TECHNICAL DATASHEET

H2A.Z Antibody - ChIP-seq Grade

Cat. No. C15210021

Type: Monoclonal **ChIP-grade** **ChIP-seq-grade**	Specificity: Human: positive. Other species: not tested
Size: 100 µg	Isotype: NA
Concentration: 1 µg/µl	Host: Rabbit
Lot No.: 001D	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 18, 2020

Description

Monoclonal antibody raised in rabbit against histone variant H2A.Z, using a KLH-conjugated synthetic peptide containing a sequence from the C-terminal part of the protein.

Applications

Applications	Suggested dilution	References
ChIP/ChIP-seq *	0.5 µg per IP	Fig 1, 2
Western Blotting	1:200	Fig 3
Immunofluorescence	1:100	Fig 4

*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. Histone variant H2A.Z is associated with the active genes.

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Validation data

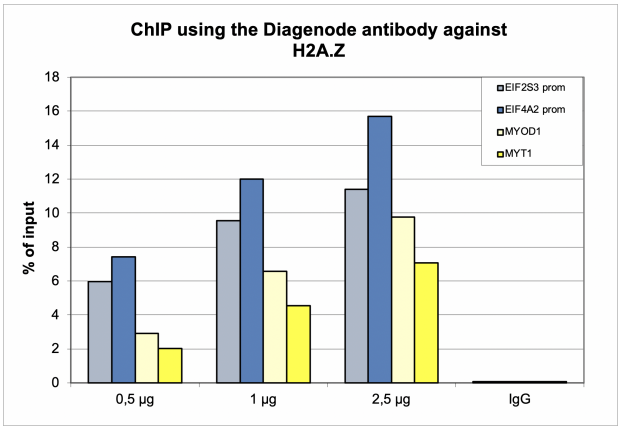

Figure 1. ChIP results obtained with the Diagenode monoclonal antibody directed against H2A.Z

ChIP was performed with the Diagenode antibody against H2A.Z (cat. No. C15210021) 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 2.5 µg per ChIP experiment was analysed. IgG (1 µg/IP) was used as negative IP control. Quantitative PCR was performed with primers for the EIF2S3 and EIF4A2 promoters, used as positive controls, and for the MYOD1 and MYT1 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).

