abcam

Product datasheet

Anti-Bmi1 antibody [F6] - ChIP Grade ab14389

敲除验证

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概述

产品名称	Anti-Bmi1抗体[F6] - ChIP Grade
描述	**小鼠单克隆抗体**[F6] to Bmi1 - ChIP Grade
特异性	Recognizes mouse Bmi-1(triplet). The clone number has been updated from (1.T.21) to (F6) both clone numbers name the same antibody clone.
经测试应用	**适用于:** ICC/IF, Flow Cyt, ChIP, WB, ICC, IP, IHC-Fr, IHC-P
种属反应性	**与反应:** Mouse, Rat, Rabbit, Cat, Human, Xenopus laevis, Zebrafish
免疫原	Recombinant fragment, corresponding to amino acids 1-202 of Mouse Bmi1
阳性对照	U2OS whole cell lysate.

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
存储溶液	pH: 7.40 Preservative: 0.05% Sodium azide Constituents: 0.276% Tris glycine, 0.87% Sodium chloride
纯度	Protein G purified
克隆	单克隆
克隆编号	F6
同种型	IgG1

应用

Our Abpromise guarantee covers the use of **ab14389** in the following tested applications.

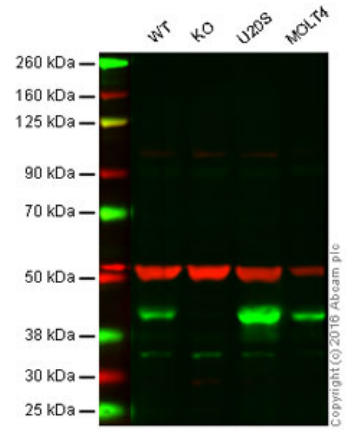
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

应用	Ab评论	说明
ICC/IF	★★★★★	Use a concentration of 0.65 µg/ml. PubMed: 19001505
Flow Cyt		Use a concentration of 0.65 µg/ml. PubMed: 19001505 ab170190 - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.
ChIP		Use at an assay dependent concentration. PubMed: 18332116
WB	★★★★☆	Use a concentration of 0.2 - 2 µg/ml. Predicted molecular weight: 37 kDa. Detects Bmi-1 in RIPA lysates from U2OS cells. U2OS cell lysate was resolved by electrophoresis, transferred to nitrocellulose and probed with ab14389 0.2ug/ml. Proteins were visualized using a goat anti-mouse IgG labeled with HRP and a chemiluminescence detection system.
ICC		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
IHC-Fr	★★★★★	Use at an assay dependent concentration. PubMed: 17210912
IHC-P	★★★★★	1/100.

靶标

功能	Component of the Polycomb group (PcG) multiprotein PRC1 complex, a complex required to maintain the transcriptionally repressive state of many genes, including Hox genes, throughout development. PcG PRC1 complex acts via chromatin remodeling and modification of histones; it mediates monoubiquitination of histone H2A 'Lys-119', rendering chromatin heritably changed in its expressibility. In the PRC1 complex, it is required to stimulate the E3 ubiquitin-protein ligase activity of RNF2/RING2.
序列相似性	Contains 1 RING-type zinc finger.
翻译后修饰	Monoubiquitinated (By similarity). May be polyubiquitinated; which does not lead to proteasomal degradation.
细胞定位	Nucleus. Cytoplasm.

图片

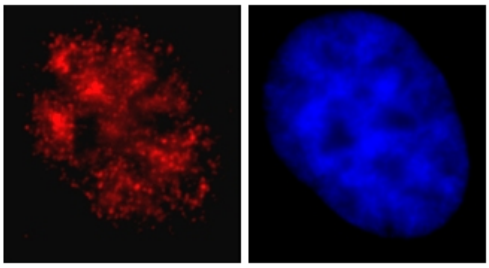


Western blot - Anti-Bmi1 antibody [1.T.21] - ChIP Grade (ab14389)

Predicted band size : 37 kDa

Lane 1: Wild-type HAP1 cell lysate (20 µg)
Lane 2: Bmi1 knockout HAP1 cell lysate)
Lane 3: U2OS cell lysate (40 ug)
Lane 4: Molt-4 cell lysate (40ug)
Lanes 1 - 4: Merged signal (red and green).
Green - ab14389 observed at 42 kDa. Red - loading control, ab176560, observed at 52 kDa.

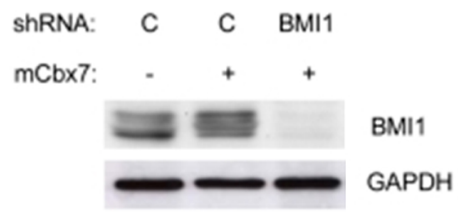
ab14389 was shown to recognize Bmi1 when Bmi1 knockout samples were used, along with additional cross-reactive bands. Wild-type and Bmi1 knockout samples were subjected to SDS-PAGE. ab14389 at a concentration of 1 µg/ml and ab176560 (loading control to alpha tubulin) diluted to 1/10000 were incubated overnight at 4°C. Blots were developed with Goat anti-Mouse IgG H&L (IRDye® 800CW) preadsorbed (ab216772) and Goat Anti-Rabbit IgG H&L (IRDye® 680RD) preadsorbed (ab216777) secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry - Anti-Bmi1 antibody [1.T.21] - ChIP Grade (ab14389)

This image is courtesy of Darin McDonald, Cross Cancer Institute

SK-N-SH cells were fixed in 4% paraformaldehyde, permeabilized in 0.55 Triton-X100 and incubated for 1 hour with ab14389 (1/200). The Bmi1 staining is shown in red. The cells were counterstained with DAPI (blue). 100x magnification.



