abcam

Product datasheet

Anti-acetyl Lysine antibody - ChIP Grade ab21623

★★★★★ 10 Abreviews 61 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 Chemical/ Small Molecule corresponding to acetyl Lysine conjugated to keyhole limpet

haemocyanin. Acetylated KLH conjugates.

General notesUse to detect acetylation of lysine.

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.

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

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 pH: 6.00

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 notesUse to detect acetylation of lysine.

Clonality Polyclonal

1

Isotype IgG

Applications

The Abpromise guarantee

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.

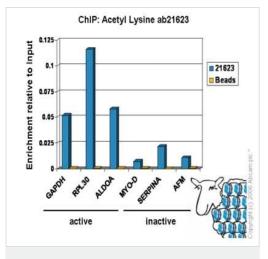
Application	Abreviews	Notes
IP	**** <u>(1)</u>	Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. PubMed: 24160175
ChIP	★★★★☆ (1)	Use 2-4 µg for 25 µg of chromatin.
ICC/IF	**** (1)	1/100 - 1/250. Subconfluent MMRU cells seeded in a 6-well plate with or without HDAC 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 antiacetylated lysine antibody at 1:100 dilution with PBSt 1 % BSA for 1 hour at room temperature.
WB	★★★★ (7)	1/1000 - 1/2500.
ELISA		1/5000.

Target

Relevance

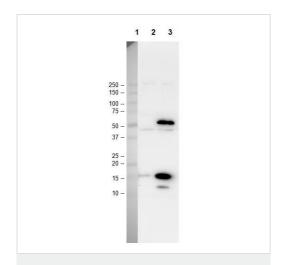
In the nucleus, DNA is tightly packed into nucleosomes generating an environment which is highly repressive towards DNA processes such as transcription. Acetylation of lysine residues within proteins has emerged as an important mechanism used by cells to overcome this repression. The acetylation of non-histone 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 nonhistone 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



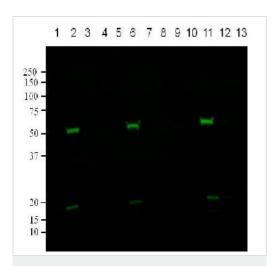
ChIP - Anti-acetyl Lysine antibody - ChIP Grade (ab21623)

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 lgG 1:10000. Performed under reducing conditions. Blocking buffer: 5% milk in PBS. Observed band sizes: 11 kDa 15kDa 45kDa 50 kDa.



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

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 camptothecin 20 ug. Lane 10 = 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 lgG(H&L)-IR680 at 1:10,000 (in green). Developed: Oddysey. 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

1 2 16-18kDa 12-14kDa

Western blot - Anti-acetyl Lysine antibody - ChIP Grade (ab21623) **All lanes :** Anti-acetyl Lysine antibody - ChIP Grade (ab21623) at $0.5 \ \mu \text{g/ml}$

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 lgG HRP at 0.25 µg/ml

Developed using the ECL technique.

Observed band size: 16-18 kDa

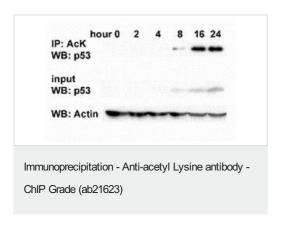
Additional bands at: 12-14 kDa. We are unsure as to the identity

of these extra bands.

Immunocytochemistry/ Immunofluorescence - Antiacetyl Lysine antibody - ChIP Grade (ab21623) 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 lyzed 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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