

Product datasheet

Anti-OTUB1 antibody [EPR13028(B)] ab175200

KO VALIDATED Recombinant RabMAb

★★★★★ [3 Abreviews](#) [10 References](#) [7 Images](#)

Overview

| | |
|----------------------------|--|
| Product name | Anti-OTUB1 antibody [EPR13028(B)] |
| Description | Rabbit monoclonal [EPR13028(B)] to OTUB1 |
| Host species | Rabbit |
| Tested applications | Suitable for: WB, IP Unsuitable for: Flow Cyt, ICC/IF or IHC-P |
| Species reactivity | Reacts with: Mouse, Rat, Human |
| Immunogen | Synthetic peptide within Human OTUB1 aa 200 to the C-terminus (Cysteine residue). The exact sequence is proprietary. Database link: Q96FW1 |
| Positive control | WB: Wild-type HAP1, HeLa, MCF7, HepG2, HEK-293T, and HEK-293 cell lysates. Rat and mouse heart tissue lysates. IP: HeLa cell lysate. |
| General notes | This product is a recombinant monoclonal antibody, which offers several advantages including: <ul style="list-style-type: none"> - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information see here . Our RabMAb [®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents . |

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 | Preservative: 0.01% Sodium azide Constituents: PBS, 0.05% BSA, 40% Glycerol (glycerin, glycerine) |
| Purity | Protein A purified |
| Clonality | Monoclonal |
| Clone number | EPR13028(B) |

Isotype

IgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab175200 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 |
|-------------|-----------|---|
| WB | ★★★★★ (2) | 1/1000 - 1/10000. Predicted molecular weight: 31 kDa. |
| IP | ★★★★★ (1) | 1/10 - 1/100. |

Application notes

Is unsuitable for Flow Cyt, ICC/IF or IHC-P.

Target

Function

Hydrolase that can remove conjugated ubiquitin from proteins and plays an important regulatory role at the level of protein turnover by preventing degradation. Regulator of T-cell anergy, a phenomenon that occurs when T-cells are rendered unresponsive to antigen rechallenge and no longer respond to their cognate antigen. Acts via its interaction with RNF128/GRAIL, a crucial inductor of CD4 T-cell anergy. Isoform 1 destabilizes RNF128, leading to prevent anergy. In contrast, isoform 2 stabilizes RNF128 and promotes anergy. Surprisingly, it regulates RNF128-mediated ubiquitination, but does not deubiquitinate polyubiquitinated RNF128. Deubiquitinates estrogen receptor alpha (ESR1). Mediates deubiquitination of 'Lys-48'-linked polyubiquitin chains, but not 'Lys-63'-linked polyubiquitin chains. Not able to cleave di-ubiquitin. Also capable of removing NEDD8 from NEDD8 conjugates, but with a much lower preference compared to 'Lys-48'-linked ubiquitin.

Tissue specificity

Isoform 1 is ubiquitous. Isoform 2 is expressed only in lymphoid tissues such as tonsils, lymph nodes and spleen, as well as peripheral blood mononuclear cells.

Sequence similarities

Belongs to the peptidase C65 family.
Contains 1 OTU domain.

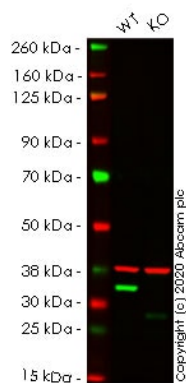
Domain

In addition to ubiquitin-binding at the Cys-91 active site, a proximal ubiquitin-binding site is also present at Cys-23. Occupancy of the active site is needed to enable tight binding to the second site. Distinct binding sites for the ubiquitins may allow to discriminate among different isopeptide linkages (i.e. 'Lys-48-', 'Lys-63'-linked polyubiquitin) in polyubiquitin substrates and achieve linkage-specific deubiquitination.

Cellular localization

Cytoplasm.

Images



Western blot - Anti-OTUB1 antibody [EPR13028(B)]
(ab175200)

All lanes : Anti-OTUB1 antibody [EPR13028(B)] (ab175200) at 1/1000 dilution

Lane 1 : Wild-type HEK-293T cell lysate

Lane 2 : OTUB1 knockout HEK-293T cell lysate

Lysates/proteins at 20 µg per lane.

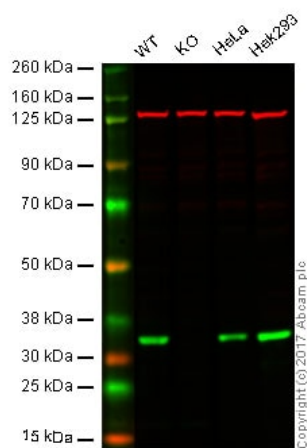
Performed under reducing conditions.

