abcam

ab303664

Anti-Ubiquitin antibody [P4D1]

Recombinant

Key facts

Isotype	IgG1
Host species	Mouse
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	P4D1
Purification technique	Affinity purification Protein A
Concentration	1.016 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

WB

Tested

Species	Human
Dilution info	1/1000
Notes	-

Species	Mouse

Dilution info	1/1000
Notes	-

Species	Rat
Dilution info	1/1000
Notes	-

IHC-P

Tested

Species	Human
Dilution info	1/20000
Notes	Perform heat-mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Species	Mouse
Dilution info	1/20000
Notes	Perform heat-mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Species	Rat
Dilution info	1/20000
Notes	Perform heat-mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/800
Notes	-

Species	Mouse
Dilution info	1/800
Notes	-

Not recommended

Species	Rat
Dilution info	-
Notes	-

IP

Not recommended

Species	Human, Mouse, Rat
Dilution info	-
Notes	-

Target data

See full target information UBB

Function	Ubiquitin. Exists either covalently attached to another protein, or free (unanchored). When covalently bound, it is conjugated to target proteins via an isopeptide bond either as a monomer (monoubiquitin), a polymer linked via different Lys residues of the ubiquitin (polyubiquitin chains) or a linear polymer linked via the initiator Met of the ubiquitin (linear polyubiquitin chains). Polyubiquitin chains, when attached to a target protein, have different functions depending on the Lys residue of the ubiquitin that is linked: Lys-6-linked may be involved in DNA repair; Lys-11-linked is involved in ERAD (endoplasmic reticulum-associated degradation) and in cell-cycle regulation; Lys-29-linked is involved in proteotoxic stress response and cell cycle; Lys-33-linked is involved in kinase modification; Lys-48-linked is involved in protein degradation via the proteasome; Lys-63-linked is involved in endocytosis, DNA-damage responses as well as in signaling processes leading to activation of the transcription factor NF-kappa-B. Linear polymer chains formed via attachment by the initiator Met lead to cell signaling. Ubiquitin is usually conjugated to Lys residues of target proteins, however, in rare cases, conjugation to Cys or Ser residues has been observed. When polyubiquitin is free (unanchored-polyubiquitin), it also has distinct roles, such as in activation of protein kinases, and in signaling.

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

What are the advantages of a recombinant monoclonal antibody?
This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity
- Long-term security of supply
- Animal-free batch production

For more information, read more on recombinant antibodies.

Want a custom formulation?
This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or conjugation for your experiments, please contact orders@abcam.com

Supplementary info

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

Activity summary	Ubiquitin is a small regulatory protein found in almost all tissues of eukaryotic organisms. It has a molecular weight of approximately 8.6 kilodaltons. It functions mechanically by attaching to proteins through a process called ubiquitination which involves the formation of an isopeptide bond. Ubiquitin molecules can form polyubiquitin chains through different lysine residues such as K48 and K63 that determine their function. These chains label substrate proteins for various fates including degradation. Ubiquitin is expressed ubiquitously in cells reflecting its essential role in maintaining protein homeostasis.
Biological function summary	The ubiquitin system plays a critical role in regulating protein turnover and quality control within cells. It is part of a larger complex known as the ubiquitin-proteasome system (UPS) which is responsible for degrading proteins that need to be turned over. This process is essential for cell cycle control response to oxidative stress and DNA repair. Ubiquitin's role in tagging proteins for degradation or signaling allows cells to respond quickly to changes in their environment and maintain balance.
Pathways	Ubiquitin functions in several important biological pathways including the Wnt and NF-kB pathways. In the Wnt signaling pathway ubiquitination modulates the stability of key components thereby affecting the pathway's overall activity. In the

	NF-kB signaling pathway ubiquitin labels inhibitor proteins for degradation which releases and activates NF-kB. These pathways highlight ubiquitin's interaction with proteins such as beta-catenin in Wnt and IkB in NF-kB illustrating how it regulates diverse cellular processes.
Associated diseases and disorders	The dysfunction of the ubiquitin system is linked to neurodegenerative diseases and cancers. Ubiquitin-related defects in protein degradation can lead to the buildup of unwanted proteins contributing to conditions like Parkinson's disease. Connections with cancer are evident as ubiquitin controls cell cycle proteins and aberrant ubiquitination may drive tumor growth and progression. The protein p53 known to be controlled by ubiquitination plays a significant role in cancer related mechanisms when dysregulated. Understanding and targeting ubiquitin-related pathways may provide new therapeutic opportunities for treating these conditions.

