

Datasheet for 600-401-J01**Histone H4 K20me1 Antibody****Overview**

Description:	Anti-Histone H4 [Monomethyl Lys20] (RABBIT) Antibody - 600-401-J01
Item No.:	600-401-J01
Size:	50 µg
Applications:	ChIP, Dot Blot, IF, Multiplex, WB
Reactivity:	Human, Mouse, C. elegans
Host Species:	Rabbit

Product Details

Background:	SET8 specifically catalyzes the mono-methylation of H4 at K20. Loss of this crucial methylation causes multiple DNA breaks, which instigates a p53-dependent DNA damage response to avoid mitosis and aberrant chromosomal activity. Therefore, this PTM is essential to genome replication and stability through S-phase. In mammalian stem cells, Xist expression blocks the formation of H4K20me1, which is one of the first examples of a direct connection between chromatin and stem cell differentiation. Anti-Histone H4 are ideal for researchers interested in Chromatin Modifiers, Chromatin Research, DNA Repair, DNA replication Transcription Translation and Splicing, Histones and Modified Histones, and Epigenetics research.
Synonyms:	rabbit anti-Histone H4 monomethyl Lys20 antibody, H4K20me1, HIST2H4B, HIST1H4H, HIST1H4I, HIST1H4J, HIST1H4K, HIST1H4L, HIST2H4, HIST2H4A, HIST1H4A, HIST1H4B, HIST1H4C, HIST1H4D, HIST1H4E, HIST1H4F, histone cluster 4, H4, histone 4, H4
Host Species:	Rabbit
Clonality:	Polyclonal
Format:	IgG

Target Details

Gene Name:	HIST4H4
Reactivity:	Human, Mouse, C. elegans
PTM Specificity:	Methylation
Immunogen Type:	Conjugated Peptide

Immunogen:	Histone H4 [Monomethyl Lys20] affinity purified antibody was prepared from whole rabbit serum produced by repeated immunizations with a synthetic monomethylated peptide surrounding Lysine 20 of human Histone H4.
Purity/Specificity:	Anti-Histone H4 [Monomethyl Lys20] was affinity purified from monospecific antiserum by immunoaffinity chromatography. This antibody reacts with human Histone H4. A BLAST analysis was used to suggest cross-reactivity with Human, mouse, and C. elegans. Predicted to react with most mammal species. Cross-reactivity with Histone H4 from other sources has not been determined.
Relevant Links:	• UniProtKB - P62805 • NCBI - NP_001029249 • GenelD - 121504

Application Details

Tested Applications:	ChIP, Dot Blot, IF, Multiplex, WB
Application Note:	Anti-Histone H4 [Monomethyl Lys20] antibody is tested for Western Blot, Dot Blot, Chromatin Immunoprecipitation, Immunofluorescence, and Immunocytochemistry. Specific conditions for reactivity should be optimized by the end user. Expect a band approximately ~13 kDa corresponding to Histone H4 protein by Western Blotting in HeLa histone prep lysate or the appropriate cell lysate or extract. Epi-Plus™ antibody production in collaboration with Novus Biologicals.
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
ChIP:	2-5µg/million cells
IF:	1:50
IHC:	1:50
WB:	1:500

Formulation

Physical State:	Liquid (sterile filtered)
Concentration:	1.12 mg/ml by UV absorbance at 280 nm
Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	0.01% (w/v) Sodium Azide
Stabilizer:	30% Glycerol

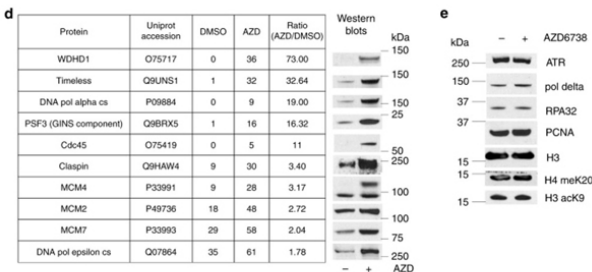
Shipping & Handling

Shipping Condition: Dry Ice

Storage Condition: Store vial at -20° C prior to opening. Aliquot contents and freeze at -20° C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use.

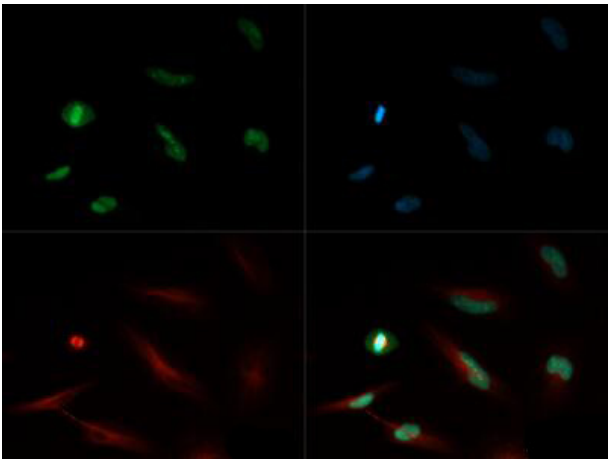
Expiration: Expiration date is one (1) year from date of receipt.

