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Product datasheet for TA347143

H2BC5 Rabbit Polyclonal Antibody

Product data:

Product Type:	Primary Antibodies
Applications:	Dot, ELISA, WB
Recommended Dilution:	ChIP/ChIP-seq (0.5-1ug/ChIP); ELISA (1:1,000); Dot blotting (1:5,000); Western blotting (1:1,000)
Reactivity:	Human
Host:	Rabbit
Isotype:	IgG
Clonality:	Polyclonal
Immunogen:	The immunogen for anti-H2BK12ac antibody: the region of histone H2B containing the acetylated lysine 12 (H2BK12ac), using a KLH-conjugated synthetic peptide.
Concentration:	lot specific
Purification:	Affinity purified polyclonal antibody in PBS containing 0.05% azide and 0.05% ProClin 300.
Conjugation:	Unconjugated
Storage:	Store at -20°C as received.
Stability:	Stable for 12 months from date of receipt.
Gene Name:	histone cluster 1, H2bd
Database Link:	NP_619790 Entrez Gene 3017 Human P62807
Background:	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. Acetylation of histone H2B is associated with active genes.

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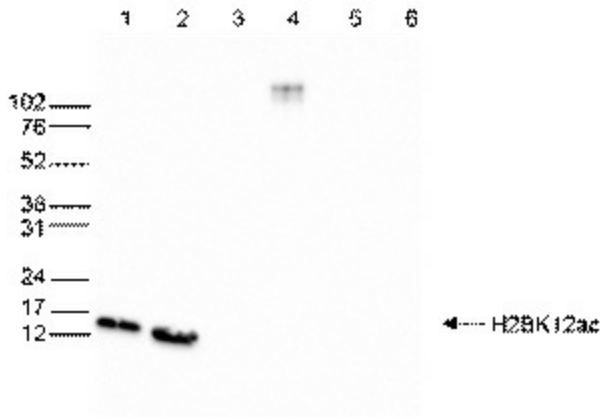
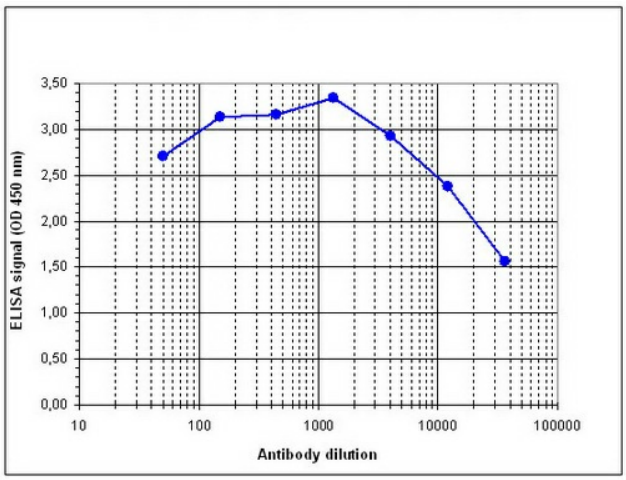
Synonyms: b; dJ221C16.6; H2B; H2B.1B; H2BFB; HIRIP2

Protein Pathways: Systemic lupus erythematosus

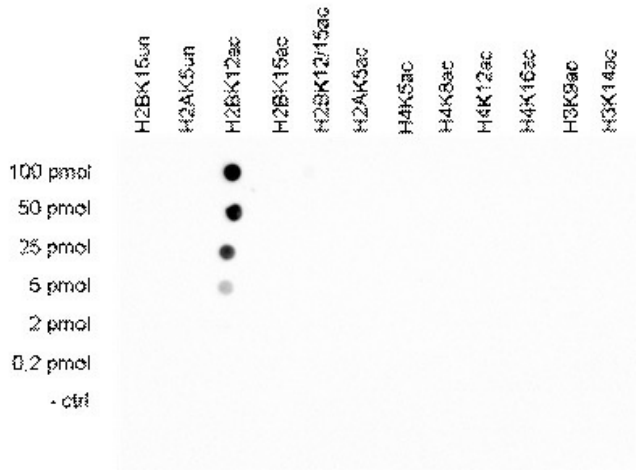
Product images:



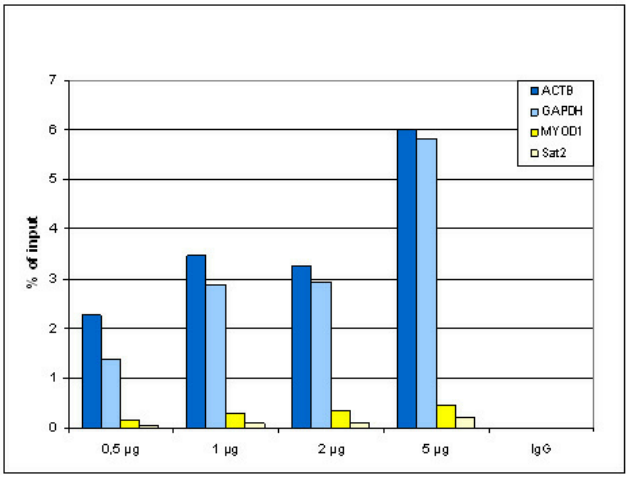
WB was performed on whole cell (25 ug, lane 1) and histone extracts (15 ug, lane 2) from HeLa cells, and on 1 ug of recombinant histone H2A, H2B, H3 and H4 (lane 3, 4, 5 and 6, respectively) using the antibody against H2BK12ac. The antibody was diluted 1:1,000 in TBS-Tween containing 5% skimmed milk. The marker (in kDa) is shown on the left.



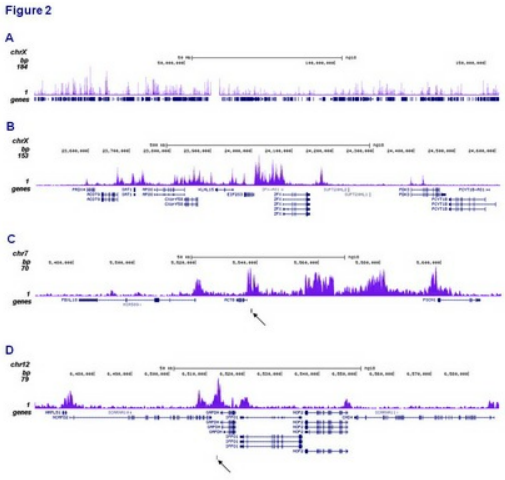
Determination of the antibody titer To determine the titer of the antibody, an ELISA was performed using a serial dilution of the antibody against H2BK12ac in antigen coated wells. The antigen used was a peptide containing the histone modification of interest. By plotting the absorbance against the antibody dilution (Figure 3), the titer of the antibody was estimated to be 1:38, 200.



A Dot Blot analysis was performed with peptides containing other histone modifications and the unmodified H2B. 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.



ChIP assays using HeLa cells (sheared chromatin from 1.5 million cells). Titration of 0.5, 1, 2 and 5ug antibody per ChIP was analysed. IgG (1 ug/IP) was used as negative control. qPCR primers were for a region ~1kb upstream of GAPDH and ACTB promoters as positive controls, and for the coding region of inactive MYOD1 gene and Sat2 satellite repeat as negative controls. Image shows the recovery, expressed as a % of input (the relative amount of IP'd DNA compared to input DNA after qPCR).



ChIP was performed on sheared chromatin from 1.5 million HeLaS3 cells using 0.5 ug antibody. The 51 bp tags were aligned to the human genome using the BWA algorithm. Image shows the enrichment along the complete sequence and a 1Mb region of the X-chromosome (A and B) and in genomic regions of chromosome 7, surrounding ACTB gene, and of chromosome 12, surrounding GAPDH gene (C and D). The position of the amplicon used for ChIP-qPCR is indicated by arrow.