Predicted band size: 31 kDa

Observed band size: 31 kDa

Lanes 1-2: Merged signal (red and green). Green - ab175200 observed at 31 kDa. Red - loading control [ab8245](#) observed at 37 kDa.

ab175200 Anti-OTUB1 antibody [EPR13028(B)] was shown to specifically react with OTUB1 in wild-type HEK-293T cells. Loss of signal was observed when knockout cell line [ab266551](#) (knockout cell lysate [ab257569](#)) was used. Wild-type and OTUB1 knockout samples were subjected to SDS-PAGE. ab175200 and Anti-GAPDH antibody [6C5] - Loading Control ([ab8245](#)) were incubated overnight at 4°C at 1 in 1000 Dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed ([ab216773](#)) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed ([ab216776](#)) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-OTUB1 antibody [EPR13028(B)]
(ab175200)

All lanes : Anti-OTUB1 antibody [EPR13028(B)] (ab175200) at 1/1000 dilution

Lane 1 : Wild-type HAP1 whole cell lysate

Lane 2 : OTUB1 knockout HAP1 whole cell lysate

Lane 3 : HeLa whole cell lysate

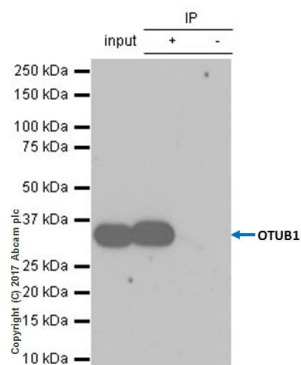
Lane 4 : HEK-293 whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 31 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab175200 observed at 35 kDa. Red - loading control, **ab18058**, observed at 130 kDa.

ab175200 was shown to specifically react with OTUB1 in wild type cells as signal was lost in OTUB1 knockout cells. Wild-type and OTUB1 knockout samples were subjected to SDS-PAGE. ab175200 and **ab18058** (Mouse anti-Vinculin loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed **ab216773** and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed **ab216776** secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



Immunoprecipitation - Anti-OTUB1 antibody [EPR13028(B)] (ab175200)

ab175200 (purified) at 1:50 dilution (2ug) immunoprecipitating OTUB1 in HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate.

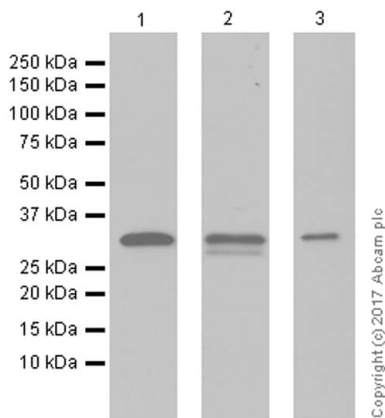
Lane 1 (input): HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate 10ug

Lane 2 (+): ab175200 & HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate

Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of ab175200 in HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate

For western blotting, VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used for detection at 1:1000 dilution.

Blocking and diluting buffer: 5% NFDM/TBST.



Western blot - Anti-OTUB1 antibody [EPR13028(B)] (ab175200)

All lanes : Anti-OTUB1 antibody [EPR13028(B)] (ab175200) at 1/1000 dilution (purified)

Lane 1 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysates

Lane 2 : Mouse heart whole tissue lysates

Lane 3 : Rat heart whole tissue lysates

Lysates/proteins at 15 µg per lane.

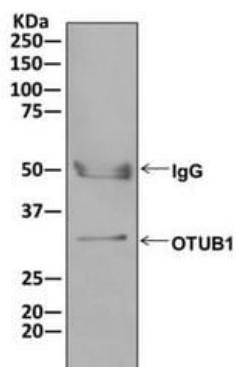
Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 31 kDa

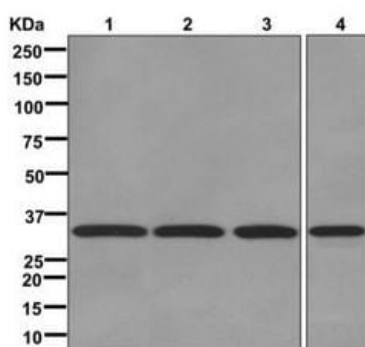
Observed band size: 31 kDa

Blocking and diluting buffer: 5% NFDM/TBST



Immunoprecipitation - Anti-OTUB1 antibody
[EPR13028(B)] (ab175200)

Western blot analysis on immunoprecipitation pellet from MCF7 cell lysate labeling OTUB1 with unpurified ab175200 at 1/10 dilution.



Western blot - Anti-OTUB1 antibody [EPR13028(B)]
(ab175200)

All lanes : Anti-OTUB1 antibody [EPR13028(B)] (ab175200) at 1/1000 dilution (unpurified)

Lane 1 : HeLa cell lysate

Lane 2 : HepG2 cell lysate

Lane 3 : 293T cell lysate

Lane 4 : MCF7 cell lysate

Lysates/proteins at 10 µg per lane.

Predicted band size: 31 kDa

Why choose a recombinant antibody?



Research with confidence
Consistent and reproducible results



Long-term and scalable supply
Recombinant technology



Success from the first experiment
Confirmed specificity



Ethical standards compliant
Animal-free production

Anti-OTUB1 antibody [EPR13028(B)] (ab175200)

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours

- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit <https://www.abcam.com/abpromise> or contact our technical team.

Terms and conditions

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors