

Product datasheet for **TA347167**

H3FA (HIST1H3A) Rabbit Polyclonal Antibody

Product data:

Product Type:	Primary Antibodies
Applications:	Dot, ELISA, IF, WB
Recommended Dilution:	ChIP (10 µl/ChIP); ELISA (1:2,000 ?? 1:3,000); Dot blotting (1:200,000); Western blotting (1:1000); Immunofluorescence (1:200)
Reactivity:	Human
Host:	Rabbit
Isotype:	IgG
Clonality:	Polyclonal
Immunogen:	The immunogen for anti-H3K9me1 antibody: histone H3 containing the monomethylated lysine 9 (H3K9me1), 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



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Background:

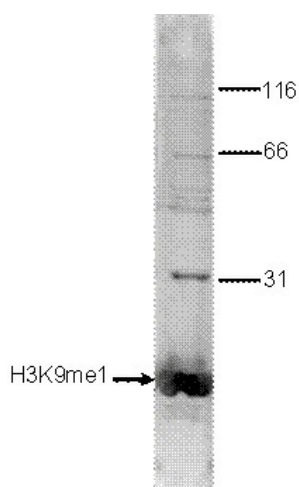
Histones are the main constituents of the protein part of chromosomes of eukaryotic cells. They are rich in the amino acids arginine and lysine and have been greatly conserved during evolution. Histones pack the DNA into tight masses of chromatin. Two core histones of each class H2A, H2B, H3 and H4 assemble 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. Methylation of histone H3K9 is associated with gene repression and heterochromatin formation, although higher levels of H3K9me1 have been found in some more active promoters.

Synonyms:

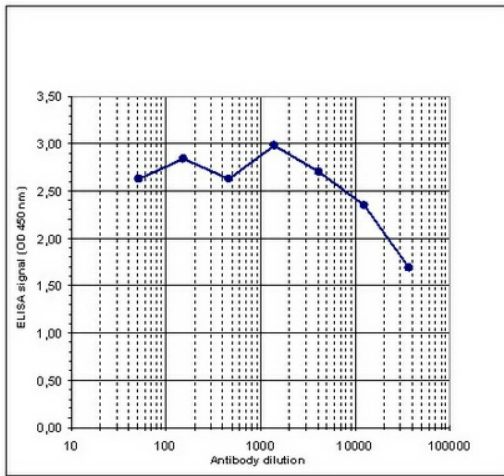
A; H3; H3FA

Protein Pathways:

Systemic lupus erythematosus

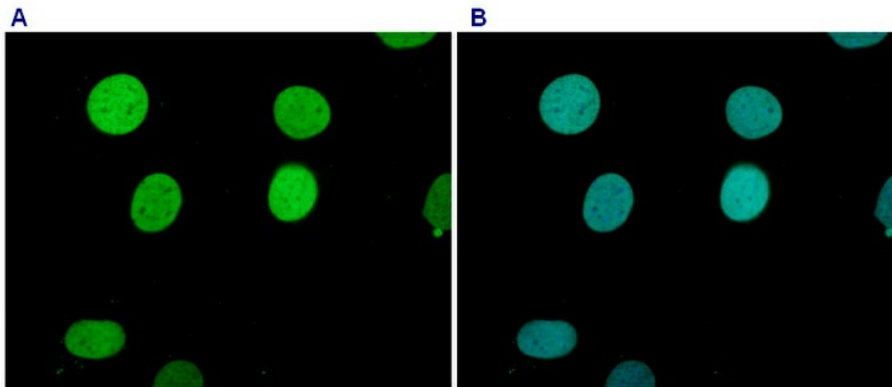
Product images:

WB using the antibody against H3K9me1 diluted 1:1000 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.

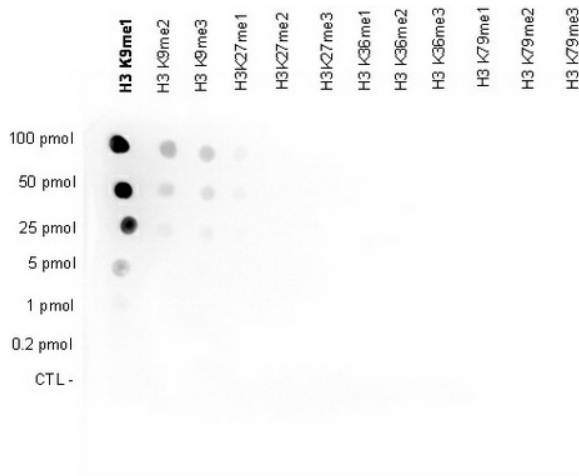


Determination of the titer To determine the titer, an ELISA was performed using a serial dilution of the antibody against H3K9me1. The antigen used was a peptide containing the histone modification of interest. By plotting the absorbance against the antibody dilution (Figure 2), the titer of the antibody was estimated to be 1:50,000.

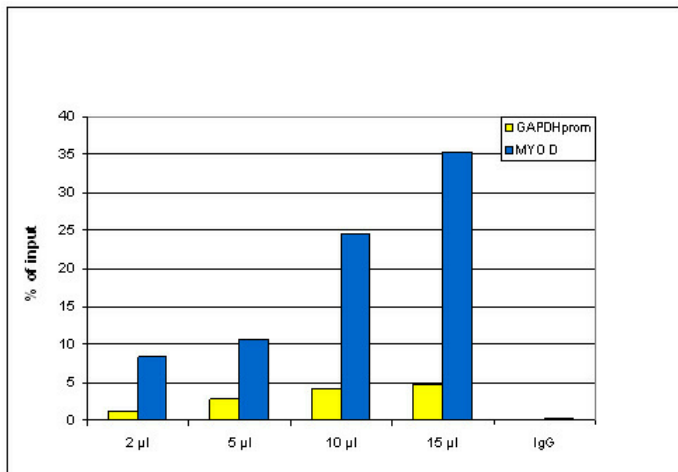
Figure 5



HeLa cells were stained with the H3K9me1 ab and with DAPI. Cells were formaldehyde fixated, permeabilized with Triton X-100 and blocked with PBS containing 2.5% BSA. A: cells were immunofluorescently labelled with the H3K9me1 antibody (diluted 1:200 and incubated for 1 hour at room temperature) followed by goat anti-rabbit antibody conjugated to DyLight. B: staining of the nuclei with DAPI, which specifically labels DNA. Both antibody and DAPI staining are restricted to the nucleus.



Dot Blot was performed to test the cross reactivity of the H3K9me1 ab with peptides containing other modifications of histone H3. Other histone modifications include di- and trimethylation of the same lysine and mono-, di- and trimethylation of lysine 27, 36 and 79. One hundred to 0.2 pmol of peptide containing the respective histone modification were spotted on a membrane. The ab was used at a dilution of 1:200,000. Image shows a high specificity of the ab for the modification of interest.



ChIP assays using human U2OS cells, IgG (5 ug/IP) was used as negative control. qPCR was performed using primers for the promoter of GAPDH and for the coding region of MYOD, a gene that is inactive at normal conditions. Image shows the recovery, expressed as a % of input (the relative amount of IP'd DNA compared to input DNA after qPCR analysis).