

Product datasheet

Anti-acetyl Lysine antibody - ChIP Grade ab21623

★★★★★ 8 Abreviews 38 References 6 Images

Overview

Product name	Anti-acetyl Lysine antibody - ChIP Grade
Description	Rabbit polyclonal to acetyl Lysine - ChIP Grade
Host species	Rabbit
Specificity	Recognises proteins acetylated on lysine residues. Tested: acetylated histone, acetylated BSA, and acetylated MBP, no reaction to the non acetylated proteins.
Tested applications	Suitable for: IP, IHC-P, ChIP, ICC/IF, WB, ELISA
Species reactivity	Reacts with: Species independent
Immunogen	Acetylated KLH conjugates.
Positive control	Subconfluent MMRU cells with or without HDAC inhibitor treatment
General notes	Use to detect acetylation of lysine.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
Storage buffer	Preservative: 0.1% Sodium azide Constituent: Tris buffered saline
Purity	Immunogen affinity purified
Purification notes	The antibody was specifically purified with immobilised acetylated lysine on agarose
Primary antibody notes	Use to detect acetylation of lysine.
Clonality	Polyclonal
Isotype	IgG

Applications

Our [Abpromise guarantee](#) covers the use of **ab21623** in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

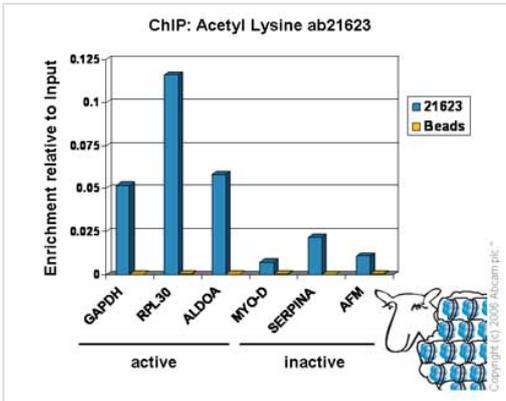
Application	Abreviews	Notes
IP	★★★★★	Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. PubMed: 24160175
ChIP	★★★★☆	Use 2-4 µg for 25 µg of chromatin.
ICC/IF		1/100 - 1/250. Subconfluent MMRU cells seeded in a 6-well plate with or without HDAC inhibitor treatment were fixed and permeabilized with 1% formaldehyde and 0.5% Triton X-100 for 10 minutes and blocked with 10 mg mL ⁻¹ BSA for 1 hour at room temperature. Cells were stained with FITC-conjugated anti-acetylated lysine antibody at 1:100 dilution with PBSt 1 % BSA for 1 hour at room temperature.
WB	★★★★☆	1/1000 - 1/2500.
ELISA		1/5000.

Target

Relevance

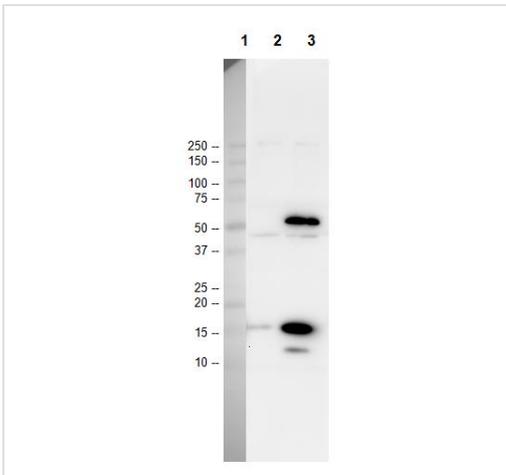
In the nucleus, DNA is tightly packed into nucleosomes generating an environment which is highly repressive towards DNA processes such as transcription. Acetylation of lysine residues within proteins has emerged as an important mechanism used by cells to overcome this repression. The acetylation of non-histone proteins such as transcription factors, as well as histones appears to be involved in this process. Acetylation may result in structural transitions as well as specific signaling within discrete chromatin domains. The role of acetylation in intracellular signaling has been inferred from the binding of acetylated peptides by the conserved bromodomain. Furthermore, recent findings suggest that bromodomain/acetylated-lysine recognition can serve as a regulatory mechanism in protein-protein interactions in numerous cellular processes such as chromatin remodeling and transcriptional activation. The reversible lysine acetylation of histones and non-histone proteins plays a vital role in the regulation of many cellular processes including chromatin dynamics and transcription, gene silencing, cell cycle progression, apoptosis, differentiation, DNA replication, DNA repair, nuclear import, and neuronal repression. More than 20 acetyltransferases and 18 deacetylases have been identified so far, but the mechanistic details of substrate selection and site specificity of these enzymes remain unclear. Over 40 transcription factors and 30 other nuclear, cytoplasmic, bacterial, and viral proteins have been shown to be acetylated in vivo.

