

BRG1/SMARCA4 Antibody: CUTANA™ Compatible (CUT&RUN)



EpiCypher®

www.epicypher.com

Catalog No. 13-2002

Lot No. 20240001-49

Pack Size 100 µL

Type Polyclonal

Target Size 185 kDa

Reactivity H, M

Host Rabbit

Format Aff. Pur. IgG

Applications CUT&RUN, WB, IP, IHC

Product Description:

This antibody meets EpiCypher's "CUTANA Compatible" criteria for performance in Cleavage Under Targets and Release Using Nuclease (CUT&RUN) and/or Cleavage Under Targets and Tagmentation (CUT&Tag) approaches to genomic mapping. Every lot of a CUTANA Compatible antibody is tested in the indicated CUTANA approach using EpiCypher optimized protocols (EpiCypher.com/resources/protocols) and determined to yield peaks that show a genomic distribution pattern consistent with reported function(s) of the target protein. BRG1 antibody produces CUT&RUN peaks primarily flanking transcription start sites (TSSs, **Figure 1**). BRG1 peaks show a large degree of overlap with BRD4 peaks (**Figure 2**), as has been reported in the literature (Conrad et al., 2017).

Immunogen:

A synthetic peptide corresponding to human BRG1 amino acids 75 to 125.

Formulation:

Antigen affinity-purified antibody (200 µg/mL) in Tris-buffered saline with 0.1% BSA and 0.09% sodium azide.

Storage and Stability:

Stable for 1 year at 4°C from date of receipt.

Application Notes:

Recommended Dilutions:

CUT&RUN: 0.5 µg **IP:** 2 - 5 µg/mg lysate

WB: 1:2,000 - 1:10,000 **IHC:** 1:250 - 1:2,000*

*Epitope retrieval with citrate buffer pH 6.0 recommended for FFPE tissue

References:

Conrad et al (2017) Mol Cell 12:42.

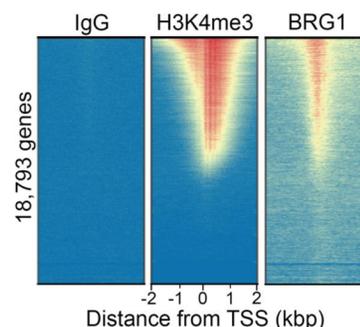


Figure 1: BRG1 enrichment at annotated TSSs in CUT&RUN. CUT&RUN was performed using 500,000 K562 cells with BRG1 antibody as well as control antibodies (IgG negative control, EpiCypher 13-0042; H3K4me3 positive control, EpiCypher 13-0041). Sequencing reads were aligned to annotated TSSs (+/- 2 kbp) of 18,793 genes. High, medium, and low signal is ranked by intensity (top to bottom) and reflected by red, yellow, and blue colors, respectively. All rows aligned relative to H3K4me3 antibody.

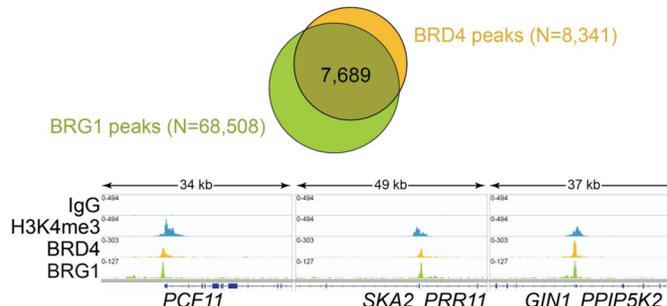


Figure 2: BRG1 CUT&RUN peak enrichment and functional overlap. The CUT&RUN data from Figure 1 was subjected to peak calling using MACS2. BRG1 peaks overlapped with BRD4 antibody CUT&RUN peaks (EpiCypher 13-2003, top), as has been demonstrated in the literature (Conrad et al., 2017). Three representative loci show BRG1 peaks in relation to control and BRD4 antibodies (bottom, Integrative Genomics Viewer).

