abcam

Product datasheet

Anti-BrdU antibody [BU1/75 (ICR1)] - BSA and Azide free ab264079

画像数 4

製品の概要

製品名 Anti-BrdU antibody [BU1/75 (ICR1)] - BSA and Azide free

製品の詳細 Rat monoclonal [BU1/75 (ICR1)] to BrdU - BSA and Azide free

由来種 Rat

特異性 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.

アプリケーション 適用あり: Flow Cyt (Intra), IHC-P, ICC/IF

種交差性 交差種: Species independent

免疫原 The details of the immunogen for this antibody are not available.

ポジティブ・コントロール ICC/IF: HeLa cells; Flow Cyt (Intra): HeLa cells; IHC-P: Rat small intestine tissue.

特記事項 ab264079 is the carrier-free version of <u>ab6326</u>.

Protocol advice:

This antibody recognizes single stranded DNA so the DNA needs to be unraveled first. This can be done with DNAse, although it doesn't give the best results. Depending on the assay, acid denaturation with 2M HCL or heat denaturation are the most successful. Please note this step is critical in any assay with this antibody and is the area that should be modified to optimize results. A detailed BrdU staining protocol is available in the Protocols tab or by clicking on this <u>link</u>.

Unstained positive control slides from mice treated with BrdU (formalin-fixed, paraffin-embedded intestine sections) are available as BrdU control slides <u>ab129956</u>.

AF488 conjugate available as <u>ab220074</u> AF647 conjugate available as <u>ab220075</u>

Please see the Associated Products tab for other available conjugates.

This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or conjugation for your experiments, please contact **orders@abcam.com**.

Our <u>carrier-free</u> antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes,

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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.

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.

The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

製品の特性

製品の状態 Liquid

保存方法 Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -

80°C. Do Not Freeze.

バッファー pH: 7.40

Constituent: PBS

BU1/75 (ICR1)

キャリア・フリー はい

精製度 IgG fraction モノクローナル

アイソタイプ IgG2a 軽鎖の種類 kappa

アプリケーション

クローン名

The Abpromise guarantee <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab264079の使用に適用されます アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
Flow Cyt (Intra)		1/25 - 1/200. Rat lgG2a, kappa monoclonal [RTK2758] (Low endotoxin, Azide Free) (ab18450), is suitable for use as an isotype control with this antibody.
IHC-P		Use a concentration of 1 - 3 μ g/ml. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

アプリケーション	Abreviews	特記事項
ICC/IF		1/250.

ターゲット情報

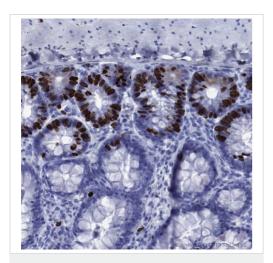
関連性

The immunocytochemical detection of bromodeoxyuridine (BrdU) incorporated into DNA is a powerful tool to study the cytokinetics of normal and neoplastic cells. In vitro or in vivo labeling of tumor cells with the thymidine analogue BrdU and the subsequent detection of incorporated BrdU with specific anti-BrdU monoclonal antibodies is an accurate and comprehensive method to quantitate the degree of DNA-synthesis. BrdU is incorporated into the newly synthezised DNA of S-phase cells may provide an estimate for the fraction of cells in S-phase. Also dynamic proliferative information such as the S-phase transit rate and the potential doubling time can be obtained, by means of bivariate BrdU/DNA flow cytometric analysis.

細胞内局在

Nuclear

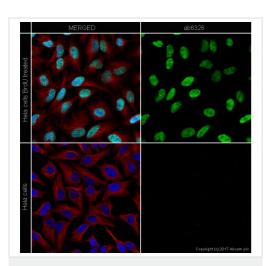
画像



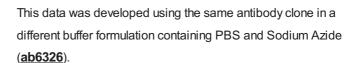
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-BrdU antibody [BU1/75 (ICR1)] - BSA and Azide free (ab264079)

IHC image of <u>ab6326</u> staining in a formalin-fixed, paraffinembedded 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 <u>ab6326</u> 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.

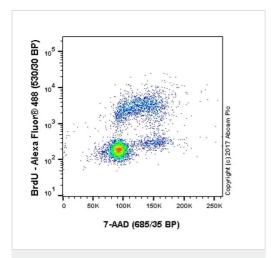
This data was developed using the same antibody clone in a different buffer formulation containing PBS and Sodium Azide (ab6326).



Immunocytochemistry/ Immunofluorescence - Anti-BrdU antibody [BU1/75 (ICR1)] - BSA and Azide free (ab264079)



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), preadsorbed 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.



Flow Cytometry (Intracellular) - Anti-BrdU antibody [BU1/75 (ICR1)] - BSA and Azide free (ab264079)

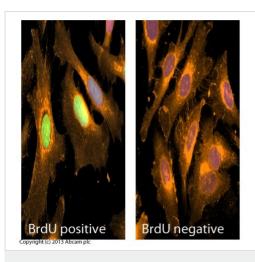
Dot plot showing BrdU-treated HeLa cells stained with <u>ab6326</u>. Cells were incubated with 10 µM BrdU for 30 minutes prior to being harvested, washed twice in 1x PBS and fixed in 70% ethanol (4°C, added drop-wise) for at least 30 minutes on ice. Once fixed, pellets were acid denatured with 2M HCl 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/1x10⁶ cells) for 30 min at 22°C. The secondary antibody used was Goat 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 >5,000 events were collected using a 50 mW Blue laser (488nm) with 530/30 and 685/35 bandpass filters.

This data was developed using the same antibody clone in a different buffer formulation containing PBS and Sodium Azide (ab6326).



Immunocytochemistry/ Immunofluorescence - Anti-BrdU antibody [BU1/75 (ICR1)] - BSA and Azide free (ab264079)

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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS and Sodium Azide (ab6326).

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