

Product datasheet

Anti-PCNA antibody ab18197

★★★★☆ 40 Abreviews 121 References 18 Images

Overview

Product name	Anti-PCNA antibody
Description	Rabbit polyclonal to PCNA
Host species	Rabbit
Tested applications	Suitable for: WB, ICC/IF, IHC-P, IHC-FoFr, Flow Cyt, IP
Species reactivity	Reacts with: Mouse, Rat, Sheep, Goat, Cow, Human, Monkey, Zebrafish, Common marmoset Predicted to work with: Dog, Xenopus laevis 
Immunogen	Synthetic peptide corresponding to Human PCNA aa 200 to the C-terminus (C terminal) conjugated to Keyhole Limpet Haemocyanin (KLH). Database link: P12004 (Peptide available as ab18602 , ab2427)
Positive control	This antibody gave a positive signal in HEK 293 (Human embryonic kidney cell line), NIH 3T3 and MEF1 (Mouse embryonic fibroblast cell lines) whole cell lysates. ICC/IF: HeLa cells. Flow Cyt: HeLa cells.

Properties

Form	Liquid
Storage instructions	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.
Storage buffer	Preservative: 0.02% Sodium Azide Constituents: 1% BSA, PBS, pH 7.4
Purity	Immunogen affinity purified
Clonality	Polyclonal
Isotype	IgG

Applications

Our [Abpromise guarantee](#) covers the use of **ab18197** in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB	★★★★☆	Use a concentration of 1 µg/ml. Detects a band of approximately 29 kDa (predicted molecular weight: 29 kDa).
ICC/IF	★★★★★	Use a concentration of 1 µg/ml.
IHC-P	★★★★★	Use at an assay dependent concentration.
IHC-FoFr		Use at an assay dependent concentration. PubMed: 21895533
Flow Cyt		Use 0.05µg for 10 ⁶ cells.
IP	★★★★★	Use at an assay dependent concentration.

Target

Function

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-aprimidinic (AP) endonuclease, APEX2 activities. Has to be loaded onto DNA in order to be able to stimulate APEX2.

Sequence similarities

Belongs to the PCNA family.

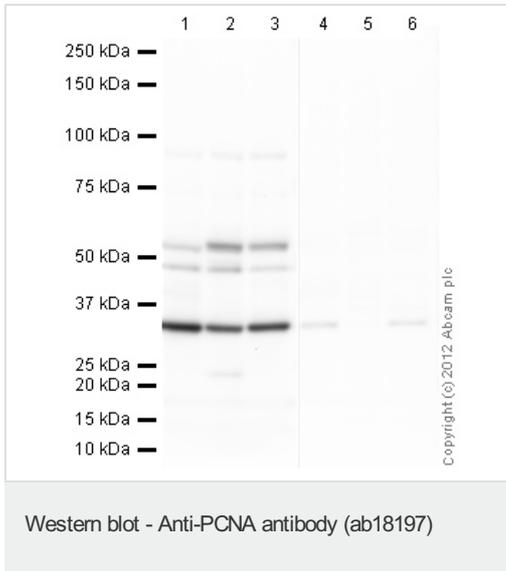
Post-translational modifications

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.

Cellular localization

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 nuclear foci in presence of oxidative DNA damaging agents.

Images



All lanes : Anti-PCNA antibody (ab18197) at 1 µg/ml

Lane 1 : HEK293 (Human embryonic kidney cell line) Whole Cell Lysate

Lane 2 : NIH 3T3 (Mouse embryonic fibroblast cell line) Whole Cell Lysate

Lane 3 : PC12 (Rat adrenal pheochromocytoma cell line) Whole Cell Lysate

Lane 4 : HEK293 (Human embryonic kidney cell line) Whole Cell Lysate with Human PCNA peptide (ab18602) at 1 µg/ml

Lane 5 : NIH 3T3 (Mouse embryonic fibroblast cell line) Whole Cell Lysate with Human PCNA peptide (ab18602) at 1 µg/ml

Lane 6 : PC12 (Rat adrenal pheochromocytoma cell line) Whole Cell Lysate with Human PCNA peptide (ab18602) at 1 µg/ml

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) preadsorbed (ab97080) at 1/5000 dilution

Developed using the ECL technique.

