abcam

Product datasheet

Anti-PCNA antibody [PC10] ab29

★★★★★ <u>72 Abreviews</u> <u>662 References</u> 画像数 13

製品の概要

製品名 Anti-PCNA antibody [PC10]

製品の詳細 Mouse monoclonal [PC10] to PCNA

由来種 Mouse

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

種交差性 交差種: Mouse, Rat, Human

交差が予測される動物種: Chicken, Cow, Pigeon, Pig, Drosophila melanogaster, Monkey,

Zebrafish, Thornback ray, Dogfish, Catshark

免疫原 Fusion protein corresponding to PCNA. Protein A-PCNA fusion protein obtained from pC2T

construct. This construct lacked 93 nucleotides at the 3' end of PCNA.

ポジティブ・コントロール WB: HeLa, HEK293, A431 whole cell lysates, PC12, SV40LT-SMC, NIH 3T3, rat liver, rat heart.

IHC-P: Human breast (Paget's disease of the nipple), mouse liver, mouse gut, rat spinal cord DRG, developing chick brain, zebrafish intestine, rat spleen, rat large intestine. IHC-Fr: mouse

embryonic brain, HeLa. Flow Cyt: HeLa.

特記事項 This antibody clone [PC10] is manufactured by Abcam.

If you require this antibody in a particular buffer formulation or a particular conjugate for your experiments, please contact <u>orders@abcam.com</u> or you can find further information <u>here</u>.

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. Avoid freeze / thaw cycle.

バッファー pH: 7.2

Preservative: 0.02% Sodium azide

1

Constituents: PBS, 6.97% L-Arginine

精製度 Protein G purified

ポリ/モノ モノクローナル

クローン名 PC10

₹I□-**₹** Sp2/0-Ag14

アイソタイプ lgG2a 軽鎖の種類 kappa

アプリケーション

The Abpromise guarantee Abpromise保証は、次のテスト済みアプリケーションにおけるab29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
IHC-P	★★★★★ (28)	1/10000 - 1/30000. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
WB	★★★★★ (20)	Use a concentration of 1 µg/ml. Detects a band of approximately 30 kDa (predicted molecular weight: 29 kDa).
ICC/IF	★★★★☆ (14)	Use a concentration of 1 - 5 µg/ml. Methanol fixation recommended
Flow Cyt (Intra)		Use 1µg for 10 ⁶ cells. ab170191 - Mouse monoclonal lgG2a, is suitable for use as an isotype control with this antibody.

ターゲット情報

機能 This protein is an auxiliary protein of DNA polymerase delta and is involved in the control of

eukaryotic DNA replication by increasing the polymerase's processibility during elongation of the

leading strand. Induces a robust stimulatory effect on the 3'-5' exonuclease and 3'-

phosphodiesterase, but not apurinic-apyrimidinic (AP) endonuclease, APEX2 activities. Has to

be loaded onto DNA in order to be able to stimulate APEX2.

配列類似性 Belongs to the PCNA family.

翻訳後修飾 Upon methyl methanesulfonate-induced DNA damage, mono-ubiquitinated by the UBE2B-RAD18

complex on Lys-164. This induces non-canonical polyubiquitination on Lys-164 through 'Lys-63' linkage of ubiquitin moieties by the E2 complex UBE2N-UBE2V2 and the E3 ligases, HLTF, RNF8 and SHPRH, which is required for DNA repair. 'Lys-63' polyubiquitination prevents genomic instability on DNA damage. Monoubiquitination at Lys-164 also takes place in undamaged proliferating cells, and is mediated by the DCX(DTL) complex, leading to enhance

PCNA-dependent translesion DNA synthesis.

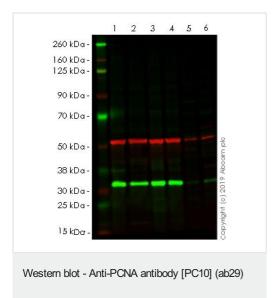
Acetylated in response to UV irradiation. Acetylation disrupts interaction with NUDT15 and $\,$

promotes degradation.

細胞内局在 Nucleus. Forms nuclear foci representing sites of ongoing DNA replication and vary in

morphology and number during S phase. Together with APEX2, is redistributed in discrete

画像



All lanes: Anti-PCNA antibody [PC10] (ab29) at 1 µg/ml

Lane 1: HeLa 20 ug

Lane 2: PC12 20 ug

Lane 3: SV40LT-SMC 20 ug

Lane 4: NIH 3T3 20 ug **Lane 5**: Rat liver 20 ug

Lane 6: Rat heart 20 ug

Secondary

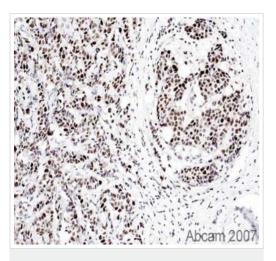
All lanes : Goat anti-Mouse IgG H&L (IRDye® 800CW) preadsorbed (ab216772) at 1/20000 dilution

Performed under reducing conditions.

Predicted band size: 29 kDa

Merged signal (red and green). Green - ab29 observed at 32 kDa. Red - loading control, **ab52866**, observed at 50 kDa.

