abcam

ab8227

Anti-beta Actin antibody

Anti-beta Actin antibody ab8227 is a rabbit polyclonal antibody that is used in beta Actin western blotting, IHC and immunofluorescence. Suitable for human, mouse and rat samples.

- Tried and trusted by researchers since 2003
Same trusted quality, new lower price

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.4 Preservative: 0.02% Sodium azide Constituents: 98.98% PBS, 1% BSA
Form	Liquid
Clonality	Polyclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Purification technique	Affinity purification Immunogen
Specificity	From Jan 2024, QC testing of replenishment batches of this polyclonal changed. All tested and expected application and reactive species combinations are still covered by our Abcam product promise. However, we no longer test all applications. For more information on a specific batch, please contact our Scientific Support who will be happy to help. 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.
Concentration	0.3 - 1 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Human		
Dilution info	0.20000-1.00000 µg/mL		
Notes	We recommend Goat Anti-Rabbit IgG H&L (Biotin) (ab6720) secondary antibody or	Goat Anti-Rabbit IgG Fc (HRP) (ab97200) secondary antibody.	Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

Expected

Species	Mouse		
Dilution info	-		
Notes	-		

Species	Rat		
Dilution info	0.20000-1.00000 µg/mL		
Notes	We recommend Goat Anti-Rabbit IgG H&L (Biotin) (ab6720) secondary antibody or	Goat Anti-Rabbit IgG Fc (HRP) (ab97200) secondary antibody.	Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

Species	Chicken	
Dilution info	0.2-1 µg/mL	
Notes	We recommend Goat Anti-Rabbit IgG H&L (Biotin) (ab6720) secondary antibod or Goat Anti-Rabbit IgG Fc (HRP) (ab97200) secondary antibody.	Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

Species	Chinese hamster	
Dilution info	0.2-1 µg/mL	
Notes	We recommend Goat Anti-Rabbit IgG H&L (Biotin) (ab6720) secondary antibod or Goat Anti-Rabbit IgG Fc (HRP) (ab97200) secondary antibody.	Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

Species	Cow	
Dilution info	0.2-1 µg/mL	
Notes	We recommend Goat Anti-Rabbit IgG H&L (Biotin) (ab6720) secondary antibod or Goat Anti-Rabbit IgG Fc (HRP) (ab97200) secondary antibody.	Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

Species	Dog
Dilution info	0.2-1 µg/mL
Notes	We recommend Goat Anti-Rabbit IgG H&L (Biotin) (ab6720) secondary antibod or Goat Anti-Rabbit IgG Fc (HRP) (ab97200) secondary antibody. Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

Species	Fish
Dilution info	0.2-1 µg/mL
Notes	We recommend Goat Anti-Rabbit IgG H&L (Biotin) (ab6720) secondary antibod or Goat Anti-Rabbit IgG Fc (HRP) (ab97200) secondary antibody. Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

Species	Rabbit
Dilution info	0.2-1 µg/mL
Notes	We recommend Goat Anti-Rabbit IgG H&L (Biotin) (ab6720) secondary antibod or Goat Anti-Rabbit IgG Fc (HRP) (ab97200) secondary antibody. Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

Species	Xenopus laevis
Dilution info	0.2-1 µg/mL
Notes	We recommend Goat Anti-Rabbit IgG H&L (Biotin) (ab6720) secondary antibod or Goat Anti-Rabbit IgG Fc (HRP) (ab97200) secondary antibody. Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

Predicted

Species	Guinea pig, Pig, Drosophila melanogaster, Monkey, Zebrafish, Rhesus monkey, Sheep
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	1/1000.00000 - 1/5000.00000
Notes	We recommend Goat Anti-Rabbit IgG H&L (HRP) (ab6721) secondary antibody. A stronger signal is observed in chemiluminescent western blot when using 2% BSA

	as the blocking agent. Milk blocking is suitable for use with fluorescent detection systems.
Species	Mouse
Dilution info	1/1000.00000 - 1/5000.00000
Notes	We recommend Goat Anti-Rabbit IgG H&L (HRP) (ab6721) secondary antibody. A stronger signal is observed in chemiluminescent western blot when using 2% BSA as the blocking agent. Milk blocking is suitable for use with fluorescent detection systems.
Species	Rat
Dilution info	1/1000.00000 - 1/5000.00000
Notes	We recommend Goat Anti-Rabbit IgG H&L (HRP) (ab6721) secondary antibody. A stronger signal is observed in chemiluminescent western blot when using 2% BSA as the blocking agent. Milk blocking is suitable for use with fluorescent detection systems.

