

Anti-H4K20me3 HIST1H4A Antibody

Catalog Number: CI1050

About HIST1H4A

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.

Overview

Product Name	Anti-H4K20me3 HIST1H4A Antibody
Reactive Species	Human, Mouse
Description	Boster Bio Anti-H4K20me3 HIST1H4A Antibody (Catalog# CI1050). Tested in ChIP, ChIP-seq, ELISA, Dot blot, WB, IF applications. This antibody reacts with Human, Mouse.
Application	ChIP, ChIP-seq, Dot blot, ELISA, IF, WB
Clonality	Polyclonal
Formulation	Affinity purified polyclonal antibody in PBS containing 0.05% azide and 0.05% ProClin 300.
Storage Instructions	Store at -20°C. For long-term storage, store at -80°C. Avoid multiple freeze-thaw cycles.
Host	Rabbit
Uniprot ID	P62805

Technical Details

Immunogen	This antibody is raised in rabbit against the region of histone H4 containing the trimethylated lysine 20 (H4K20me3), using a KLH-conjugated synthetic peptide.
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western blot. Boster recommends high sensitivity ChIP-seq Kit (CK1001 & CK1002) for Chromatin Immunoprecipitation.
Form	Liquid
Concentration	0.5-1mg/ml, actual concentration vary by lot. Use suggested dilution ratio to decide dilution procedure.
Purification	Affinity purified

Suggested Dilutions

Dilute the sample so that the expected range of concentrations fall within the detection range of this kit.

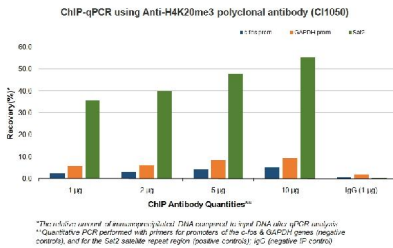
If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples.

Some PubMed article(s) citing the expression level of this target are as follows:

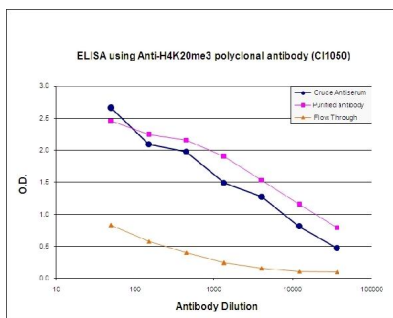
Boster Bio's internal QC testing used:

User needs to optimize the dilution ratio for this antibody.

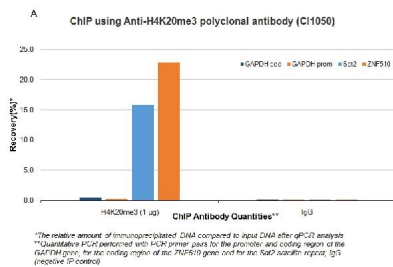
Anti-H4K20me3 HIST1H4A Antibody (CI1050) Images



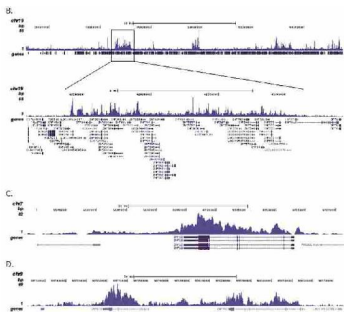
ChIP assays were performed using human HeLa cells, Anti-H4K20me3 polyclonal antibody (Catalog # CI1050) and optimized PCR primer sets for qPCR. A titration of the antibody consisting of 1, 2, 5, and 10 ug per ChIP experiment was analysed. IgG (1 ug/IP) was used as negative IP control. QPCR was performed with primers for promoters of the active genes c-fos and GAPDH, used as negative controls, and for the Sat2 satellite repeat region used as a positive control.



To determine the titer of the antibody, an ELISA was performed using a serial dilution of Anti-H4K20me3 polyclonal antibody (Catalog # CI1050), crude serum and flow through in antigen coated wells. The antigen used was a peptide containing the histone modification of interest. By plotting the absorbance against the antibody dilution, the titer of the antibody was estimated to be 1:7,400.



ChIP was performed with 1 ug of Anti-H4K20me3 polyclonal antibody (Catalog # CI1050) on sheared chromatin from 1 million HeLaS3 cells. The IP DNA was analysed by QPCR with optimized PCR primer pairs for the promoter and coding region of the active GAPDH gene, for the coding region of the ZNF510 gene and for the Sat2 satellite repeat (figure 2A). The IP DNA was subsequently analysed on an Illumina Genome Analyzer. Library preparation, cluster generation and sequencing were performed according to the manufacturer instructions. The 36 bp tags were aligned to the human genome using the ELAND algorithm. Figure 2B shows the signal distribution along the long arm of chromosome 19 and a zoomin to an enriched region containing several ZNF repeat genes. Figure 2C and D show the enrichment at ZNF12 and ZNF510 on chromosome 7 and 9, respectively. These results clearly show an enrichment of H4K20me3 at ZNF repeat genes.

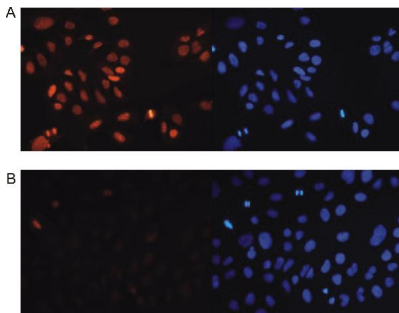


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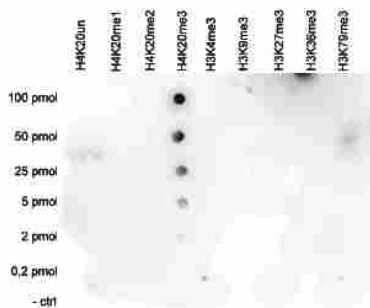
containing several ZNF repeat genes. Figure 2C and D show the enrichment at ZNF12 and ZNF510 on chromosome 7 and 9, respectively. These results clearly show an enrichment of H4K20me3 at ZNF repeat genes.



Western blot analysis of H4K20me3 expression in histone extracts from HeLa cells (15 ug). H4K20me3 was detected using Anti-H4K20me3 polyclonal antibody (Catalog # CI1050) at 1/1000 dilution.



Immunofluorescence images stained on Human osteosarcoma (U2OS) cells: (Figure A left) Cells stained with anti-H4K20me3 polyclonal antibody (Catalog # CI1050) at 1/300. (Figure A right) Nuclei stained with DAPI. (Figure B) staining of the cells with the H4K20me3 antibody after incubation of the antibody with 5 ng/ul blocking peptide.



A Dot Blot analysis was performed to test the cross reactivity of Anti-H4K20me3 polyclonal antibody (Catalog # CI1050) with peptides containing other histone modifications and the unmodified H4K20. One hundred to 0.2 pmol of the respective peptides were spotted on a membrane. The antibody was used at a dilution of 1:20,000. This figure shows a high specificity of the antibody for the modification of interest.

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