

Anti-pan methyl Lysine antibody - ChIP Grade ab7315

★★★★☆ [2 Abreviews](#) [27 References](#) [5 Images](#)

Overview

Product name	Anti-pan methyl Lysine antibody - ChIP Grade
Description	Rabbit polyclonal to pan methyl Lysine - ChIP Grade
Host species	Rabbit
Specificity	ab7315 recognises Histone H3 di-methyl K4, di-methyl K9 and di-methyl K27 in WB. Non-histone samples have not been tested, thus, we do not know if ab7315 would work on non-histone samples.
Tested applications	Suitable for: IP, ChIP, ELISA, WB, ICC/IF, IHC-P
Species reactivity	Reacts with: Species independent
Immunogen	Full length native protein (purified). This information is proprietary to Abcam and/or its suppliers.
General notes	<p>This antibody will be extremely useful in the study of the regulation of transcription by methylation. Has also been successfully used in CHIP in both human and yeast.</p> <p>The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing.</p> <p>If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As</p>

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Add glycerol to a final volume of 50% for extra stability and aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.40 Preservative: 0.02% Sodium azide Constituent: PBS

Batches of this product that have a concentration < 1mg/ml may have BSA added as a stabilising agent. If you would like information about the formulation of a specific lot, please contact our scientific support team who will be happy to help.

Purity	Immunogen affinity purified
Primary antibody notes	This antibody will be extremely useful in the study of the regulation of transcription by methylation. Has also been successfully used in CHIP in both human and yeast.
Clonality	Polyclonal
Isotype	IgG

Applications

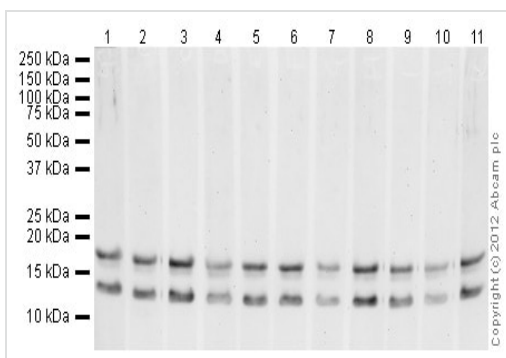
The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab7315 in the following tested applications. The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IP		Use at an assay dependent concentration.
ChIP	★★★★★ (1)	Use at an assay dependent concentration. Every new batch of this antibody is tested at Abcam in ChIP.
ELISA		Use at an assay dependent concentration.
WB	★★★★★ (1)	1/1000 - 1/2000. Predicted molecular weight: 14-17 kDa.
ICC/IF		Use a concentration of 5 µg/ml.
IHC-P		Use a concentration of 1 µg/ml. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

Target

Relevance Lysine methylation occurs in three distinct states, having either one (me1), two (me2) or three (me3) methyl groups attached to the amine group of the lysine side chain. In eukaryotes, histone H3 trimethylated at lysine 4 (H3K4me3) is associated with active chromatin and gene expression.

Images



Western blot - Anti-pan methyl Lysine antibody - ChIP Grade (ab7315)

All lanes : Anti-pan methyl Lysine antibody - ChIP Grade (ab7315) at 1 µg/ml

Lane 1 : Calf Thymus Histone Preparation Nuclear Lysate

Lane 2 : Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (unmodified) peptide (**ab7228**) at 0.5 µg/ml

Lane 3 : Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (mono methyl K4) peptide (**ab1340**) at 0.5 µg/ml

Lane 4 : Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (di methyl K4) peptide (**ab7768**) at 0.5 µg/ml

Lane 5 : Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (tri methyl K4) peptide (**ab1342**) at 0.5 µg/ml

Lane 6 : Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (mono methyl K9) peptide (**ab1771**) at 0.5 µg/ml

Lane 7 : Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (di methyl K9) peptide (**ab1772**) at 0.5 µg/ml

Lane 8 : Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (tri methyl K9) peptide (**ab1773**) at 0.5 µg/ml

Lane 9 : Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (mono methyl K27) peptide (**ab1780**) at 0.5 µg/ml

Lane 10 : Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (di methyl K27) peptide (**ab1781**) at 0.5 µg/ml

Lane 11 : Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (tri methyl K27) peptide (**ab1782**) at 0.5 µg/ml

Lysates/proteins at 0.5 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) preadsorbed (**ab97080**) at 1/5000 dilution

Developed using the ECL technique.

