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H3K4ac polyclonal antibody - Classic

Purified rabbit polyclonal Antibody

Catalog # ADN10292

Specification

H3K4ac polyclonal antibody - Classic - Product Information

Application	**CHIP, E, DB, WB, IF**
Primary Accession	Q93081
Reactivity	**Human**
Host	**Rabbit**
Clonality	**Polyclonal**

H3K4ac polyclonal antibody - Classic - Additional Information

Target/Specificity
H3K4ac

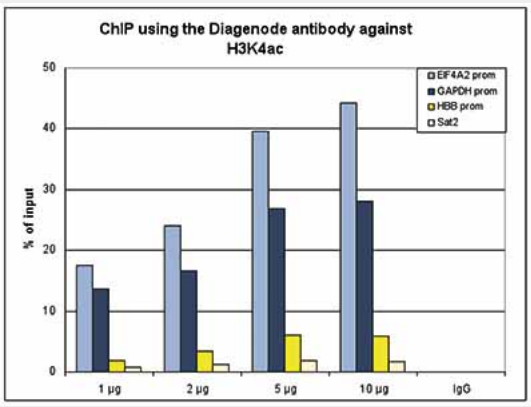
Precautions
H3K4ac polyclonal antibody - Classic is for research use only and not for use in diagnostic or therapeutic procedures.

H3K4ac polyclonal antibody - Classic - Protein Information

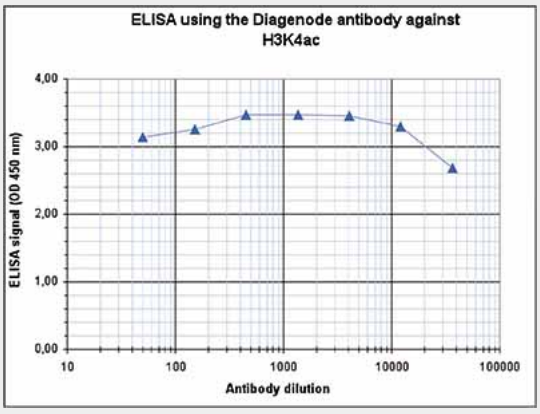
H3K4ac polyclonal antibody - Classic - Protocols

Provided below are standard protocols that you may find useful for product applications.

- Western Blot
- Blocking Peptides
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- Cell Culture



ChIP assays were performed using human HeLa cells, the Diagenode antibody against H3K4ac (Cat. No. ADN10292) and optimized PCR primer sets for qPCR. ChIP was performed with the “iDeal ChIP-seq” kit (Cat. No. C01010051), using sheared chromatin from 4 million cells. A titration of the antibody consisting of 1, 2, 5 and 10 µg per ChIP experiment was analysed. IgG (1 µg/IP) was used as negative IP control. QPCR was performed with primers for the GAPDH and EIF4A2 promoters, used as positive controls, and for the HBB promoter 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).



To determine the titer of the antibody, an

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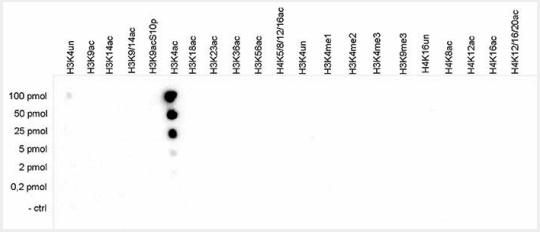
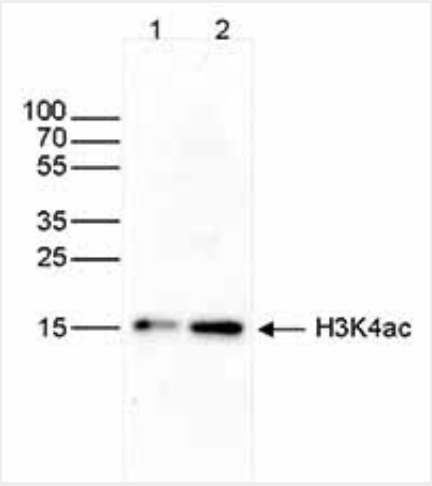
ELISA was performed using a serial dilution of the Diagenode antibody directed against H3K4ac (Cat. No. ADN10292) in antigen coated wells. The antigen used was a peptide containing the histone modification of interest. By plotting the absorbance against the antibody dilution (figure 3), the titer of the purified antibody was estimated to be 1:197,500.



A Dot Blot analysis was performed to test the cross reactivity of the Diagenode antibody against H3K4ac (Cat. No. ADN10292) with peptides containing other histone H3 and H4 modifications and the unmodified H3K4 sequence. One hundred to 0.2 pmol of the respective peptides were spotted on a membrane. The antibody was used at a dilution of 1:5,000. Figure 4 shows a high specificity of the antibody for the modification of interest.



Whole cell (25 µg, lane 1) and histone extracts (15 µg, lane 2) from HeLa cells were analysed by Western blot using the Diagenode antibody against H3K4ac (Cat. No. ADN10292), diluted 1:1,000 in TBSTween containing 5% BSA. The marker (in kDa) is shown on the left, the position of the protein of interest is indicated on the right.

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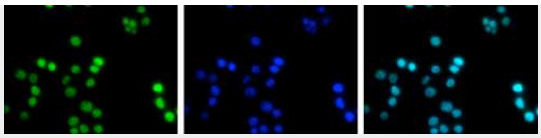

HeLa cells were stained with the Diagenode antibody against H3K4ac (Cat. No. ADN10292) and with DAPI. Cells were fixed with 4% formaldehyde for 10’ and blocked with PBS/TX-100 containing 5% normal goat serum and 1% BSA. The cells were immunofluorescently labeled with the H3K4ac antibody (left) diluted 1:200 in blocking solution followed by an anti-rabbit antibody conjugated to Alexa488. The middle panel shows staining of the nuclei with DAPI. A merge of the two stainings is shown on the right.