

AbFlex® BRD4 antibody (rAb)

Catalog Nos: 91301, 91302

Application(s): ChIP, ChIP-Seq, CUT&RUN

Reactivity: Human

Quantities: 100 µg, 10 µg

Purification: Protein A Chromatography

Host: Rabbit

Isotype: IgG

Concentration: 1 µg/µl

Molecular Weight: 180 kDa

Background: AbFlex® antibodies are recombinant antibodies (rAbs) that have been generated using defined DNA sequences to produce highly specific, reproducible antibodies. Each AbFlex antibody contains a 6xHis Tag, a Biotinylation Tag for enzymatic biotin conjugation using the biotin ligase, BirA, and a sortase recognition motif (LPXTG) to attach a variety of labels directly to the antibody including fluorophores, enzymatic substrates (HRP, AP), peptides, drugs as well as solid supports. AbFlex® BRD4 antibody was expressed in 293 cells and contains rabbit immunoglobulin heavy and light chains.

BRD4 (Bromodomain-containing protein 4) belongs to the BET subclass of proteins, which are characterized by two N-terminal bromodomains and one ET (Extra Terminal) domain. BRDs associate with chromatin through their bromodomains that recognize acetylated histone lysine residues. bromodomains function as 'readers' of these epigenetic histone marks and regulate chromatin structure and gene expression by linking associated proteins to the acetylated nucleosomal targets. The ET domain functions as a protein binding motif and exerts atypical serine-kinase activity. The BET family consists of at least four members in mouse and human, BRD2 (also referred to as FSRG1, RING3), BRD3 (FSRG2, ORFX), BRD4 (FSRG4, MCAP/HUNK1), and BRDT (FSRG3, BRD6). BRD proteins are related to the female Sterile Homeotic protein gene in Drosophila, a gene required maternally for proper expression of other homeotic genes, such as Ubx, which is involved in pattern formation. BRD4 has been identified recently as a therapeutic target in many cancers, including acute myeloid leukemia, multiple myeloma, Burkitt's lymphoma, NUT midline carcinoma, colon cancer, and breast cancer. BRD4 regulates the transcription of oncogenes, HIV, and human papilloma virus (HPV). It has been shown to bind and phosphorylate RNA pol II, which implicates its involvement in the regulation of eukaryotic transcription. It shows binding specificity for acetylated H3K9, H3K9/ K14, H4K5, H4K8, H4K12, H4K5/K8, H4K5/K12, H4K8/K12, H4K12/K16, H4K12/K16/K20 and H4K5/K8/K12/K16, as well as acetylated RelA-K310.

Immunogen: This antibody was raised against a recombinant protein comprising amino acids 149-284 of human BRD4.

Buffer: Purified IgG in 140 mM Hepes, pH 7.5, 70 mM NaCl, 32 mM NaOAc, 0.035% sodium azide, 30% glycerol. Sodium azide is highly toxic.

Application Notes:

Applications Validated by Active Motif:

ChIP-Seq: 4 µg per ChIP

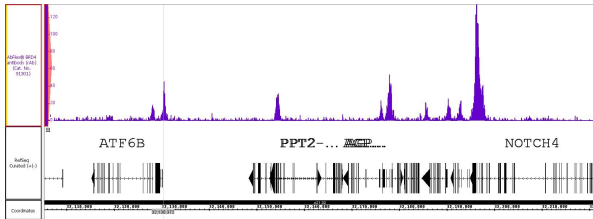
CUT&RUN: 1 µl per 50 µl reaction

Storage and Guarantee: Some products may be shipped at room temperature. This will not affect their stability or performance. Avoid repeated freeze/thaw cycles by aliquoting items into single-use fractions for storage at -20°C for up to 2 years. Keep all reagents on ice when not in storage. This product is guaranteed for 12 months from date of receipt.

This product is for research use only and is not for use in diagnostic procedures.

AbFlex® BRD4 recombinant antibody (rAb) tested by ChIP-Seq

Chromatin immunoprecipitation (ChIP) was performed using the ChIP-IT® High Sensitivity Kit (Cat. No. 53040) with 30 µg of HCT-15 cells (human colorectal adenocarcinoma line chromatin) and 4 µg of BRD4 antibody. ChIP DNA was sequenced on the Illumina NextSeq and 14.5 million sequence tags were mapped to identify BRD4 binding sites on chromosome 19.



AbFlex® BRD4 antibody (rAb) tested by CUT&RUN

CUT&RUN was performed using 500,000 K562 nuclei and sequenced using 38 base-pair, paired-end reads on the Illumina NovaSeq. Data was collected from 32 million reads, and BRD4 data is shown for Chromosome 5.

