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Product datasheet

Anti-Histone H3 (di methyl K9) antibody [Y49] - ChIP Grade ab32521

יעלאעבע RabMAb

★★★★★ 1 Abreviews 17 References 画像数6

製品の概要

製品名 Anti-Histone H3 (di methyl K9) antibody [Y49] - ChIP Grade

製品の詳細 Rabbit monoclonal [Y49] to Histone H3 (di methyl K9) - ChIP Grade

由来種 Rabbit

特異性 The antibody only detects Histone H3 dimethylated on Lysine 9.

アプリケーション 適用あり: ICC/IF, WB, Flow Cyt (Intra), ChIP

適用なし: IHC or IP

種交差性 交差種: Human

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

交差が予測される動物種: Mouse, Rat 🔷

ポジティブ・コントロール WB: HeLa cell lysate. ICC/IF: HeLa cells. Flow Cyt (intra): HeLa cells. ChIP: Chromatin prepared

from HeLa cells.

特記事項 This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility

- Improved sensitivity and specificity

- Long-term security of supply

- Animal-free production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb patents**.

製品の特性

製品の状態

保存方法 Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Avoid freeze / thaw cycle.

バッファー pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 9% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA, 50% Tissue culture

supernatant

精製度 Protein A purified

ポリ/モノ モノクローナル

クローン名 Y49 アイソタイプ IgG

アプリケーション

The Abpromise guarantee <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab32521の使用に適用されますアプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
ICC/IF	**** <u>(1)</u>	1/500.
WB		1/1000. Detects a band of approximately 17 kDa (predicted molecular weight: 15 kDa).
Flow Cyt (Intra)		1/220.
ChIP		Use 5 µg for 25 µg of chromatin.

追加情報

Is unsuitable for IHC or IP.

ターゲット情報

機能 Core component of nucleosome. Nucleosomes wrap and compact DNA into chromatin, limiting

DNA accessibility to the cellular machineries which require DNA as a template. Histones thereby play a central role in transcription regulation, DNA repair, DNA replication and chromosomal stability. DNA accessibility is regulated via a complex set of post-translational modifications of

histones, also called histone code, and nucleosome remodeling.

配列類似性 Belongs to the histone H3 family.

発生段階 Expressed during S phase, then expression strongly decreases as cell division slows down

during the process of differentiation.

翻訳後修飾 Acetylation is generally linked to gene activation. Acetylation on Lys-10 (H3K9ac) impairs

methylation at Arg-9 (H3R8me2s). Acetylation on Lys-19 (H3K18ac) and Lys-24 (H3K24ac)

favors methylation at Arg-18 (H3R17me).

Citrullination at Arg-9 (H3R8ci) and/or Arg-18 (H3R17ci) by PADI4 impairs methylation and

represses transcription.

 $Asymmetric\ dimethylation\ at\ Arg-18\ (H3R17me2a)\ by\ CARM1\ is\ linked\ to\ gene\ activation.$

 $Symmetric\ dimethylation\ at\ Arg-9\ (H3R8me2s)\ by\ PRMT5\ is\ linked\ to\ gene\ repression.$

Asymmetric dimethylation at Arg-3 (H3R2me2a) by PRMT6 is linked to gene repression and is mutually exclusive with H3 Lys-5 methylation (H3K4me2 and H3K4me3). H3R2me2a is present at the 3' of genes regardless of their transcription state and is enriched on inactive promoters, while

it is absent on active promoters.

Methylation at Lys-5 (H3K4me), Lys-37 (H3K36me) and Lys-80 (H3K79me) are linked to gene activation. Methylation at Lys-5 (H3K4me) facilitates subsequent acetylation of H3 and H4. Methylation at Lys-80 (H3K79me) is associated with DNA double-strand break (DSB) responses

and is a specific target for TP53BP1. Methylation at Lys-10 (H3K9me) and Lys-28 (H3K27me) are linked to gene repression. Methylation at Lys-10 (H3K9me) is a specific target for HP1 proteins (CBX1, CBX3 and CBX5) and prevents subsequent phosphorylation at Ser-11 (H3S10ph) and acetylation of H3 and H4. Methylation at Lys-5 (H3K4me) and Lys-80 (H3K79me) require preliminary monoubiquitination of H2B at 'Lys-120'. Methylation at Lys-10 (H3K9me) and Lys-28 (H3K27me) are enriched in inactive X chromosome chromatin.

