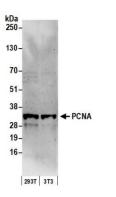
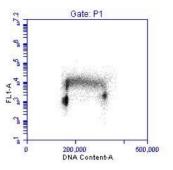
## **PCNA Antibody**

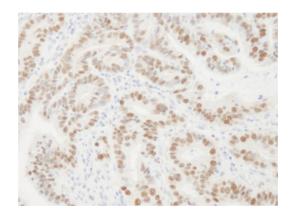
| Rabbit Polyclo   |        |  |         | 512004  |  |  |
|--|--------|--|---------|---|--|--|
| Antigen Affinity Purifie   |        |  | tein ID | P12004  |  |  |
| Catalog No.  | A300-2 |  | nelD    | 5111  | BETHYL   |  |
| Lot No.  | A300   | 276A-1   |         |   |  |  |
| APPLICATIONS   |        | WB, IP, IHC, F   |         |   |  |  |
| SPECIES REACTIVITY   |        | Human, Mouse   |         |   |  |  |
| PRESUMED REACTIVITY  |        | Based on 100% sequence identity, this antibody is predicted to react with Rat, Chicken and<br>Bovine   |         |   |  |  |
| AMOUNT   |        | 100 μΙ   |         |   |  |  |
| CONCENTRATION  |        | 1000 μg/ml   |         |   |  |  |
| STORAGE/SHELF LIFE   |        | 2 - 8° C / 1 year from date of receipt   |         |   |  |  |
| PHYSICAL STATE   |        | Liquid   |         |   |  |  |
| BUFFER   |        | Tris-citrate/phosphate buffer, pH 7 to 8 containing 0.09% Sodium Azide   |         |   |  |  |
| ISOTYPE  |        | IgG  |         |   |  |  |
| ORIGIN   |        | USA  |         |   |  |  |
| PRODUCTION<br>PROCEDURES   |        | Antibody was affinity purified using an epitope specific to PCNA immobilized on solid support.   |         |   |  |  |
|  |        | The epitope recognized by A300–276A maps to a region between residues 75 and 125 of human proliferating cell nuclear antigen using the numbering given in SwissProt entry P12004 (GeneID 5111).  |         |   |  |  |
|  |        | Immunoglobulin concentration was determined by extinction coefficient: absorbance at 280 nm of 1.4 equals 1.0 mg of IgG.   |         |   |  |  |
|  |        | Centrifuge tube to remove product from lid. Optimal working dilutions should be determined experimentally by the investigator. Prepare working dilution immediately before use.  |         |   |  |  |
|  |        | Western Blot   | 1:1     | ,000 - 1:10,000                                     |  |  |
|  |        | Immunoprecipitatio   | n 2-    | 5 µg/mg lysate                                      |  |  |
|  |        | Immunohistochemi   |         | 2,000 – 1:10,000. Epitor<br>commended for FFPE tise | be retrieval with citrate buffer pH 6.0 is sue sections. |  |
|  |        | Flow Cytometry   | 0.0     | )3 µg per 1 X 10^6 cells                            | in a 150 µl volume                                       |  |
| APPLICATION NOTES  |        | Western blot of immunoprecipitates performed using Normal Pig Serum (Cat. No. S100–020),<br>Goat anti–Rabbit Light Chain HRP Conjugate (Cat. No. A120–113P) and 4–20% SDS–PAGE<br>(link to IP–western blot protocol in Additional Info section below). |         |   |  |  |
| IHC HUMAN CONTROLS   |        | Western blot of lysates performed using standard western blot reagents and 4–20% SDS-PAGE.<br>Breast Carcinoma, Colon Carcinoma, Ovarian Carcinoma, Prostate Carcinoma, Stomach<br>Adenocarcinoma, Testicular Seminoma                                 |         |   |  |  |
| IHC MOUSE CONTROLS   |        | Squamous Cell Carcinoma, Teratoma  |         |   |  |  |
| ADDITIONAL INFO  |        | https://www.bethyl.com/product/A300-276A<br>Use the link above to view SDS, a current list of citations, and other product specific information.<br>IP-western blot protocol: https://www.bethyl.com/content/protocol_IP_WB                            |         |   |  |  |
|  |        |  |         |   |  |  |
| This document cortifies that this product has met all of the quality control standards defined by Pothyl Laboratories. Inc |        |  |         |   |  |  |

This document certifies that this product has met all of the quality control standards defined by Bethyl Laboratories, Inc. Eric McIntush, PhD | Chief Scientific Officer Date: June 21, 2019

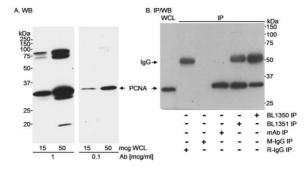




Detection of human and mouse PCNA by western blot. Samples: Whole cell lysate (50 µg) from HEK293T and mouse NIH 3T3 cells prepared using NETN lysis buffer. Antibody: Affinity purified rabbit anti-PCNA antibody A300-276A (lot A300-276A-1) used for WB at 0.1 µg/ml. Detection: Chemiluminescence with an exposure time of 3 minutes. **Detection of PCNA Versus DNA Content.** Asynchronous Jurkat cells were fixed and permeabilized in a sequential treatment of FACS buffer (PBS, 0.5% triton-X-100, 0.5mM EDTA, 1% BSA) and 100% methanol.  $1 \times 10^{6}$  cells were stained with  $0.03 \mu g$  anti-PCNA [A300-276A]. Secondary detection was performed with FITC conjugated goat F(ab')2 anti-rabbit antibody [A120-114F], and DNA stained with



**Detection of human PCNA by immunohistochemistry.** *Sample:* FFPE section of human stomach carcinoma. *Antibody:* Affinity purified rabbit anti- PCNA (Cat. No. A300-276A Lot1) used at a dilution of 1:10,000 (0.1 µg/ml). *Detection:* DAB



## Detection of human PCNA by western blot and

**immunoprecipitation.** *Samples:* A. Whole cell lysate (WCL) from A-431 cells. B. WCL (500 µg for IP; 30 µg for WCL lane) from MDA-MB-468 cells. *Antibodies:* A. Affinity purified rabbit anti-PCNA antibody BL1350 (Cat. No. A300-276A) used at the indicated concentrations for WB. B. PCNA was immunoprecipitated using BL1350, BL1351 (Cat. No. A300-277A) or a mouse monoclonal antibody to PCNA (mAb). Control mock IP was performed using normal mouse IgG (M-IgG) and normal rabbit IgG (R-IgG). immunoprecipitatesd PCNA was blotted using a mixture of BL1350 and BL1351 (each at 1 µg/ml). *Detection:* Chemiluminescence with an exposure time of 2 minutes (A) or 5 seconds (B).