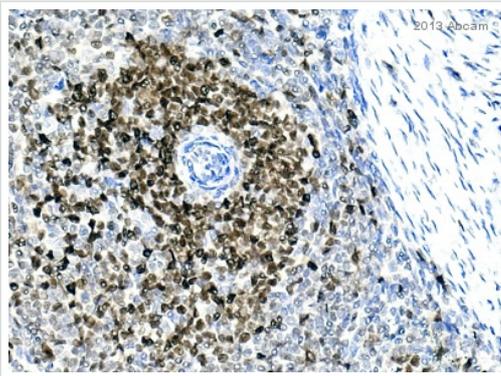
Performed under reducing conditions.

Predicted band size: 29 kDa

Observed band size: 29 kDa

Additional bands at: 48 kDa, 52 kDa. We are unsure as to the identity of these extra bands.

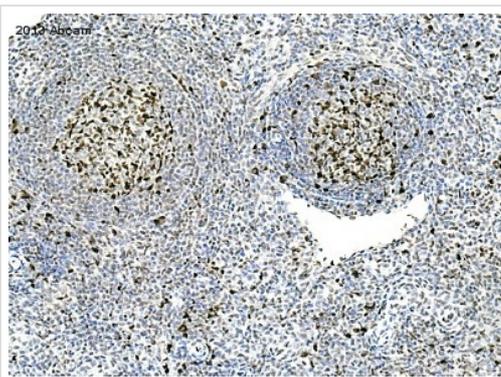
Exposure time: 1 minute



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PCNA antibody (ab18197)

Image courtesy of Carl Hobbs, Kings College London, U.K.

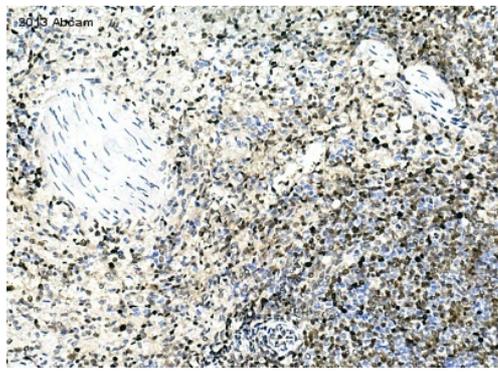
ab18197 staining PCNA in tissue sections of the goat spleen by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections). Tissue was fixed with formaldehyde and a heat mediated antigen retrieval step was performed. Samples were then blocked with 1% B.S.A. for 10 minutes at 21°C followed by incubation with the primary antibody for 2 hours at 1/4000. A biotin-conjugated goat anti-rabbit polyclonal was used as secondary antibody at a 1/250 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PCNA antibody (ab18197)

Image courtesy of Carl Hobbs, Kings College London, U.K.

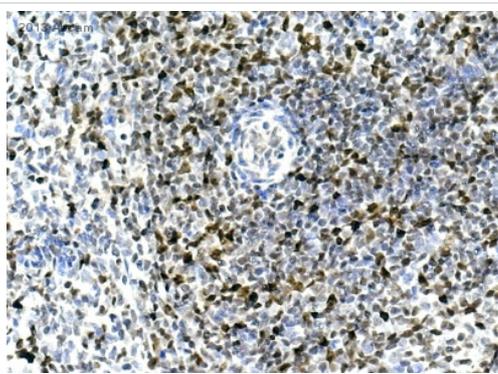
ab18197 staining PCNA in tissue sections of the marmoset spleen by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections). Tissue was fixed with formaldehyde and a heat mediated antigen retrieval step was performed. Samples were then blocked with 1% B.S.A. for 10 minutes at 21°C followed by incubation with the primary antibody for 2 hours at 1/6000. A biotin-conjugated goat anti-rabbit polyclonal was used as secondary antibody at a 1/250 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PCNA antibody (ab18197)

Image courtesy of Carl Hobbs, King College London, U.K.