Expected

Species	Chinese hamster
Dilution info	1/1000.00000 - 1/5000.00000
Notes	We recommend Goat Anti-Rabbit IgG H&L (HRP) (ab6721) secondary antibody. A stronger signal is observed in chemiluminescent western blot when using 2% BSA as the blocking agent. Milk blocking is suitable for use with fluorescent detection systems.
Species	Fish
Dilution info	1/1000.00000 - 1/5000.00000
Notes	We recommend Goat Anti-Rabbit IgG H&L (HRP) (ab6721) secondary antibody. A stronger signal is observed in chemiluminescent western blot when using 2% BSA as the blocking agent. Milk blocking is suitable for use with fluorescent detection systems.
Species	Xenopus laevis
Dilution info	1/1000.00000 - 1/5000.00000
Notes	We recommend Goat Anti-Rabbit IgG H&L (HRP) (ab6721) secondary antibody. A stronger signal is observed in chemiluminescent western blot when using 2% BSA as the blocking agent. Milk blocking is suitable for use with fluorescent detection systems.
Species	Dog

Dilution info	1/1000.00000 - 1/5000.00000
Notes	We recommend Goat Anti-Rabbit IgG H&L (HRP) (ab6721) secondary antibody. A stronger signal is observed in chemiluminescent western blot when using 2% BSA as the blocking agent. Milk blocking is suitable for use with fluorescent detection systems.

Species	Cow
Dilution info	1/1000.00000 - 1/5000.00000
Notes	We recommend Goat Anti-Rabbit IgG H&L (HRP) (ab6721) secondary antibody. A stronger signal is observed in chemiluminescent western blot when using 2% BSA as the blocking agent. Milk blocking is suitable for use with fluorescent detection systems.

Species	Chicken
Dilution info	1/1000.00000 - 1/5000.00000
Notes	We recommend Goat Anti-Rabbit IgG H&L (HRP) (ab6721) secondary antibody. A stronger signal is observed in chemiluminescent western blot when using 2% BSA as the blocking agent. Milk blocking is suitable for use with fluorescent detection systems.

Species	Rabbit
Dilution info	1/1000.00000 - 1/5000.00000
Notes	We recommend Goat Anti-Rabbit IgG H&L (HRP) (ab6721) secondary antibody. A stronger signal is observed in chemiluminescent western blot when using 2% BSA as the blocking agent. Milk blocking is suitable for use with fluorescent detection systems.

Predicted

Species	Guinea pig, Pig, Drosophila melanogaster, Monkey, Zebrafish, Rhesus monkey, Sheep
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Rat
Dilution info	1 µg/mL

Notes	-

Species	Human
Dilution info	1 µg/mL
Notes	-

Species	Mouse
Dilution info	1 µg/mL
Notes	-

Expected

Species	Chicken
Dilution info	1 µg/mL
Notes	-

Species	Chinese hamster
Dilution info	1 µg/mL
Notes	-

Species	Cow
Dilution info	1 µg/mL
Notes	-

Species	Dog
Dilution info	1 µg/mL
Notes	-

Species	Fish
Dilution info	1 µg/mL
Notes	-

Species	Rabbit
Dilution info	1 µg/mL
Notes	-

Species	Xenopus laevis
Dilution info	1 µg/mL
Notes	-

Predicted

Species	Guinea pig, Pig, Drosophila melanogaster, Monkey, Zebrafish, Rhesus monkey, Sheep
Dilution info	-
Notes	-

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

For western blot, milk blocking is suitable for use with fluorescent detection systems. For western blot using chemiluminescent (ECL) systems we recommend BSA blocking.
Abcam recommended secondaries - Goat Anti-Rabbit HRP (ab205718) and Goat Anti-Rabbit Alexa Fluor® 488 (ab150077). Or search our wide range of secondary antibodies for use with your experiment.

