abcam

ab179434

# Anti-Ubiquitin (linkage-specific K63) antibody [EPR8590-448]

Anti-Ubiquitin (linkage-specific K63) antibody [EPR8590-448] (ab179434) is a rabbit monoclonal antibody detecting Ubiquitin in **Western Blot, Flow Cytometry (Intra), IHC-P**. Suitable for **Human, Mouse, Rat**.

- Biophysical QC for unrivalled batch-batch consistency
- Over 80 publications

Recombinant | RabMAb | 20ul selling size

## Key facts

| | |
|---|---|
| **Isotype** | IgG |
| **Host species** | Rabbit |
| **Storage buffer** | 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** | EPR8590-448 |
| **Purification technique** | Affinity purification Protein A |
| **Concentration** | 2.258 mg/mL The concentration of this product may be batch-dependent<br>Batch concentration finder → |

## Reactivity data

### WB

**Tested**

| | |
|---|---|
| **Species** | Recombinant full length protein |
| **Dilution info** | 1/1000 |
| **Notes** | - |

| | |
|---|---|
| **Species** | Mouse |
| **Dilution info** | 1/1000 |
| **Notes** | - |

| | |
|---|---|
| **Species** | Rat |
| **Dilution info** | 1/1000 |
| **Notes** | - |

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/1000 |
| **Notes** | - |

## Flow Cyt (Intra)

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/210 |
| **Notes** | - |

### Expected

| | |
|---|---|
| **Species** | Mouse, Rat |
| **Dilution info** | Use at an assay dependent concentration. |
| **Notes** | - |

### Not recommended

| | |
|---|---|
| **Species** | Recombinant full length protein |
| **Dilution info** | - |
| **Notes** | - |

## IHC-P

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/250 - 1/500 |
| **Notes** | **For unpurified, use 1/100 - 1/250.**<br>Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. |

### Expected

| | |
|---|---|
| **Species** | Mouse, Rat |
| **Dilution info** | Use at an assay dependent concentration. |
| **Notes** | - |

### Not recommended

| | |
|---|---|
| **Species** | Recombinant full length protein |
| **Dilution info** | - |
| **Notes** | - |

# Target data

See full target information UBC Linkage-specific K63

**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. During ubiquitination, the acceptor ubiquitin is positioned in the active site via direct interaction with the E2 ubiquitin-conjugating enzymes such as UBE2R2 (PubMed:38326650). As a monoubiquitin, its C-terminal glycine is recognized as a C-degron by Cul2-RING (CRL2) E3 ubiquitin-protein ligase complexes (PubMed:39548056).

## Storage

| | |
|---|---|
| **Shipped at conditions** | Conditional Ambient |
| **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 is this antibody validated in?**
Anti-Ubiquitin (linkage-specific K63) antibody [EPR8590-448] (ab179434) is a rabbit recombinant monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Intra), Immunohistochemistry (IHC-P) in Human, Mouse, Rat samples.

**What is the molecular weight of Ubiquitin?**
Anti-Ubiquitin (linkage-specific K63) [EPR8590-448] (ab179434) specifically detects a band for Ubiquitin (UniProt: P0CG48) at a molecular weight of 77kDa.

**Trusted by the scientific community**
Anti-Ubiquitin (linkage-specific K63) [EPR8590-448] (ab179434) was first used in a scientific publication in 2013 and has been cited over 80 times in peer-reviewed journals.

**Reviewed by scientists**
Anti-Ubiquitin (linkage-specific K63) [EPR8590-448] (ab179434) has over 5 independent reviews from customers.