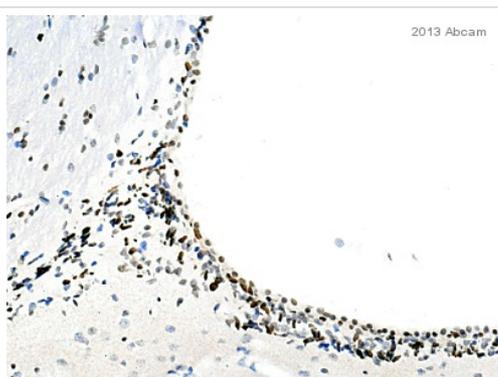
ab18197 staining PCNA in tissue sections of the cow spleen by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections). Tissue was fixed with formaldehyde and a heat mediated antigen retrieval step was performed. Samples were then blocked with 1% B.S.A. for 10 minutes at 21°C followed by incubation with the primary antibody for 2 hours at 1/4000. A biotin-conjugated goat anti-rabbit polyclonal was used as secondary antibody at a 1/250 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PCNA antibody (ab18197)

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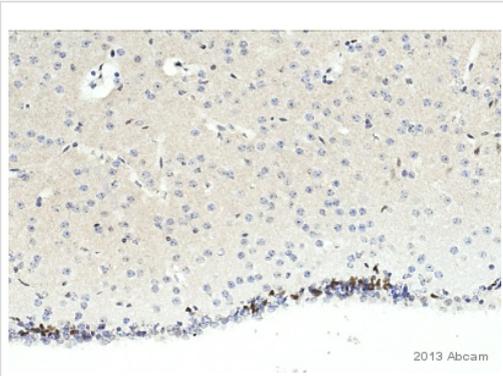
ab18197 staining PCNA in tissue sections of the sheep spleen by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections). Tissue was fixed with formaldehyde and a heat mediated antigen retrieval step was performed. Samples were then blocked with 1% B.S.A. for 10 minutes at 21°C followed by incubation with the primary antibody for 2 hours at 1/6000. A biotin-conjugated goat anti-rabbit polyclonal was used as secondary antibody at a 1/250 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PCNA antibody (ab18197)

Image courtesy of Carl Hobbs, Kings College London, U.K.

ab18197 staining PCNA in tissue sections of the rat brain by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections). Tissue was fixed with formaldehyde and a heat mediated antigen retrieval step was performed. Samples were then blocked with 1% B.S.A. for 10 minutes at 21°C followed by incubation with the primary antibody for 2 hours at 1/10000. A biotin-conjugated goat anti-rabbit polyclonal was used as secondary antibody at a 1/250 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PCNA antibody (ab18197)

Image courtesy of Carl Hobbs, Kings College London, U.K.

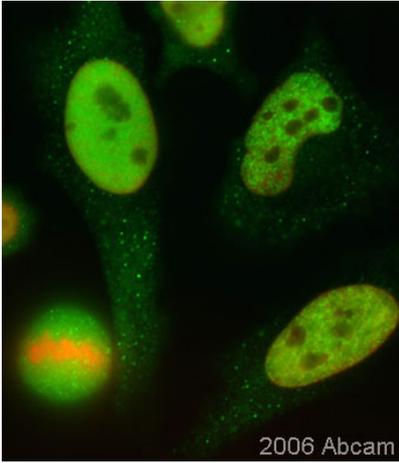
ab18197 staining PCNA in tissue sections of the mouse brain by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections). Tissue was fixed with formaldehyde and a heat mediated antigen retrieval step was performed. Samples were then blocked with 1% B.S.A. for 10 minutes at 21°C followed by incubation with the primary antibody for 2 hours at 1/6000. A biotin-conjugated goat anti-rabbit polyclonal was used as secondary antibody at a 1/250 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PCNA antibody (ab18197)

Image Courtesy of Carl Hobbs, Kings College London, U.K.