Abcam is leading the way to address reproducibility in scientific research with our highly validated recombinant monoclonal and recombinant multiclonal antibodies. Search & select one of Abcam's thousands of recombinant alternatives to eliminate batch-variability and unnecessary animal use.

If you do not find a host species to meet your needs, our catalogue and custom Chimeric range provides scientists the specificity of Abcam's RabMAbs in the species backbone of your choice. Remember to also review our range of edited cell lines, proteins and biochemicals relevant to your target that may help you further your research goals.

Abcam antibodies are extensively validated in a wide range of species and applications, so please check the reagent specifications meet your scientific needs before purchasing. If you have any questions or bespoke requirements, simply visit the Contact Us page to send us an inquiry or contact our Support Team ahead of purchase.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	Beta actin also known as beta cytoplasmic actin plays a central role in cell structure and motility. It is part of the actin protein family and is widely expressed in eukaryotic cells. The molecular weight of beta actin is approximately 42 kDa. It contributes to the formation of the cytoskeleton and participates in various cellular processes including movement and stability. Actin is abundant in all cell types providing structural integrity and flexibility.
Biological function summary	Beta actin contributes to the maintenance of cell shape and is an important player in cell division and muscle contraction. It forms part of a larger actin filaments network often associating with other proteins to form the actin cytoskeleton complex. This complex supports cellular processes such as signaling intracellular trafficking and positioning of organelles. The dynamic polymerization and depolymerization of actin filaments are critical for cellular functions.
Pathways	Beta actin functions in the regulation of important biological pathways such as the Rho/Rac/Cdc42 signaling pathway and the Wnt signaling pathway. These pathways are essential in numerous cellular activities including cell morphology and gene transcription. Beta actin closely interacts with proteins like myosin and tropomyosin which facilitate its role in muscle contraction and cell division and proteins such as Rac and Cdc42 which help govern cytoskeletal dynamics and cellular responses to extracellular stimuli.
Associated diseases and disorders	Beta actin has links to cancers and muscular dystrophies. Aberrations in actin dynamics can result in tumor cell migration and metastasis making it a component of interest in cancer research. Additionally mutations or dysregulation in actin-associated proteins may contribute to muscular dystrophies affecting muscle function and strength. Beta actin's interactions with proteins like dystrophin involved in maintaining muscle integrity further highlight its relevance in both biological functions and disease contexts.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

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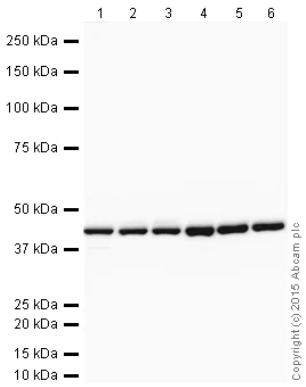
19 product images



Western blot - Anti-beta Actin antibody (ab8227)

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 ab8227 overnight at 4°C. Antibody binding was detected using ab205718, and visualised using ECL development solution ab133406.

All lanes:
Western blot - Anti-beta Actin antibody (ab8227) at 1 µg/mL

Lane 1:
Liver (Human) Tissue Lysate at 10 µg

Lane 2:
Liver (Mouse) Tissue Lysate at 10 µg

Lane 3:
Liver (Rat) Tissue Lysate at 10 µg

Lane 4:
HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate at 10 µg

Lane 5:
NIH 3T3 (Mouse embryonic fibroblast cell line) Whole Cell Lysate at 10 µg

Lane 6:
PC12 (Rat adrenal pheochromocytoma cell line) Whole Cell Lysate at 10 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab205718) at 1/50000 dilution

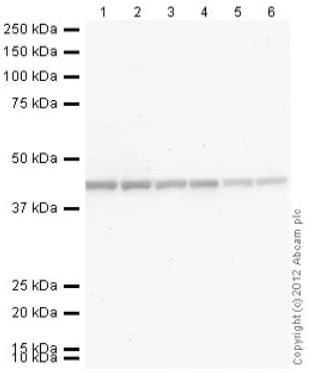
Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 36 kDa

