

Anti-PCNA Rabbit Monoclonal Antibody

Catalog # ABO13291

Product Information

Application	WB, IHC, IF, ICC, IP, FC
Primary Accession	P12004
Host	Rabbit
Isotype	Rabbit IgG
Reactivity	Rat, Human, Mouse
Clonality	Monoclonal
Format	Liquid
Description	Anti-PCNA Rabbit Monoclonal Antibody . Tested in WB, IHC, ICC/IF, IP, Flow Cytometry applications. This antibody reacts with Human, Mouse, Rat.

Additional Information

Gene ID	5111
Other Names	Proliferating cell nuclear antigen, PCNA, Cyclin, PCNA
Calculated MW	28769
Application Details	WB 1:3000-1:10000 IHC 1:50-1:200 ICC/IF 1:100-1:500 IP 1:50-1:100 FC 1:200-1:500
Subcellular Localization	Nucleus. Colocalizes with CREBBP, EP300 and POLD1 to sites of DNA damage (PubMed:24939902). 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..
Source	Eukaryota
Contents	Rabbit IgG in phosphate buffered saline, pH 7.4, 150mM NaCl, 0.02% sodium azide and 50% glycerol, 0.4-0.5mg/ml BSA.
Clone Names	Clone: DO-16
Immunogen	A synthesized peptide derived from human PCNA
Purification	Affinity-chromatography
Storage	Store at -20°C for one year. For short term storage and frequent use, store at 4°C for up to one month. Avoid repeated freeze-thaw cycles.

Protein Information

Name	PCNA
Function	Confers DNA tethering and processivity to DNA polymerases and other proteins (PubMed: 24695737 , PubMed: 24939902 , PubMed: 35585232). Auxiliary protein of DNA polymerase delta and epsilon, is involved in the control of DNA replication by increasing the polymerases' processivity during elongation of the leading strand (PubMed: 35585232). 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. Plays a key role in DNA damage response (DDR) by being conveniently positioned at the replication fork to coordinate DNA replication with DNA repair and DNA damage tolerance pathways (PubMed: 24939902). Acts as a loading platform to recruit DDR proteins that allow completion of DNA replication after DNA damage and promote postreplication repair: monoubiquitinated PCNA leads to recruitment of translesion (TLS) polymerases, while 'Lys-63'-linked polyubiquitination of PCNA is involved in error-free pathway and employs recombination mechanisms to synthesize across the lesion (PubMed: 24695737).
Cellular Location	Nucleus. Note=Colocalizes with CREBBP, EP300 and POLD1 to sites of DNA damage (PubMed:24939902). Forms nuclear foci representing sites of ongoing DNA replication and vary in morphology and number during S phase (PubMed:15543136). Co-localizes with SMARCA5/SNF2H and BAZ1B/WSTF at replication foci during S phase (PubMed:15543136). Together with APEX2, is redistributed in discrete nuclear foci in presence of oxidative DNA damaging agents

Images

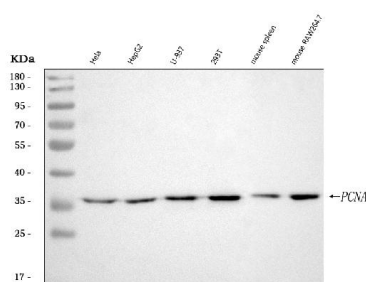


Figure 1. Western blot analysis of PCNA using anti-PCNA antibody (M00125-1).

Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.

Lane 1: human Hela whole cell lysates,
Lane 2: human HepG2 whole cell lysates,
Lane 3: human U-937 whole cell lysates,
Lane 4: human 293T whole cell lysates,
Lane 5: mouse spleen tissue lysates,
Lane 6: rat RAW264.7 tissue lysates.

After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-PCNA antigen affinity purified monoclonal antibody (Catalog # M00125-1) at 1:3000 overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for PCNA at approximately 36 kDa. The expected band size for PCNA

is at 29 kDa.

Immunoprecipitate (IP) analysis using the Antibody at 1:50 dilution. (wb at 1:3K dilution)

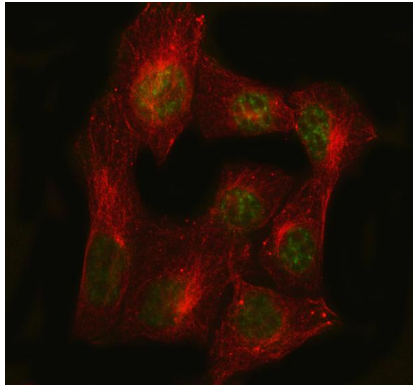
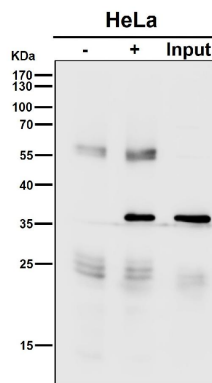


Figure 3. IF analysis of PCNA using anti-PCNA antibody (M00125-1) and anti-Beta Tubulin antibody (M01857-3). PCNA was detected in immunocytochemical section of U2OS cell. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated at 1:100 with rabbit anti-PCNA Antibody (M00125-1) and mouse anti-Beta Tubulin antibody (M01857-3) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Rabbit IgG (BA1127) and Dylight550-conjugated Anti- mouse IgG Secondary Antibody (red)(Catalog#BA1133) were used as secondary antibody at 1:500 dilution and incubated for 30 minutes at 37°C. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

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