# abcam

## Product datasheet

## Anti-acetyl Lysine antibody [1C6] ab22550

## ★★★★★ <u>5 Abreviews</u> <u>15 References</u> 4 Images

#### Overview

Product name Anti-acetyl Lysine antibody [1C6]

**Description** Mouse monoclonal [1C6] to acetyl Lysine

Host species Mouse

**Specificity** ab22550 recognises proteins with acetylated lysine.

Tested applications Suitable for: ChIP, ICC/IF, WB

Species reactivity Reacts with: Species independent

Immunogen Synthetic peptide: sequence surrounding the acetylated lysine 9 of histone H3

Positive control ChIP: HeLa cell lysate. ICC: MCF7 cell line. WB: HeLa, COS7 and C2C12 cell lysates.

General notes

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 or -

80°C. Avoid freeze / thaw cycle.

**Storage buffer** Preservative: 0.05% Sodium azide

Constituents: PBS, 0.1% BSA

Purity Protein G purified

**Clonality** Monoclonal

Clone number 1C6
Isotype IgG

#### **Applications**

## The Abpromise guarantee

Our Abpromise guarantee covers the use of ab22550 in the following tested applications.

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

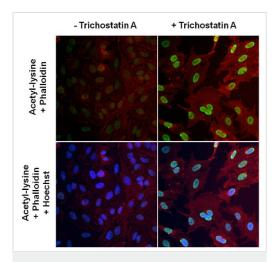
Application	Abreviews	Notes
ChIP	<b>★★★★</b> <u>(1)</u>	Use a concentration of 10 µg/ml.
ICC/IF		1/100 - 1/500.
WB	★★★★☆ (3)	1/500 - 1/2000.

### **Target**

#### Relevance

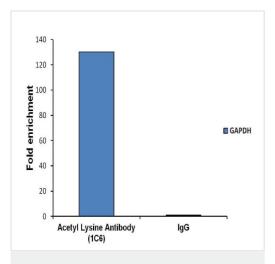
In the nucleus, DNA is tightly packed into nucleosomes generating an environment which is highly repressive towards DNA processes such as transcription. Acetylation of lysine residues within proteins has emerged as an important mechanism used by cells to overcome this repression. The acetylation of non-histone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



Immunocytochemistry/ Immunofluorescence - Antiacetyl Lysine antibody [1C6] (ab22550)

Immunoflouroescence analysis of HeLa Cells labelling lysine acetylated proteins with ab22550. Formalin fixed cells were permeabalized with 0.1& Triton X-100 in TBS for 10 mins at room temperature and subsequently blocked with BSA at room temperature for 15 mins. Cells were then probed with ab22550 at 1/100 for 1 hour at room temperature. The secondary used was a DyLight® 488 goat anti-mouse used at 1/400 for 30 minutes at room temperature. Additional counterstains used were F-actin with a DyLight® 554 Phalloidin and Neuclei stained using a Hoechst 33342 conjugate. Image was taken at X20 magnification.

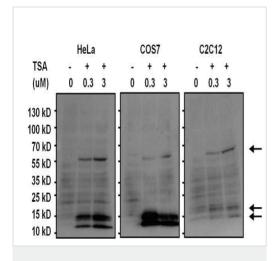


ChIP - Anti-acetyl Lysine antibody [1C6] (ab22550)

Chromatin Co-Immunoprecipitation (ChIP) analysis using ab22550 binding acetylated lysines in 10E+06 LNCaP cells. Protein binding was detected using real-time PCR.

Positive control: Fold enrichment of ab22550.

Negative Control: Non-specific lgG.



Western blot - Anti-acetyl Lysine antibody [1C6] (ab22550)

**All lanes :** Anti-acetyl Lysine antibody [1C6] (ab22550) at 1/1000 dilution

Lane 1: HeLa (untreated) cell lysate

Lane 2: HeLa (treated with 0.3 uM TSA, 16h) cell lysate

Lane 3: HeLa (treated with 3 uM TSA, 16h) cell lysate

Lane 4: COS7 (untreated) cell lysate

Lane 5: COS7 (treated with 0.3 uM TSA, 16h) cell lysate

Lane 6: COS7 (treated with 3 uM TSA, 16h) cell lysate

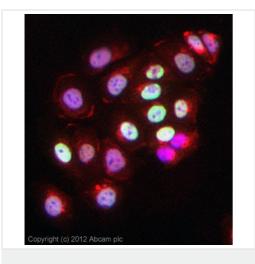
Lane 7: C2C12 (untreated) cell lysate

Lane 8: C2C12 (treated with 0.3 uM TSA, 16h) cell lysate

Lane 9: C2C12 (treated with 3 uM TSA, 16h) cell lysate

Lysates/proteins at 50 µg per lane.

Western blot analysis of lysine acetylated proteins from cells left untreated (DMSO only) or cells treated with 0.3uM or 3uM of Trichostatin A (TSA) for 16 hours was performed by loading 50 µg of the indicated whole cell lysates per well and 10 µL of PageRuler Prestained Protein Ladder onto a 4-20% Tris-HCl polyacrylamide gel. Proteins were transferred to a PVDF membrane and blocked with 5% BSA/TBST for at least 1 hour. The membrane was probed with an Acetyl Lysine monoclonal antibody at a dilution of 1:1000 overnight at 4°C on a rocking platform, washed in TBS-0.1%Tween-20, and probed with a goat anti-mouse IgG-HRP secondary antibody at a dilution of 1:20,000 for 1 hour. Chemiluminescent detection was performed using SuperSignal West Pico.



Immunocytochemistry/ Immunofluorescence - Antiacetyl Lysine antibody [1C6] (ab22550)

ICC/IF image of ab22550 stained MCF7 cells. The cells were 4% formaldehye fixed (10 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 (ab22550, 5μg/ml) overnight at +4°C. The secondary antibody (green) was ab96879, DyLight® 488 goat anti-mouse lgG (H+L) used at a 1/250 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.

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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- Replacement or refund for products not performing as stated on the datasheet
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- We investigate all quality concerns to ensure our products perform to the highest standards

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