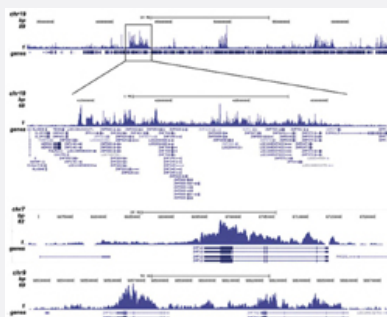
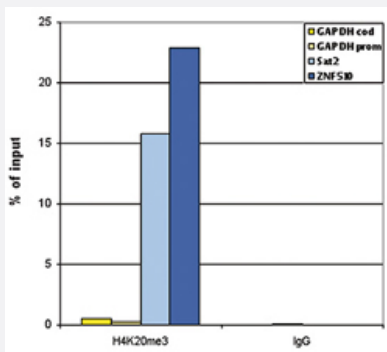
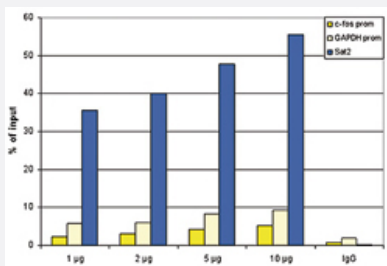


Histone H4 (K20me3) polyclonal antibody

Catalog # PAB31329

Size 50 ug

Applications



ChIP

ChIP assays were performed using human HeLa cells. 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. The figure shows the recovery, expressed as a % of input (the relative amount of immunoprecipitated DNA compared to input DNA after qPCR analysis).

ChIP

ChIP was performed on sheared chromatin from 1 million HeLaS3 cells. The IP'd 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.

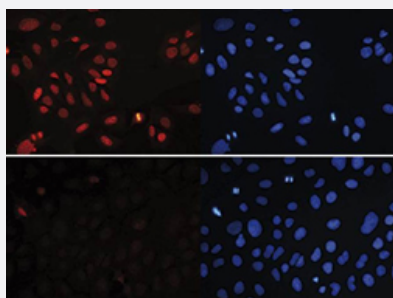
ChIP-Seq

The figure shows the signal distribution along the long arm of chromosome 19 and a zoomin to an enriched region containing several ZNF repeat genes and the enrichment at ZNF12 and ZNF510 on chromosome 7 and 9, respectively. These results clearly show an enrichment of H4K20me3 at ZNF repeat genes.



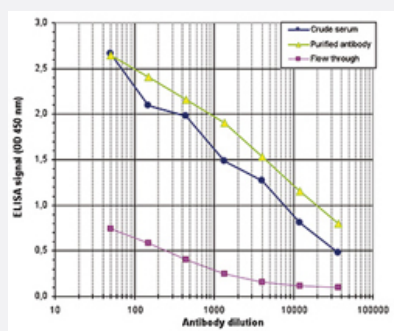
Western Blot (Cell lysate)

Western Blot (Cell lysate) analysis of 15 ug histone extracts of HeLa cells.



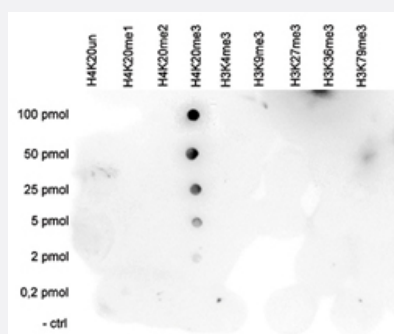
Immunofluorescence

Immunofluorescent staining of human osteosarcoma (U2OS) cell line with antibody followed by anti-rabbit antibody conjugated to Alexa568 (left) or with DAPI (right). The bottom panel shows staining of with antibody after incubation of the antibody with blocking peptide.



Enzyme-linked Immunoabsorbent Assay

ELISA is a quantitative method used to determine the titer of the antibody using a serial dilution of antibody against Histone H4 (K20me3), 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 1:7400.



Dot Blot

Cross reactivity test using the Histone H4 (K20me3) antibody. Dot Blot analysis was performed 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 1:20000. The figure shows a high specificity of the antibody for the modification of interest.

Specification

Product Description

Rabbit polyclonal antibody raised against synthetic peptide of Histone H4 (K20me3).

Immunogen

A synthetic peptide (conjugated with KLH) corresponding to Histone H4, trimethylated at lysine 20.

Host

Rabbit

Reactivity	Human, Mouse
Form	Liquid
Purification	Affinity purification
Recommend Usage	ELISA (1:100) Western Blot (1:1000) ChIP (1-2 ug/IP) Dot Blot (1:20000) Immunofluorescence (1:300) The optimal working dilution should be determined by the end user.
Storage Buffer	In PBS (0.05% sodium azide, 0.05% proclin 300).
Storage Instruction	Store at -20°C. For long term storage store at -80°C. Aliquot to avoid repeated freezing and thawing.
Note	This product contains sodium azide: a POISONOUS AND HAZARDOUS SUBSTANCE which should be handled by trained staff only.

