abcam

Product datasheet

Anti-beta Actin antibody [mAbcam 8224] - Loading Control ab8224

★★★★★ 35 Abreviews 472 References 10 图像

概述

产品名称	Anti-beta Actin抗体[mAbcam 8224] - Loading Control
描述	小鼠单克隆抗体[mAbcam 8224] to beta Actin - Loading Control
宿主	Mouse
特异性	Recognises a single band at 42kD representing beta Actin. The immunogen used for this product shares 77% homology with Gamma actin/actin cytoplasmic 2. Cross-reactivity with this protein has not been confirmed experimentally.
经测试应用	**适用于:** WB, IHC-P, ICC/IF **不适用于:** Flow Cyt (Intra)
种属反应性	**与反应:** Mouse, Rat, Human, Xenopus laevis, Drosophila melanogaster, Schizosaccharomyces pombe **预测可用于:** Rabbit, Chicken, Cow, Cat, Dog, Pig, Chinese hamster, Other species
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers. (Peptide available as **ab13772**)
阳性对照	WB: A431; HEK293; NIH3T3; PC12 whole cell lysates; Xenopus embryo lysate; Drosophila lysate; S. pombe lysate. ICC/IF: HeLa cells. IHC/P: Human colon (FFPE)
常规说明	This monoclonal antibody to beta actin works well as a protein loading control in Western blot for a broad range of species including Xenopus, Drosophila and S. pombe. This antibody clone [mAbcam 8224] is manufactured by Abcam. If you require this antibody in a particular buffer formulation or a particular conjugate for your experiments, please contact **orders@abcam.com** or you can find further information **here**. 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

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Store at -20°C or -80°C. Avoid freeze / thaw cycle.
存储溶液	pH: 7.40 Preservative: 0.02% Sodium azide Constituents: PBS, 6.97% L-Arginine
纯度	Protein G purified
Primary antibody说明	This clone works well as a loading control for Xenopus, Drosophila, S. cerevisiae and S.pombe. We recommend using ab8224 instead of **ab8226** for these species.
克隆	单克隆
克隆编号	mAbcam 8224
骨髓瘤	Sp2/0-Ag14
同种型	IgG1
轻链类型	kappa

应用

The Abpromise guarantee **Abpromise™承诺保证使用ab8224于以下的经测试应用**

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB	★★★★★ (22)	Use a concentration of 1 µg/ml. Detects a band of approximately 42 kDa (predicted molecular weight: 42 kDa).Can be blocked with **Human beta Actin peptide (ab13772)**. This antibody has been designed for use as a loading control and is ideal for this purpose. Block membrane for 1 hr in 5%BSA. Incubate antibody in TBST for one hour or more.
IHC-P	★★★★★ (7)	Use a concentration of 1 µg/ml. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
ICC/IF	★★★★★ (6)	Use a concentration of 1 µg/ml.

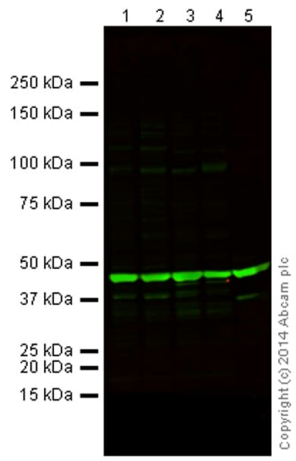
应用说明 Is unsuitable for Flow Cyt (Intra).

靶标

功能	Actins are highly conserved proteins that are involved in various types of cell motility and are ubiquitously expressed in all eukaryotic cells.
疾病相关	Defects in ACTB are a cause of dystonia juvenile-onset (DYTJ) [MIM:607371]. DYTJ is a form of dystonia with juvenile onset. Dystonia is defined by the presence of sustained involuntary muscle contraction, often leading to abnormal postures. DYTJ patients manifest progressive, generalized, dopa-unresponsive dystonia, developmental malformations and sensory hearing loss.
序列相似性	Belongs to the actin family.
翻译后修饰	ISGylated.

细胞定位	Cytoplasm > cytoskeleton. Localized in cytoplasmic mRNP granules containing untranslated mRNAs.

