

H3K9me3 Antibody - ChIP-seq Grade

Cat. No. C15210014

Type: Monoclonal ChIP-seq grade	Specificity: Human, wide range expected.
Size: 100 µg	Isotype: NA
Concentration: 1 µg/µl	Host: Rabbit
Lot No.: 001	Purity: Affinity purified polyclonal antibody.
Storage buffer: PBS containing 50% glycerol, 1% BSA and 0.09% azide.	Storage conditions: 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.	

Last Data Sheet Update: August 11, 2020

Description

Monoclonal antibody raised in rabbit against the region of histone H3 containing the trimethylated lysine 9 (H3K9me3), using a KLH-conjugated synthetic peptide.

Applications

Applications	Suggested dilution	References
ChIP/ChIP-seq *	1 µg per IP	Fig 1, 2
Western Blotting	1:1,000	Fig 3

*Please note that the optimal antibody amount per IP should be determined by the end-user. We recommend testing 0.5 - 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 is dynamically regulated by respectively histone methyl transferases and histone demethylases. Trimethylation of histone H3K9 is associated with inactive genomic regions and with zinc finger repeats.

Validation data

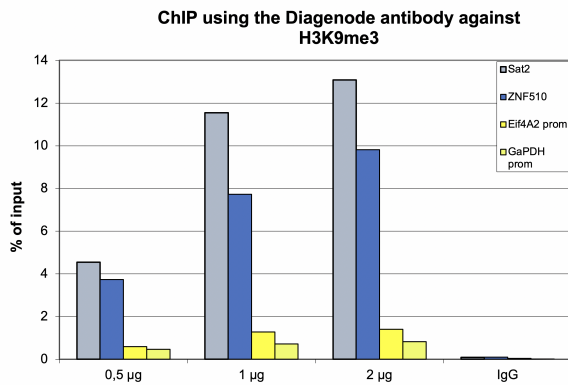


Figure 1. ChIP results obtained with the Diagenode monoclonal antibody directed against H3K9me3

ChIP was performed with the Diagenode antibody against H3K9me3 (cat. No. C15210014) on sheared chromatin from 500,000 HeLaS3 cells using the “iDeal ChIP-seq” kit (cat. No. C01010051). A titration of the antibody consisting of 0.5, 1, and 2 µg per ChIP experiment was analysed. IgG (1 µg/IP) was used as negative IP control. Quantitative PCR was performed with primers for the ZNF510 gene and the Sat2 satellite repeat, used as positive controls, and for the promoters of the GAPDH and EIF4A2 genes, used as negative controls. The graph shows the recovery, expressed as a % of input (the relative amount of immunoprecipitated DNA compared to input DNA after qPCR analysis).

