EZH2 CUTANA™ CUT&RUN Antibody

Catalog No	13-2026	**Type**	Polyclonal
Lot No	22070001-90	**Host**	Rabbit
Pack Size	100 µL	**Concentration**	1,000 µg/mL
Applications	CUT&RUN, IHC, IP, WB	**Reactivity**	Human, Mouse

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. EZH2 catalyzes histone H3 lysine 27 trimethylation (H3K27me3), which leads to transcriptional repression of target genes [1]. EZH2 antibody produces CUT&RUN peaks above background primarily in intronic and intergenic regions (**Figure 1**) that overlap with H3K27me3 (**Figure 2**), consistent with its known role as the histone methyltransferase catalytic subunit of the Polycomb Repressive complex 2 (PRC2).

TECHNICAL INFORMATION

Immunogen	Between amino acids 696 and 746
Storage	Stable for 1 year at 4°C from date of receipt
Formulation	Antigen affinity-purified antibody in Tris-citrate/phosphate buffer pH 7-8, 0.09% sodium azide

RECOMMENDED DILUTION

CUT&RUN	0.5 µg per reaction	**Immunoprecipitation**	2 - 10 µg/mg lysate
Immunohistochemistry	1:500 - 1:2,000	**Western Blot**	1:500 - 1:2,500

Epitope retrieval with citrate buffer pH 6.0 is recommended for FFPE tissue sections

GENE & PROTEIN INFORMATION

UniProt ID	Q15910
Gene Name	enhancer of zeste 2 polycomb repressive complex 2
Protein Name	Histone-lysine N-methyltransferase EZH2
Target Size	85 kDa
Alternate Names	enhancer of zeste homolog 2, ENX1, ENX-1, EZH2b, KMT6, KMT6A, WVS, WVS2

REFERENCES

[1] Cao et al. *Science* (2002). PMID: 12351676

VALIDATION DATA

CUT&RUN Methods

CUT&RUN was performed on 500k HeLa cells with 0.5 µg of either EZH2 or IgG negative control (EpiCypher 13-0042) antibodies using the CUTANA™ 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 CUTANA™ 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 5.1 million reads (IgG), 5.1 million reads (EZH2), and 5.7 million reads (H3K27me3). Data were aligned to the hg19 genome using Bowtie2. Data were filtered to remove duplicates, multi-aligned reads, and ENCODE DAC Exclusion List regions.

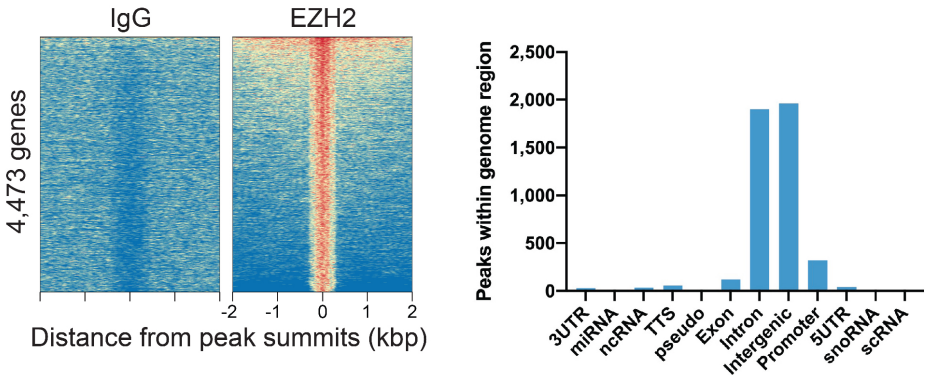

FIGURE 1 EZH2 peaks in CUT&RUN. CUT&RUN was performed as described above. Peaks were called using MACS2. Heatmaps show EZH2 peaks relative to IgG negative control antibody in aligned rows ranked by intensity (top to bottom) and color such that red indicates high localized enrichment and blue denotes background signal (**left**). The number of peaks that fall into distinct classes of functionally annotated genomic regions are shown (**right**).

