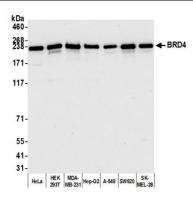
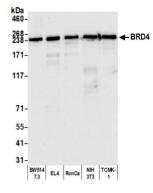
BRD4 Antibody

| Rabbit Polyclonal Antigen Affinity Purified Catalog No. A301–985A50 Lot No. A301–985A50–8 | | Protein ID GenelD | NP_490597.1 23476 | | BETHYL LABORATORIES, INC | |
|--|--|---|---|--|-----------------------------|------------------|
| APPLICATIONS | | WB, IP, IHC | | | | |
| SPECIES REACTIVITY | | Human, Mouse | | | | |
| AMOUNT | | 50 µl | | | | |
| 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 | | Antibody was affinity purified using an epitope specific to BRD4 immobilized on solid support. | | | | |
| PROCEDURES | | The epitope recognized by A301-985A50 maps to a region between residue 1312 and 1362 of human bromodomain-containing protein 4 using the numbering given in entry NP_490597.1 (GeneID 23476). 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 | 1:2 | 2,000 - 1:10,000 | | |
| | | Immunoprecipit | tation 2 - | 5 µg/mg lysate | | |
| | | Immunohistoch | | ,000 – 1:5,000. Epitope rei commended for FFPE tissue | | buffer pH 6.0 is |
| APPLICATION NOTES | | Western blot of immunoprecipitates performed using Normal Pig Serum (Cat. No. S100–020), Goat anti–Rabbit Light Chain HRP Conjugate (Cat. No. A120–113P) and 4–8% SDS–PAGE (link to IP–western blot protocol in Additional Info section below). | | | | |
| | | | of lysates performed using standard western blot reagents and 4–8% SDS–PAGE. ma, Ovarian Carcinoma | | | |
| IHC MOUSE CONTROLS | | Renal Cell Carcinoma | | | | |
| ADDITIONAL INFO | | https://www.bethyl.com/product/A301-985A50 Use the link above to view SDS, a current list of citations, and other product specific information. IP-western blot protocol https://www.bethyl.com/content/protocol_IP_WB | | | | |

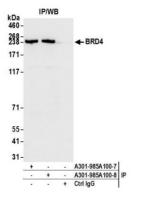
This document certifies that this product has met all of the quality control standards defined by Bethyl Laboratories, Inc. Eric McIntush, PhD | Chief Scientific Officer Date: December 20, 2019



Detection of human BRD4 by western blot. *Samples:* Whole cell lysate (10 µg) from HeLa, HEK293T, MDA-MB-231, Hep-G2, A-549, SW620, and SK-MEL-28 cells prepared using NETN lysis buffer. *Antibody:* Affinity purified rabbit anti-BRD4 antibody A301-985A50 (lot A301-985A50-8) used for WB at 0.04 µg/ml. *Detection:* Chemiluminescence with an exposure time of 10 seconds.

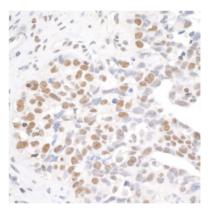


Detection of mouse BRD4 by western blot. Samples: Whole cell lysate (15 μ g) from BW5147.3, EL4, RenCa, NIH 3T3, and TCMK-1 cells prepared using NETN lysis buffer. Antibody: Affinity purified rabbit anti-BRD4 antibody A301-985A50 (lot A301-985A50-8) used for WB at 0.04 μ g/ml. Detection: Chemiluminescence with an exposure time of 3 seconds.



Detection of human BRD4 by western blot of

immunoprecipitates. *Samples:* Whole cell lysate (1.0 mg per IP reaction; 20% of IP loaded) from HeLa cells prepared using NETN lysis buffer. *Antibodies:* Affinity purified rabbit anti-BRD4 antibody A301–985A50 (lot A301–985A50–8) used for IP at 6 µg per reaction. BRD4 was also immunoprecipitated by a previous lot of this antibody (lot A301–985A50–7). For blotting immunoprecipitated BRD4, rabbit anti-BRD4 recombinant monoclonal antibody A700–004 was used at 1:1000. *Detection:* Chemiluminescence with an exposure time of 1 second.

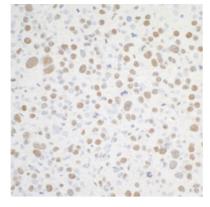


Detection of human BRD4 by immunohistochemistry. *Sample:* FFPE section of human ovarian carcinoma. *Antibody:* Affinity purified rabbit anti-BRD4 (Cat. No. A301-985A50 lot 8) used at a dilution of 1:5,000 (0.2 µg/ml). *Detection:* DAB



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3



Detection of mouse BRD4 by immunohistochemistry. *Sample:* FFPE section of mouse renal cell carcinoma. *Antibody:* Affinity purified rabbit anti-BRD4 (Cat. No. A301-985A50 lot 8) used at a dilution of 1:5,000 (0.2 µg/ml). *Detection:* DAB

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