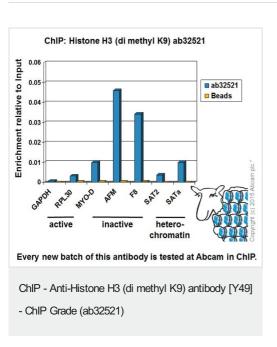
Phosphorylated at Thr-4 (H3T3ph) by GSG2/haspin during prophase and dephosphorylated during anaphase. Phosphorylation at Ser-11 (H3S10ph) by AURKB is crucial for chromosome condensation and cell-cycle progression during mitosis and meiosis. In addition phosphorylation at Ser-11 (H3S10ph) by RPS6KA4 and RPS6KA5 is important during interphase because it enables the transcription of genes following external stimulation, like mitogens, stress, growth factors or UV irradiation and result in the activation of genes, such as c-fos and c-jun. Phosphorylation at Ser-11 (H3S10ph), which is linked to gene activation, prevents methylation at Lys-10 (H3K9me) but facilitates acetylation of H3 and H4. Phosphorylation at Ser-11 (H3S10ph) by AURKB mediates the dissociation of HP1 proteins (CBX1, CBX3 and CBX5) from heterochromatin. Phosphorylation at Ser-11 (H3S10ph) is also an essential regulatory mechanism for neoplastic cell transformation. Phosphorylated at Ser-29 (H3S28ph) by MLTK isoform 1, RPS6KA5 or AURKB during mitosis or upon ultraviolet B irradiation. Phosphorylation at Thr-7 (H3T6ph) by PRKCBB is a specific tag for epigenetic transcriptional activation that prevents demethylation of Lys-5 (H3K4me) by LSD1/KDM1A. At centromeres, specifically phosphorylated at Thr-12 (H3T11ph) from prophase to early anaphase, by DAPK3 and PKN1. Phosphorylation at Thr-12 (H3T11ph) by PKN1 is a specific tag for epigenetic transcriptional activation that promotes demethylation of Lys-10 (H3K9me) by KDM4C/JMJD2C. Phosphorylation at Tyr-42 (H3Y41ph) by JAK2 promotes exclusion of CBX5 (HP1 alpha) from chromatin.

Monoubiquitinated by RAG1 in lymphoid cells, monoubiquitination is required for V(D)J recombination (By similarity). Ubiquitinated by the CUL4-DDB-RBX1 complex in response to ultraviolet irradiation. This may weaken the interaction between histones and DNA and facilitate DNA accessibility to repair proteins.

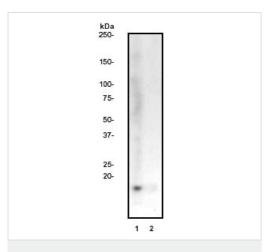
Nucleus. Chromosome.

細胞内局在

画像



Chromatin was prepared from HeLa cells according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10 minutes. The ChIP was performed with 25µg of chromatin, 5µg of ab32521 (blue), and 20µl of Protein A/G sepharose beads. No antibody was added to the beads control (yellow). The immunoprecipitated DNA was quantified by real time PCR (Taqman approach for active and inactive loci, Sybr green approach for heterochromatic loci). Primers and probes are located in the first kb of the transcribed region.



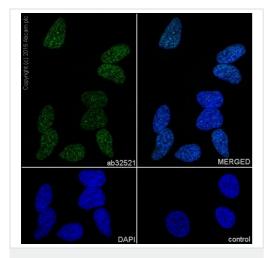
Western blot - Anti-Histone H3 (di methyl K9) antibody [Y49] - ChIP Grade (ab32521)

All lanes : Anti-Histone H3 (di methyl K9) antibody [Y49] - ChIP Grade (ab32521) at 1/1000 dilution

Lane 1: HeLa cell lysate

Lane 2: recombinant Histone H3

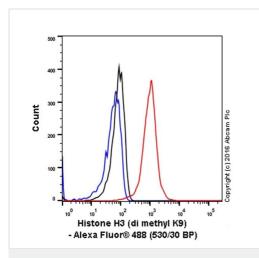
Predicted band size: 15 kDa **Observed band size:** 17 kDa



Immunocytochemistry/ Immunofluorescence - Anti-Histone H3 (di methyl K9) antibody [Y49] - ChIP Grade (ab32521)

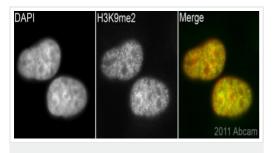
Immunocytochemistry/Immunofluorescence analysis of HeLa (human cervix adenocarcinoma) labelling Histone H3 (di methyl K9) with purified ab32521 at 1/500. Cells were fixed with 4% PFA and permeabilized with 0.1% Triton X-100. An Alexa Fluor[®] 488-conjugated goat anti-rabbit lgG (1/1000) was used as the secondary antibody (Ab150077). Nuclei counterstained with DAPI (blue).

Control: PBS only



Flow Cytometry (Intracellular) - Anti-Histone H3 (di methyl K9) antibody [Y49] - ChIP Grade (ab32521)

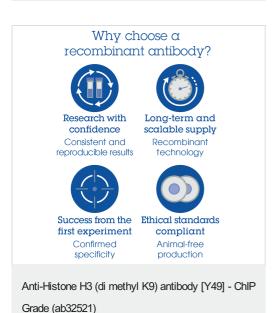
Intracellular Flow Cytometry analysis of HeLa (human cervix adenocarcinoma) cells labeling Histone H3 (di methyl K9) with purified ab32521 at 1/220 dilution (10ug/mL) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit lgG (Alexa Fluorr®488) (1/2000 dilution) was used as the secondary antibody. Rabbit monoclonal lgG (Black) was used as the isotype control, cells without incubation with primary antibody and secondary antibody (Blue) were used as the unlabeled control.



Immunocytochemistry/ Immunofluorescence - Anti-Histone H3 (di methyl K9) antibody [Y49] - ChIP Grade (ab32521)

Image courtesy of an Abreview submitted by Dr. Kirk McManus, Univ. of Manitoba/Cancer Care MICB, Canada

ab32521 (1/500) staining Histone H3 di-methyl K9 in HeLa cells (green). Cells were fixed in paraformaldehyde, permeabilised with 0.5% Triton X100 and counterstained with DAPI in order to highlight the nucleus (red). For further experimental details, please refer to Abreview.



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