TP53/p53 CUTANA™ CUT&RUN Antibody

Catalog No	13-2015	**Type**	Polyclonal
Lot No	22070001-81	**Host**	Rabbit
Pack Size	100 µL	**Concentration**	1,000 µg/mL
Applications	CUT&RUN, IP, WB	**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. TP53/p53 acts as a tumor suppressor by promoting growth arrest or apoptosis and is mutated in over 50% of cancers. It forms a complex with NF-Y, which binds to CCAAT box-containing promoters [1]. TP53/p53 antibody produces CUT&RUN peaks above background primarily in promoter regions (**Figure 1**) that are enriched with known TP53/p53 DNA-binding motifs (**Figure 2**).

TECHNICAL INFORMATION

Immunogen	Between amino acids 325 and 375
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	**Western Blot**	1:2,000 - 1:10,000
Immunoprecipitation	2-10 µg/mg lysate		

GENE & PROTEIN INFORMATION

UniProt ID	P04637
Gene Name	TP53
Protein Name	Cellular tumor antigen p53
Target Size	43 kDa
Alternate Names	antigen NY-CO-13, BCC7, LFS1, mutant tumor protein p53, P53, p53 tumor suppressor, TRP53

REFERENCES

[1] Benatti et al. *Nucleic Acids Res* (2008). PMID: 18187512

VALIDATION DATA

CUT&RUN Methods

CUT&RUN was performed on 500k HeLa cells with 0.5 µg of either TP53/p53 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 5.9 million reads (TP53). Data were aligned to the hg19 genome using Bowtie2. Data were filtered to remove duplicates, multi-aligned reads, and ENCODE DAC Exclusion List regions.

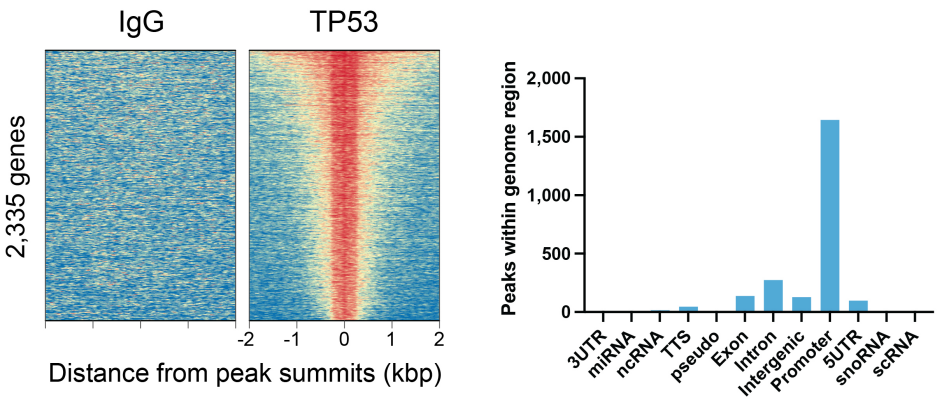

FIGURE 1 TP53/p53 peaks in CUT&RUN. CUT&RUN was performed as described above. Peaks were called using MACS2. Heatmaps show TP53/p53 peaks relative to IgG negative control antibody in aligned rows ranked by intensity (top to bottom) and colored such that red indicates high localized enrichment and blue denotes background signal (**left**). The number of peaks that fall into distinct classes of functionally annotated genomic regions are shown (**right**).