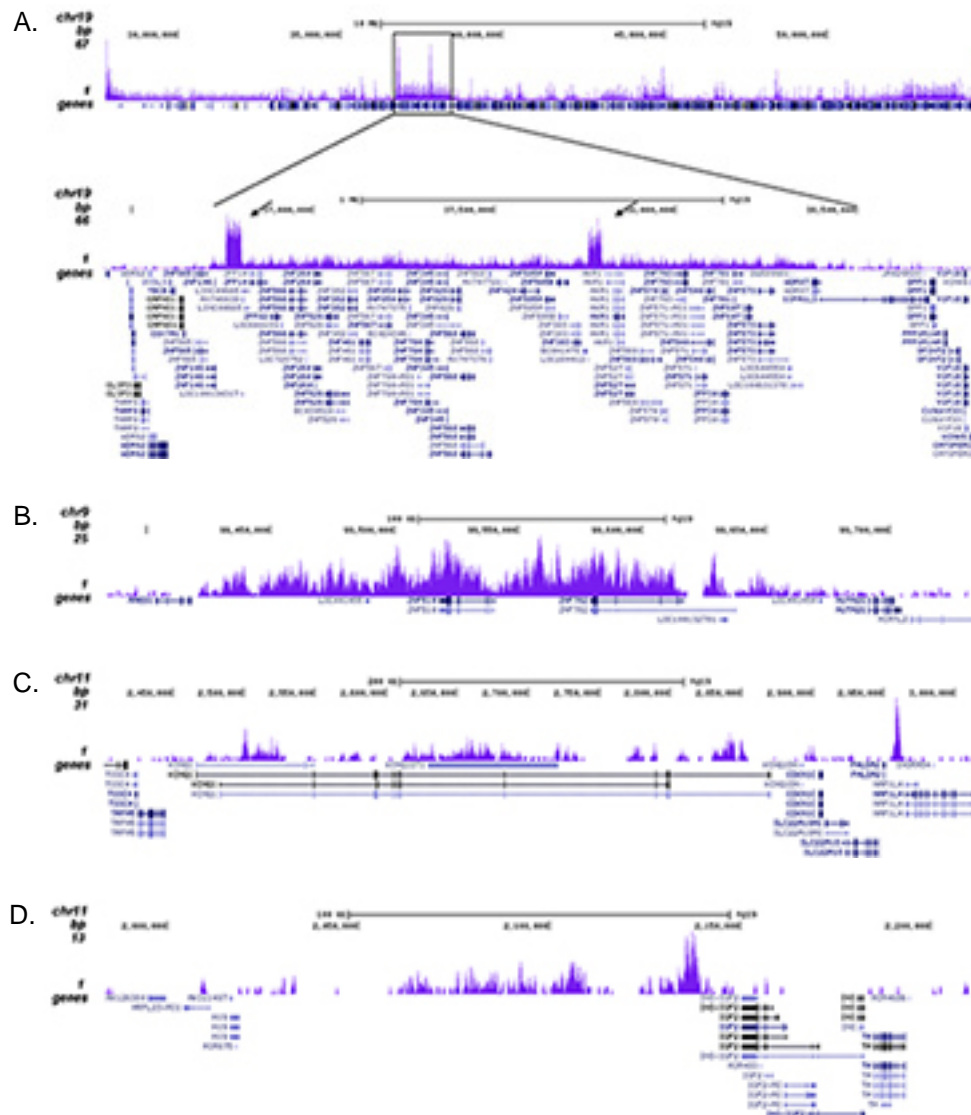


Figure 2. ChIP-seq results obtained with the Diagenode antibody directed against H3K9me3

ChIP was performed on sheared chromatin from 500,000 HeLaS3 cells using 1 µg of the Diagenode antibody against H3K9me3 (cat. No. C15210014) as described above. The IP'd DNA was subsequently analysed on an Illumina NovaSeq. Library preparation, cluster generation and sequencing were performed according to the manufacturer's instructions. The 51 bp tags were aligned to the human genome using the BWA algorithm. Figure 2A shows the signal distribution along the long arm of chromosome 19 and a zoomin to an enriched region containing several ZNF repeat genes. The arrows indicate two satellite repeat regions which exhibit a stronger signal. Figures 2B, C and D show the enrichment genomic regions surrounding the ZNF510 positive control target and at the KCNQ1 and H19 imprinted genes, respectively.

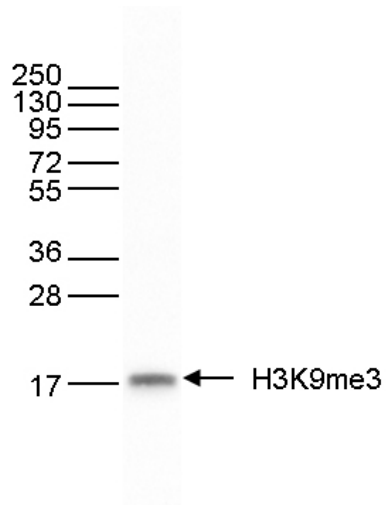


Figure 3. Western blot analysis using the Diagenode monoclonal antibody directed against H3K9me3

Western blot was performed on whole cell extracts (40 μ g) from HeLa cells using the Diagenode antibody against H3K9me3 (cat. No. C15210014). The antibody was diluted 1:1,000 in TBS-Tween containing 5% skimmed milk. The position of the protein of interest is shown on the right, the marker (in kDa) is shown on the left.