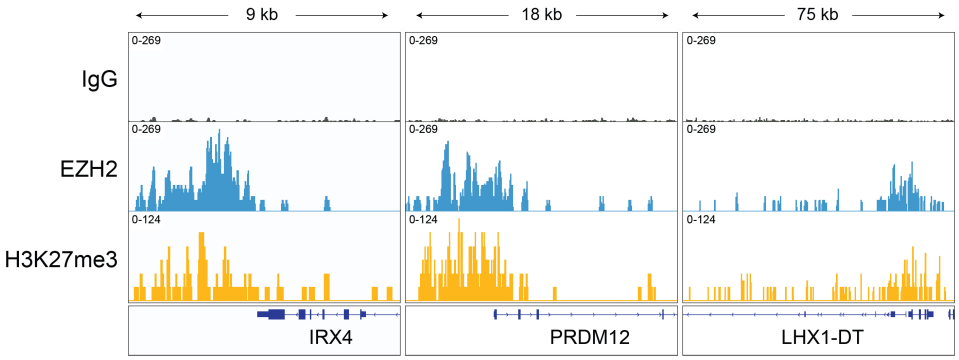

FIGURE 2 EZH2 CUT&RUN representative browser tracks. CUT&RUN was performed as described above. Gene browser shots were generated using the Integrative Genomics Viewer (IGV, Broad Institute). Three representative loci show overlap between EZH2 and H3K27me3 peaks.

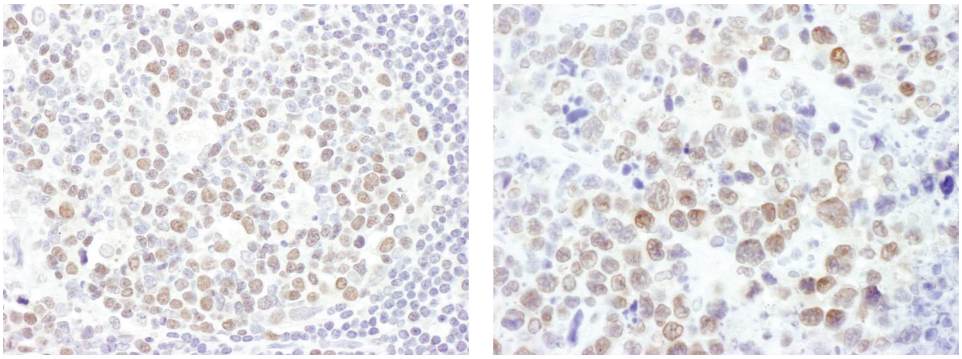

FIGURE 3 Immunohistochemistry data. FFPE sections of human ovarian carcinoma (**left**) and mouse plasmacytoma (**right**) using EZH2 antibody at a dilution of 1:1,000.

VALIDATION DATA

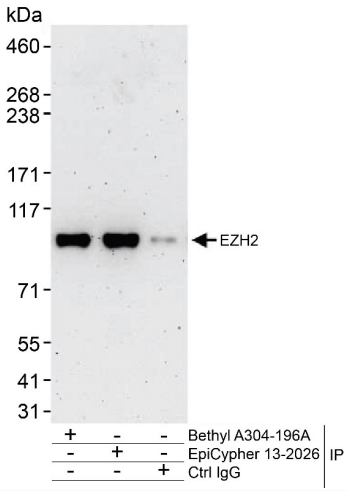

FIGURE 4 Immunoprecipitation data. EpiCypher EZH2 antibody (6 µg/mg lysate) was used to immunoprecipitate whole cell lysates (1 mg, 20% of IP loaded) isolated from HEK293T cells. A negative control IgG antibody and positive control antibodies targeting a different EZH2 epitope (Bethyl Laboratories) were also used to demonstrate specificity of the IP. For blotting immunoprecipitates, EpiCypher EZH2 antibody was used at a 1:1,000 dilution.

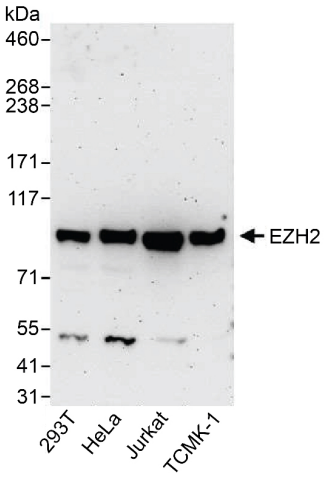

FIGURE 5 Western blot data. Western analysis of EZH2 in whole cell extracts from HEK293T, HeLa, Jurkat, and mouse TCMK-1 cells. Fifty micrograms of lysate was resolved via SDS-PAGE and detected with a 1:1,000 dilution of EZH2 antibody.