

H3K36me2 antibody

Cat. No. C15200182

Lot:	001–12
Size:	10 µg / 50 µg
Type:	Monoclonal, ChIP-grade, CUT&Tag-grade
Isotype:	IgG1
Source:	Mouse
Concentration:	1 µg/µl

Specificity:	Human: positive Other species: not tested
Purity:	Protein A purified monoclonal antibody
Storage buffer:	PBS containing 0.05% azide.

Storage: Store at -20°C; for long storage, store at -80°C. Avoid multiple freeze-thaw cycles.

Precautions: This product is for research use only. Not for use in diagnostic or therapeutic procedures.

Description: Monoclonal antibody raised in mouse against histone H3 dimethylated at lysine 36 (H3K36me2), using a KLH-conjugated synthetic peptide.

Applications

Applications	Suggested dilution	References
ChIP*	1–2 µg per ChIP	Fig 1
CUT&Tag	1 µg	Fig 2
ELISA	1:3,000	Fig 3
Western blotting	1:1,000 – 1:2,000	

*Please note that the optimal antibody amount per IP should be determined by the end-user. We recommend testing 1-5 µg per IP.

Target description

Histones are the main constituents of the protein part of chromosomes of eukaryotic cells. They are rich in the amino acids arginine and lysine and have been greatly conserved during evolution. Histones pack the DNA into tight masses of chromatin. Two core histones of each class (H2A, H2B, H3, and H4) assemble and are wrapped by 146 base pairs of DNA to form one octameric nucleosome. Histone tails undergo numerous post-translational modifications, which either directly or indirectly alter chromatin structure to facilitate transcriptional activation or repression or other nuclear processes. In addition to the genetic code, combinations of the different histone modifications reveal the so-called “histone code”. Histone methylation and demethylation are dynamically regulated by histone methyl transferases and histone demethylases, respectively.

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Results

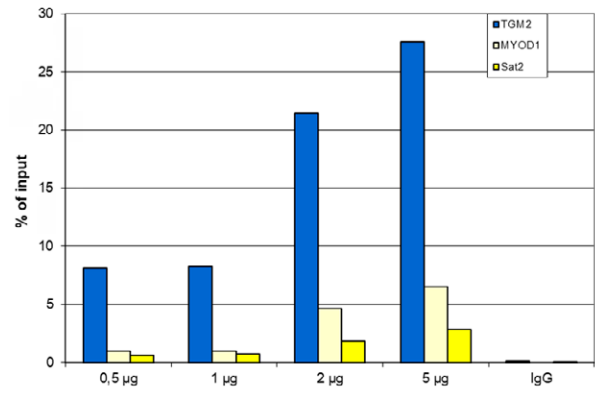


Figure 1: ChIP results obtained with the monoclonal antibody directed against H3K36me2

ChIP assays were performed using human HeLa cells, a monoclonal antibody against H3K36me2 (cat. No. C15200182), and optimized PCR primer pairs for qPCR. ChIP was performed on sheared chromatin from 1 million cells. A titration consisting of 0.5, 1, 2 and 5 µg of antibody per ChIP experiment was analyzed. IgG (1 µg/IP) was used as a negative IP control. Quantitative PCR was performed with primers specific for a genomic region upstream of the TGM2 gene, used as a positive control, and for the inactive MYOD1 gene and the Sat2 satellite repeat, used as negative controls. Figure 1 shows the recovery, expressed as a % of input (the relative amount of immunoprecipitated DNA compared to input DNA after qPCR analysis).

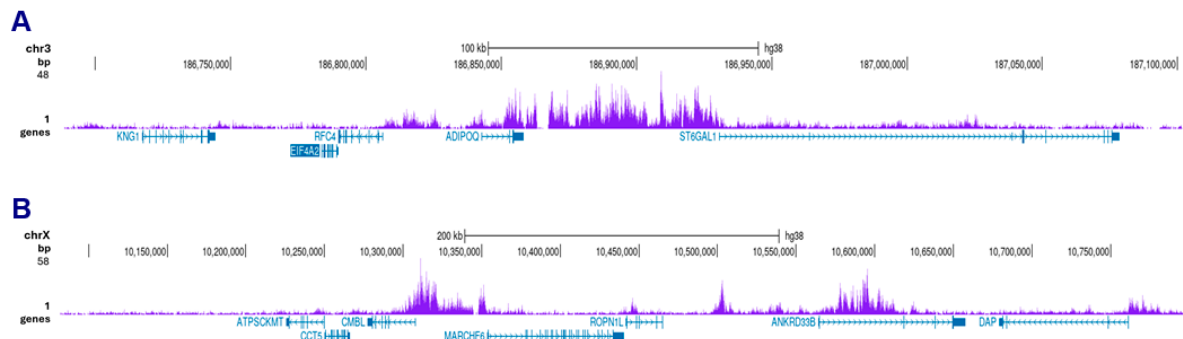


Figure 2: Cut&Tag results obtained with the monoclonal antibody directed against H3K36me2

Cut&Tag was performed on 50,000 K562 cells using 1 µg of the monoclonal antibody against H3K36me2 (cat. No. C15200182), the pA-Tn5 transposase (C01070001) and the iDeal Cut&Tag kit (cat. No. C01070021). The libraries were subsequently analyzed on an Illumina NovaSeq sequencer (2x50 paired-end reads) according to the manufacturer's instructions. The tags were aligned to the human genome (hg38) using the BWA algorithm. Figure 2 shows the peak distribution in two genomic regions on chromosome 3 and X (figure 2A and 2B, respectively).

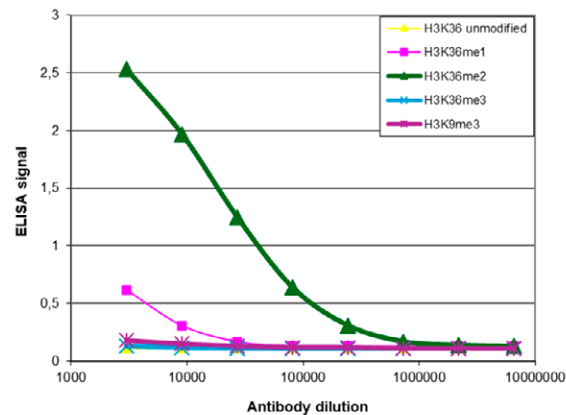


Figure 3: Cross reactivity of the monoclonal antibody directed against H3K36me2

To test the specificity, an ELISA was performed using a serial dilution of the monoclonal antibody against H3K36me2 (cat. No. C15200182). The wells were coated with peptides containing the unmodified H3K36 region as well as the mono-, di-, and trimethylated H3K36 and the trimethylated H3K9. Figure 3 shows a high specificity of the antibody for the peptide containing the modification of interest.