CHD3 CUTANA™ CUT&RUN Antibody

Catalog No	13-2009	**Type**	Polyclonal
Lot No	21013001-43	**Host**	Rabbit
Pack Size	100 µL	**Concentration**	1000 µg/mL
Applications	CUT&RUN, IP, IHC	**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. CHD3 is an understudied member of the nucleosome remodeling and deacetylase (NuRD) complex. CHD3 antibody produces CUT&RUN peaks above background (**Figure 1**) within gene promoters, intergenic regions, and introns (**Figures 1-2**).

TECHNICAL INFORMATION

Immunogen	Between amino acids 1950 and 2000
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 - 5 µg/mg lysate
Immunohistochemistry	1:500 - 1:2,000		

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

GENE & PROTEIN INFORMATION

UniProt ID	Q12873
Gene Name	CHD3
Protein Name	Chromodomain-helicase-DNA-binding protein 3
Target Size	227 kDa
Alternate Names	ATP-dependent helicase CHD3, Mi-2 autoantigen 240 kDa protein, Mi2-alpha, Zinc finger helicase (hZFH), CHD-3

REFERENCES

VALIDATION DATA

CUT&RUN Methods

CUT&RUN was performed on 500k K562 cells with 0.5 µg of either CHD3, H3K4me3 positive control (EpiCypher 13-0041), 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 2.4 million reads (IgG), 4.1 million reads (H3K4me3), and 5.0 million reads (CHD3). Data were aligned to the hg19 genome using Bowtie2. Data were filtered to remove duplicates, multi-aligned reads, and ENCODE DAC Exclusion List regions.

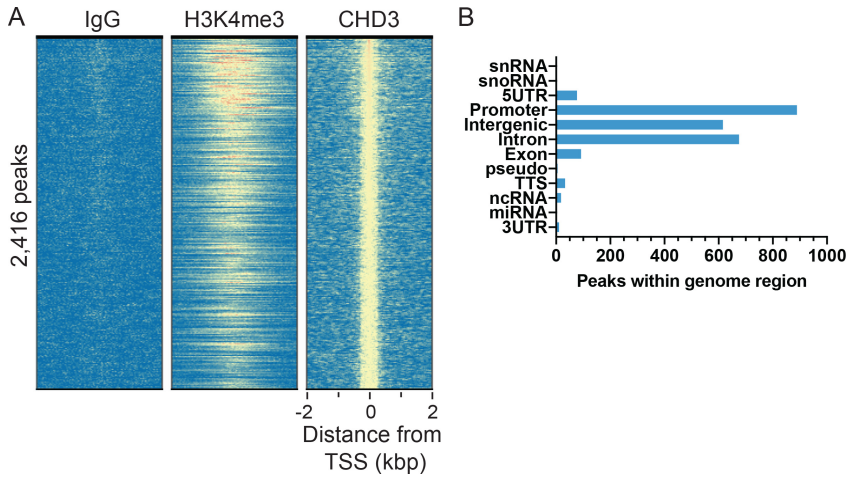

FIGURE 1 CHD3 peaks in CUT&RUN. CUT&RUN was performed as described above. Peaks were called with MACS2. (**A**) Heatmaps show CHD3 peaks relative to IgG negative control antibody in aligned rows ranked by intensity (top to bottom) and colored such that red indicates high localized enrichment and blue denotes background signal. (**B**) The number of peaks that fall into distinct classes of functionally annotated genomic regions are shown.

