

PCNA (human), functional

10-151 20 μ g, 10-152 100 μ g

PCNA (Proliferating cell nuclear antigen) is a homotrimeric protein (261 aa; 29 kDa) known to act as a co-factor for DNA polymerase δ, which is responsible for leading strand DNA replication. PCNA was originally identified as an antigen that is expressed in the nuclei of cells during the DNA synthesis phase of the cell cycle. Crystal structure data suggests that a PCNA homotrimer ring encircles and slides along the DNA double helix. Multiple proteins involved in DNA replication, DNA repair, and cell cycle control bind to PCNA rather than directly associates with DNA, thus facilitating rapid processing of DNA. PCNA is a useful marker for DNA synthesis and some cancers. It is highly conserved among most amimals.

Applications confirmed:

- 1. Functional studies on DNA replication, recombination and repair. (Ref 2, 3, 5, 6, 7, 8, 9, 10).
- 2. Identification of proteins interacting with PCNA by using PCNA -conjugated resin. (Ref 1, 5)
- 3. Ubiquitination targets (Ref 4, 9, 10).
- 4 SDS-PAGE (Fig. 1). 5. Western blot (Fig. 2) . 6. Dot blot. 7. ELISA. Not tested for other applications.

Source: Human PCNA was over-expressed in *E. coli* as a recombinant full-size protein without any tag and highly purified.

Form: 1.0 mg/ml in 25 mM HEPES (pH 7.9), 1 mM EDTA,

0.01% NP40, 1 mM DTT, 2 ug/ml leupeptin, 0.1 mM PMSF, 75 mM NaCl, 50% glycerol.

Storage: Sent at 4°C or -20°C. Upon arrival spin-down and store at -20°C (or at -80°C for longer storage)

Purity: Greater than 98% purity as determined by SDS-PAGE (Fig.1).

Data Link: Swiss-Prot P12004 (human), P04961 (rat), P17918 (mouse), Q9PTP1 (Zebrafish).

References: This product has been used in the following References.

- 1. Ohta S. et al (2002) A proteomics approach to identify proliferating cell nuclear antigen (PCNA)-binding proteins in human cell lysates. Identification of the human CHL12/RFCs2-5 complex as a novel PCNA-binding protein. J Biol Chem **277**: 40362-40367 **PMID**: 12171929.
- Iida T. et al (2002) "PCNA clamp facilitates action of DNA cytosine methyltransferase 1 on hemimethylated DNA. Genes Cells 7: 997-1007 PMID: <u>12354094</u>.
- 3. Shiomi Y, et al (2004) The reconstituted human Chl12-RFC complex functions as a second PCNA loader. Genes Cells. 9:279-90. PMID: 15066120.
- 4.. Watanabe K, et al. (2004) Rad18 guides pol eta to replication stalling sites through physical interaction and PCNA monoubiquitination. EMBO J. 23:3886-96 PMID: 15359278.
- 5. Tsurimoto T, et al. (2005) Human Werner helicase interacting protein 1 (WRNIP1) functions as a novel modulator for DNA polymerase delta. Genes Cells. 10:13-22. PMID 15670210



- 6. Nishitani H, et al. (2006) Two E3 ubiquitin ligases, SCF-Skp2 and DDB1-Cul4, target human Cdt1 for proteolysis. EMBO J. **25**:1126-36. **PMID**: <u>16482215</u>.
 - 7. Shiomi Y, et al. (2007) A second proliferating cell nuclear antigen loader complex, Ctf18-replication factor C, stimulates DNA polymerase eta activity. J Biol Chem. **282**:20906-14. **PMID**: <u>17545166</u>.
- Masuda Y, et al. (2007) Dynamics of human replication factors in the elongation phase of DNA replication. Nucleic Acids Res. 35:6904-16. PMID: 17932049.
- Tomida J, et al. (2008) DNA damage-induced ubiquitylation of RFC2 subunit of replication factor C complex. J Biol Chem. 283:9071-9. PMID: 18245774.
- 10.Tsuji Y, et al. (2008) Recognition of forked and single-stranded DNA structures by human RAD18 complexed with RAD6B protein triggers its recruitment to stalled replication forks. Genes Cells. 13:343-54. PMID: 18363965.

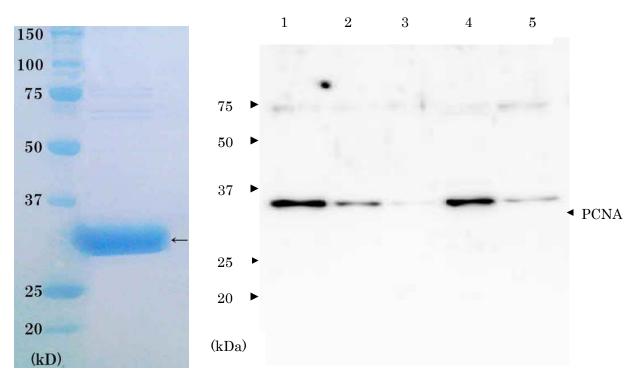


Fig. 1. SDS-PAGE anlysis of purified PCNA protein.

Fig. 2 Western Bloting of PCNA. Lane 1; Purified PCNA (3 ng). Lane 2; Purified PCNA (1 ng). Lane 3; Purified PCNA (0.3 ng). Lane 4; Crude extract of Hela cells (10 μ g). Lane 5; Crude extract of HeLa cells (2 μ g) . Primary antibody is anti-PCNA antibody, BioAcademia # 70-080.

Related product; #70-080 Anti- PCNA antibody, rabbit polyclonal