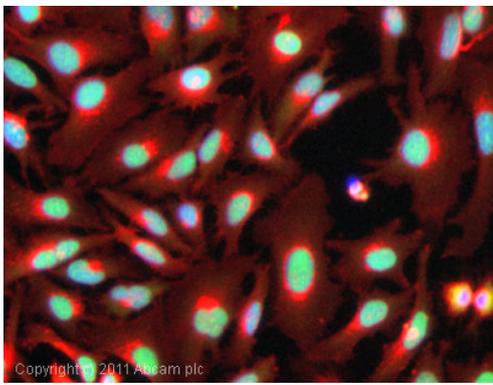
ab18197 staining PCNA in Monkey COS cell pellet by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections). Tissue was fixed with formaldehyde and a heat mediated antigen retrieval step was performed. Samples were then blocked with 1% B.S.A. for 10 minutes at 21°C followed by incubation with the primary antibody for 2 hours at 1/4000. A biotin-conjugated goat anti-rabbit polyclonal was used as secondary antibody at a 1/250 dilution.



Immunocytochemistry/ Immunofluorescence - Anti-PCNA antibody (ab18197)

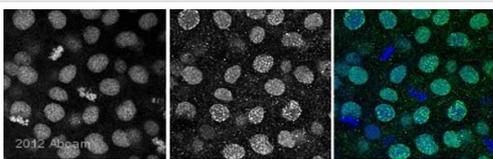
This image is courtesy of an Abreview submitted by Dr Kirk McManus

ab18197, at a 1/5000 dilution, staining PCNA in asynchronous HeLa cells. Cells were counter-stained with DAPI (red). For more information please refer to Abreview.



Immunocytochemistry/ Immunofluorescence - Anti-PCNA antibody (ab18197)

ICC/IF image of ab18197 stained HeLa cells. The cells were 100% methanol fixed (5 min) and 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 (ab18197, 1µg/ml) overnight at +4°C. The secondary antibody (green) was DyLight® 488 goat anti-rabbit IgG - H&L, pre-adsorbed (ab96899) 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.

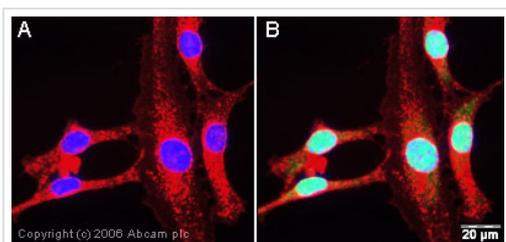


Immunocytochemistry/ Immunofluorescence - Anti-PCNA antibody (ab18197)

Image courtesy of Hank Farr by Abreview.

ab18197 staining PCNA in Zebrafish gastrula embryos by Immunocytochemistry/ Immunofluorescence (wholemout). Zebrafish embryos were fixed overnight at 4°C when they had reached 60% epiboly. Cells were fixed in formaldehyde, permeabilized using Proteinase K, blocked with 2% goat serum for 2 hours at 20°C and then incubated with ab18197 at a 1/500 dilution for 16 hours at 4°C. The secondary used was an Alexa-Fluor 488 conjugated goat anti-rabbit polyclonal used at a 1/500 dilution. Cells were post-fixed in PFA for 20 minutes at room temperature after extensive washing of the secondary antibody.

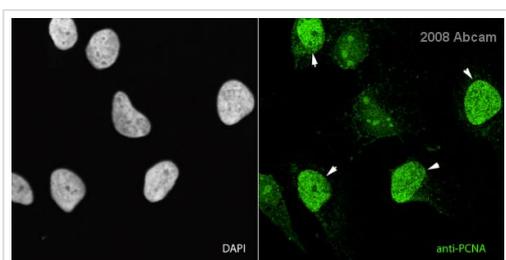
The left panel shows DAPI stained nuclei, the center panel is PCNA staining, and the right panel is the merged image.



Immunocytochemistry/ Immunofluorescence - Anti-PCNA antibody (ab18197)

ICC/IF image of ab18197 stained NIH/3T3 cells. The cells were methanol fixed (5 min) and incubated with the antibody (ab18197, 1 µg/ml) for 1h at room temperature. The secondary antibody (green) was Alexa Fluor® 488 goat anti-rabbit IgG (H+L) used at a 1/1000 dilution for 1h. Image-iT™ FX Signal Enhancer was used as the primary blocking agent, 5% BSA (in TBS-T) was used for all other blocking steps. DAPI was used to stain the cell nuclei (blue). Alexa Fluor® 594 WGA was used to label plasma membranes (red).

