

BRM/SMARCA2 CUTANA™ CUT&RUN Antibody

Catalog No	13-2006	Туре	Polyclonal
Lot No	21013001-40	Host	Rabbit
Pack Size	100 μL	Concentration	200 μg/mL
Applications	CUT&RUN, WB, IP	Reactivity	Human

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 approach using EpiCypher optimized protocols (epicypher.com/protocols) and determined to yield peaks that show a genomic distribution pattern consistent with reported function(s) of the target protein. BRM/SMARCA2 antibody produces CUT&RUN peaks above background (Figure 1) localized to gene transcription start sites (Figures 1-2), consistent with its known role as the ATP-dependent helicase subunit of the SWI/SNF chromatin remodeler complex [1].

TECHNICAL INFORMATION

Immunogen Between amino acids 1 and 50

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

Formulation Antigen affinity-purified antibody in Tris-buffered saline, 0.1% BSA, 0.09% sodium azide

RECOMMENDED DILUTION

CUT&RUN 0.5 µg per reaction **Western Blot** 1:2,000 - 1:10,000

Immunoprecipitation 2 - 5 μg/mg lysate

GENE & PROTEIN INFORMATION

UniProt ID P51531
Gene Name SMARCA2

Protein Name Probable global transcription activator SNF2L2

Target Size 181 kDa

Alternate Names BAF190B, SNF2A, SNF2L2, ATP-dependent helicase SMARCA2, BRG1-associated factor 190B

(BAF190B), Protein bahma homolog (hBRM), SNF2-alpha, SWI-SNF-related matrix-associated

actin-dependent regulator of chromatin subfamily A member 2

REFERENCES

[1] Raab et al. Epigenetics Chromatin (2017). PMID: 29273066

CUT&RUN Methods

CUT&RUN was performed on 500k native or fixed (0.1% formaldehyde, 1 min) K562 cells with 0.5 µg of either BRM, H3K4me3 positive control (EpiCypher 13-0041), or IgG negative control (EpiCypher 13-0042) antibodies using the CUTANATM ChIC/CUT&RUN Kit v2.0 (EpiCypher 14-1048). Library preparation was performed with 5 ng of DNA (or the total amount recovered if less than 5 ng) using the CUTANATM CUT&RUN Library Prep Kit (EpiCypher 14-1001/14-1002). Both kit protocols were adapted for high throughput Tecan liquid handling. Libraries were run on an Illumina NextSeq2000 with paired-end sequencing (2x50 bp). Sample sequencing depth was 10.2 million reads (BRM native), 10.0 million reads (BRM 0.1% fixation), 4.1 million reads (H3K4me3), and 2.4 million reads (IgG). Data were aligned to the hg19 genome using Bowtie2. Data were filtered to remove duplicates, multi-aligned reads, and ENCODE DAC Exclusion List regions.

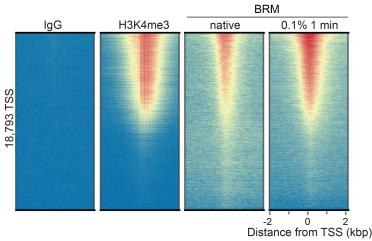
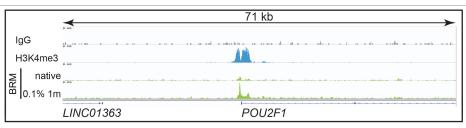


FIGURE 1 BRM peaks in CUT&RUN. CUT&RUN was performed as described above. Peaks were called with MACS2. Heatmaps show BRM peaks relative to IgG negative control antibody and H3K4me3 positive control antibody in aligned rows ranked by intensity (top to bottom) and colored such that red indicates high localized enrichment and blue denotes background signal. All rows aligned to BRM antibody with moderate fixation (0.1% formaldehyde, 1 min), which improved signal vs. native conditions.



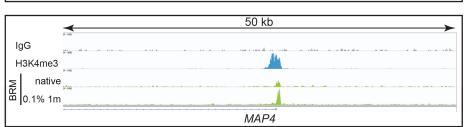


FIGURE 2 BRM CUT&RUN representative browser tracks. CUT&RUN was performed as described above. Gene browser shots were generated using the Integrative Genomics Viewer (IGV, Broad Institute). Two gene loci show overlap of BRM and H3K4me3 peaks, consistent with the reported function of BRM as a member of the SWI/SNF chromatin remodeler complex [1]. Improved signal recovery with moderate fixation (0.1% formaldehyde, 1 min) is notable.

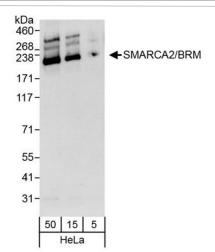


FIGURE 3 Western blot data. Whole cell lysates were isolated from HeLa cells using NETN lysis buffer. The indicated amounts (μg) of lysate were loaded onto a 4-8% SDS-PAGE gel and analyzed under standard western blot conditions using BRM antibody (0.04 μg/mL).

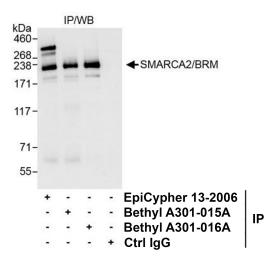


FIGURE 4 Immunoprecipitation data. EpiCypher BRM antibody (3 μ g) was used to immunoprecipitate whole cell lysates isolated from HeLa cells using NETN lysis buffer (1 mg per IP). A negative control IgG antibody and positive control antibodies to various BRM epitopes (Bethyl Laboratories) were also used to demonstrate the specificity of the IP. Immunoprecipitates were loaded onto a 4-8% SDS-PAGE gel (20% of IP loaded) and probed via western blot with EpiCypher BRM antibody (1.0 μ g/mL).

This product is provided for commercial sale under license from Bethyl Laboratories, Inc.