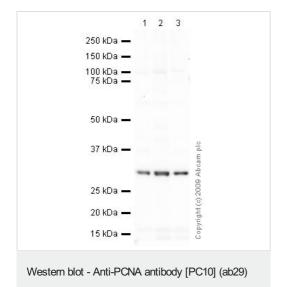
All samples were subjected to SDS-PAGE. The membrane was blocked with 3% NF Milk. Ab29 and <u>ab52866</u>(Rabbit anti alpha Tubulin loading control) were incubated overnight at 4°C at 1 ug/mL and 1/20000 dilution respectively. Blots were developed with Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed <u>ab216772</u> and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed <u>ab216777</u> secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



Mouse monoclonal [PC10] to PCNA - Proliferation Marker (ab29) used in immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections; 1/6000 for 2h at RT) on human tissue sections (Paget's disease of the nipple). Antigen retrieval step: Heat mediated. Blocking step: 1% BSA for 10 mins at RT. Incubation time: Secondary Antibody: Biotin conjugated goat anti mouse Igs (1/200).

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PCNA antibody [PC10] (ab29)

This image is courtesy of Carl Hobbs, King's College London, United Kingdom



All lanes: Anti-PCNA antibody [PC10] (ab29) at 5 µg/ml

Lane 1 : HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate

Lane 2 : HEK293 (Human embryonic kidney cell line) Whole Cell Lysate

Lane 3 : A431 (Human epithelial carcinoma cell line) Whole Cell Lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat polyclonal to Mouse lgG - H&L - Pre-Adsorbed (HRP) at 1/3000 dilution

Performed under reducing conditions.

Predicted band size: 29 kDa

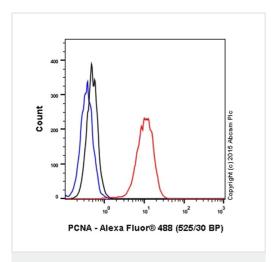
Observed band size: 29 kDa

Exposure time: 4 minutes

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Immunocytochemistry/ Immunofluorescence - Anti-PCNA antibody [PC10] (ab29)

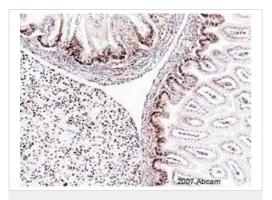
ab29 stained in HeLa cells. Cells were fixed with 100% methanol (5 min) at room temperature and incubated with PBS containing 10% goat serum, 0.3 M glycine, 1% BSA and 0.1% triton for 1h at room temperature to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody ab29 at 5 μ g/ml and <u>ab6046</u> (Rabbit polyclonal to beta tubulin - Loading Control) at 1/1000 dilution overnight at +4°C. The secondary antibodies were <u>ab150117</u> (pseudo-colored red) and <u>ab150080</u> (colored green) used at 1 μ g/ml for 1 hour at room temperature. DAPI was used to stain the cell nuclei (colored blue) at a concentration of 1.43 μ M for 1 hour at room temperature.



Flow Cytometry (Intracellular) - Anti-PCNA antibody [PC10] (ab29)

Overlay histogram showing HeLa cells stained with ab29 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween 20 for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab29, 0.1µg/1x10⁶ cells) for 30 min at 22°C. The secondary antibody used was Alexa Fluor[®] 488 goat anti-mouse IgG (H&L) (ab150113) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse IgG2a [ICIGG2A] (ab91361, 0.1µg/1x10⁶ cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control.

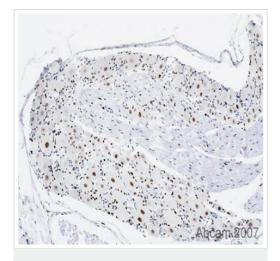
Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PCNA antibody [PC10] (ab29)

This image is courtesy of an abreview submitted by Carl Hobbs (King's College London, United Kingdom)

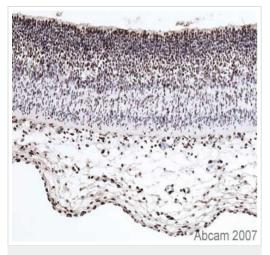
ab29 at 1/6000 staining mouse embryo (day 17) liver and gut tissue sections by IHC-P. The tissue was formaldehyde fixed and a heat mediated antigen retrieval step in Tris buffer was performed. The tissue was blocked before incubation with the antibody for 2 hours. A biotinylated goat polyclonal antibody was used as the secondary.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PCNA antibody [PC10] (ab29)

This image is courtesy of Carl Hobbs, King's College London, United Kingdom

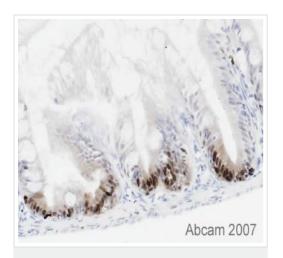
Immunohistochemistical staining (Formaldehyde/PFA-fixed paraffin-embedded sections) for PCNA antibody [PC10] - Proliferation Marker (ab29) on Rat Tissue sections (adult spinal cord DRG). Antigen retrieval step: Heat mediated. Blocking step: 1% BSA for 10 mins at RT. Primary Antibody used at 1/6000 for 2 minutes at RT. Secondary Antibody: Biotin labelled goat anti mouse lgs (1/200).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PCNA antibody [PC10] (ab29)