Observed band size: 42 kDa

Exposure time: 30s



Western blot - Anti-beta Actin antibody (ab8227)

All lanes:
Western blot - Anti-beta Actin antibody (ab8227) at 1 µg/mL

All lanes:
HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate at 10 µg

Secondary

Lanes 1 - 2:
Western blot - Donkey Anti-Rabbit IgG H&L (Alkaline Phosphatase) (ab97061) at 1/5000 dilution

Lanes 3 - 4:
Western blot - Donkey Anti-Rabbit IgG H&L (Alkaline Phosphatase) (ab97061) at 1/20000 dilution

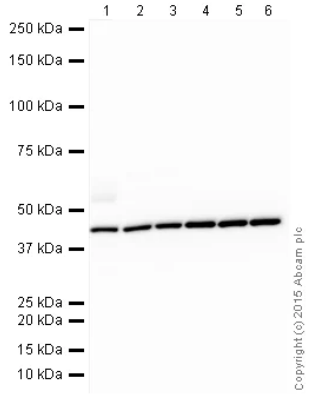
Lanes 5 - 6:
Western blot - Donkey Anti-Rabbit IgG H&L (Alkaline Phosphatase) (ab97061) at 1/50000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 36 kDa

Exposure time: 3min



Western blot - Anti-beta Actin antibody (ab8227)

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 ab8227 overnight at 4°C. Antibody binding was detected using ab205722, and visualised using ECL development solution ab133406.

All lanes:
Western blot - Anti-beta Actin antibody (ab8227) at 1 µg/mL

Lane 1:
Liver (Human) Tissue Lysate at 10 µg

Lane 2:
Liver (Mouse) Tissue Lysate at 10 µg

Lane 3:
Liver (Rat) Tissue Lysate at 10 µg

Lane 4:
HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate at 10 µg

Lane 5:
NIH 3T3 (Mouse embryonic fibroblast cell line) Whole Cell Lysate at 10 µg

Lane 6:
PC12 (Rat adrenal pheochromocytoma cell line) Whole Cell Lysate at 10 µg

Secondary

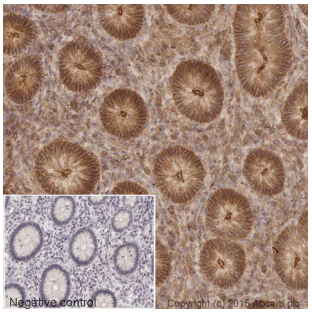
All lanes:
Western blot - Donkey Anti-Rabbit IgG H&L (HRP) (ab205722) at 1/10000 dilution
Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 36 kDa

Observed band size: 42 kDa

Exposure time: 10s



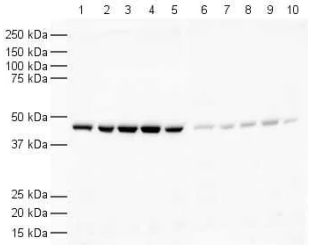
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Actin antibody (ab8227)

IHC image of beta actin staining in a section of formalin-fixed paraffin-embedded normal human colon*. The section was pre-treated using pressure cooker heat mediated antigen retrieval with sodium citrate buffer (pH6) for 30mins. The section was then incubated with ab8227, 1/1000 dilution, for 15 mins at room temperature. A goat anti-rabbit biotinylated secondary antibody (ab6720, 1/1000 dilution) was used to detect the primary, and visualized using an HRP conjugated ABC system. Streptavidin HRP was used, ab7403 at a 1/10000 dilution. DAB was used as the chromogen (ab103723), diluted 1/100 and incubated for 10min at room temperature. The section was then counterstained with haematoxylin and mounted with DPX.

The inset negative control image is taken from an identical assay without primary antibody.

