SP1 CUTANA™ CUT&RUN Antibody

Catalog No	13-2024	**Type**	Polyclonal
Lot No	22056001-82	**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. SP1 is the original member of the Sp1-like family of zinc-finger transcription factors. It binds GC-rich motifs with high affinity [1] and is involved in expressing genes related to cell growth, cell-cycle progression, survival, and tumorigenesis [2]. SP1 antibody produces CUT&RUN peaks above background primarily in intronic and promoter regions (**Figure 1**) that overlap with known SP1 DNA-binding motifs (**Figure 2**).

TECHNICAL INFORMATION

Immunogen	Between amino acids 750 and 785
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	1:2000-1:10,000	**Western Blot**	1:2,000 - 1:10,000

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

GENE & PROTEIN INFORMATION

UniProt ID	P08047
Gene Name	SP1
Protein Name	Transcription factor Sp1
Target Size	81 kDa
Alternate Names	specificity protein 1, transcription factor Sp1, TSFP1

REFERENCES

[1] Raiber et al. *Nucleic Acids Res* (2012). PMID: 22021377

VALIDATION DATA

CUT&RUN Methods

CUT&RUN was performed on 500k HeLa cells with 0.5 µg of either SP1 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) and 5.3 million reads (SP1). Data were aligned to the hg19 genome using Bowtie2. Data were filtered to remove duplicates, multi-aligned reads, and ENCODE DAC Exclusion List regions.

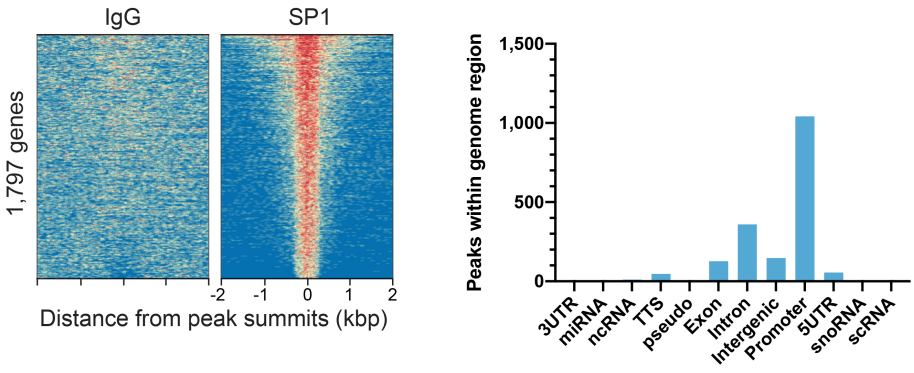

FIGURE 1 SP1 peaks in CUT&RUN. CUT&RUN was performed as described above. Peaks were called with MACS2. Heatmaps show SP1 peaks relative to IgG negative control antibody in aligned rows ranked by intensity (top to bottom) and colored such that red indicates high localized enrichment and blue denotes background signal (**left**). The number of peaks that fall into distinct classes of functionally annotated genomic regions are shown (**right**).

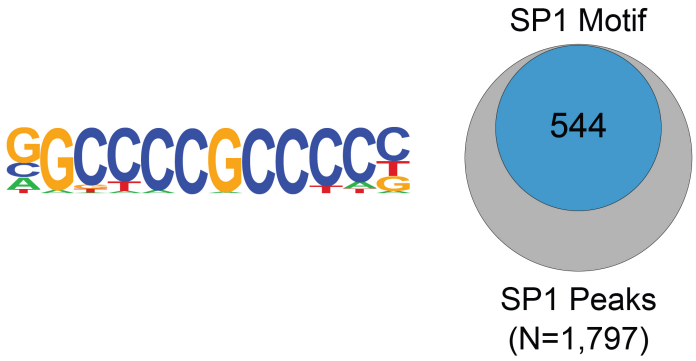

FIGURE 2 SP1 transcription factor binding motif analysis in CUT&RUN. Sp1(Zf) /Promoter/Homer consensus motif, represented as a sequence logo position weight matrix, was the top called motif significantly enriched under SP1 CUT&RUN peaks (**left**). The number of SP1 peaks containing the consensus motif from the left panel is represented by a Venn Diagram (**right**).

