 minutes prior to being harvested with 1X trypsin-EDTA washed twice in PBS containing 1% BSA and fixed in 70% ethanol (added drop-wise) for at least 30 minutes on ice. Once fixed pellets were acid denatured with HCl/Triton X-100 for 30 minutes at room temperature and then neutralised with sodium tetraborate.
Pelleted cells were re-suspended in Tween/BSA/PBS to which primary antibody was then added (0.1 µg in 0.5% Tween 20 (v/v) plus 1% BSA in PBSA) and incubated for 30 minutes at room temperature. Secondary Alexa Fluor® 488-conjugated Goat anti-Rat IgG (H+L) was used at 1/500 and incubated for 30 minutes at room temperature in the dark. Cells were pelleted once more and resuspended in PBS containing 5 µg/mL propidium iodide.
Gating Strategy: Based on forward and side scatter cells were gated into the region used for analysis.



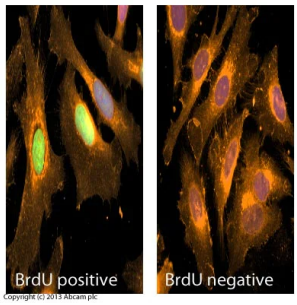
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Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BrdU antibody [BU1/75 (ICR1)] - Proliferation Marker (ab6326)

BrdU Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of mouse lung tissue using rat Anti-BrdU antibody

ab6326 staining BrdU in mouse lung tissue sections by Immunohistochemistry (Formalin/PFA-fixed parafin-embedded sections). Tissue samples were heat mediated with Citrate buffer for 40min. The sections were blocked with 10% donkey serum for 1 hour at 22°C. Samples were incubated with the primary antibody (1/200 in 10% Donkey serum-PBS 0.2% Triton) at 4°C for 16 hours. Cy™3 AffiniPure Donkey Anti-Rat IgG (H+L) (1/400 dilution) was used as the secondary antibody.
Representative confocal images of terminal bronchioles in mouse lungs taken at day 5 following naphthalene injection.

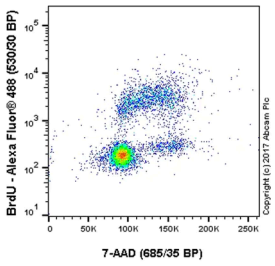
Colors label CC10 (red) FoxJ1 (green) BrdU (yellow) and nuclear DNA (blue).



Immunocytochemistry/ Immunofluorescence - Anti-BrdU antibody [BU1/75 (ICR1)] - Proliferation Marker (ab6326)

BrdU Immunocytochemistry/ Immunofluorescence staining using rat Anti-BrdU antibody

ICC/IF image of ab6326 stained HeLa cells both BrdU treated (left image) and normal cells (right image). The cells were 100% methanol fixed (5 min) and then subjected to acid hydrolysis using 2M HCL in 0.1% PBS-Tween for 30 minutes at room temperature to denature the DNA. They were then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab6326 10µg/ml) overnight at +4°C. The secondary antibody (green) was Goat Anti-Rat IgG H&L (DyLight® 488) preadsorbed (ab98420) used at a 1/250 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM. Positive staining can be seen in the BrdU treated cells but not in the normal cells demonstrating specificity for BrdU.



Flow Cytometry (Intracellular) - Anti-BrdU antibody [BU1/75 (ICR1)] - Proliferation Marker (ab6326)

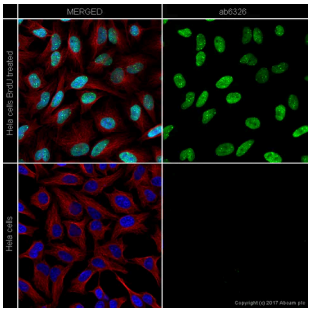
BrdU Flow Cytometry (Intracellular) staining of HeLa cells using rat Anti-BrdU antibody

Dot plot showing BrdU-treated HeLa cells stained with ab6326. Cells were incubated with 10 µM BrdU for 30 minutes prior to being harvested washed twice in 1x PBS and fixed in 70% ethanol (4°C added drop-wise) for at least 30 minutes on ice. Once fixed pellets were acid denatured with 2M HCl for 30 minutes at 22°C and then neutralised with borate buffer (0.1M pH8.5).

