

H3T3p monoclonal antibody

Cat. No. C15210001

Type: Monoclonal ChIP-grade

Source: Rabbit

Lot #: 001

Size: 100 µg/100 µl

Concentration: 1 µg/µl

Specificity: Human: positive

Other species: not tested

Purity: Protein A purified monoclonal antibody in PBS containing 50% glycerol, 1% BSA and 0.09% 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 rabbit against histone H3, phosphorylated at threonine 3 (H3T3p), using a KLH-conjugated synthetic peptide.

Applications

	Suggested dilution	Results
ChIP*	0.5 - 1 µg/ChIP	Fig 1
Western blotting	1:10,000	Fig 2
Immunofluorescence	1:1,000	Fig 3

* Please note that the optimal antibody amount per ChIP should be determined by the end-user. We recommend testing 0.5-5 µg per ChIP.

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.

Results

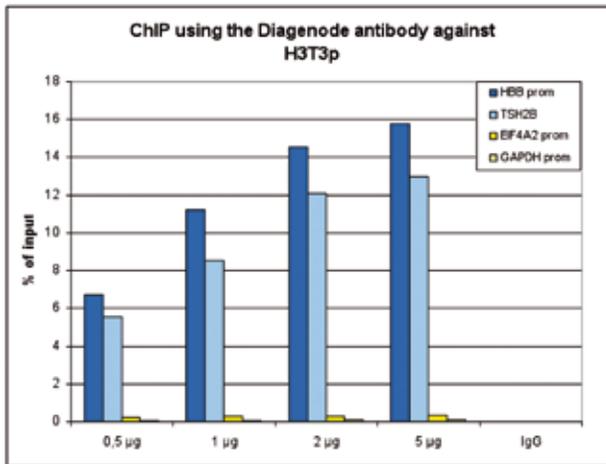


Figure 1. ChIP results obtained with the Diagenode monoclonal antibody directed against H3T3p

ChIP assays were performed using HeLa cells, the Diagenode antibody against H3T3p (cat. No. C15210001) and optimized PCR primer sets for qPCR. ChIP was performed with the "iDeal ChIP-seq" kit (cat. No. C01010051), using 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 optimized primers for the TSH2B and HBB genes, used as positive controls, and for the promoters of the EIF4A2 and GAPDH genes, 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).

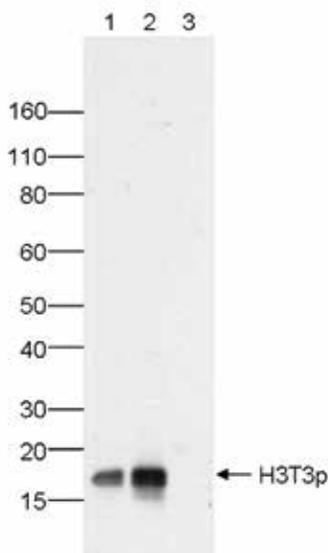


Figure 2. Western blot analysis using the Diagenode monoclonal antibody directed against H3T3p

Histone extracts from HeLa cells (lane 1) or HeLa cells treated with nocodazole (lane 2), as well as recombinant H3 (lane 3) were analysed by Western blot using the Diagenode monoclonal antibody against H3T3p (Cat. No. C15210001) diluted 1:10,000 in TBS-Tween containing 5% skimmed milk. The position of the protein of interest is indicated on the right; the marker (in kDa) is shown on the left.

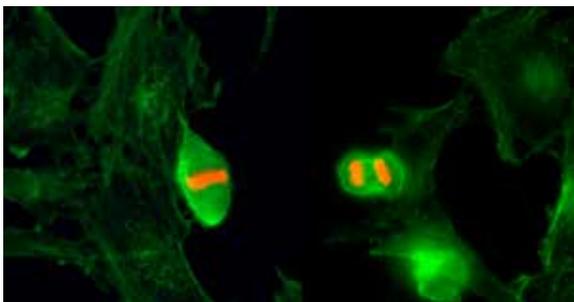


Figure 3. Immunofluorescence using the Diagenode monoclonal antibody directed against H3T3p

HeLa cells were stained with the Diagenode antibody against H3T3p (Cat. No. C15210001, red) diluted 1:1,000. Actin was stained with fluorescein phalloidin (green).

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