

### **Product datasheet for TA347184**

#### OriGene Technologies, Inc.

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# H3FA (HIST1H3A) Rabbit Polyclonal Antibody

#### **Product data:**

Product Type: Primary Antibodies

Applications: Dot, ELISA, IF, WB

**Recommended Dilution:** ChIP (2 μg/ChIP); ELISA (1:100 - 1:500); Dot blotting (1:20,000); Western blotting (1:1,000);

Immunofluorescence (1:1,000)

Reactivity: Human
Host: Rabbit
Isotype: IgG

Clonality: Polyclonal

Immunogen: The immunogen for anti-H3K27me1 antibody: histone H3 containing the monomethylated

lysine 27 (H3K27me1), 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: NP 003520

Entrez Gene 8350 Human

P68431

**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.

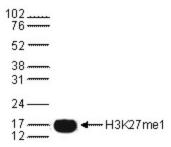




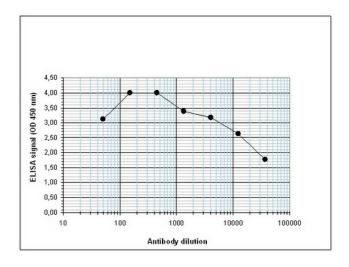
Synonyms: A; H3; H3FA

**Protein Pathways:** Systemic lupus erythematosus

# **Product images:**

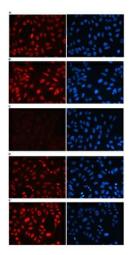


WB using the antibody against H3K27me1 diluted 1:1,000 in TBS-Tween containing 5% skimmed milk. The position of the protein of interest is indicated on the right; the marker (in kDa) is shown on the left.

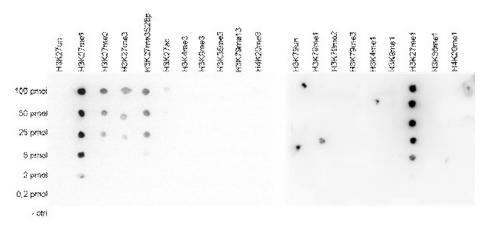


Determination of the antibody titer To determine the titer of the antibody, an ELISA was performed using a serial dilution of the antibody against H3K27me1, crude serum and flow through. The antigen used was a peptide containing the histone modification of interest. By plotting the absorbance against the antibody dilution (Figure 1), the titer of the purified antibody was estimated to be 1:32, 900.

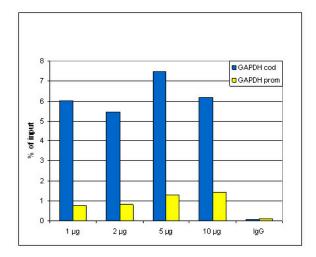




Human U2OS cells were fixed with 4% formaldehyde for 20' and blocked with PBS/TX-100 containing 5% normal goat serum. A: cells were labeled with the H3K27me1 antibody (left) at 1:1,000 followed by an anti-rabbit conjugated to Alexa568 or with DAPI (right). B, C, D and E: staining of the cells with the H3K27me1 antibody after incubation of the antibody with 2 ng/ul blocking peptide containing the unmodified and the mono-, di- and trimethylated H3K27, respectively.



A Dot Blot analysis was performed to test the cross reactivity of the antibody against H3K27me1 with peptides containing other modifications and unmodified sequences of histone H3 and H4. One hundred to 0.2 pmol of the peptide containing the respective histone modification were spotted on a membrane. The antibody was used at a dilution of 1:20,000. Image shows a high specificity of the antibody for the modification of interest.



ChIP assays using HeLa cells: ChIP" kit, using sheared chromatin from 100,000 cells. A titration of the antibody consisting of 1, 2, 5 and 10 ug per ChIP experiment was analysed. IgG (2 ug/IP) was used as negative control. qPCR primers were for the promoter and the coding region of the active gene GAPDH negative and a positive control target, respectively. Image shows the recovery, expressed as a % of input (the relative amount of IP'd DNA compared to input DNA after qPCR analysis).