**Other related products**
We have a range of other formats of antibody clone [EPR8590-448] also available for your convenience: ab179434, Alexa Fluor® 647 - ab192489, Carrier free - ab271929, PE - ab305677, APC - ab305678, HRP - ab305679, Alkaline Phosphatase - ab308969, Alexa Fluor® 594 - ab310741, Alexa Fluor® 555 - ab312270, Alexa Fluor® 568 - ab312761, Alexa Fluor® 750 - ab321221

**Patented technology**
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

**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.

**Trial sizes available!**
Test your antibody or perform pre-screening before committing to a larger quantity. Sold in 20µl. Discover our selection of trial-size antibodies.

## 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**

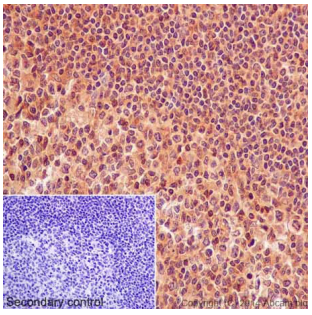
We do not recommend this combination. It is not covered by our product promise.

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.

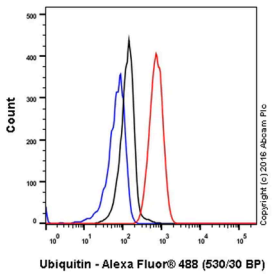
## 7 product images




### Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ubiquitin (linkage-specific K63) antibody [EPR8590-448] (ab179434)

Ubiquitin (linkage-specific K63) Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Ubiquitin (linkage-specific K63) antibody

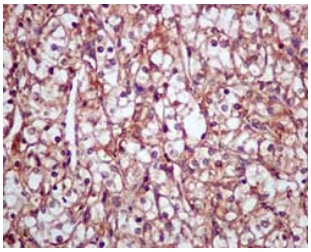
Immunohistochemical staining of paraffin embedded human tonsil with purified ab179434 at a dilution of 1/500. The secondary antibody used is a HRP conjugated goat anti-rabbit (ab97051 1/500). The sample is counter-stained with hematoxylin. Antigen retrieval was perfomed using Tris-EDTA buffer pH 9.0. PBS was used instead of the primary antibody as the negative control and is shown in the inset.




## Flow Cytometry (Intracellular) - Anti-Ubiquitin (linkage-specific K63) antibody [EPR8590-448] (ab179434)

Ubiquitin (linkage-specific K63) Flow Cytometry (Intracellular) staining using rabbit Anti-Ubiquitin (linkage-specific K63) antibody

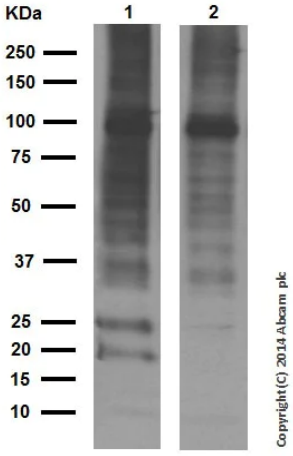
Intracellular Flow Cytometry analysis of HeLa (human cervix adenocarcinoma) cells labelling Ubiquitin (linkage-specific K63) with purified ab179434 at 1/210 (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. An Alexa Fluor®488-conjugated goat anti-rabbit IgG (1/2000) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.



## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ubiquitin (linkage-specific K63) antibody [EPR8590-448] (ab179434)

Ubiquitin (linkage-specific K63) Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Ubiquitin (linkage-specific K63) antibody

Immunohistochemical analysis of paraffin-embedded human kidney carcinoma tissue labeling K63-linkage specific polyubiquitin with unpurified ab179434 at 1/100 dilution.