Images



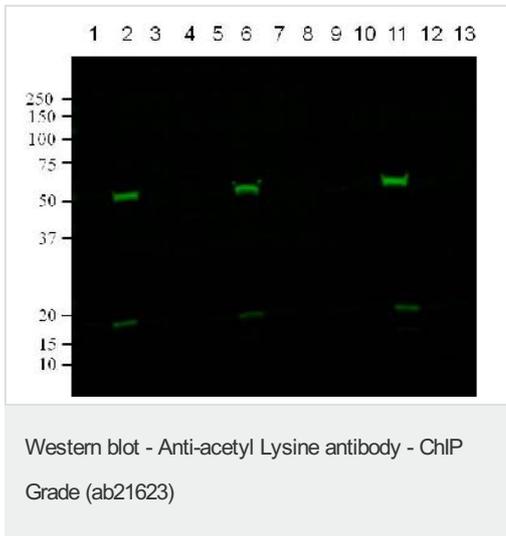
ChIP - Anti-acetyl Lysine antibody - ChIP Grade (ab21623)

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, 2µg of ab21623 (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.



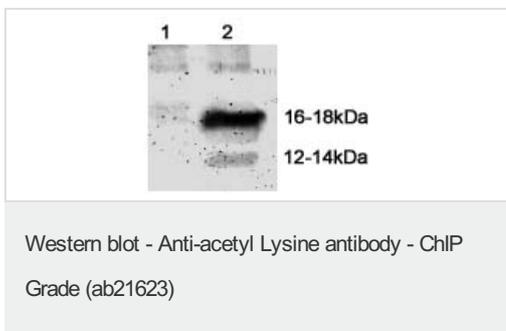
Western blot - Anti-acetyl Lysine antibody - ChIP Grade (ab21623)

Primary: All Lanes: Anti acetyl Lysine antibody (ab21623) at 1:1000. Lane 1: Marker. Lane 2: HeLa cells vehicle-treated (ab139414). Lane 3: HeLa cells, trichostatin A-treated (ab139414). Lysates at 20 ug/lane . Secondary: All Lanes: Goat anti-Rabbit IgG 1:10000. Performed under reducing conditions. Blocking buffer: 5% milk in PBS. Observed band sizes: 11 kDa 15kDa 45kDa 50 kDa.



Lane 1 = Extract of Mcf7 cells incubated with vehicle ? 20 ug. Lane 2 = Extract of Mcf7 cells incubated with trichostatin A ? 20 ug. Lane 3 = Extract of Mcf7 cells incubated with EX527 ? 20 ug. Lane 4 = Extract of Mcf7 cells incubated with nicotinamide ? 20 ug. Lane 5 = Extract of Mcf7 cells incubated with camptothecin ? 20 ug. Lane 6 = Extract of Mcf7 cells incubated with camptothecin and trichostatin A ? 20 ug. Lane 7 = Extract of Mcf7 cells incubated with camptothecin and EX527 ? 20 ug. Lane 8 = Extract of Mcf7 cells incubated with camptothecin and nicotinamide ? 20 ug. Lane 9 = Extract of 293T cells incubated with vehicle ? 20 ug. Lane 10 = Extract of 293T cells incubated with camptothecin ? 20 ug. Lane 11 = Extract of 293T cells incubated with camptothecin and trichostatin A ? 20 ug. Lane 12 = Extract of 293T cells incubated with camptothecin and EX527 ? 20 ug. Lane 13 = Extract of 293T cells incubated with camptothecin and nicotinamide ? 20 ug