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Applications Key: CHIP: Chromatin immunoprecipitation; CUT&RUN: Cleavage Under Targets and Release Using Nuclease; CUT&Tag: Cleavage Under Targets and Tagmentation; E: ELISA; FACS: Flow cytometry; ICC: Immunocytochemistry; IF: Immunofluorescence; IHC: Immunohistochemistry; IP: Immunoprecipitation; L: Luminex; WB: Western Blot. **Reactivity Key:** B: Bovine; Ce: C. elegans; Ch: Chicken; Dm: Drosophila; Eu: Eukaryote; H: Human; M: Mouse; Ma: Mammal; R: Rat; Sc: S.cerevesiae; Sp: S. pombe; WR: Wide Range (predicted); X: Xenopus; Z: Zebrafish

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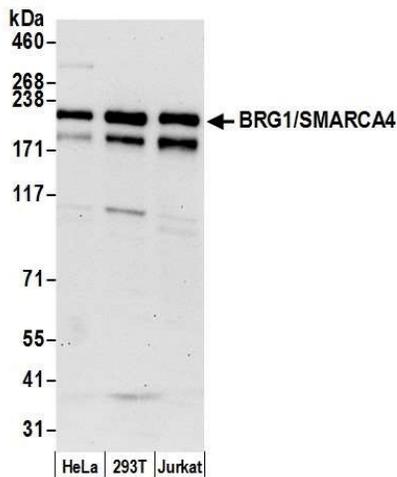


Figure 3: Western blot detection of human BRG1. Whole cell lysates were isolated from HeLa, HEK293T ("293T"), and Jurkat cells using NETN lysis buffer. Lysates (15 µg) were loaded onto 4-8% SDS-PAGE gel and analyzed under standard western blot conditions using BRG1 antibody (0.1 µg/mL).

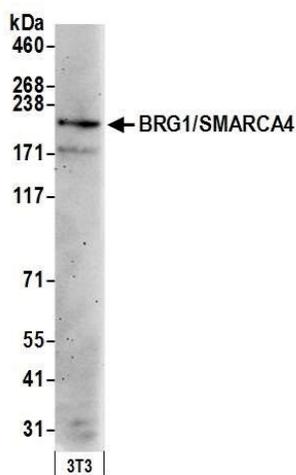


Figure 4: Western blot detection of mouse BRG1. Whole cell lysates were isolated from NIH 3T3 ("3T3") cells using NETN lysis buffer. Lysates (15 µg) were loaded onto 4-8% SDS-PAGE gel and analyzed under standard western blot conditions using BRG1 antibody (0.1 µg/mL).

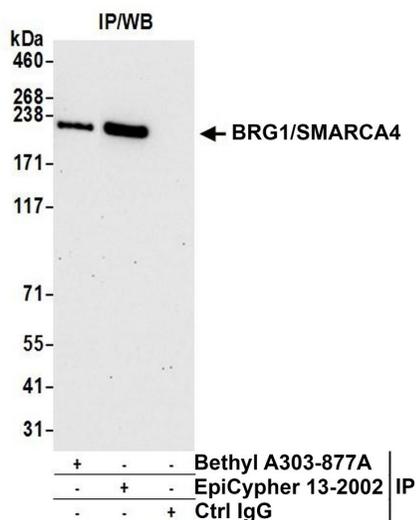


Figure 5: Immunoprecipitation of human BRG1. EpiCypher BRG1 antibody (3 µg) was used to immunoprecipitate whole cell lysates isolated from HeLa cells using NETN lysis buffer (1 mg per IP). A negative control IgG antibody and positive control antibody to a different BRG1/SMARCA4 epitope (Bethyl Laboratories) were also used to demonstrate specificity of the IP. Immunoprecipitates were loaded onto 4-20% SDS-PAGE gel (20% of IP loaded) and probed via western blot with EpiCypher BRG1 antibody (1 µg/mL).

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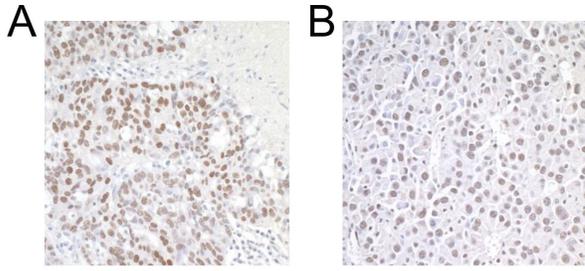


Figure 6: Immunohistochemistry detection of human and mouse BRG1. A) FFPE section of human ovarian carcinoma examined using BRG1 antibody (1:1,000 dilution, 0.2 $\mu\text{g}/\text{mL}$). **B)** FFPE section of mouse renal cell carcinoma examined using BRG1 antibody (1:1,000 dilution, 0.2 $\mu\text{g}/\text{mL}$).

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