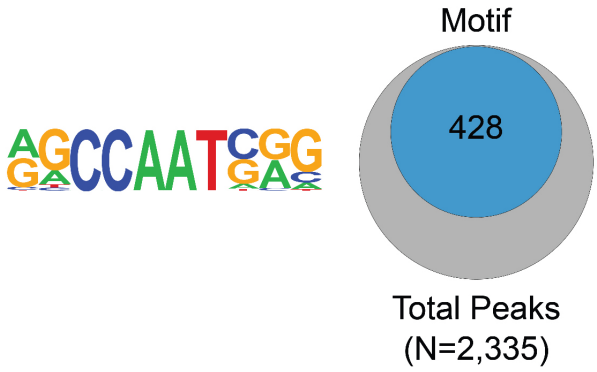

FIGURE 2 TP53/p53 transcription factor binding motif analysis in CUT&RUN. NFY (CCAAT)/Promoter/Homer consensus motif, represented as a sequence logo position weight matrix, was the top called motif significantly enriched under TP53/p53 CUT&RUN peaks (**left**). The number of TP53/p53 peaks containing the consensus motif from the left panel is represented by a Venn Diagram (**right**).

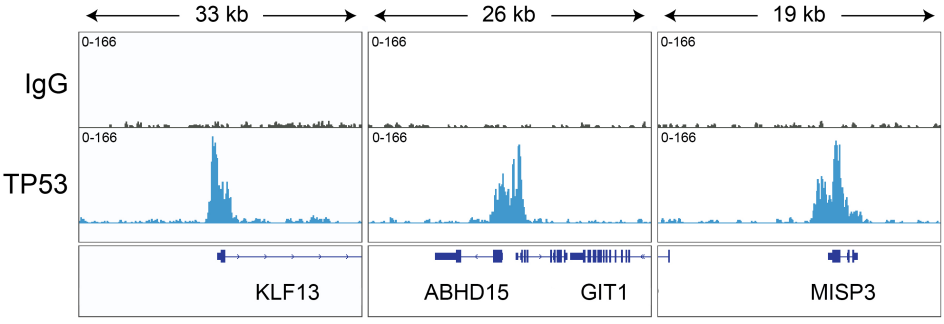

FIGURE 3 TP53/p53 CUT&RUN representative browser tracks. CUT&RUN was performed as described above. Gene browser shots were generated using the Integrative Genomics Viewer (IGV, Broad Institute). Three representative loci of the top called peaks are shown.

VALIDATION DATA

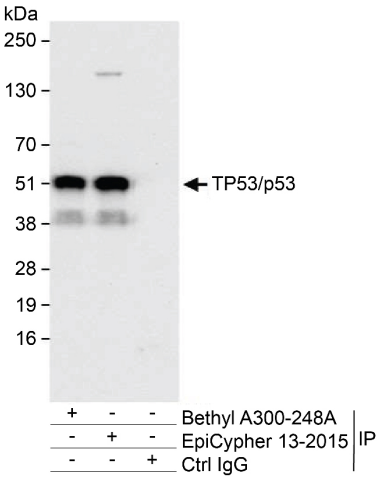

FIGURE 4 Immunoprecipitation data. EpiCypher TP53/p53 antibody (6 µg/reaction) was used to immunoprecipitate whole cell lysates (1 mg, 20% of IP loaded) isolated from HEK293T cells. A negative control IgG antibody and positive control antibodies targeting the same epitope (Bethyl Laboratories) were also used to demonstrate specificity of the IP. For blotting immunoprecipitates, EpiCypher TP53/p53 antibody was used at a 1:2,500 dilution.

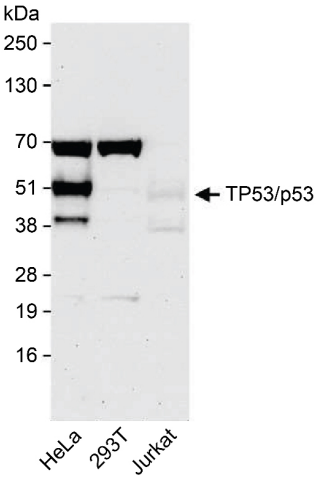

FIGURE 5 Western blot data. Western analysis of TP53/p53 in whole cell extracts from HeLa, HEK293T, and Jurkat cells. Fifty micrograms of lysate was resolved via SDS-PAGE and detected with a 1:10,000 dilution of TP53/p53 antibody.