abcam

Product datasheet

Histone H3 (K4 methylation) Panel (mono methyl K4, di methyl K4, tri methyl K4) ab103938

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Overview

Product name Histone H3 (K4 methylation) Panel (mono methyl K4, di methyl K4, tri methyl K4)

Product overview ab103938 is a Histone H3 (K4 methylation) Panel designed for the validation and characterization of the methylation state of Histone H3 on K4. Methylation of the canonical core histones can contribute to the formation of transcriptionally active and inactive chromatin in response to various signalling pathways and is a central modification for regulating epigenetic transitions in chromatin. Methylation of Histone H3 K4 is associtated with euchromatin and active genes.

Notes **Explore our range of antibody sample panels** designed to provide you with a variety of trial-size antibodies in a convenient and cost-effective format.

Carrier-free formulations of our recombinant antibodies are available and ready to use for multiplex assays. Please refer to the 'Associated products' section below.

Properties

Storage instructions Store at -20°C. Please refer to protocols.

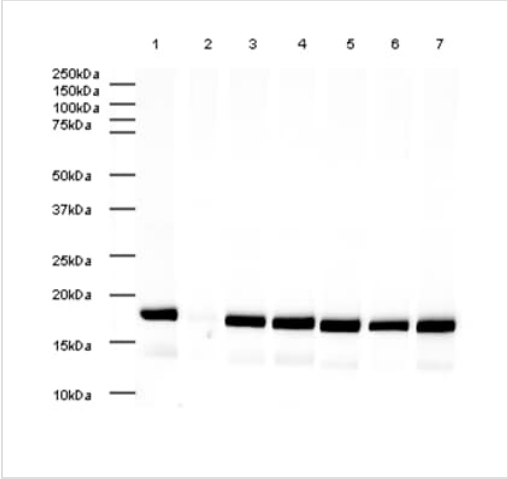
Components	1 units
ab32356 - Anti-Histone H3 (di methyl K4) antibody [Y47] - ChIP Grade	1 x 25µl
ab8895 - Anti-Histone H3 (mono methyl K4) antibody - ChIP Grade	1 x 25µg
ab213224 - Anti-Histone H3 (tri methyl K4) antibody [EPR20551-225] - ChIP Grade	1 x 25µl
ab176842 - Rabbit monoclonal to Histone H3 [EPR16987]	1 x 10µl

Function 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.

Sequence similarities Belongs to the histone H3 family.

Developmental stage	Expressed during S phase, then expression strongly decreases as cell division slows down during the process of differentiation.
Post-translational modifications	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.
Cellular localization	Nucleus. Chromosome.

Images



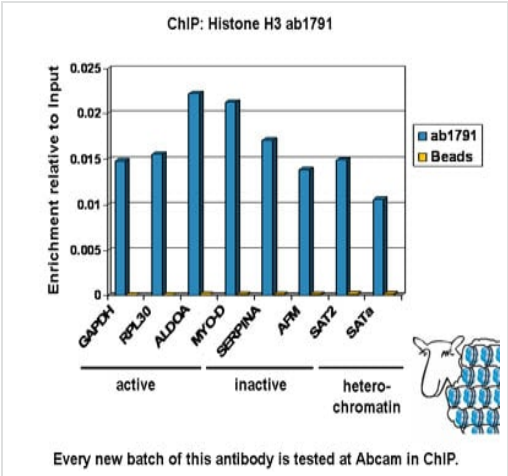
Western blot - Histone H3 (K4 Methylation) Panel (ab103938)

All lanes : Anti-Histone H3 (mono methyl K4) antibody - ChIP Grade (**ab8895**) at 1/500 dilution

Lane 1 : Calf thymus histone lysate
Lane 2 : Calf thymus histone lysate with Human Histone H3 (mono methyl K4) peptide (**ab1340**) at 1 µg/ml
Lane 3 : Calf thymus histone lysate with Human Histone H3 (di methyl K4) peptide (**ab7768**) at 1 µg/ml
Lane 4 : Calf thymus histone lysate with Human Histone H3 (tri methyl K4) peptide (**ab1342**) at 1 µg/ml
Lane 5 : Calf thymus histone lysate with Human Histone H3 (mono methyl K9) peptide (**ab1771**) at 1 µg/ml
Lane 6 : Calf thymus histone lysate with Human Histone H3 (mono methyl K27) peptide (**ab1780**) at 1 µg/ml
Lane 7 : Calf thymus histone lysate with Human Histone H3 (unmodified) peptide (**ab2903**)