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

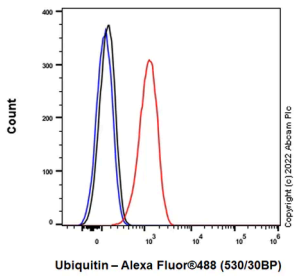
We do not recommend this combination. It is not covered by our product promise.

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.

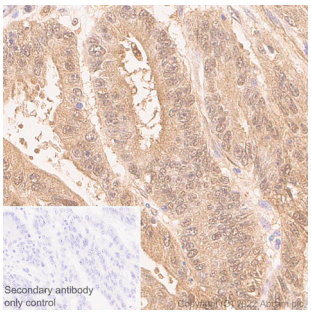
7 product images



Flow Cytometry (Intracellular) - Anti-Ubiquitin antibody [P4D1] (ab303664)

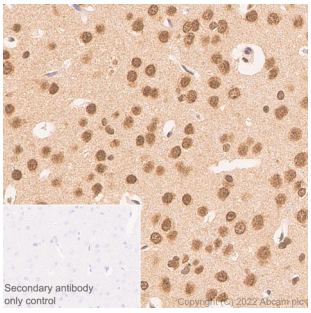
Ubiquitin Flow Cytometry (Intracellular) staining using mouse Anti-Ubiquitin antibody

Flow cytometric (Intracellular) overlay histogram showing 4% paraformaldehyde fixed, 90% methanol permeabilized NIH/3T3 (mouse embryonic fibroblast) cells labeling Ubiquitin with ab303664 at 1/800 dilution (0.1µg) (red line). Isotype control antibody (black line) was a mouse monoclonal IgG used at the same concentration and conditions as the primary antibody. Unlabeled sample (blue line) was also used as a control.



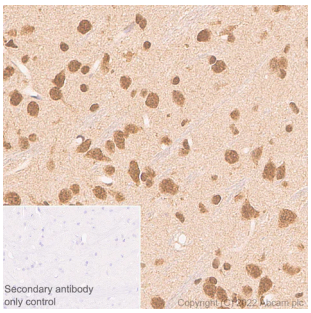
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ubiquitin antibody [P4D1] (ab303664)

Immunohistochemical analysis of paraffin-embedded human colon cancer tissue labeling Ubiquitin with ab303664 at 1/20000 (0.044 µg/ml) followed by a ready to use Leica DS9800 (Bond™ Polymer Refine Detection kit). Nuclear staining on human colon cancer. The section was incubated with ab303664 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Secondary antibody only control: Primary diluent was used instead of primary antibody, followed by a ready to use secondary antibody Leica DS9800 (Bond™ Polymer Refine Detection kit). Heat mediated antigen retrieval with citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins.



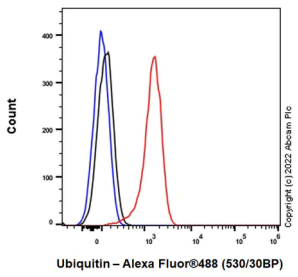
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ubiquitin antibody [P4D1] (ab303664)

Immunohistochemical analysis of paraffin-embedded rat cerebrum tissue labeling Ubiquitin with ab303664 at 1/20000 (0.044 µg/ml) followed by a ready to use Leica DS9800 (Bond™ Polymer Refine Detection kit). Nuclear staining on rat cerebrum. The section was incubated with ab303664 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Secondary antibody only control: Primary diluent was used instead of primary antibody, followed by a ready to use secondary antibody Leica DS9800 (Bond™ Polymer Refine Detection kit). Heat mediated antigen retrieval with citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins.