Samples were washed and incubated in 1x PBS / 10% normal goat serum to block non-specific protein-protein interactions followed by the antibody (ab6326 1µg/1x10^6 cells) for 30 min at 22°C. The secondary antibody used wasGoat Anti-Rat IgG H&L (Alexa Fluor® 488) preadsorbed (ab150165) at 1/2000 dilution for 30 min at 22°C.

7-AAD was added to cells 20 min prior to data acquisition.

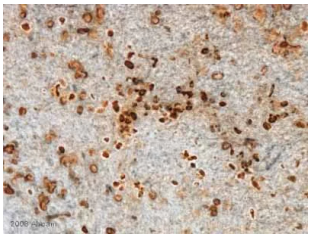
Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) with 530/30 and 685/35 bandpass filters.



Immunocytochemistry/ Immunofluorescence - Anti-BrdU antibody [BU1/75 (ICR1)] - Proliferation Marker (ab6326)

BrdU Immunocytochemistry/ Immunofluorescence staining of HeLa cells using rat Anti-BrdU antibody

ab6326 staining BrdU in Hela cells. Untreated and BrdU treated (10uM for 24 hours) cells. The cells were fixed with 100% methanol (5 min) and then subjected to acid hydrolysis using 2M HCL in 0.1% PBS-Tween for 30 minutes at room temperature to denature the DNA. They were then permeabilized with 0.1% PBS-Tween for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1%PBS-Tween for 1hr. The cells were then incubated overnight at 4°C with ab6326 at 1µg/ml and ab7291 Mouse monoclonal [DM1A] to alpha Tubulin - Loading Control. Cells were then incubated with ab150165 Goat polyclonal Secondary Antibody to Rat IgG - H&L (Alexa Fluor® 488) pre-adsorbed at 1/1000 dilution (shown in green) and ab150120 Goat polyclonal Secondary Antibody to Mouse IgG - H&L (Alexa Fluor® 594) pre-adsorbed at 1/1000 dilution (shown in pseudocolour 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.

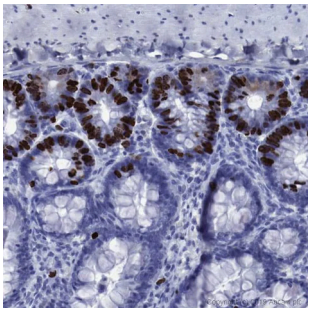


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Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BrdU antibody [BU1/75 (ICR1)] - Proliferation Marker (ab6326)

BrdU Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of rat brain tissue using rat Anti-BrdU antibody

ab6326 staining cultured cells of rat brain tissue by ICC. The sample was PFA fixed and permeabilized in 1M HCl prior to blocking with 5% serum for 1 hour at 25°C. The primary antibody was diluted 1/500 and incubated with the sample for 16 hours at 25°C. A biotinylated rabbit anti-rat IgG antibody diluted 1/200 was used as the secondary.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BrdU antibody [BU1/75 (ICR1)] - Proliferation Marker (ab6326)

BrdU Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of rat small intestine using rat Anti-BrdU antibody

IHC image of ab6326 staining in a formalin-fixed paraffin-embedded rat small intestine BrdU tissue section. The section was pre-treated using pressure cooker heat mediated antigen retrieval with sodium citrate buffer (pH6) for 30mins and incubated overnight at +4°C with ab6326 at 3 ug/ml. A goat anti-rat biotinylated secondary antibody was used to detect the primary and visualized using an HRP conjugated ABC system. The section was counterstained with haematoxylin and mounted with DPX.
For other IHC staining systems (automated and non-automated) users should optimize variable parameters such as antigen retrieval conditions primary antibody concentration and antibody incubation times.