This image is courtesy of Carl Hobbs, King's College London, United Kingdom

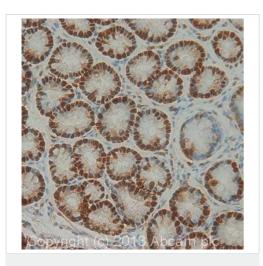
Mouse monoclonal [PC10] to PCNA - Proliferation Marker (ab29) used in immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections; 1/6000 for 2h at RT) on E6 developing chick brain). Antigen retrieval step: Heat mediated. Blocking step: 1% BSA for 10 mins at RT. Incubation time: Secondary Antibody: Biotin conjugated goat anti mouse lgs (1/200). NB: This image shows developing brain/overlying skin.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PCNA antibody [PC10] (ab29)

This image is courtesy of Carl Hobbs, King's College London, United Kingdom

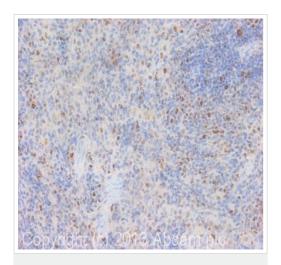
Mouse monoclonal [PC10] to PCNA - Proliferation Marker (ab29) used in immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections;1/6000 for 2h at RT) on intestine of adult zebrafish). Antigen retrieval step: Heat mediated. Blocking step: 1% BSA for 10 mins at RT. Incubation time: Secondary Antibody: Biotin conjugated goat anti mouse lgs (1/200). NB: The crypt nuclei on this image of zebrafish intestine, are positive for the PCNA/PC10 clone conforming to accepted localisation data for PCNA in other species.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PCNA antibody [PC10] (ab29)

IHC image of PCNA staining in rat large intestine formalin fixed paraffin embedded tissue section, performed on a Leica Bond system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab29, 0.025µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

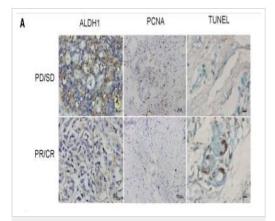
For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PCNA antibody [PC10] (ab29)

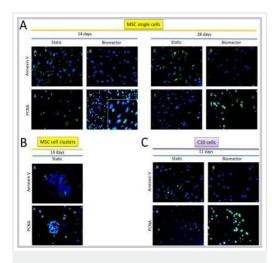
IHC image of PCNA staining in rat spleen formalin fixed paraffin embedded tissue section, performed on a Leica Bond system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab29, 1/30,000 dilution for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PCNA antibody [PC10] (ab29)

Gong, C. et al PLoS One. 2010 Dec 20;5(12):e15630. doi: 10.1371/journal.pone.0015630 Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PCNA antibody [PC10] (ab29)

Crabbe et al PLoS One. 2015 May 11;10(5):e0126846. doi: 10.1371/journal.pone.0126846. eCollection 2015 Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

Aldehyde dehydrogenase (ALDH1) expression correlates with clinical outcome of breast cancer patients

(A) Immunohistochemical staining shows tumors with poor clinical response (progressive or stable disease, PD/SD) to neo-adjuvant chemotherapy express high ALDH1 (>20% positive cancer cells) in pre-chemotherapy samples, and tumors with partal response (PR) express low ALDH1 (≤20% positive cancer cells). High proliferating cell nuclear antigen (PCNA)(>25% positive cancer cells) and poor apoptosis are observed in tumors with PD/SD after neo-adjuvant chemotherapy. Representative images of ALDH1 (×200), PCNA (×200) and Terminal deoxynucleotidyl transferase (TdT)-mediated dUTP labeling (TUNEL,×400).

PCNA is detected using ab29 at 1/100 dilution.

(From Figure 1A of Gong et al)

Annexin V and PCNA staining of decellularized lung scaffolds recellularized with (A) MSCs in static versus bioreactor conditions for 14 (panels A, B for annexin V, and a, b for PCNA) and 28 days (panels C, D for annexin V, and c, d for PCNA) (single cells), (B) MSC cell clusters in static conditions at 14 days (panel A for annexin V, and a for PCNA), (C) C10 cells in static (panel A for annexin V, a for PCNA) versus bioreactor (panel B for annexin V, b for PCNA) conditions for 11 days.

An inset in Fig 4Ab with higher magnification is shown to demonstrate that a majority of the cells stained positive for PCNA. Cell nuclei are labeled in blue; marker of interest is labeled in green. Magnifications are 400x. Overlap of cell nucleus and marker of interest can generate green or white color. For each condition, images are representative of the entire lung.

MSCs = Bone marrow-derived mouse mesenchymal stromal (stem) cells.

PCNA is detected using ab29 at 1/1000 dilution.

(From Figure 4 of Crabbe et al)

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