

PCNA siRNA (h): sc-29440

BACKGROUND

The proliferating cell nuclear antigen (PCNA), a protein synthesized in early G₁ and S phases of the cell cycle, functions in cell cycle progression, DNA replication and DNA repair. In early S phase, PCNA exhibits granular distribution and is absent from the nucleoli; however, in late S phase, it relocates to the nucleoli. PCNA exists in two basic forms: one involved in ongoing DNA replication, which localizes specifically to the nucleus, and a second, soluble form, not implicated in constant synthesis. Interestingly, the latter form degrades in the presence of organic solvents, rendering it undetectable by histological methods in tissues using organic fixatives, and thus also providing a method of visualizing only the synthesizing form.

REFERENCES

1. Bravo, R., et al. 1987. Existence of two populations of cyclin/proliferating cell nuclear antigen during the cell cycle: association with DNA replication sites. *J. Cell Biol.* 105: 1549-1554.
2. Waseem, N.H. and Lane, D.P. 1990. Monoclonal antibody analysis of the proliferating cell nuclear antigen (PCNA). Structural conservation and the detection of a nucleolar form. *J. Cell Sci.* 96: 121-129.
3. Woods, A.L., et al. 1991. The assessment of proliferating cell nuclear antigen (PCNA) immunostaining in primary gastrointestinal lymphomas and its relationship to histological grade, S+G₂+M phase fraction (flow cytometric analysis) and prognosis. *Histopathology* 19: 21-27.
4. Baida, A., et al. 2003. Germline mutations at microsatellite loci in homozygous and heterozygous mutants for mismatch repair and PCNA genes in *Drosophila*. *DNA Repair* 2: 827-833.

CHROMOSOMAL LOCATION

Genetic locus: PCNA (human) mapping to 20p13.

PRODUCT

PCNA siRNA (h) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PCNA shRNA Plasmid (h): sc-29440-SH and PCNA shRNA (h) Lentiviral Particles: sc-29440-V as alternate gene silencing products.

For independent verification of PCNA (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-29440A, sc-29440B and sc-29440C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNAses and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

PCNA siRNA (h) is recommended for the inhibition of PCNA expression in human cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 μ M in 66 μ l. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

PCNA (PC10): sc-56 is recommended as a control antibody for monitoring of PCNA gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker[™] Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz[®] Mounting Medium: sc-24941 or UltraCruz[®] Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor PCNA gene expression knockdown using RT-PCR Primer: PCNA (h)-PR: sc-29440-PR (20 μ l, 449 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Khanal, S. and Galloway, D.A. 2019. High-risk human papillomavirus oncogenes disrupt the Fanconi anemia DNA repair pathway by impairing localization and de-ubiquitination of FancD2. *PLoS Pathog.* 15: e1007442.
2. Kundu, K., et al. 2019. Inhibition of the NKp44-PCNA immune checkpoint using a mAb to PCNA. *Cancer Immunol. Res.* 7: 1120-1134.
3. Lu, S., et al. 2021. Proliferating cell nuclear antigen directly interacts with androgen receptor and enhances androgen receptor-mediated signaling. *Int. J. Oncol.* 59: 41.
4. Su, X., et al. 2021. PCNA inhibition enhances the cytotoxicity of β -lapachone in NQO1-positive cancer cells by augmentation of oxidative stress-induced DNA damage. *Cancer Lett.* 519: 304-314.

RESEARCH USE

For research use only, not for use in diagnostic procedures.