图片



Western blot - Anti-beta Actin antibody [mAbcam 8224] - Loading Control (ab8224)

All lanes : Anti-beta Actin antibody [mAbcam 8224] - Loading Control (ab8224) at 1 µg/ml

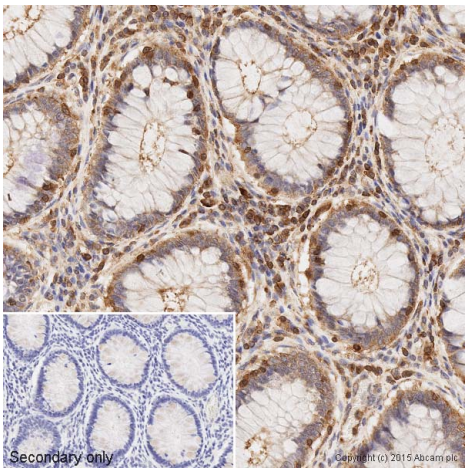
Lane 1 : A431 (Human epithelial carcinoma cell line) Whole Cell Lysate
Lane 2 : HEK293 (Human embryonic kidney cell line) Whole Cell Lysate
Lane 3 : NIH 3T3 (Mouse embryonic fibroblast cell line) Whole Cell Lysate
Lane 4 : PC12 (Rat adrenal pheochromocytoma cell line) Whole Cell Lysate
Lane 5 : Skeletal Muscle (Human) Tissue Lysate - adult normal tissue

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Mouse IgG H&L (Alexa Fluor® 790) (**ab175783**) at 1/10000 dilution

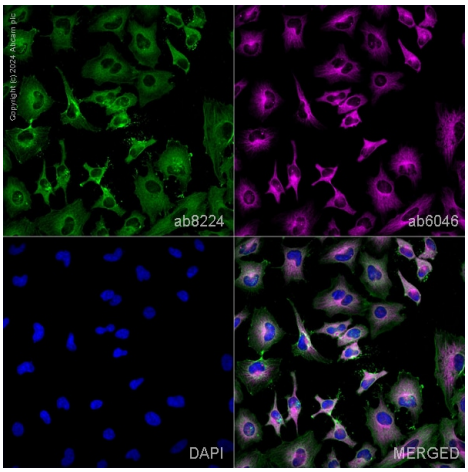
Predicted band size: 42 kDa
Observed band size: 42 kDa

This blot was produced using a 4-12% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 50 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 5% Milk before being incubated with ab8224 overnight at 4°C. Antibody binding was detected using a goat **anti-mouse Alexa Fluor 790** (**ab175783**) at a 1:10,000 dilution for 1hr at room temperature and then imaged using the Licor Odyssey CLx.



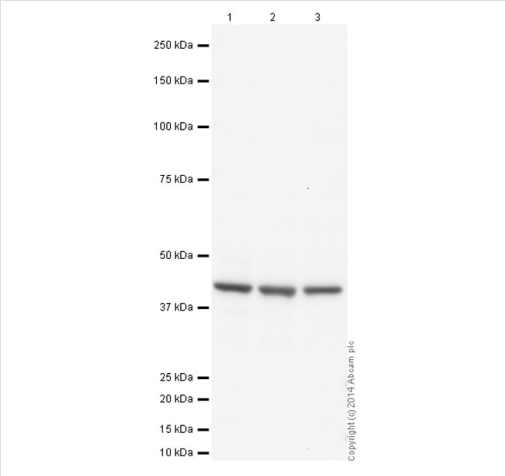
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Actin antibody [mAbcam 8224] - Loading Control (ab8224)

IHC image of ab8224 staining beta Actin in human colon formalin fixed paraffin embedded tissue sections*, performed on a Leica Bond. 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 ab8224, 1µg/ml working concentration, 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. No primary antibody was used in the secondary only control (shown on the inset).
For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.
*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre



Immunocytochemistry/ Immunofluorescence - Anti-beta Actin antibody [mAbcam 8224] - Loading Control (ab8224)

ab8224 staining beta Actin in HeLa cells. The cells were fixed with 100% methanol (5 min), permeabilized with 0.1% PBS-Tween for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1%PBS-Tween for 1h. The cells were then incubated overnight at 4°C with ab8224 at 1µg/ml and **ab6046**, Rabbit polyclonal to beta Tubulin - Loading Control. Cells were then incubated with **ab150117**, Goat polyclonal Secondary Antibody to Mouse IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (shown in green) and **ab150080**, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 594) at 1/1000 dilution (shown in pseudocolour magenta). Nuclear DNA was labelled with DAPI (shown in blue). Image was acquired with a high-content analyser (Operetta CLS, Perkin Elmer) and a maximum intensity projection of confocal sections is shown.



Western blot - Anti-beta Actin antibody [mAbcam 8224] - Loading Control (ab8224)

All lanes : Anti-beta Actin antibody [mAbcam 8224] - Loading Control (ab8224) at 1 µg/ml

Lane 1 : HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate
Lane 2 : NIH 3T3 (Mouse embryonic fibroblast cell line) Whole Cell Lysate
Lane 3 : PC12 (Rat adrenal pheochromocytoma cell line) Whole Cell Lysate

Lysates/proteins at 10 µg per lane.