Western blot - Anti-Bmi1 antibody [1.T.21] - ChIP Grade (ab14389)

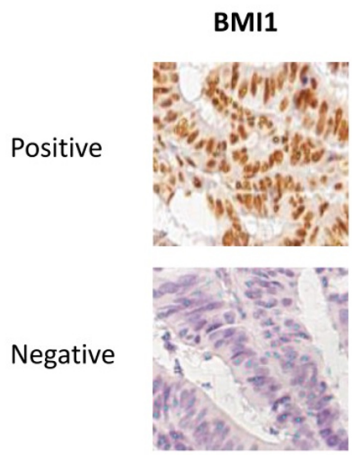
Image from PLoS One. 2009; 4(7): e6380. Fig 4D, doi: 10.1371/journal.pone.0006380

Predicted band size : 37 kDa

Image from PLoS One. 2009; 4(7): e6380. Fig 4D, doi: 10.1371/journal.pone.0006380

Cells were lysed in RIPA buffer. Samples (25 µg) of total protein were separated by SDS-polyacrylamide gel electrophoresis gel (PAGE) in a 12% gel and transferred onto a nitrocellulose membrane. This was followed by an incubation with mouse anti-BMI1 (ab14389) and a sheep anti-mouse HRP conjugated antibody diluted 1:2000 was used as a secondary antibody. HRP conjugated anti-GAPDH antibody (ab9482) was used as a loading control.

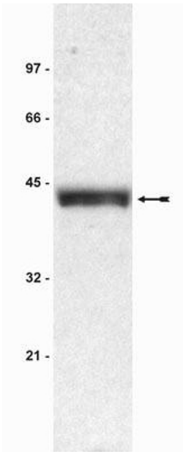
The mCbx7 expressing cells were infected with lentivirus-based shRNA vectors targeting BMI1 or an irrelevant control (C).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bmi1 antibody [1.T.21] - ChIP Grade (ab14389)

Image from PLoS One. 2014; 9(9): e108265. Fig 2, doi: 10.1371/journal.pone.0108265

Formalin-fixed paraffin-embedded (FFPE) colorectal cancer tissues and normal colorectal tissues. Endogenous peroxidase was blocked by incubating the sections in a 0.3% solution of hydrogen peroxide (in PBS) for 20 min. Antigen retrieval was performed by heating the sections for 10 min at 95°C in a citrate buffer. Tissues were incubated with ab14389 overnight (16 hrs). Staining was visualized using the Dako REAL EnVision Detection System, Peroxidase/DAB+, Rabbit/Mouse (Dako). The stained TMA sections were scanned using a 20x magnification on the semi-automated Ariol system (Leica Microsystems, Wetzlar, Germany). Tumor cell areas (tumor tissues) and colon epithelium (in normal tissues) were identified for positive (indicated by yellow dots) and negative (blue dots) nuclei in tumor cores. TMA slides were scanned using a 20x magnification. Shown are positively stained tumor cores (*top image*) and negative tumor cores (*bottom image*).



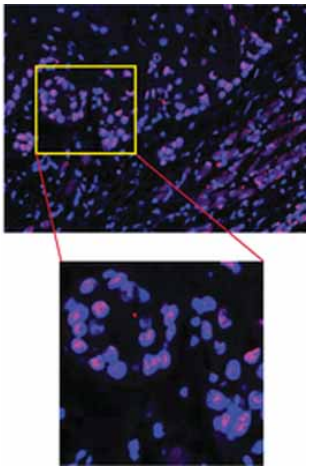
Western blot - Anti-Bmi1 antibody [1.T.21] - ChIP Grade (ab14389)

Developed using the ECL technique

Performed under reducing conditions.

Predicted band size : 37 kDa

Western blot analysis of RIPA lysates from U2OS cells labelling Bmi-1 with ab14389 at 0.2µg . An IgG (HRP) goat anti-mouse was used as the secondary antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bmi1 antibody [1.T.21] - ChIP Grade (ab14389)

Image from Ganguli-Indra G et al, PLoS One. 2009;4(4):e5367. Epub 2009 Apr 28, Fig 2.

ab14389 staining Bmi1 in human head and neck squamous cell cancers (HNSCC) by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections).
Sections were deparaffinized in xylene, dehydrated through graded alcohols, and placed in 0.1% hydrogen peroxide to quench any endogenous peroxidase activity. A 5 minute, 750 W microwave pretreatment in citrate buffer (pH 6.0) was repeated 4 times and followed by treatment with 10% normal rabbit serum for 30 minutes to block nonspecific antibody binding. The slides were then incubated with ab14389 at a 1/100. Primary antibody incubation was followed by three washes with PBST and incubation with fluorescently-labeled Cy2 (1/250) secondary antibody for 2 hours. Nuclei were counterstained with DAPI. Finally, sections were rinsed with PBST, dehydrated through sequential washes in 50%, 70%, 95%, and 100% ethanol and then cleared in xylene. Slides were mounted with DPX mounting media and allowed to dry overnight.

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