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



Western blot - Anti-beta Actin antibody (ab8227)

All lanes:
Western blot - Anti-beta Actin antibody (ab8227) at 1/1000 dilution

Lanes 1 and 6:
HeLa nuclear lysate at 20 µg

Lanes 2 and 7:
HeLa whole cell lysate at 20 µg

Lanes 3 and 8:
A431 cell lysate at 20 µg

Lane 4:
Jurkat cell lysate at 20 µg

Lanes 10 and 5:
HEK 293 cell lysate at 20 µg

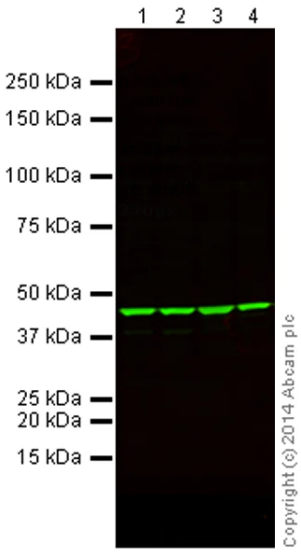
Lane 9:
Jurkate cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab6721) at 1/5000 dilution
Predicted band size: 41 kDa

Observed band size: 41.7 kDa

Exposure time: 5s



Western blot - Anti-beta Actin antibody (ab8227)

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 ab8227 overnight at 4°C. Antibody binding was detected using Goat Anti-Rabbit IgG H&L (Alexa Fluor® 790) (ab175781) secondary antibody at a 1:10,000 dilution for 1hr at room temperature and then imaged using the Licor Odyssey CLx.

All lanes:
Western blot - Anti-beta Actin antibody (ab8227) at 1/1000 dilution

Lane 1:
A431 (human epidermoid carcinoma cell line) Whole Cell Lysate at 20 µg

Lane 2:
HEK-293 (human epithelial cell line from embryonic kidney) Whole Cell Lysate at 20 µg

Lane 3:
NIH/3T3 (mouse embryo fibroblast cell line) Whole Cell Lysate at 20 µg

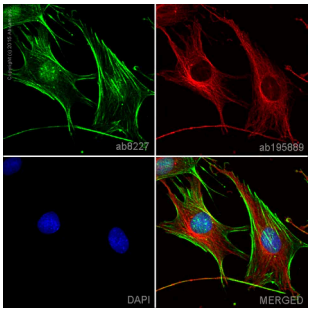
Lane 4:
PC-12 (rat adrenal gland pheochromocytoma cell line) Whole Cell Lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (Alexa Fluor® 790) (ab175781) at 1/10000 dilution

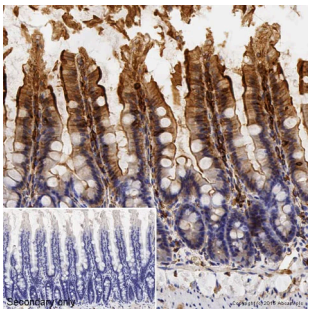
Predicted band size: 41 kDa

Observed band size: 42 kDa



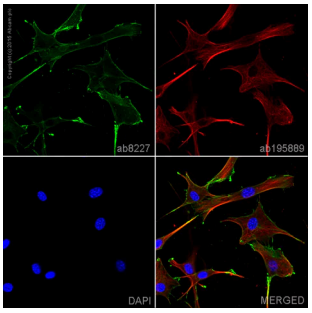
Immunocytochemistry/ Immunofluorescence - Anti-beta Actin antibody (ab8227)

ab8227 staining beta Actin in SV40LT-SMC (rat aortic smooth muscle cells transfected with SV40). The cells were fixed with 4% formaldehyde (10min), permeabilized with 0.1% Triton X-100 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 ab8227 at 1µg/ml (detected with ab150081, Alexa Fluor® 488 Goat anti-Rabbit, 1µg/ml, shown in green); and ab195889, Mouse monoclonal [DM1A] to alpha Tubulin - Microtubule Marker (Alexa Fluor® 594), at 1/250 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



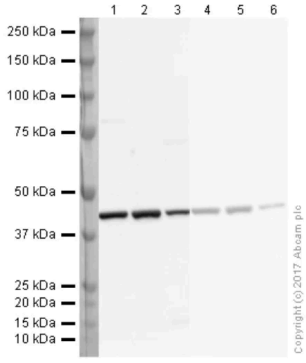
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Actin antibody (ab8227)

IHC image of ab8227 staining beta Actin in rat small intestine formalin fixed paraffin embedded tissue sections, performed on a Leica Bond. The section was pre-treated using heat mediated antigen retrieval with EDTA (epitope retrieval solution 2) for 20 mins. The section was then incubated with ab8227, 0.2µg/ml, 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.



