

OriGene Technologies, Inc.

9620 Medical Center Drive, Ste 200 Rockville, MD 20850, US Phone: +1-888-267-4436 https://www.origene.com techsupport@origene.com EU: info-de@origene.com CN: techsupport@origene.cn

Product datasheet for TA347191

H3FA (HIST1H3A) Rabbit Polyclonal Antibody

Product data:

Product Type:	Primary Antibodies
Applications:	Dot, ELISA, WB
Recommended Dilution:	ChIP/ChIP-seq (2 ug/ChIP); ELISA (1:500 ~1:1,000); Dot blotting (1:50,000); Western blotting (1:500)
Reactivity:	Human, Mouse
Host:	Rabbit
lsotype:	lgG
Clonality:	Polyclonal
Immunogen:	The immunogen for anti-H3K79me3 antibody: histone H3 containing the trimethylated lysine 79 (H3K79me3), 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, H3a
Database Link:	<u>NP_003520</u> <u>Entrez Gene 360198 MouseEntrez Gene 8350 Human</u> <u>P68431</u>



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GRIGENE H3FA (HIST1H3A) Rabbit Polyclonal Antibody – TA347191

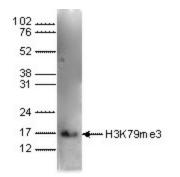
Background: Histones are the main constituents of the protein part of chromosomes of eukaryotic cells. They are rich in the amino acids arginine and lysine and have been greatly conserved during evolution. Histones pack the DNA into tight masses of chromatin. Two core histones of each class H2A, H2B, 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 H3 on K79 was shown to be more present at active promotors than at silent promotors.

Synonyms: A; H3; H3FA

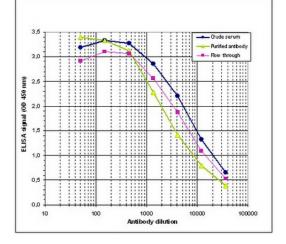
Protein Pathways:

Systemic lupus erythematosus

Product images:

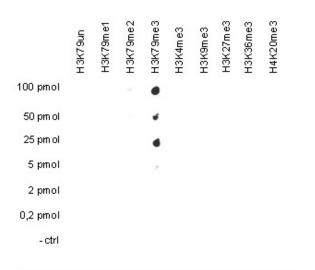


WB using the antibody against H3K79me3, diluted 1:1,000 in TBS-Tween containing 5% skimmed milk. The molecular weight marker is shown on the right; the location of the protein of interest is indicated on the left.

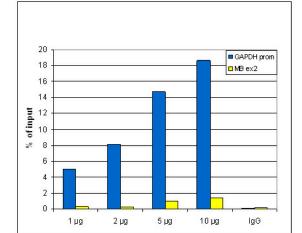


Determination of the antibody titer To determine the titer of the antibody, an ELISA was performed using a serial dilution of antibody against H3K79me3, crude serum and flow through in antigen coated wells. The antigen used was a peptide containing the histone modification of interest. By plotting the absorbance against the antibody dilution (Figure 3), the titer of the antibody was estimated to be 1:3, 500.

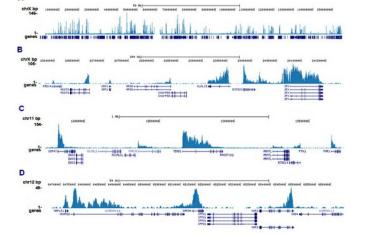
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A Dot Blot analysis was performed to test the cross reactivity of the antibody against H3K79me3 with peptides containing other histone modifications and the unmodified H3K79. One hundred to 0.2 pmol of the respective peptides were spotted on a membrane. The antibody was used at a dilution of 1:20,000. Figure 4 shows a high specificity of the antibody for the modification of interest.



ChIP was performed with the ab on sheared chromatin from 1 million HeLaS3 cells using the "Auto Histone ChIP experiment was analysed. IgG (5 ug/IP) was used as negative control. qPCR primers were for the GAPDH promoter and for exon 2 of the inactive myoglobin gene. Image shows the recovery, expressed as a % of input (the relative amount of IP'd DNA compared to input DNA after qPCR). These results are in accordance with the observation that H3K79me3 shows a preference for active promoters.



ChIP was performed as described above using 2 ug of the ab against H3K79me3. The IP'd DNA was analysed on an Illumina Genome Analyzer. The 36 bp tags were aligned to the human genome using the ELAND algorithm. Image shows the peak distribution along the complete sequence and a 600 kb region of the Xchromosome (figure 2A and B), in a 2 Mb region from chromosome 11 (figure 2C), and in a 100 kb region surrounding the GAPDH positive control gene (figure 2D).

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