Images



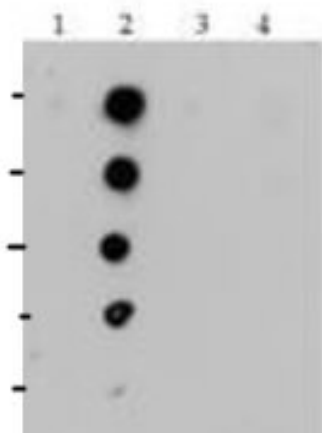
Western Blot

(d, e) 293T cells were treated with AZD6738 or DMSO for 30 min and then fractionated. Nuclease-insoluble fraction was analyzed by mass spectrometry showing increase of the proteins involved in replication. Western blots confirm the MS results. While ATR and RPA34 accumulate in the chromatin after treatments with DNA damaging agents, neither of these proteins were recruited to the chromatin in cells treated with ATRi alone in Fig. 1e. [p/n 600-401-I72 H3, acK9; p/n 600-401-J01, H4 meK20] Fig 1. PMID: 29123096



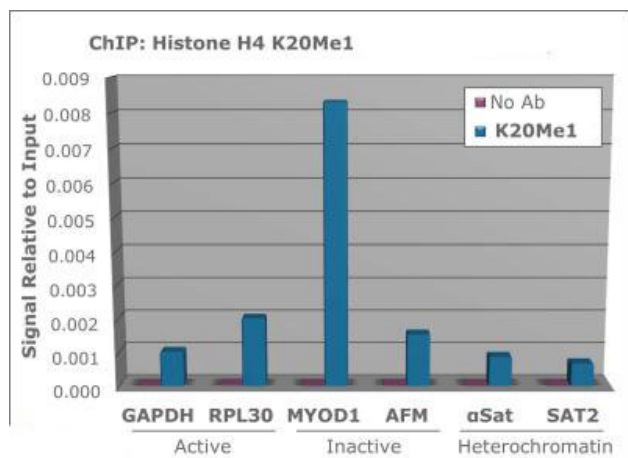
Immunofluorescence Microscopy

Immunofluorescence of Rabbit Anti-Histone H4 [Monomethyl Lys20] Antibody. Tissue: HeLa cells. Fixation: 0.5% PFA. Antigen retrieval: Not required. Primary antibody: Histone H4 [Monomethyl Lys20] antibody at a 1:500 dilution for 1 h at RT. Secondary antibody: FITC secondary antibody at 1:10,000 for 45 min at RT. Localization: Histone H4 [Monomethyl Lys20] is nuclear and chromosomal. Staining: Histone H4 [Monomethyl Lys20] is expressed in green, nuclei and alpha-tubulin are counterstained with DAPI (blue) and Dylight 550 (red).



Dot Blot

Dot Blot of Rabbit Histone H4 [Monomethyl Lys20] Antibody.
 Antigen: Lane 1: unmodified. Lane 2: K20me1. Lane 3: K20me2. Lane 4: K20me3. Load: 0.6, 1, 3, 6, and 10 picomoles of peptide. Primary antibody: Histone H4 [Monomethyl Lys20] antibody at 2 µg/ml for 45 min at 4°C. Secondary antibody: Dylight™488 rabbit secondary antibody at 1:10,000 for 45 min at RT. Block: 5% BLOTTO overnight at 4°C.



ChIP

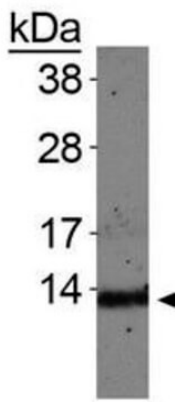
Chromatin Immunoprecipitation Rabbit Anti-Histone H4 [Monomethyl Lys20] Antibody. Chromatin from one million formaldehyde cross-linked HeLa cells was used with 2µg of Anti-Histone H4K20me1 and 20ul of magnetic IgG beads per immunoprecipitation. A no antibody (No Ab) control was also used. Immunoprecipitated DNA was quantified using quantitative real-time PCR and SYBR green dye, then normalized to the non-precipitated input chromatin, which is equal to one.

250>
 150>
 100>
 75>
 50>
 37>
 25>
 20>
 15>
 10>



Western Blot

Western Blot of Rabbit Anti-Histone H4 [Monomethyl Lys20] Antibody. Lane 1: C. elegans embryo lysate. Load: 30 µg per lane. Primary antibody: Histone H4 [Monomethyl Lys20] at 1:500 for overnight at 4°C. Secondary antibody: IRDye800™ rabbit secondary antibody at 1:10,000 for 45 min at RT. Block: 5% BLOTTO overnight at 4°C. Predicted/Observed size: ~13 kDa. Other band(s): None.

**Western Blot**

Western Blot of Rabbit Anti-Histone H4 [Monomethyl Lys20] Antibody. Lane 1: NIH-3T3 histone preps. Load: 30 µg per lane. Primary antibody: Histone H4 [Monomethyl Lys20] at 1:500 for overnight at 4°C. Secondary antibody: IRDye800™ rabbit secondary antibody at 1:10,000 for 45 min at RT. Block: 5% BLOTTO overnight at 4°C. Predicted/Observed size: ~13 kDa. Other band(s): None.

References

- Moiseeva et al. ATR kinase inhibition induces unscheduled origin firing through a Cdc7-dependent association between GINS and And-1. *Nature Communications* (2017)

Disclaimer

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