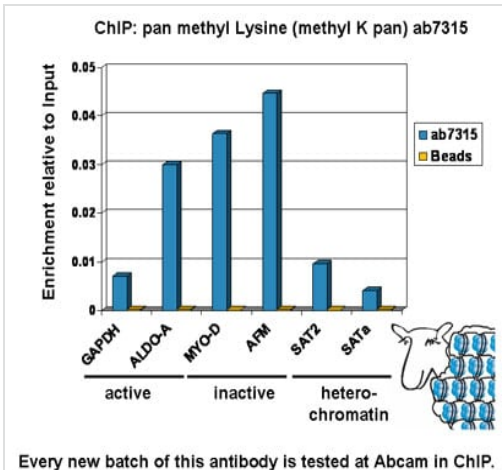
Performed under reducing conditions.

Predicted band size: 14-17 kDa

Exposure time: 20 minutes

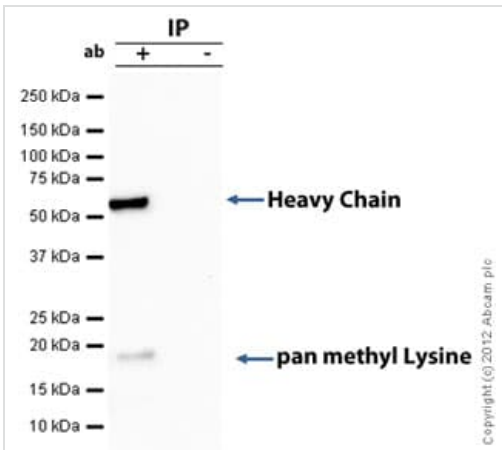
Expected molecular weights: H3 = 17 kDa; H4 = 14 kDa This image shows that the main epitopes recognized by ab7315 are the

di methylated lysine residues. This can be seen in lanes 4, 7 and 10 where the activity of the antibody is quenched by the immunizing peptides (**ab7768**, **ab1772**, **ab1781**).



ChIP - Anti-pan methyl Lysine antibody - ChIP Grade (ab7315)

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, 6.5µl of ab7315 (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.



Immunoprecipitation - Anti-pan methyl Lysine antibody - ChIP Grade (ab7315)

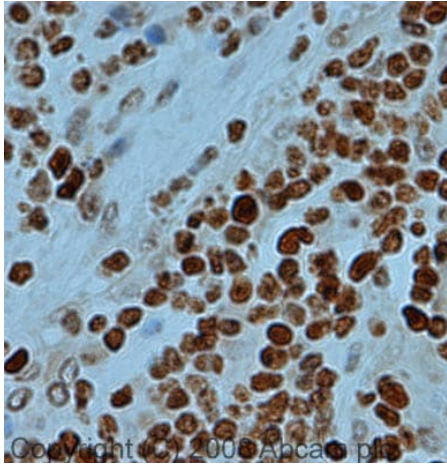
pan methyl Lysine was immunoprecipitated using 0.5mg HeLa whole cell extract, 5µg of it polyclonal to pan methyl Lysine and 50µl of protein G magnetic beads (+). No antibody was added to the control (-).

The antibody was incubated under agitation with Protein G beads for 10min, HeLa whole cell extract lysate diluted in RIPA buffer was added to each sample and incubated for a further 10min under agitation.

Proteins were eluted by addition of 40µl SDS loading buffer and incubated for 10min at 70°C; 10µl of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab7315.

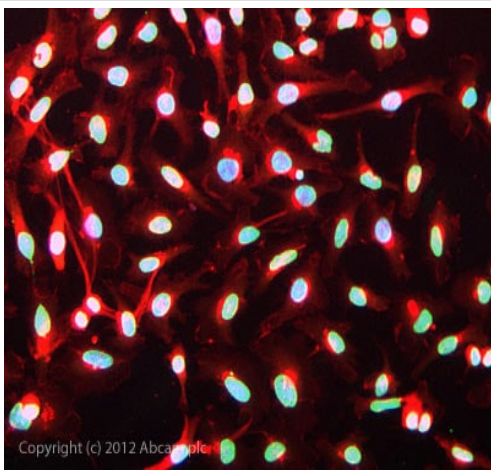
Secondary: Clean blot (HRP conjugate) at 1/1000 dilution.

Band: 17kDa: pan methyl Lysine.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-pan methyl Lysine antibody - ChIP Grade (ab7315)

IHC image of pan methyl Lysine (methyl K pan) staining in human breast carcinoma FFPE section, performed on a Bond™ system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab7315, 1µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.



Immunocytochemistry/ Immunofluorescence - Anti-pan methyl Lysine antibody - ChIP Grade (ab7315)

ICC/IF image of ab7315 stained HeLa cells. The cells were 100% methanol fixed (5 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody ab7315 at 5µg/ml overnight at +4°C. The secondary antibody (green) was DyLight® 488 goat anti- rabbit (**ab96899**) IgG (H+L) used at a 1/1000 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.

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