

H3K9ac monoclonal antibody

Cat. No. C15200185 (MAb-185-050)

Type: Monoclonal ChIP-grade

IgG isotype: IgG2b

Source: Mouse

Lot #: 001-11

Size: 50 µg/ 50 µl

Concentration: 1 µg/µl

Specificity: Human: positive
Other species: not tested

Purity: Protein A purified monoclonal antibody in 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 acetylated at lysine 9 (H3K9ac), using a KLH-conjugated synthetic peptide.

Applications

	Suggested dilution	Results
ChIP*	0.5 - 1 µg/ChIP	Fig 1
ELISA	1:3,000	Fig 2
Western blotting	1:1,000 - 1:2,000	
IF	1:500	Fig 3

*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 is dynamically regulated by respectively histone methyl transferases and histone demethylases.

Results

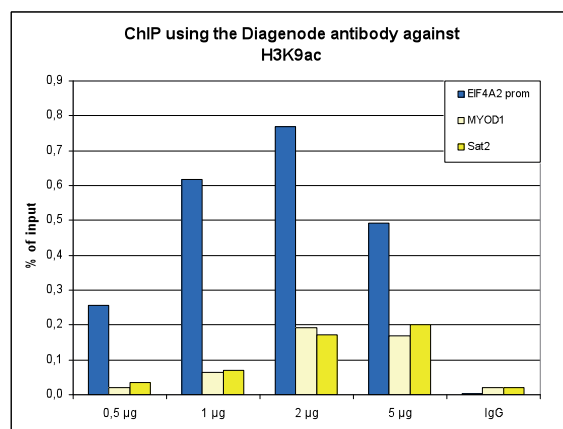


Figure 1. ChIP results obtained with the Diagenode monoclonal antibody directed against H3K9ac

ChIP assays were performed using human HeLa cells, the Diagenode monoclonal antibody against H3K9ac (Cat. No. C15200185) and optimized PCR primer pairs for qPCR. ChIP was performed with the "Auto Histone ChIP-seq" kit (Cat. No. C01010020), 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 primers specific for the promoter of the EIF4A2 gene, used as 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].

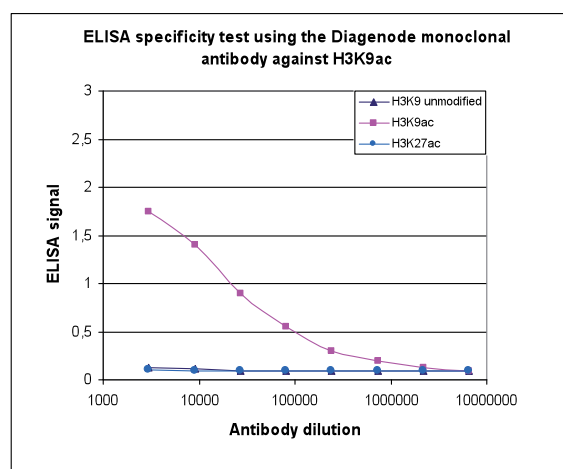


Figure 2. Cross reactivity of the Diagenode monoclonal antibody directed against H3K9ac

To test the specificity an ELISA was performed using a serial dilution of the Diagenode monoclonal antibody against H3K9ac (Cat. No. C15200185). The wells were coated with peptides containing the unmodified H3K9 region as well as the acetylated H3K9 and the acetylated H3K27. Figure 2 shows a high specificity of the antibody for the peptide containing the modification of interest.

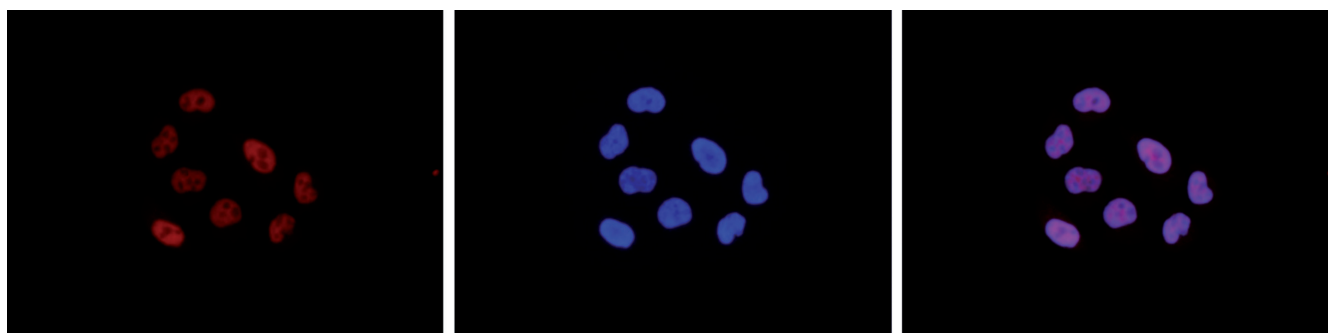


Figure 3. Immunofluorescence using the Diagenode monoclonal antibody directed against H3K9ac

HeLa cells were stained with the Diagenode antibody against H3K9ac (Cat. No. C15200185) and with DAPI. Cells were fixed with 4% formaldehyde for 10' and blocked with PBS/TX-100 containing 5% normal goat serum and 1% BSA. The cells were immunofluorescently labelled with the H3K9ac antibody (left) diluted 1:500 in blocking solution followed by an anti-mouse antibody conjugated to Alexa594. The middle panel shows staining of the nuclei with DAPI. A merge of the two stainings is shown on the right.

Diagenode sa. BELGIUM | EUROPE

LIEGE SCIENCE PARK
Rue Bois Saint-Jean, 3
4102 Seraing (Ougrée) - Belgium
Tel: +32 4 364 20 50
Fax: +32 4 364 20 51
orders@diagenode.com
info@diagenode.com

Diagenode Inc. USA | NORTH AMERICA

400 Morris Avenue, Suite 101
Denville, NJ 07834 - USA
Tel: +1 862 209-4680
Fax: +1 862 209-4681
orders.na@diagenode.com
info.na@diagenode.com

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