Immunocytochemistry/ Immunofluorescence - Anti-beta Actin antibody (ab8227)

ab8227 staining beta Actin in NIH/3T3 (mouse embryo fibroblast cell line) cells. The cells were fixed with 100% methanol (5min), permeabilized with 0.1% Triton X-100 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 ab8227 at 1µg/ml (shown in green) and ab195889, Mouse monoclonal [DM1A] to alpha Tubulin - Microtubule Marker (Alexa Fluor® 594), at 1/250 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



Western blot - Anti-beta Actin antibody (ab8227)

Lanes 1-3: Blocked with 2% BSA

Lanes 4-6: Blocked with 3% Milk

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 or 3% Milk before being incubated with ab8227 overnight at 4°C. Antibody binding was detected using an anti rabbit HRP secondary antibody, and visualised using ECL development solution ab133406

All lanes:
Western blot - Anti-beta Actin antibody (ab8227) at 1 µg/mL

Lane 1:
HeLa (human epithelial cell line from cervix adenocarcinoma) whole cell lysate (blocked with 2% BSA) at 10 µg

Lane 2:
NIH/3T3 (mouse embryo fibroblast cell line) whole cell lysate (blocked with 2% BSA) at 10 µg

Lane 3:
Rat Liver tissue lysate (blocked with 2% BSA) at 10 µg

Lane 4:
HeLa whole cell lysate (blocked with 3% Milk) at 10 µg

Lane 5:
NIH/3T3 whole cell lysate (blocked with 3% Milk) at 10 µg

Lane 6:
Rat Liver tissue lysate (blocked with 3% Milk) at 10 µg

Secondary

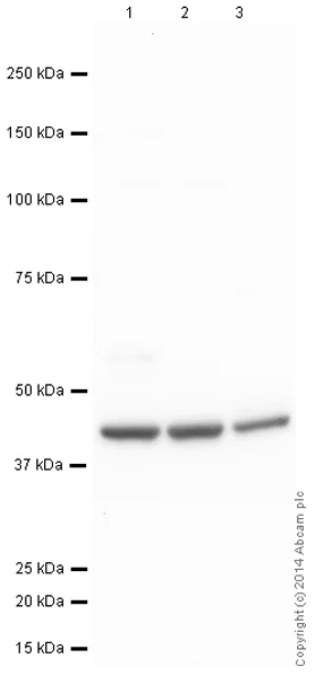
All lanes:
Goat Anti-Rabbit IgG H&L (HRP) at 1/50000 dilution
Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 41 kDa

Observed band size: 45 kDa

Exposure time: 10s



Western blot - Anti-beta Actin antibody (ab8227)

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 ab8227 overnight at 4°C. Antibody binding was detected using an anti-rabbit HRP secondary antibody (ab6721), and visualised using ECL development solution ab133406.

All lanes:
Western blot - Anti-beta Actin antibody (ab8227) at 0.1 µg/mL

Lane 1:
HeLa (human epithelial cell line from cervix adenocarcinoma) Whole Cell Lysate at 10 µg

Lane 2:
NIH/3T3 (mouse embryonic fibroblast cell line) Whole Cell Lysate at 10 µg

Lane 3:
Liver (Rat) Tissue Lysate at 10 µg

Secondary

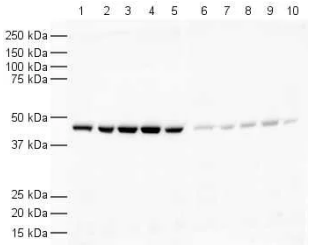
All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/50000 dilution

Performed under reducing conditions.