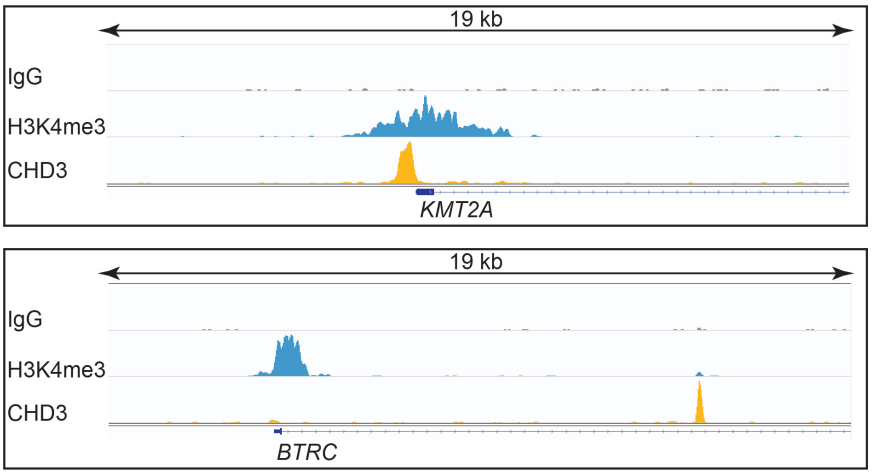

FIGURE 2 CHD3 CUT&RUN representative browser tracks. CUT&RUN was performed as described above. Gene browser shots were generated using the Integrative Genomics Viewer (IGV, Broad Institute). Two gene loci showing CHD3 peaks at a gene promoter (top) and an intronic region (bottom) are representative of the functional annotation analysis of CHD3 peak localization in CUT&RUN (Figure 1).

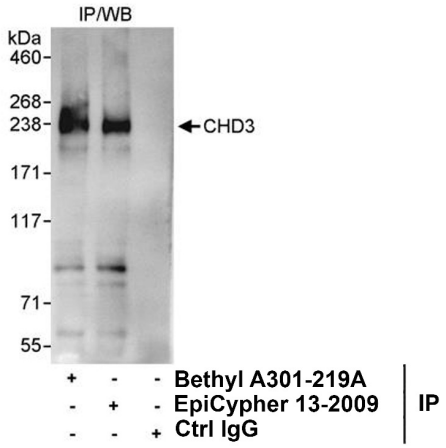

FIGURE 3 Immunoprecipitation data. EpiCypher CHD3 antibody (3 µg) was used to immunoprecipitate whole cell lysates isolated from HeLa cells using NETN lysis buffer (1.0 mg per IP). A negative control IgG antibody and positive control antibody to a different CHD3 epitope (Bethyl Laboratories) were also used to demonstrate specificity of the IP. Immunoprecipitates were loaded onto a 4-8% SDS-PAGE gel (20% of IP loaded) and probed via western blot with Bethyl A301-219A antibody (1.0 µg/mL).

VALIDATION DATA

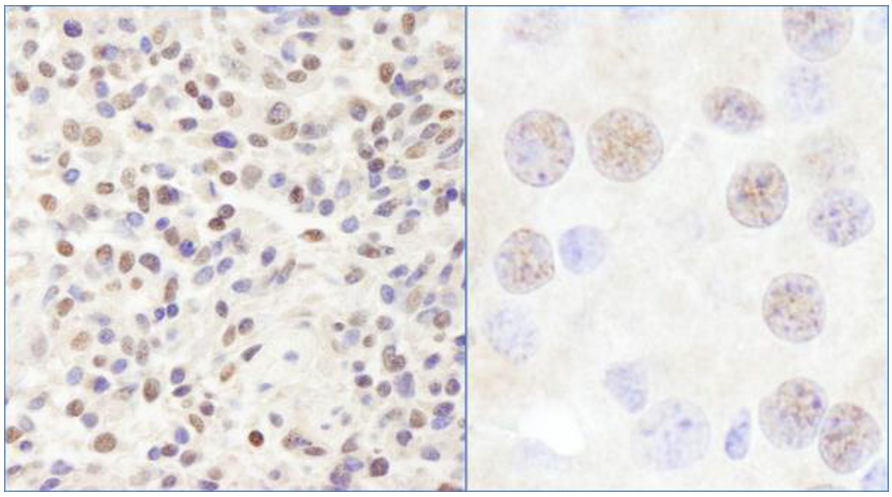

FIGURE 4 **Immunohistochemistry data.** FFPE section of human Ewing sarcoma (**left**) and mouse renal cell carcinoma (**right**) using CHD3 antibody at a dilution of 1:1,000.