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Product datasheet for TA347166

H3FA (HIST1H3A) Rabbit Polyclonal Antibody

Product data:

Product Type:	Primary Antibodies
Applications:	Dot, ELISA, WB
Recommended Dilution:	ChIP/ChIP-seq (10 µl/ChIP); ELISA (Titer: 1:100 ?? 1:500); Dot blotting (1:100,000); Western blotting (1:1,000)
Reactivity:	Human
Host:	Rabbit
Isotype:	IgG
Clonality:	Polyclonal
Immunogen:	The immunogen for anti-H3K36me3 antibody: histone H3 containing the trimethylated lysine 36 (H3K36me3), using a KLH-conjugated synthetic peptide.
Concentration:	lot specific
Purification:	Whole antiserum from rabbit containing 0.05% azide.
Conjugation:	Unconjugated
Storage:	Store at -20°C as received.
Stability:	Stable for 12 months from date of receipt.
Gene Name:	histone cluster 1, H3a
Database Link:	NP_003520 Entrez Gene 8350 Human P68431

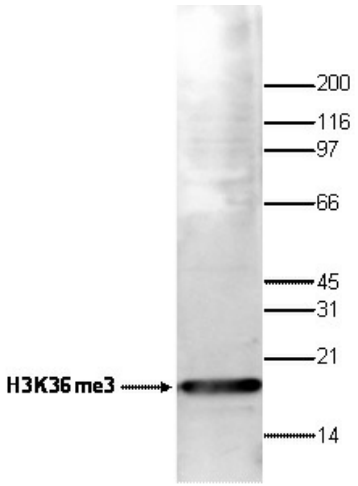
View online »

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. Trimethylation of histone H3K36 is preferentially present at active genes.

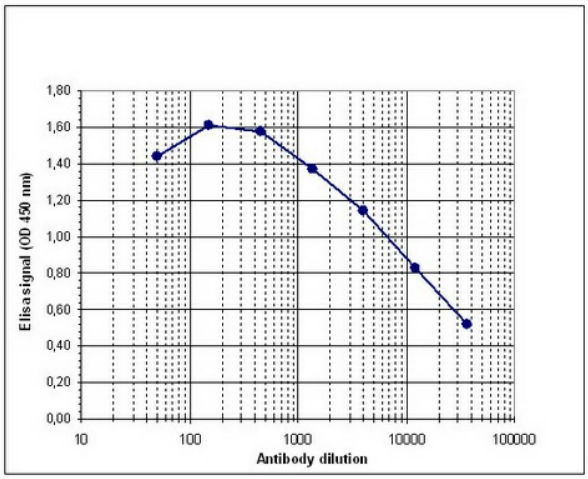
Synonyms: A; H3; H3FA

Protein Pathways: Systemic lupus erythematosus

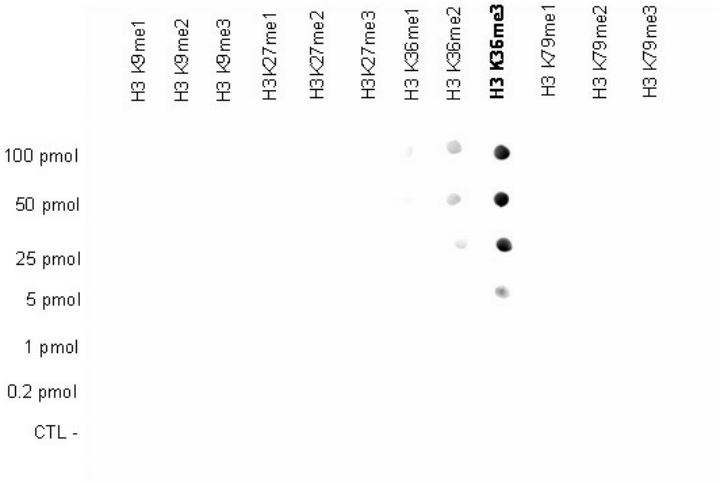
Product images:



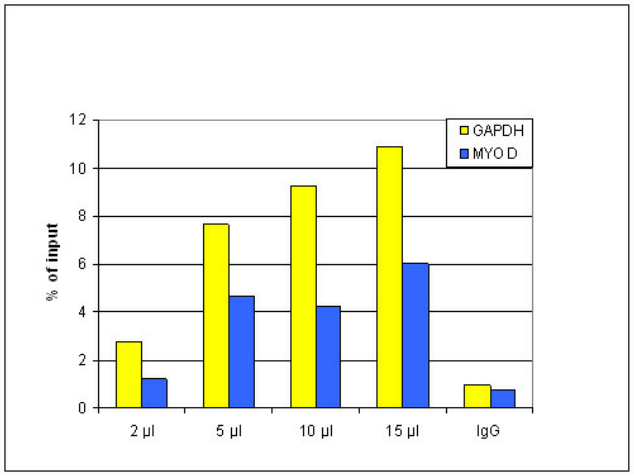
WB using the antibody against H3K36me3 diluted 1:1,000 in TBS-Tween containing 5% skimmed milk. The position of the protein of interest is indicated on the left; the marker (in kDa) is shown on the right.



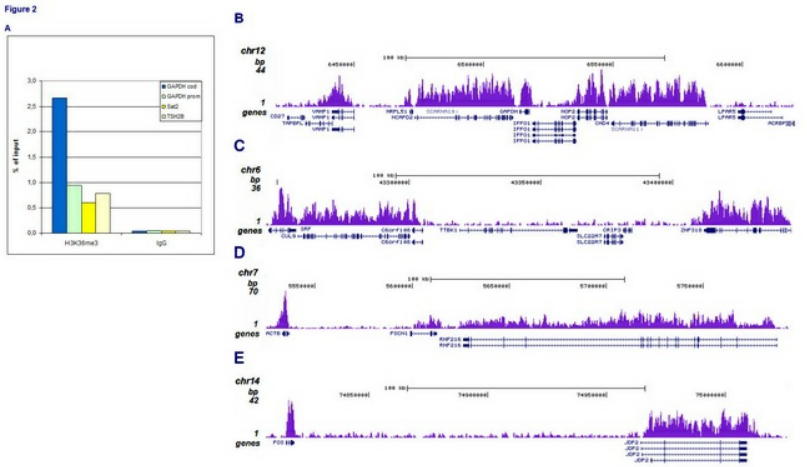
Determination of the titer To determine the titer, an ELISA was performed using a serial dilution of the antibody against H3K36me3. The antigen used was a peptide containing the histone modification of interest. By plotting the absorbance against the antibody dilution (Figure 3), the titer of the antibody was estimated to be 1:12, 700.



Dot Blot analysis was performed to test the cross reactivity of the ab with peptides containing other modifications of histone H3. Other histone modifications include mono- and dimethylation of the same lysine and mono-, di- and trimethylation of lysine 9, 27 and 79. One hundred to 0.2 pmol of peptide containing the respective histone modification were spotted on a membrane. The antibody was used at 1:100,000. Image shows a high specificity of the antibody for the modification of interest.



ChIP assays using human U2OS cells. IgG (5 ug/IP) was negative control. qPCR was performed using primer sets for the housekeeping gene GAPDH and for myogenic differentiation gene (MYOD). Image shows the recovery, expressed as a % of input (the relative amount of IP'd DNA compared to input DNA after qPCR analysis).



ChIP was performed with 5 ul ab on sheared chromatin from 1 million HeLaS3 cells. IgG (2 ug/IP) was negative control. The IP'd DNA was analysed by qPCR with primers for the coding and promoter region of active GAPDH, for the coding region of inactive TSH2B and for the Sat2 (A). The IP'd DNA was analysed. The 36 bp tags were aligned to the human genome using the ELAND algorithm. B shows the results in 200 kb regions of chromosome 12 (including the GAPDH positive control), 6 and 7 and 14. Results show an enrichment of the H3K36me3 at active genes.