Predicted band size: 41 kDa

Observed band size: 42 kDa

Exposure time: 1min



Western blot - Anti-beta Actin antibody (ab8227)

All lanes:
Western blot - Anti-beta Actin antibody (ab8227) at 1/1000 dilution

Lane 1:
HeLa (human epithelial cell line from cervix adenocarcinoma) nuclear lysate at 20 µg

Lanes 2 and 7:
HeLa whole cell lysate at 20 µg

Lane 3:
A431 (human epidermoid carcinoma cell line) cell lysate at 20 µg

Lane 4:
Jurkat (human T cell leukemia cell line from peripheral blood) cell lysate at 20 µg

Lane 5:
HEK-293 (human epithelial cell line from embryonic kidney) cell lysate at 20 µg

Lane 6:
HeLa nuclear lysate at 20 µg

Lane 8:
A431 cell lysate at 20 µg

Lane 9:
Jurkat cell lysate at 20 µg

Lane 10:
HEK-293 cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab6721) at 1/5000 dilution

Predicted band size: 41 kDa

Observed band size: 41.7 kDa

Exposure time: 5s

Western blot - Anti-beta Actin antibody (ab8227)

All lanes:
Western blot - Anti-beta Actin antibody (ab8227) at 1/1000 dilution

Lane 1:
HeLa (Human epithelial cell line from cervix adenocarcinoma) cells at 20 µg

Lane 2:
NIH/3T3 (Mouse embryonic fibroblast cell line) cells at 20 µg

Lane 3:
Fish Liver at 20 µg

Lane 4:
Rabbit Liver at 20 µg

Lane 5:

MDCK (Canine kidney cell line) cells at 20 µg

Lane 6:
EBTr (cow trachea) cells at 20 µg

Lane 7:
SL-29 (chicken day 11 embryo) cells at 20 µg

Lane 8:
CHO (Chinese hamster ovary cell line) cells at 20 µg

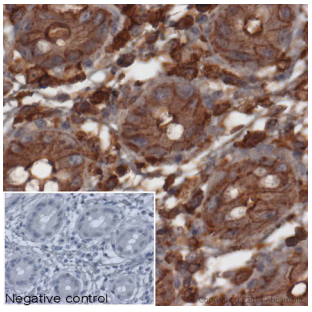
Lane 9:
Xenopus laevis embryo at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab6721) at 1/5000 dilution
Predicted band size: 41 kDa

Observed band size: 41.7 kDa

Exposure time: 30s

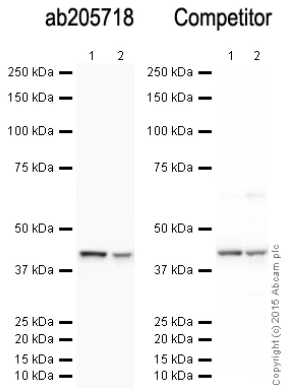


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Actin antibody (ab8227)

IHC image of beta Actin staining in normal human colon, formalin-fixed and paraffin-embedded tissue*. The section was pre-treated using pressure cooker heat mediated antigen retrieval with sodium citrate buffer (pH6) for 30mins. The section was incubated with ab8227, 3µg/ml overnight at +4°C. A anti-rabbit HRP secondary antibody (ab97200, 1/200 dilution) was used for 1hr at room temperature. The section was counterstained with haematoxylin and mounted with DPX.

The inset negative control image is taken from an identical assay without primary antibody.

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



Western blot - Anti-beta Actin antibody (ab8227)

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 ab8227 overnight at 4°C. Antibody binding was detected using ab205718 (Left Image) and a competitor secondary (Right Image), and visualised using ECL development solution ab133406.

All lanes:
Western blot - Anti-beta Actin antibody (ab8227) at 1 µg/mL

Lane 1:
Liver (Mouse) Tissue Lysate at 10 µg

Lane 2:
Liver (Rat) Tissue Lysate at 10 µg

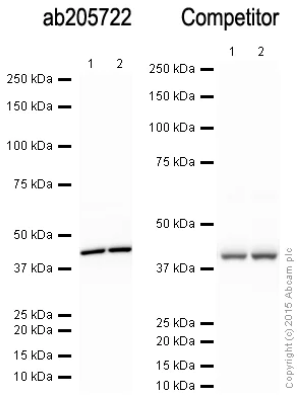
Secondary

All lanes:
ab205718 (Left Image) at 1/20,000 and a competitor secondary (Right Image) at 1/50,000. Notice the increased background of the competitor product.
Performed under reducing conditions.

