

Product datasheet

Anti-beta Actin antibody [AC-15] ab6276

KO VALIDATED

★★★★★ [94 Abreviews](#) [2211 References](#) [6 Images](#)

Overview

Product name	Anti-beta Actin antibody [AC-15]
Description	Mouse monoclonal [AC-15] to beta Actin
Host species	Mouse
Tested applications	Suitable for: ICC/IF, WB
Species reactivity	Reacts with: Mouse, Rat, Cow, Dog, Human, African green monkey, Chinese hamster Does not react with: Drosophila melanogaster, Dictyostelium discoideum
Immunogen	Synthetic peptide corresponding to beta Actin aa 1-100 (N terminal) conjugated to keyhole limpet haemocyanin. Slightly modified β -cytoplasmic actin N-terminal peptide, Ac-Asp-Asp-Asp-Ile-Ala-Ala-Leu-Val-Ile-Asp-Asn-Gly-Ser-Gly-Lys, conjugated to KLH. Run BLAST with Expasy Run BLAST with NCBI
Epitope	N-terminal of the beta isoform of actin.
Positive control	ICC/IF: SV40LT-SMC cells WB: HAP1, HeLa, Jurkat, A431, HEK-293, COS-7, NIH/3T3, PC-12, Rat2, CHO, MDBK and MDCK cell lysates.
General notes	<p>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.</p> <p>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</p>

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 long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.40 Preservative: 0.097% Sodium azide Constituent: PBS
Purity	Affinity purified

Purification notes	Purified from hybridoma cell culture.
Clonality	Monoclonal
Clone number	AC-15
Isotype	IgG1

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab6276 in the following tested applications.

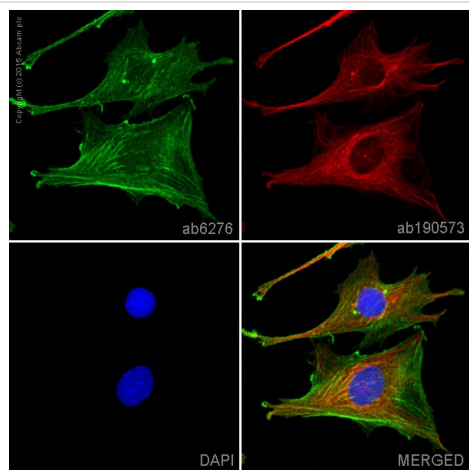
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
ICC/IF	★★★★★ (9)	Use a concentration of 5 µg/ml.
WB	★★★★★ (76)	1/5000 - 1/16000. Predicted molecular weight: 42 kDa.

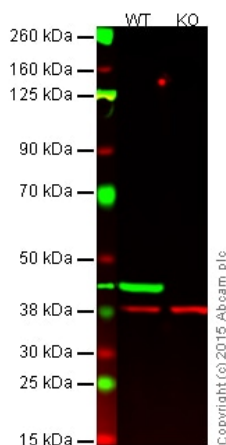
Target

Function	Actins are highly conserved proteins that are involved in various types of cell motility and are ubiquitously expressed in all eukaryotic cells.
Involvement in disease	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.
Sequence similarities	Belongs to the actin family.
Post-translational modifications	ISGylated.
Cellular localization	Cytoplasm > cytoskeleton. Localized in cytoplasmic mRNP granules containing untranslated mRNAs.

Images



Immunocytochemistry/ Immunofluorescence - Anti-beta Actin antibody [AC-15] (ab6276)



Western blot - Anti-beta Actin antibody [AC-15] (ab6276)

ab6276 staining beta Actin in SV40LT-SMC cells. The cells were fixed with 4% formaldehyde (10 min), permeabilized in 0.1% Triton X-100 for 5 minutes and then blocked in 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab6276 at a working concentration of 5µg/ml and **ab190573**, Rabbit monoclonal [EP1332Y] to alpha Tubulin (Alexa Fluor® 647, shown in red) at 1/250 overnight at +4°C, followed by a further incubation at room temperature for 1h with an **anti-mouse AlexaFluor® 488 (ab150117)** at 2 µg/ml (shown in green). Nuclear DNA was labelled in blue with DAPI.

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

All lanes : Anti-beta Actin antibody [AC-15] (ab6276) at 1/5000 dilution

Lane 1 : Wild-type HAP1 cell lysate (20 µg)

Lane 2 : Beta actin knockout HAP1 cell lysate (20 µg)

Secondary

All lanes : Goat anti-Mouse IgG H&L (IRDye® 800CW) preadsorbed (**ab216772**) at 1/10000 dilution

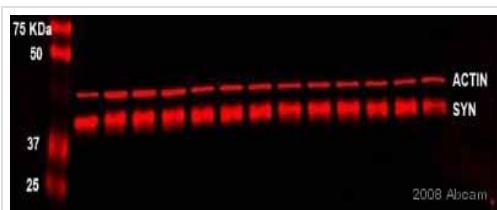
Predicted band size: 42 kDa

Lane 1: Wild-type HAP1 cell lysate (20 µg)

Lane 2: Beta actin knockout HAP1 cell lysate (20 µg)

Lanes 1 and 2: Merged signal (red and green). Green - beta actin, ab6276 observed at 42 kDa. Red - loading control, **ab181602** observed at 37 kDa.