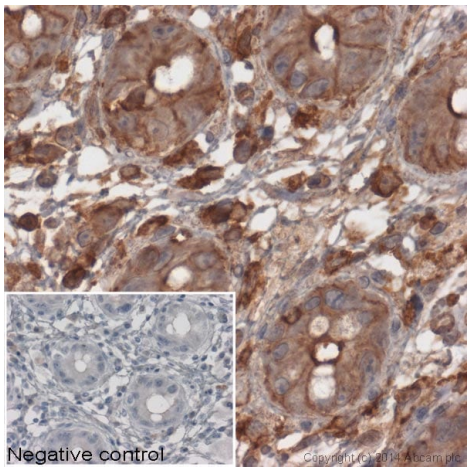
Secondary
All lanes : Goat Anti-Mouse IgG H&L (HRP) preadsorbed (**ab97040**) at 1/50000 dilution

Performed under reducing conditions.

Predicted band size: 42 kDa
Observed band size: 42 kDa

Exposure time: 3 minutes

This blot was produced using a 4-12% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 50 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 2% Bovine Serum Albumin before being incubated with ab8224 overnight at 4°C. Antibody binding was detected using an anti-mouse antibody conjugated to HRP, and visualised using ECL development solution **ab133406**

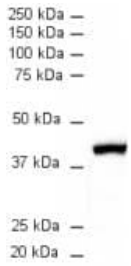


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Actin antibody [mAbcam 8224] - Loading Control (ab8224)

IHC image of beta actin staining in human colon formalin fixed paraffin embedded tissue section*. The section was pre-treated using pressure cooker heat mediated antigen retrieval with sodium citrate buffer (pH6) for 30mins. The section was incubated with ab8224, 3µg/ml overnight at +4°C. A goat **anti-mouse HRP**-conjugated secondary antibody (**ab6789**, 1/2000 dilution) was used for 1hr at room temperature. The section was counterstained with haematoxylin and mounted with DPX.

The inset negative control image is secondary-only at 1/500 dilution.

*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre



Western blot - Anti-beta Actin antibody [mAbcam 8224] - Loading Control (ab8224)

Anti-beta Actin antibody [mAbcam 8224] - Loading Control (ab8224) + Xenopus embryo lysate at 20 µg

Secondary
Rabbit Anti-Mouse IgG H&L (HRP) (**ab6728**)

Developed using the ECL technique.

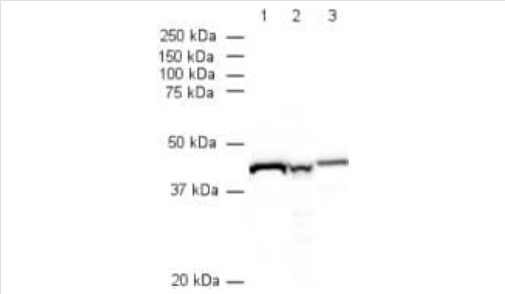
Performed under reducing conditions.

Predicted band size: 42 kDa

ab8224 used on Xenopus embryo lysate (20 ug of lysate/lane).

Secondary
Rabbit polyclonal **anti-mouse HRP** was used as the secondary antibody (**ab6728**) and developed using the ECL technique.
Performed under reducing conditions.

Predicted band size : 42kD



Western blot - Anti-beta Actin antibody [mAbcam 8224] - Loading Control (ab8224)

All lanes : Anti-beta Actin antibody [mAbcam 8224] - Loading Control (ab8224) at 1 µg/ml

Lane 1 : Drosophila lysate
Lane 2 : S. pombe lysate
Lane 3 : S. cerevisiae lysate (Actin 1 - please see note)

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Rabbit Anti-Mouse IgG H&L (HRP) (**ab6728**) at 1/5000 dilution

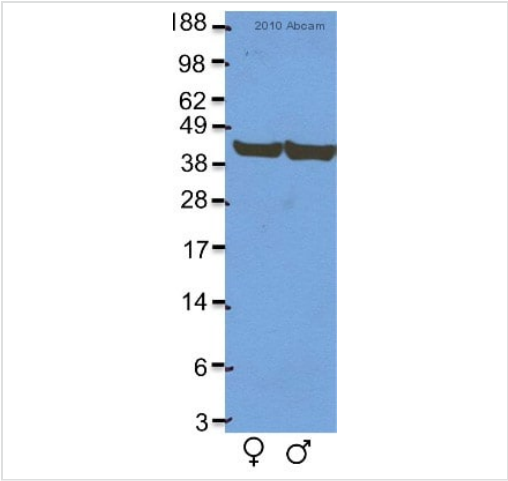
Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 42 kDa
Observed band size: 42 kDa

Note: although *S. cerevisiae* is not known to express beta Actin, Abcam believes that the band on lane 3 corresponds to Actin 1 (Swissprot ID: P60010, based on sequence similarity).