## Western blot - Anti-Ubiquitin (linkage-specific K63) antibody [EPR8590-448] (ab179434)

Blocking buffer: 5% NFDM/TBST
Dilution buffer: 5% NFDM/TBST

All lanes:
Western blot - Anti-Ubiquitin (linkage-specific K63) antibody [EPR8590-448] (ab179434) at 1/1000 dilution

Lane 1:
HEK-293 (human epithelial cell line from embryonic kidney) cell lysate at 20 µg

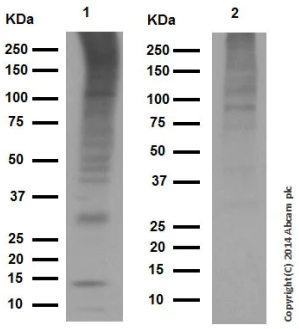
Lane 2:
HeLa (human epithelial cell line from cervix adenocarcinoma) cell lysate at 20 µg

Secondary

All lanes:
HRP goat anti-rabbit (H+L) at 1/1000 dilution

Predicted band size: 26 kDa

Observed band size: 16-300 kDa




## Western blot - Anti-Ubiquitin (linkage-specific K63) antibody [EPR8590-448] (ab179434)

Blocking buffer: 5% NFDM/TBST
Dilution buffer: 5% NFDM/TBST

All lanes:
Western blot - Anti-Ubiquitin (linkage-specific K63) antibody [EPR8590-448] (ab179434) at 1/1000 dilution

Lane 1:
Mouse brain tissue lysate at 20 µg

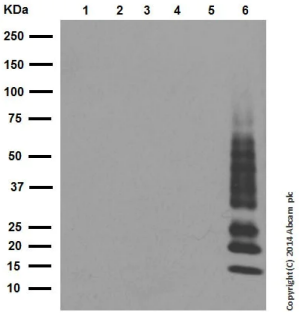
Lane 2:
Rat brain tissue lysate at 20 µg

Secondary

All lanes:
HRP goat anti-rabbit (H+L) at 1/1000 dilution

Predicted band size: 26 kDa

Observed band size: 16-300 kDa




## Western blot - Anti-Ubiquitin (linkage-specific K63) antibody [EPR8590-448] (ab179434)

Blocking buffer: 5% NFDM/TBST
Dilution buffer: 5% NFDM/TBST

All lanes:
Western blot - Anti-Ubiquitin (linkage-specific K63) antibody [EPR8590-448] (ab179434) at 1/5000 dilution

Lane 1:
K6-linked-Ub2 recombinant protein at 20 µg

Lane 2:
K11-linked-Ub2 recombinant protein at 20 µg

Lane 3:
K29-linked-Ub2 recombinant protein at 20 µg

Lane 4:
K33-linked-Ub2 recombinant protein at 20 µg

Lane 5:
K48-linked-Ub2 recombinant protein at 20 µg

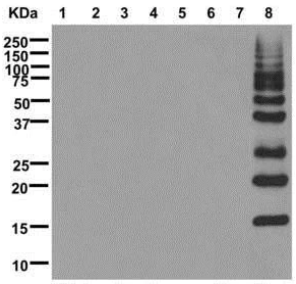
Lane 6:
K63-linked-Ub2 recombinant protein at 20 µg

Secondary

All lanes:
HRP goat anti-rabbit (H+L) at 1/1000 dilution

Predicted band size: 26 kDa

Observed band size: 16-70 kDa




## Western blot - Anti-Ubiquitin (linkage-specific K63) antibody [EPR8590-448] (ab179434)

All lanes:
Western blot - Anti-Ubiquitin (linkage-specific K63) antibody [EPR8590-448] (ab179434) at 1/1000 dilution

Lane 1:
Ubiquitin recombinant protein at 0.02 µg

Lane 2:
K6-linked-Ub2 recombinant protein at 0.02 µg

Lane 3:
K11-linked-Ub2 recombinant protein at 0.02 µg

Lane 4:
K27-linked-Ub2 recombinant protein at 0.02 µg

Lane 5:
K29-linked-Ub2 recombinant protein at 0.02 µg

Lane 6:
K33-linked-Ub2 recombinant protein at 0.02 µg

Lane 7:
K48-linked-Ub2-7 recombinant protein at 0.02 µg

Lane 8:
K63-linked-Ub2-7 recombinant protein at 0.02 µg

Predicted band size: 26 kDa

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