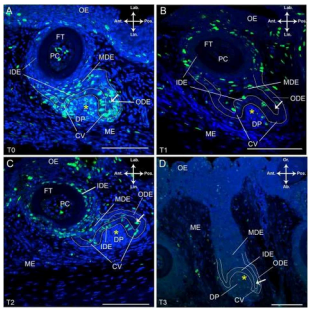


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Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BrdU antibody [BU1/75 (ICR1)] - Proliferation Marker (ab6326)

BrdU Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rat Anti-BrdU antibody

Immunohistological staining of frontal (A-C) and sagittal (D) sections through a tooth family on the lower jaw of *S. salar* with BrdU labelled cells in green and DAPI counterstain in blue. The tooth families presented are in a similar state of development: a young functional tooth and its successor in morphogenesis stage. (A) Pulsed specimen, T0; (B) Chase time T1, one week after BrdU administration; (C) Chase time T2, two weeks after BrdU administration and (D) Chase time T3, four weeks after BrdU administration. The white arrow indicates single cells within the middle dental epithelium (MDE) enclosed by the IDE, cervical loop and ODE of the replacement tooth. Abbreviations: Ab: aboral; Ant: anterior; CV: cervical loop; DP: dental papilla; FT: functional tooth; IDE: inner dental epithelium; Lab: labial; Lin: lingual; MDE: middle dental epithelium; ME: mesenchyme; OE: oral epithelium; ODE: outer dental epithelium; Or: oral; PC: pulp cavity; Pos: posterior; yellow asterisk: replacement tooth; scale bars: 100 µm.
Immunohistological staining for BrdU on the paraffin sections: Rehydration through a decreasing ethanol series, chromatin precipitation in hydrochloric acid, block in 3% BSA/ 1% milk powder, exposure to primary antibody (ab6326) and secondary antibody (polyclonal anti-rat Alexa Fluor® 488), DAPI counterstaining (1µl/ml). Immunofluorescence was visualized on a NIKON eclips TE2000-S confocal laser-scanning microscope. Adobe Illustrator CS5, Adobe Photoshop CS5 and Fiji were used to process bright field and fluorescent images of the immunostained sections.

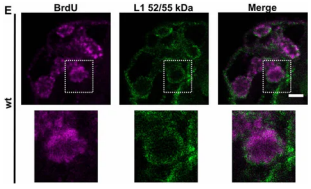


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Immunocytochemistry/ Immunofluorescence - Anti-BrdU antibody [BU1/75 (ICR1)] - Proliferation Marker (ab6326)

BrdU Immunocytochemistry/ Immunofluorescence staining using rat Anti-BrdU antibody

Confocal immunofluorescence sections (~0.3 µm thick) showing the localization of the different factors in HEK 293 cells infected with Ad5 wt. Double labeling for BrdU (ab6326, magenta) and L1 52/55 kDa (green). Dotted rectangles indicate the areas shown at higher magnification in the bottom row. Bars: 3 µm. Monolayer HEK293 cells grown in cover glasses were infected with Ad5 wt. The infection was synchronized by incubating the cells for 30 min at 4°C and then 30 min at 37°C. Then, the inoculums were removed and medium was added. For BrdU labeling, after 18 h at 37°C the medium was changed by medium containing 25 µg/ml BrdU (5-Bromo-2'-deoxyuridine), followed by another change at 25 hpi. Incubation with BrdU proceeded at 37°C. After 36 hpi, the medium was removed and 4% paraformaldehyde in PBS was added to the cells during 10 min. After 3 rinses with PBS, cover glasses were incubated with a mixture of 0.5% saponin and 10% FBS in PBS for 10 min. Samples were incubated with the primary antibody in 0.5% saponin and 2% FBS in PBS during 45 min. After three more rinses, incubation with secondary antibodies diluted in 0.5% saponin and 2% FBS in PBS was carried out in darkness. After a final rinse with PBS, cover glasses were mounted on glass slides. Antifade reagent was allowed to dry overnight before sample observation. All incubations were carried out at room temperature. Images were taken using a confocal multispectral Leica TCS SP5 system.

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