SDS PAGE performed under reducing conditions (100mM DTT ? Sample heated at 50°C). Primary : Lanes 1-13: Rabbit anti acetyl Lysine antibody (ab21623) at 1/500 dilution. Secondary : Lanes 1-13: Goat anti rabbit IgG(H&L)-IR680 at 1:10,000 (in green). Developed: Odyssey. Blocking: in 5% Milk + PBS for 3 hours at RT. Primary antibody: in 5% BSA + 50mM Tris pH 7.5 + 150 mM NaCl + 0.05% Tween-20. Secondary antibody: in 5% Milk + PBS + 0.1% Tween-20 + 0.01%SDS for 2 hour at RT. Predicted band size : multiple. Observed band size : multiple



All lanes : Anti-acetyl Lysine antibody - ChIP Grade (ab21623) at 0.5 µg/ml

Lane 1 : Untreated Human melanoma cell lysate

Lane 2 : TSA-treated Human melanoma cell lysate

Lysates/proteins at 75 µg per lane.

Secondary

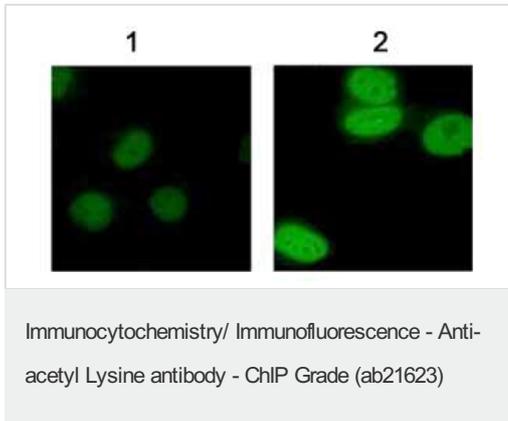
All lanes : Goat anti-rabbit IgG HRP at 0.25 µg/ml

Developed using the ECL technique.

Observed band size: 16-18 kDa

[why is the actual band size different from the predicted?](#)

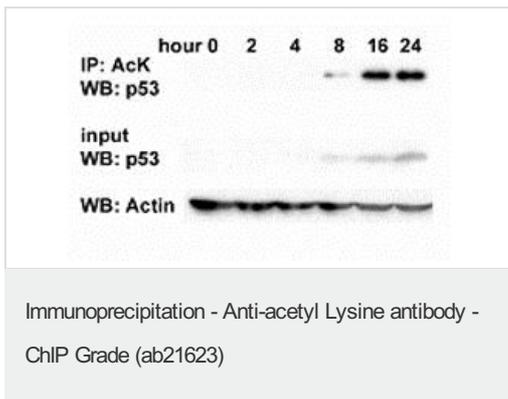
Additional bands at: 12-14 kDa. We are unsure as to the identity of these extra bands.



Immunofluorescent staining of Human melanoma cells, using Rabbit polyclonal to acetyl Lysine (ab21623) at 1:100 dilution.

1: Untreated cells

2: TSA treated cells



p53 acetylation upon Doxorubicin treatment in human melanoma cells (MMRU cells). MMRU cells were treated with 0.5 ug/ml Dox for various times and lysed for whole protein. Immunoprecipitation was performed with ab21623. Western blot was performed to detect the immunoprecipitated p53 with anti-p53 antibody.

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