Ab6276 was shown to specifically react with beta actin in wild-type HAP1 cells. No band was observed when beta actin knockout samples were used. Wild-type and beta actin knockout samples were subjected to SDS-PAGE. ab6276 (beta actin) and **ab181602** (loading control to GAPDH) were diluted 1/5000 and 1/10 000 and 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 1/10 000 dilution for 1 h at room temperature before imaging.

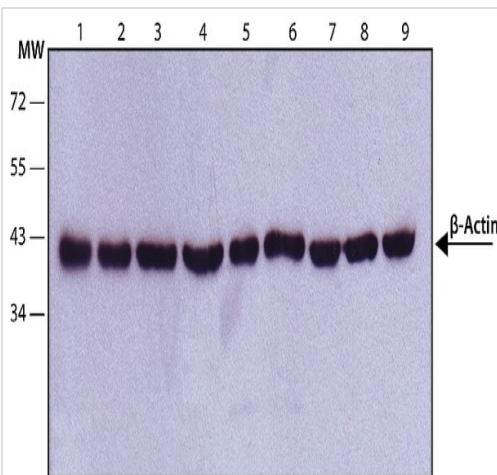


Western blot - Anti-beta Actin antibody [AC-15]
(ab6276)

This image is courtesy of an Abreview submitted by Dr Mark Elliott

Western Blot of ab6276 (used as loading control) with whole tissue lysate of human grey matter from BA20 (temporal cortex). Ab6276 was diluted 1/50000 and incubated with the sample for 16 hours at 4°C. 5 µg of lysate was loaded onto the gel, which was blocked with 5% milk for 1 hour at 20°C. An Alexa Fluor® 680 conjugated goat anti-mouse antibody, diluted 1/5000, was used as the secondary.

Bands below actin in image are synaptophysin (SYN).



Western blot - Anti-beta Actin antibody [AC-15]
(ab6276)

All lanes : Anti-beta Actin antibody [AC-15] (ab6276) at 1 µg/ml

Lane 1 : HeLa (human epithelial cell line from cervix adenocarcinoma) cell lysate

Lane 2 : Jurkat (human T cell leukemia cell line from peripheral blood) cell lysate

Lane 3 : COS-7 (african green monkey kidney fibroblast-like cell line) cell lysate

Lane 4 : NIH/3T3 (mouse embryonic fibroblast cell line) cell lysate

Lane 5 : PC-12 (rat adrenal gland pheochromocytoma cell line) cell lysate

Lane 6 : Rat2 (rat fibroblast cell line) cell lysate

Lane 7 : CHO (chinese hamster ovary cell line) cell lysate

Lane 8 : MDBK (bovine kidney cell line) cell lysate

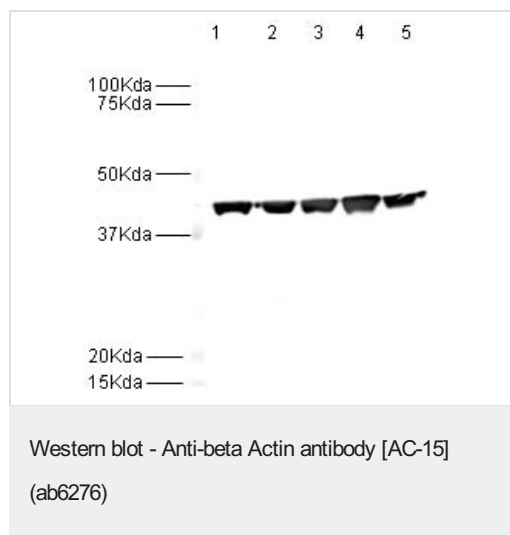
Lane 9 : MDCK (canine kidney cell line) cell lysate

Secondary

All lanes : Goat Anti-Mouse IgG-Peroxidase

Developed using the ECL technique.

Predicted band size: 42 kDa



All lanes : Anti-beta Actin antibody [AC-15] (ab6276) at 1/5000 dilution

Lane 1 : HeLa nuclear

Lane 2 : HeLa whole cell lysate

Lane 3 : A431 cell lysate

Lane 4 : Jurkat cell lysate

Lane 5 : HEK293 cell lysate

Lysates/proteins at 20 µg per lane.

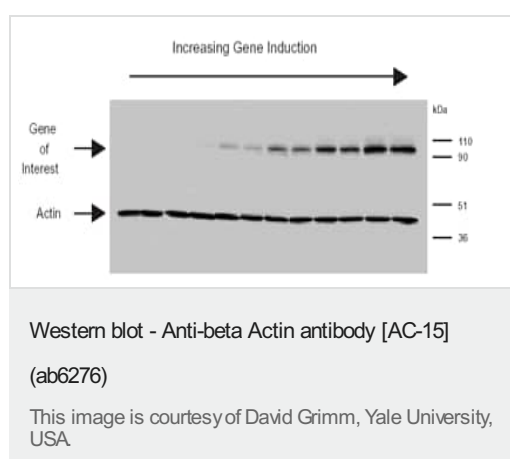
Secondary

All lanes : Alexa Fluor anti mouse at 1/5000 dilution

Performed under reducing conditions.

Predicted band size: 42 kDa

Observed band size: 42 kDa



MDCK cells induced with increasing amounts of doxycycline to control expression of the gene of interest. All cells were normalized for loading with an albumin protein standard assay. Anti-beta actin (ab6276) was used at a concentration of 1:5000 in a milk blocking solution. B-actin blotting confirms the albumin assay in showing that an equal amount of lysate was loaded in each lane.

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