Applications

● ChIP

ChIP assays were performed using human HeLa cells. 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. The figure shows the recovery, expressed as a % of input (the relative amount of immunoprecipitated DNA compared to input DNA after qPCR analysis).

● ChIP

ChIP was performed on sheared chromatin from 1 million HeLaS3 cells. The IP'd 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.

● ChIP-Seq

The figure shows the signal distribution along the long arm of chromosome 19 and a zoomin to an enriched region containing several ZNF repeat genes and 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 (Cell lysate)

Western Blot (Cell lysate) analysis of 15 ug histone extracts of HeLa cells.

- Immunofluorescence

Immunofluorescent staining of human osteosarcoma (U2OS) cell line with antibody followed by anti-rabbit antibody conjugated to Alexa568 (left) or with DAPI (right). The bottom panel shows staining of with antibody after incubation of the antibody with blocking peptide.

- Enzyme-linked Immunoabsorbent Assay

ELISA is a quantitative method used to determine the titer of the antibody using a serial dilution of antibody against Histone H4 (K20me3), 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 1:7400.

- Dot Blot

Cross reactivity test using the Histone H4 (K20me3) antibody.

Dot Blot analysis was performed 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 1:20000. The figure shows a high specificity of the antibody for the modification of interest.

Gene Info — HIST1H4A

Entrez GeneID [8359](#)

Protein Accession# [P62805](#)

Gene Name HIST1H4A

Gene Alias H4/a, H4FA

Gene Description histone cluster 1, H4a

Omim ID [602822](#)

Gene Ontology [Hyperlink](#)

Gene Summary Histones are basic nuclear proteins that are responsible for the nucleosome structure of the chromosomal fiber in eukaryotes. Two molecules of each of the four core histones (H2A, H2B, H3, and H4) form an octamer, around which approximately 146 bp of DNA is wrapped in repeating units, called nucleosomes. The linker histone, H1, interacts with linker DNA between nucleosomes and functions in the compaction of chromatin into higher order structures. This gene is intronless and encodes a member of the histone H4 family. Transcripts from this gene lack polyA tails but instead contain a palindromic termination element. This gene is found in the large histone gene cluster on chromosome 6. [provided by RefSeq]

Other Designations H4 histone family, member A|histone 1, H4a

Publication Reference

- [Decoupling of DNA methylation and activity of intergenic LINE-1 promoters in colorectal cancer.](#)

Vafadar-Isfahani N, Parr C, McMillan LE, Sanner J, Yeo Z, Saddington S, Peacock O, Cruickshanks HA, Meehan RR, Lund JN, Tufarelli C.

Epigenetics 2017 Jun; 12(6):465.

Application: ChIP, Human, HCT-116, MCF-7, T-47D, RKO cells

- [Heat shock represses rRNA synthesis by inactivation of TIF-IA and lncRNA-dependent changes in nucleosome positioning.](#)

Zhao Z, Dammert MA, Hoppe S, Bierhoff H, Grummt I.

Nucleic Acids Research 2016 Sep; 44(17):8144.

Application: ChIP, WB-Ce, Mouse, NIH/3T3 cells

- [DOT1L Activity Promotes Proliferation and Protects Cortical Neural Stem Cells from Activation of ATF4-DDIT3-Mediated ER Stress In Vitro.](#)

Roidl D, Hellbach N, Bovio PP, Villarreal A, Heidrich S, Nestel S, Grüning BA, Boenisch U, Vogel T.

Stem Cells 2016 Jan; 34(1):233.

Application: ChIP-Seq, WB-Ce, Mouse, Mouse cortical cells

- [Use of a mouse in vitro fertilization model to understand the developmental origins of health and disease hypothesis.](#)

Feuer SK, Liu X, Donjacour A, Lin W, Simbulan RK, Giritharan G, Plane LD, Kolahi K, Ameri K, Maltepe E, Rinaudo PF.

Endocrinology 2014 May; 155(5):1956.

Application: ChIP, Mouse, Mouse blastocysts

- [Expression of a large LINE-1-driven antisense RNA is linked to epigenetic silencing of the metastasis suppressor gene TFPI-2 in cancer.](#)

Cruickshanks HA, Vafadar-Isfahani N, Dunican DS, Lee A, Sproul D, Lund JN, Meehan RR, Tufarelli C.

Nucleic Acids Research 2013 Aug; 41(14):6857.

Application: ChIP, Human, CaCo-2, HCT-116, MCF-7, T-47D cells

- [Overexpression of facioscapulohumeral muscular dystrophy region gene 1 causes primary defects in myogenic stem cells.](#)

Xynos A, Neguembor MV, Caccia R, Licastro D, Nonis A, Di Serio C, Stupka E, Gabellini D.

Journal of Cell Science 2013 May; 126(Pt 10):2236.

- [Systemic lupus erythematosus](#)