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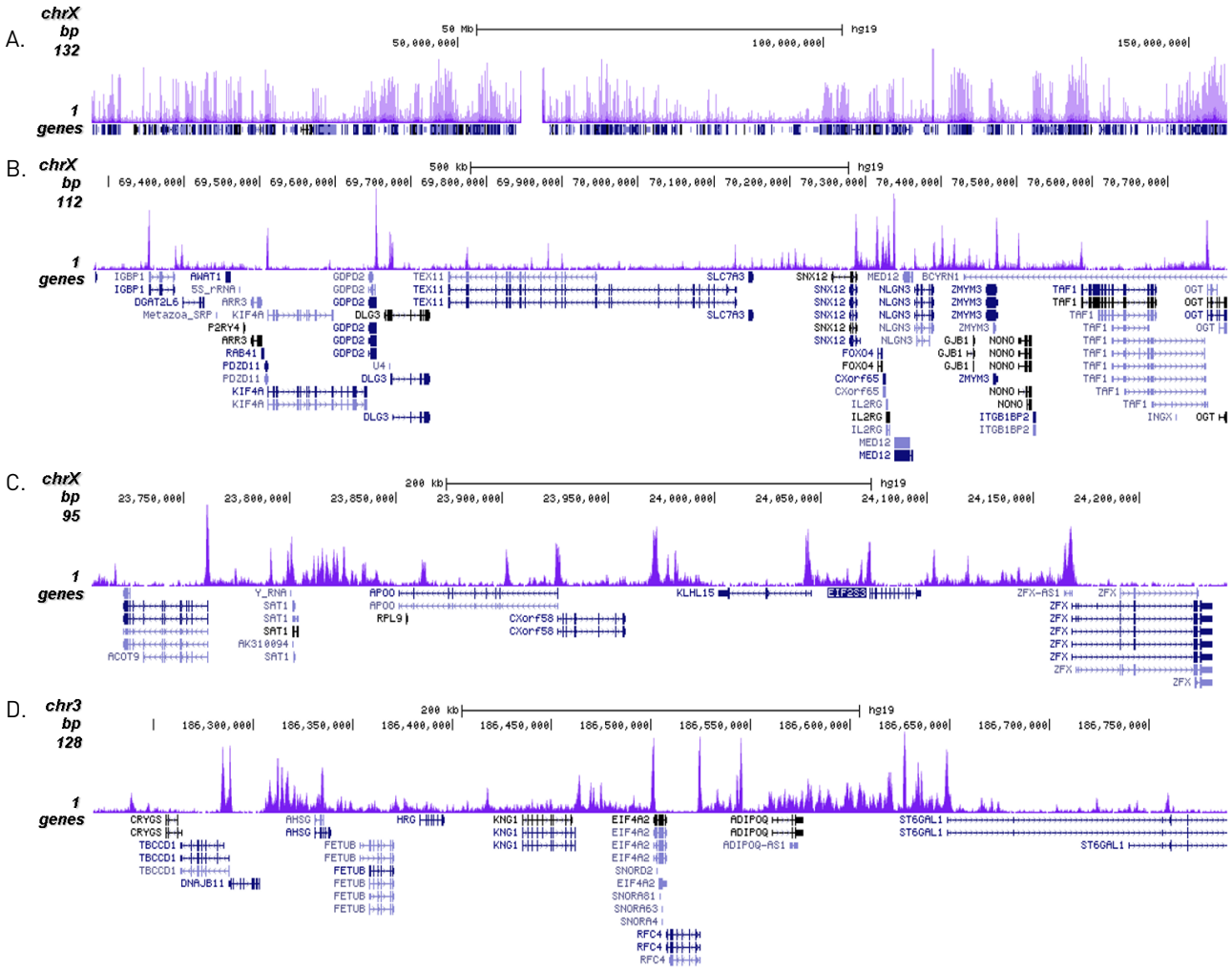

Figure 2. ChIP-seq results obtained with the Diagenode monoclonal antibody directed against H2A.Z
ChIP was performed on sheared chromatin from 500,000 HeLaS3 cells using 0.5 µg of the Diagenode antibody against H2A.Z (cat. No. C15210021) as described above. The IP'd DNA was subsequently analysed on an 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 2 shows the H2A.Z signal along the complete sequence and 1.5 Mb region of the human X-chromosome (figure 2A and B), and in two genomic regions surrounding the EIF2S3 and EIF4A2 positive controls (figure 2C and D).

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Figure 3. Western blot analysis using the Diagenode monoclonal antibody directed against H2A.Z
Western blot was performed on whole cell extracts (40 µg) from HeLa cells using the Diagenode antibody against H2A.Z (cat. No. C15210021). The antibody was diluted 1:200 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.

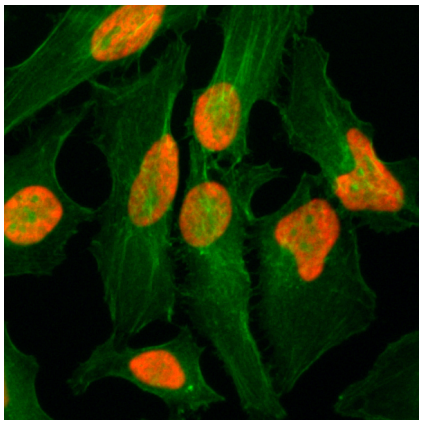

Figure 4. Immunofluorescence using the Diagenode monoclonal antibody directed against H2A.Z
HeLa cells were stained with the Diagenode antibody against H2A.Z (cat. No. C15210021, red) diluted 1:100. Actin was stained with fluorescein phalladoin (green).

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