CHD4 CUTANA™ CUT&RUN Antibody

Catalog No	13-2016	**Type**	Polyclonal
Lot No	22070001-82	**Host**	Rabbit
Pack Size	100 µL	**Concentration**	1,000 µg/mL
Applications	CUT&RUN, 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. CHD4 is a member of the nucleosome remodeling and deacetylase (NuRD) complex. Within the complex, CHD4 remodels chromatin by deacetylating histones [1]. CHD4 antibody produces CUT&RUN peaks above background (**Figure 1**) within gene promoters, intergenic, and intronic regions (**Figures 1-2**).

TECHNICAL INFORMATION

Immunogen	Between amino acids 25 and 75
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**	1:10,000 - 1:25,000
Immunoprecipitation	2-10 µg/mg lysate		

GENE & PROTEIN INFORMATION

UniProt ID	Q14839
Gene Name	CHD4
Protein Name	Chromodomain-helicase-DNA-binding protein 4
Target Size	218 kDa
Alternate Names	ATP-dependent helicase CHD4, CHD-4, Mi-2b, Mi2-beta, Mi-2 autoantigen 218 kDa protein, Mi-2b

REFERENCES

[1] Tong et al. *Nature* (1998). PMID: 9804427

VALIDATION DATA

CUT&RUN Methods

CUT&RUN was performed on 500k HeLa cells with 0.5 µg of either CHD4 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 5.1 was million reads (IgG), 17.4 million reads (CHD4), and 6.2 million reads (H3K4me3). Data were aligned to the hg19 genome using Bowtie2. Data were filtered to remove duplicates, multi-aligned reads, and ENCODE DAC Exclusion List regions.

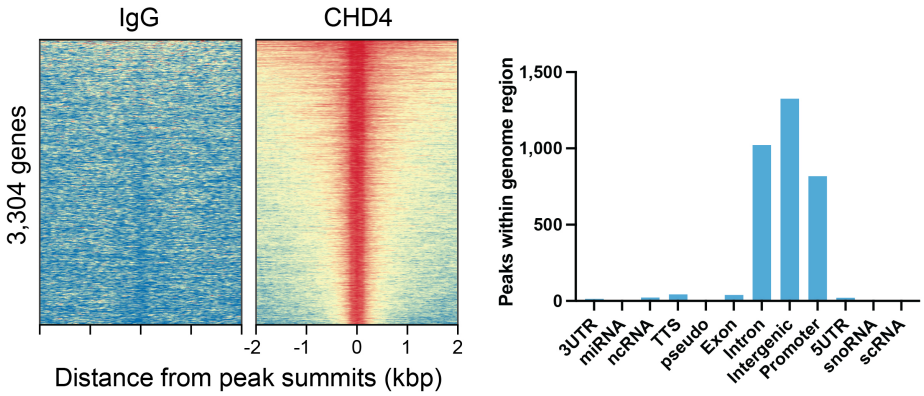

FIGURE 1 CHD4 peaks in CUT&RUN. CUT&RUN was performed as described above. Peaks were called using MACS2. Heatmaps show CHD4 peaks relative to IgG negative control antibody in aligned rows ranked by intensity (top to bottom) and colored such that red indicates high localized enrichment and blue denotes background signal (**left**). The number of peaks that fall into distinct classes of functionally annotated genomic regions are shown (**right**).

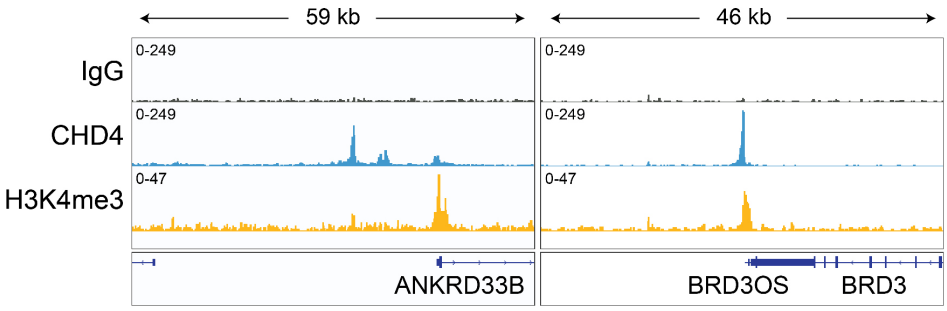

FIGURE 2 CHD4 CUT&RUN representative browser tracks. CUT&RUN was performed as described above. Gene browser shots were generated using the Integrative Genomics Viewer (IGV, Broad Institute). Two gene loci show CHD4 peaks at intergenic (**left panel**) and promoter (**right panel**) regions, which is representative of the functional annotation analysis of CHD4 peak localization in CUT&RUN (**Figure 1**).

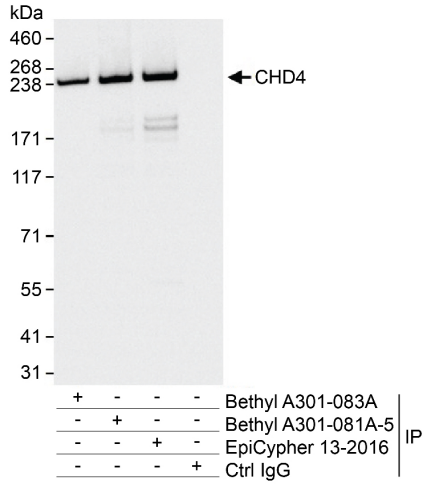

FIGURE 3 Immunoprecipitation data. EpiCypher CHD4 antibody (6 µg/reaction) was used to immunoprecipitate whole cell lysates (1 mg, 20% of IP loaded) isolated from HEK293T cells. A negative control IgG antibody and positive control antibodies targeting CHD4 (Bethyl Laboratories) were also used to demonstrate specificity of the IP. EpiCypher 13-2016 and Bethyl A301-081A-5 target the same epitope, while Bethyl A301-083A targets a different epitope (between amino acids 1625 and 1675). For blotting immunoprecipitates, EpiCypher CHD4 antibody was used at a 1:25,000 dilution.

VALIDATION DATA

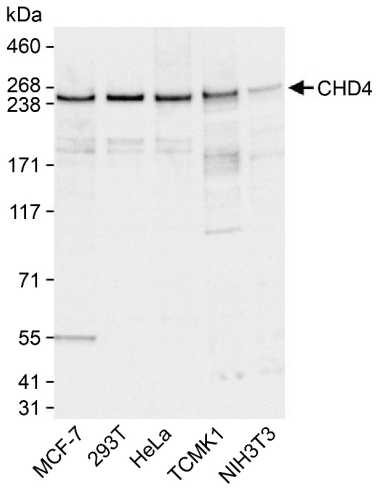

FIGURE 4 Western blot data. Western analysis of CHD4 in whole cell extracts from MCF-7, HEK293T, HeLa, TCMK1, and NIH3T3 cells. Fifty micrograms of lysate was resolved via SDS-PAGE and detected with a 1:25,000 dilution of CHD4 antibody.