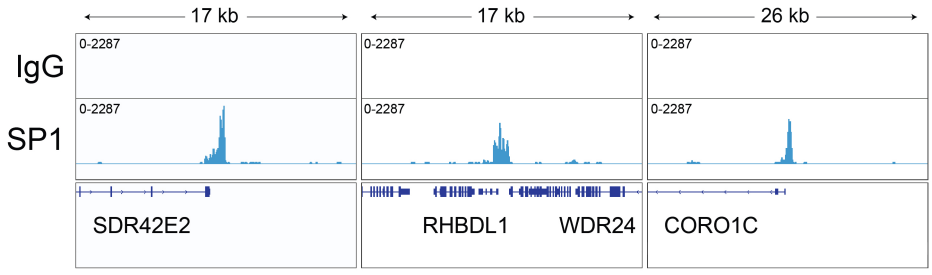

FIGURE 3 SP1 CUT&RUN representative browser tracks. CUT&RUN was performed as described above. Gene browser shots were generated using the Integrative Genomics Viewer (IGV, Broad Institute). Three representative loci of the top called peaks are shown.

VALIDATION DATA

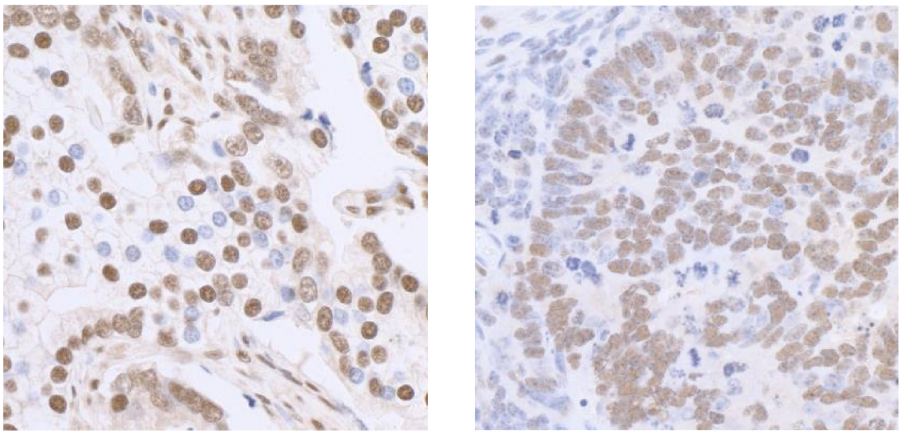

FIGURE 4 Immunohistochemistry data. FFPE sections of human breast carcinoma (**left**) and mouse renal cell carcinoma (**right**) using SP1 antibody at a dilution of 1:5,000.

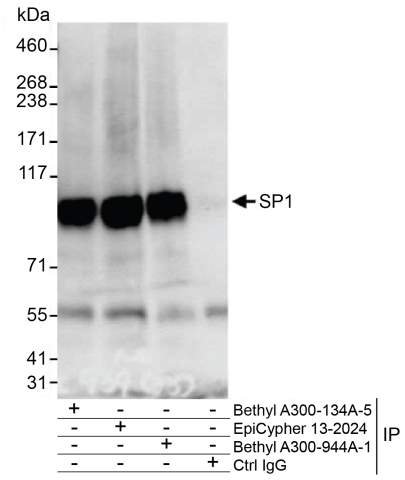

FIGURE 5 Immunoprecipitation data. EpiCypher SP1 antibody (6 µg) was used to immunoprecipitate whole cell lysates (0.5 - 1 mg, 20% of IP loaded) isolated from HeLa cells using NETN lysis buffer. A negative control IgG antibody and positive control antibodies targeting SP1 (Bethyl Laboratories) were also used to demonstrate specificity of the IP. EpiCypher 13-2024 and Bethyl A300-134A-5 target the same epitope, while Bethyl A300-944A-1 targets a different epitope (between amino acids 735 and 785). For blotting immunoprecipitates, EpiCypher SP1 antibody was used at a 1:1,000 dilution.

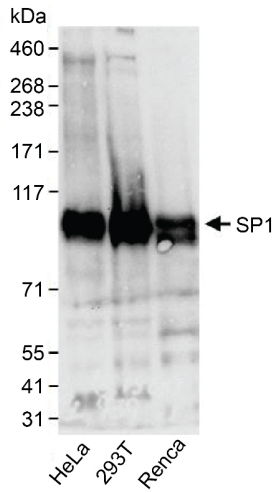

FIGURE 6 Western blot data. Western analysis of SP1 in whole cell extracts from HeLa, HEK293T, and mouse Renca cells. Fifty micrograms of lysate was resolved via SDS-PAGE and detected with 1:10,000 dilution of SP1 antibody.