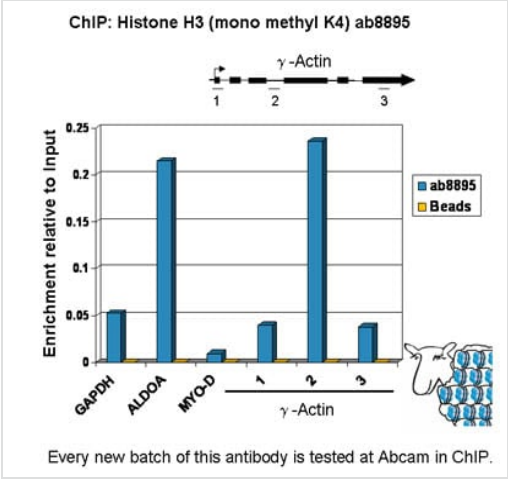
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab6721**) at 1/5000 dilution

ab8895 is specific for mono-methylated Lysine 4 of histone H3 and does not recognize di- or tri-methyl Lysine 4 nor methylation at Lysine 9. This is shown in lane 2 where the activity of the antibody is specifically blocked by the addition of the immunizing peptide (**ab1340**).



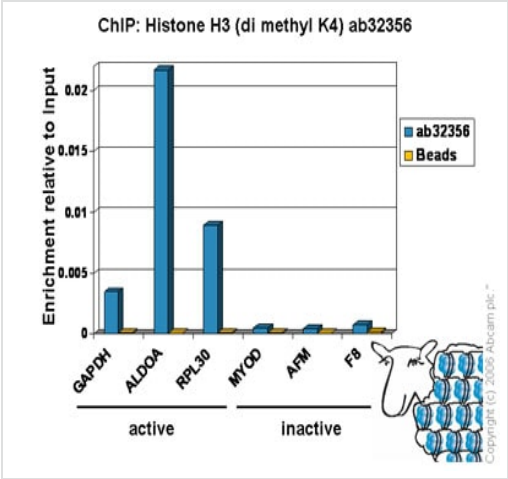
ChIP - Histone H3 (K4 Methylation) Panel (ab103938)

Chromatin was prepared from U2OS cells according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10 min. The ChIP was performed with 25 µg of chromatin, 2 µg of **ab1791** (blue), and 20 µl 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). Primers and probes are located in the first kb of the transcribed region.



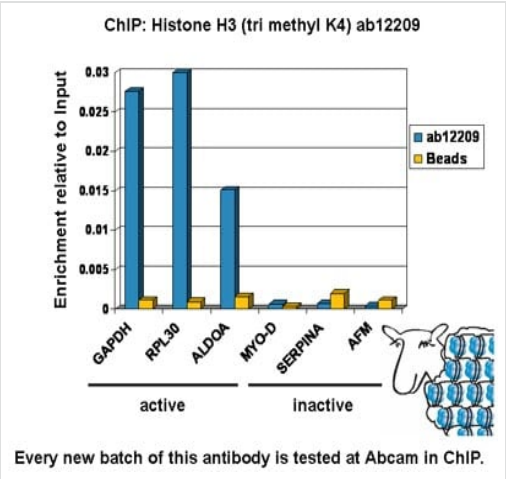
ChIP - Histone H3 (K4 Methylation) Panel (ab103938)

Chromatin was prepared from U2OS cells according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10min. The ChIP was performed with 25µg of chromatin, 2µg of **ab8895** (blue), and 20µl of Protein A/G sepharose beads. No antibody was added to the beads control (yellow). The immunoprecipitated DNA was quantified on the GAPDH and ALDOA (active) and MYO-D (inactive) promoters and over the y-Actin gene (active). Schematic diagram of the y-Actin gene is shown on the top of the figure. Black boxes represent exons and thin lines represent introns. PCR products are depicted as bars under the gene.



ChIP - Histone H3 (K4 Methylation) Panel (ab103938)

Chromatin was prepared from Hela cells according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10min. The ChIP was performed with 25µg of chromatin, 8µl of **ab32356** (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). Primers and probes are located in the first kb of the transcribed region.



ChIP - Histone H3 (K4 Methylation) Panel (ab103938)

Chromatin was prepared from U2OS cells according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10min. The ChIP was performed with 25 µg of chromatin, 2 µg of **ab12209** (blue), and 20 µl 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). Primers and probes are located in the first kb of the transcribed region.

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