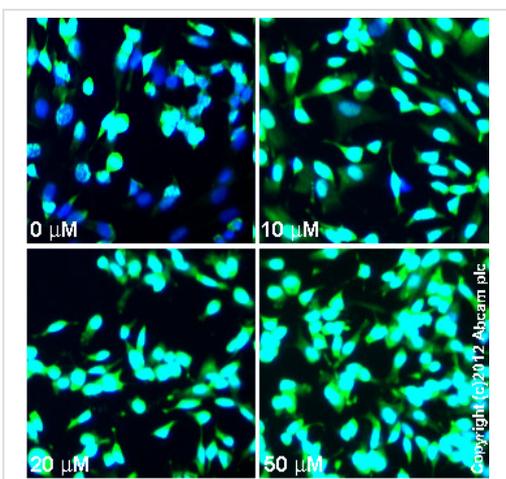
Panel A does not show the Alexa Fluor® 488 channel, Panel B shows the specific nuclear staining by ab18197.



Immunocytochemistry/ Immunofluorescence - Anti-PCNA antibody (ab18197)

This image is courtesy of an abreview submitted by Dr Catherine Green

ab18197, at a 1/2000 dilution staining PCNA in MRC5 Sv40 transformed fibroblasts. Cells were counterstained with DAPI (blue). For more information please refer to Abreview.



Immunocytochemistry/ Immunofluorescence - Anti-PCNA antibody (ab18197)

ab18197 staining PCNA in SK-N-SH cells treated with KN-62 (ab120421), by ICC/IF. Increase in PCNA nuclear expression correlates with increased concentration of KN-62, as described in literature.

The cells were incubated at 37°C for 24h in media containing different concentrations of ab120421 (KN-62) in DMSO, fixed with 100% methanol for 5 minutes at -20°C and blocked with PBS containing 10% goat serum, 0.3 M glycine, 1% BSA and 0.1% tween for 2h at room temperature. Staining of the treated cells with ab18197 (1 µg/ml) was performed overnight at 4°C in PBS containing 1% BSA and 0.1% tween. A DyLight 488 goat anti-rabbit polyclonal antibody (ab96899) at 1/250 dilution was used as the secondary antibody. Nuclei were counterstained with DAPI and are shown in blue.



Immunoprecipitation - Anti-PCNA antibody

(ab18197)

This image is courtesy of an Abreview submitted by Antibody Solutions Ltd

ab18197 staining rat PC12 whole cell lysate by Immunoprecipitation.

ab18197 was incubated with the lysate (at a concentration of 5µg/ml) and a Protein A matrix for 12 hours at 4°C to achieve immunoprecipitation. 400µg of protein were present in the lysate input.

Lane order: PCNA IP (lane 1), Control IP (lane 2), Input 5% (lane 3)

This antibody immunoprecipitates a product of ~34 kDa which is consistent with the mobility of PCNA on SDS-PAGE

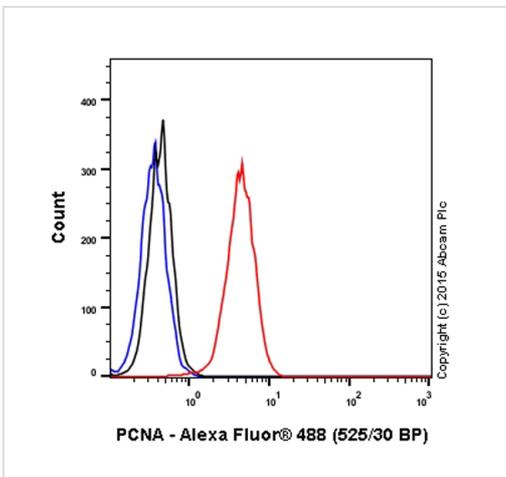
ab18197 was also used for the western blot step, at a contraction of 1µg/ml



Western blot - Anti-PCNA antibody (ab18197)



Western blot - Anti-PCNA antibody (ab18197)



Flow Cytometry - Anti-PCNA antibody (ab18197)

Overlay histogram showing HeLa cells stained with ab18197 (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 (ab18197, 0.05µg/1x10⁶ cells) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit IgG (H&L) (ab150081) at 1/4000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (polyclonal) (ab171870, 0.05µg/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.

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