Predicted band size: 36 kDa

Observed band size: 42 kDa

Exposure time: 5s



Western blot - Anti-beta Actin antibody (ab8227)

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 ab8227 overnight at 4°C. Antibody binding was detected using ab205722 (Left Image) and a competitor secondary (Right Image), and visualised using ECL development solution ab133406.

All lanes:
Western blot - Anti-beta Actin antibody (ab8227) at 1 µg/mL

Lane 1:
Liver (Mouse) Tissue Lysate at 10 µg

Lane 2:
Liver (Rat) Tissue Lysate at 10 µg

Secondary

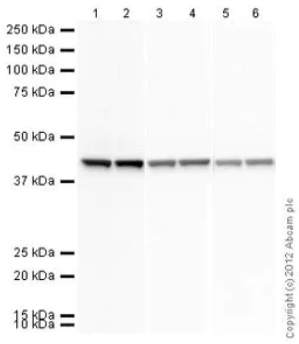
All lanes:
ab205722 (Left Image) at 1/10,000 and a competitor secondary (Right Image) at 1/10,000. Notice the decreased signal of the competitor product.

Performed under reducing conditions.

Predicted band size: 36 kDa

Observed band size: 42 kDa

Exposure time: 10s



Western blot - Anti-beta Actin antibody (ab8227)

Lanes 1 - 6:
Western blot - Anti-beta Actin antibody (ab8227) at 1 µg/mL

Lanes 1 - 2:
Western blot - Anti-GNAT2 antibody (ab97501) at 1/2000 dilution

All lanes:
HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate at 10 µg

Secondary

Lanes 3 - 4:

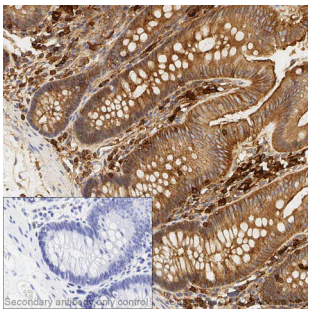
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/10000 dilution

Lanes 5 - 6:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution
Developed using the ECL technique.

Performed under reducing conditions.

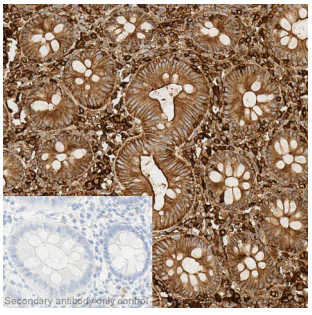
Predicted band size: 36 kDa

Exposure time: 10s



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Actin antibody (ab8227)

Immunohistochemical analysis of formalin fixed paraffin embedded human colon labelling beta actin with ab8227 at a concentration of 2 µg/ml. The immunostaining was performed on a Leica Biosystems BOND® RX instrument with a Bond™ Polymer Refine Detection kit. Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution 2) for 20 mins. ab8227 anti-beta actin antibody was incubated for 30 mins at room temperature. Sections were counterstained with Hematoxylin. Image inset shows absence of staining in secondary antibody only control.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Actin antibody (ab8227)

Immunohistochemical analysis of formalin fixed paraffin embedded human colon labelling beta actin with ab8227 at a concentration of 1 µg/ml. The immunostaining was performed on a Ventana DISCOVERY ULTRA (Roche Tissue Diagnostics) instrument with an OptiView DAB IHC Detection Kit. Heat mediated antigen retrieval was conducted for 32 mins at 100°C with ULTRA cell conditioning solution (CC1, pH 8.5). ab8227 anti-beta actin antibody was incubated at 37°C for 16 mins. Sections were counterstained with Hematoxylin II. Image inset shows absence of staining in secondary antibody only control.