Secondary antibody - rabbit **anti-mouse HRP** (**ab6728**)



Western blot - Anti-beta Actin antibody [mAbcam 8224] - Loading Control (ab8224)

This image is courtesy of an anonymous Abreview

All lanes : Anti-beta Actin antibody [mAbcam 8224] - Loading Control (ab8224) at 1/1000 dilution

Lane 1 : Fruit fly (Drosophila melanogastor) whole cell lysate - Female
Lane 2 : Fruit fly (Drosophila melanogastor) whole cell lysate - Male

Lysates/proteins at 100 µg per lane.

Secondary
All lanes : An HRP-conjugated Sheep polyclonal to mouse IgG at 1/10000 dilution

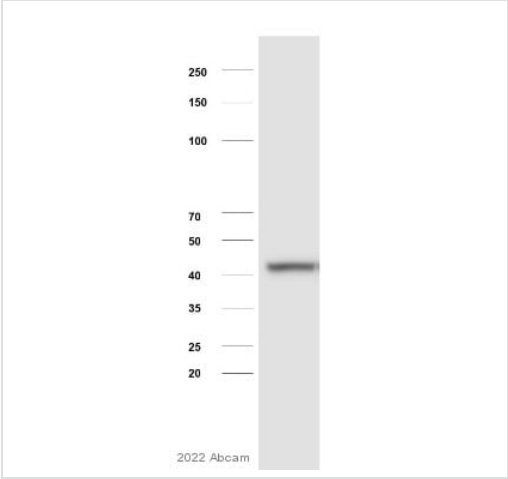
Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 42 kDa

Exposure time: 2 minutes

Blocking step: 5% Milk for 1 hour at 20°C.



Western blot - Anti-beta Actin antibody [mAbcam 8224] - Loading Control (ab8224)

This image is courtesy of an Abreview submitted by Joel Ohana

Anti-beta Actin antibody [mAbcam 8224] - Loading Control (ab8224) at 1/10000 dilution + Mouse CT26 cells at 40 µg

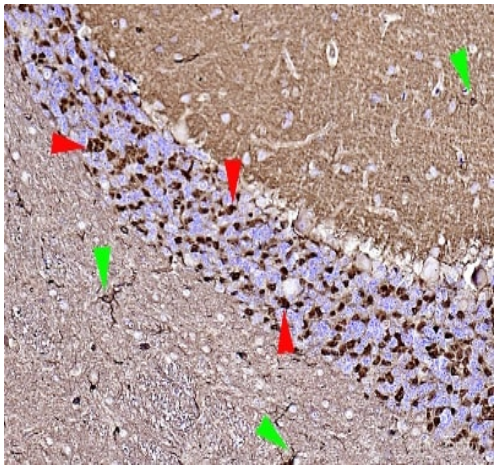
Secondary
Anti-mouse IgG, HRP-linked Antibody at 1/10000 dilution

Developed using the ECL technique.

Predicted band size: 42 kDa
Observed band size: 42 kDa

Exposure time: 6 minutes

Western blot analysis using ab8224 at 1:1000 on Mouse CT26 cells. Blocking agent and dilution buffer was 5% milk in TBST



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Actin antibody [mAbcam 8224] - Loading Control (ab8224)

This image is courtesy of an abreview submitted by Carl Hobbs, King's College London, United Kingdom

Immunohistochemistical detection of beta Actin using antibody [mAbcam 8224] - Loading Control on formaldehyde-fixed paraffin-embedded rat cerebellum sections. Antigen retrieval step: heat mediated Citric acid pH6 buffer. Permeabilization: No. Blocking step: 1% BSA for 10 mins @ rt°C. Primary antibody dilution 1/1000 for 2 hours in TBS/BSA/azide. Secondary Antibody: anti Mouse Igs conjugated to biotin (1/200). beta Actin appears to be particularly enriched not only in the glomeruli of the Granule cell layer (indicated by red arrowheads) but also in Microglia (indicated by green arrowheads); All positive microglia appear to be ramified thus not presumed to be activated.

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