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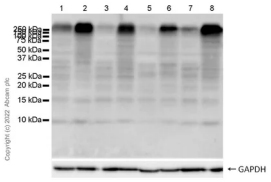
Immunohistochemical analysis of paraffin-embedded mouse cerebrum tissue labeling Ubiquitin with ab303664 at 1/20000 (0.044 µg/ml) followed by a ready to use Leica DS9800 (Bond™ Polymer Refine Detection kit). Nuclear staining on mouse cerebrum. The section was incubated with ab303664 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Secondary antibody only control: Primary diluent was used instead of primary antibody, followed by a ready to use secondary antibody Leica DS9800 (Bond™ Polymer Refine Detection kit). Heat mediated antigen retrieval with citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins.



Flow Cytometry (Intracellular) - Anti-Ubiquitin antibody [P4D1] (ab303664)

Ubiquitin Flow Cytometry (Intracellular) staining using mouse Anti-Ubiquitin antibody

Flow cytometric (Intracellular) overlay histogram showing 4% paraformaldehyde fixed, 90% methanol permeabilized NIH/3T3 (mouse embryonic fibroblast) cells labeling Ubiquitin with ab303664 at 1/800 dilution (0.1µg) (red line). Isotype control antibody (black line) was a mouse monoclonal IgG used at the same concentration and conditions as the primary antibody. Unlabeled sample (blue line) was also used as a control.



Western blot - Anti-Ubiquitin antibody [P4D1] (ab303664)

Ubiquitin Western blot staining using mouse Anti-Ubiquitin antibody

Blocking and diluting buffer and concentration: 5% NFDM/TBST Exposure time: 26 seconds
All lanes:
Western blot - Anti-Ubiquitin antibody [P4D1] (ab303664) at 1/1000 dilution

Lane 1:
Untreated 293T (human embryonic kidney epithelial cell), whole cell lysate at 20 µg

Lane 2:
293T treated with 10µM MG-132 (ab141003) for 4 hours, whole cell lysate at 20 µg

Lane 3:
Untreated HeLa (human cervix adenocarcinoma epithelial cell), whole cell lysate at 20 µg

Lane 4:
HeLa treated with 10µM MG-132 (ab141003) for 6 hours, whole cell lysate at 20 µg

Lane 5:
Untreated MCF7 (human breast adenocarcinoma epithelial cell), whole cell lysate at 20 µg

Lane 6:
MCF7 treated with 10µM MG-132 (ab141003) for 2 hours, whole cell lysate at 20 µg

Lane 7:
Untreated NIH/3T3 (mouse embryonic fibroblast), whole cell lysate at 20 µg

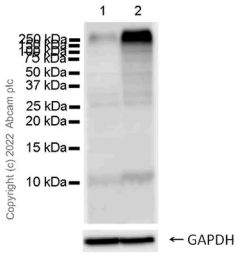
Lane 8:
NIH/3T3 treated with 20µM MG-132 (ab141003) for 5 hours, whole cell lysate at 20 µg

Secondary

All lanes:
Peroxidase-Conjugated Goat anti-Mouse IgG (H+L) at 1/10000 dilution

Observed band size: 8 kDa, 200 kDa

Exposure time: 26s



Western blot - Anti-Ubiquitin antibody [P4D1] (ab303664)

Blocking and diluting buffer and concentration: 5% NFDM/TBST Exposure time: 3 seconds
All lanes:
Western blot - Anti-Ubiquitin antibody [P4D1] (ab303664) at 1/1000 dilution

Lane 1:
Untreated PC-12 (rat adrenal gland pheochromocytoma), whole cell lysate at 20 µg

Lane 2:
PC-12 treated with 10µM MG-132 (ab141003) for 4 hours, whole cell lysate at 20 µg

Secondary

All lanes:
Peroxidase-Conjugated Goat anti-Mouse IgG (H+L) at 1/10000 dilution

Observed band size: 8 kDa, 200 kDa

Exposure time: 3s

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.