EGFR CUTANA™ CUT&RUN Antibody

Catalog No	13-2018	**Type**	Polyclonal
Lot No	22070001-84	**Host**	Rabbit
Pack Size	100 µL	**Concentration**	1,000 µg/mL
Applications	CUT&RUN, IHC, WB	**Reactivity**	Human, Monkey (predicted)

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. EGFR is a tyrosine kinase receptor for epidermal growth factor (EGF) and other ligands of the EGF family. Activated EGFR can carry out its biological effects via the cytoplasmic pathway or nuclear pathway. The cytoplasmic pathway can cause tumorigenesis, metastasis, high proliferation, and resistance to chemotherapy and radiation. The nuclear pathway causes membrane EGFR to translocate into the nucleus and interact with other DNA-binding transcription factors, which can activate gene expression [1]. EGFR antibody produces CUT&RUN peaks above background (**Figure 1**) within gene promoters, intronic, and intergenic regions (**Figures 1-2**).

TECHNICAL INFORMATION

Immunogen	Between amino acids 1100 and 1150
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 - 1:50,000
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	P00533
Gene Name	EGFR
Protein Name	Epidermal growth factor receptor
Target Size	134 kDa
Alternate Names	Proto-oncogene c-ErbB-1, Receptor tyrosine-protein kinase erbB-1, ERBB, ERBB1, HER1, mENA,

REFERENCES

[1] Hung et al. *Nuc Acids Res* (2008). PMID: 18586824

VALIDATION DATA

CUT&RUN Methods

CUT&RUN was performed on 500k HeLa cells with 0.5 µg of either EGFR 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 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), 9.7 million reads (EGFR), 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.

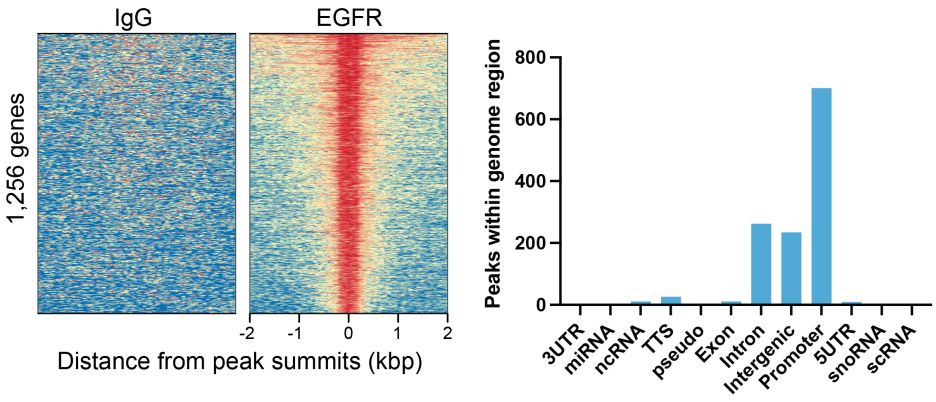

FIGURE 1 EGFR peaks in CUT&RUN. CUT&RUN was performed as described above. Peaks were called using MACS2. Heatmaps show EGFR peaks relative to IgG negative control antibody in aligned rows ranked by intensity (top to bottom) and colored such that red indicates high localized enrichment and blue denotes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

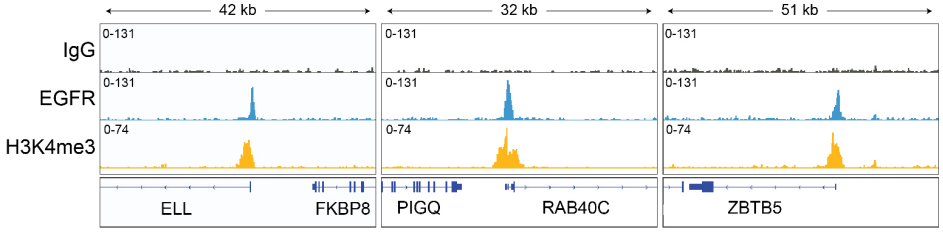

FIGURE 2 EGFR CUT&RUN representative browser tracks. CUT&RUN was performed as described above. Gene browser shots were generated using the Integrative Genomics Viewer (IGV, Broad Institute). Three gene loci show EGFR peaks at promoters, which is representative of the functional annotation analysis of EGFR peak localization in CUT&RUN (**Figure 1**).

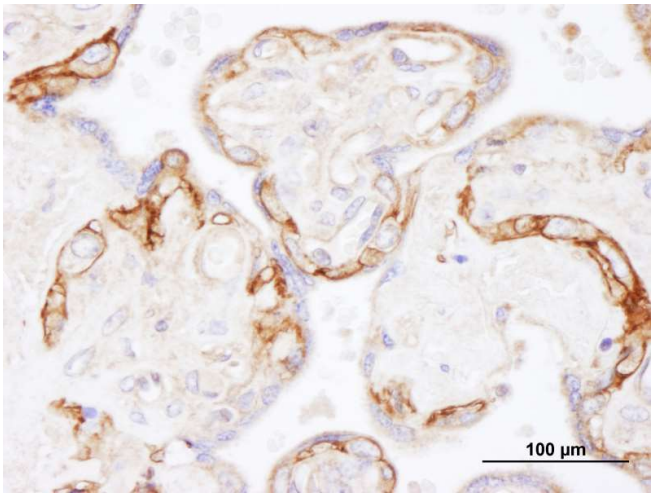

FIGURE 3 Immunohistochemistry data. FFPE section of human placenta using EGFR antibody at a dilution of 1:1,000.

VALIDATION DATA

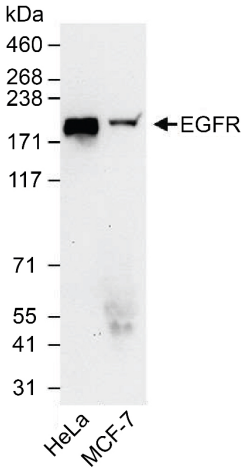

FIGURE 4 **Western blot data.** Western analysis of EGFR in whole cell extracts from HeLa and MCF-7 cells. Fifty micrograms of lysate was resolved via SDS-PAGE and detected with a 1:50,000 dilution of EGFR antibody.