abcam

## Product datasheet

# Anti-Ubiquitin antibody [EPR8830] - BSA and Azide free ab230145

Recombinant RabMAb

9 Images

### Overview

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|---|---|
| **Product name** | Anti-Ubiquitin antibody [EPR8830] - BSA and Azide free |
| **Description** | Rabbit monoclonal [EPR8830] to Ubiquitin - BSA and Azide free |
| **Host species** | Rabbit |
| **Tested applications** | **Suitable for:** WB, IHC-P, Flow Cyt, ICC/IF |
| **Species reactivity** | **Reacts with:** Mouse, Rat, Human |
| **Immunogen** | Synthetic peptide within Human Ubiquitin aa 1-100. The exact sequence is proprietary. (Peptide available as ab220157) |
| **General notes** | The formulation and the concentration of this product is compatible for metal-conjugation for mass cytometry (CyTOF®).<br>ab230145 is a PBS-only buffer format of ab134953. Please refer to ab134953 for recommended dilutions, protocols, and image data.<br>Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.<br>Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMab® patents.<br>This product is a recombinant rabbit monoclonal antibody. |

### Properties

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| **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** | Constituent: PBS |
| **Purity** | Protein A purified |
| **Clonality** | Monoclonal |

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| **Clone number** | EPR8830 |
| **Isotype** | IgG |

## Applications

Our Abpromise guarantee covers the use of **ab230145** 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 | | Use at an assay dependent concentration. Predicted molecular weight: 8 kDa.Can be blocked with Ubiquitin peptide (ab220157).<br>Monoubiquitin molecular weight |
| IHC-P | | Use at an assay dependent concentration.<br>See IHC antigen retrieval protocols. |
| Flow Cyt | | Use at an assay dependent concentration.<br>ab199376 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody. |
| ICC/IF | | Use at an assay dependent concentration. |

## Target

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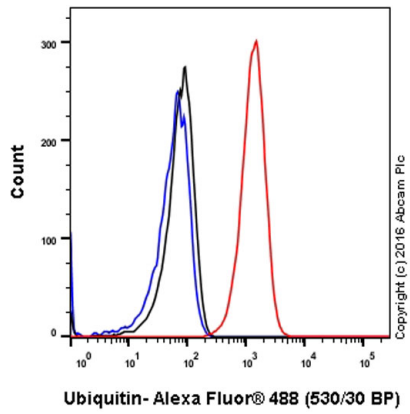
| **Relevance** | Function: Ubiquitin exists either covalently attached to another protein, or free (unanchored). When covalently bound, it is conjugated to target proteins via an isopeptide bond either as a monomer (monoubiquitin), a polymer linked via different Lys residues of the ubiquitin (polyubiquitin 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 lysosomal degradation; 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. Similarity: Belongs to the ubiquitin family. Contains 3 ubiquitin-like domains. |
| **Cellular localization** | Cell Membrane, Cytoplasmic and Nuclear |

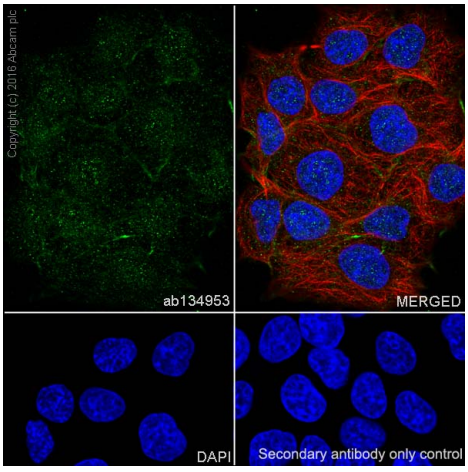
## Images




Flow Cytometry - Anti-Ubiquitin antibody [EPR8830] - BSA and Azide free (ab230145)

Flow Cytometry analysis of JAR (Human placenta choriocarcinoma cell line) cells labeling Ubiquitin with purified ab134953 at 1:70 dilution (10 ug/ml) (red). Cells were fixed with 4% Paraformaldehyde. A Goat anti rabbit IgG (Alexa Fluor® 488) secondary antibody was used at 1:2000 dilution. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).

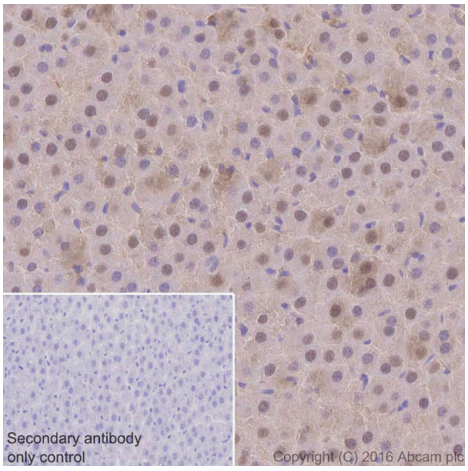
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab134953).




Immunocytochemistry/ Immunofluorescence - Anti-Ubiquitin antibody [EPR8830] - BSA and Azide free (ab230145)

Immunocytochemistry/ Immunofluorescence analysis of JAR (Human placenta choriocarcinoma cell line) cells labeling Ubiquitin with Purified ab134953 at 1:100 dilution (7.2µg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5 µg/ml). ab150077 Goat anti rabbit IgG(Alexa Fluor® 488) was used as the secondary antibody at 1:1000 dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.

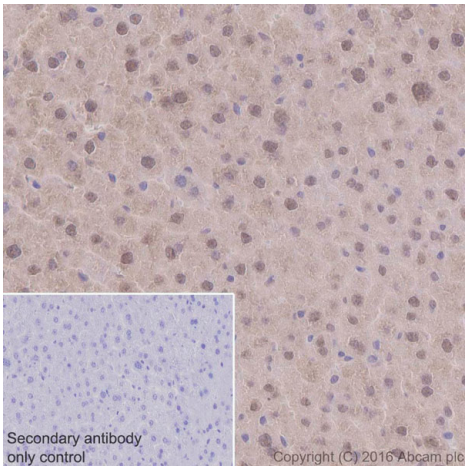
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab134953).




Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ubiquitin antibody [EPR8830] - BSA and Azide free (ab230145)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Rat liver tissue sections labeling Ubiquitin with Purified ab134953 at 1:800 dilution (0.9 µg/ml). Heat mediated antigen retrieval was performed using Perform heat mediated antigen retrieval using EDTA Buffer, PH9. Tissue was counterstained with Hematoxylin. ab97051 Goat Anti-Rabbit IgG H&L (HRP)
secondary antibody was used at 1:500 dilution. PBS instead of the primary antibody was used as the negative control.

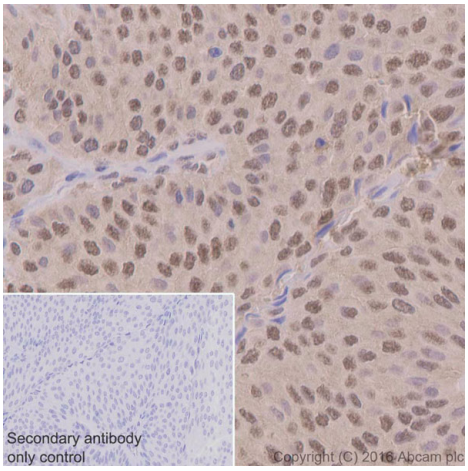
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab134953).




Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ubiquitin antibody [EPR8830] - BSA and Azide free (ab230145)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Mouse liver tissue sections labeling Ubiquitin with Purified ab134953 at 1:800 dilution (0.9 µg/ml). Heat mediated antigen retrieval was performed using Perform heat mediated antigen retrieval using EDTA Buffer, PH9. Tissue was counterstained with Hematoxylin. ab97051 Goat Anti-Rabbit IgG H&L (HRP)
secondary antibody was used at 1:500 dilution. PBS instead of the primary antibody was used as the negative control.

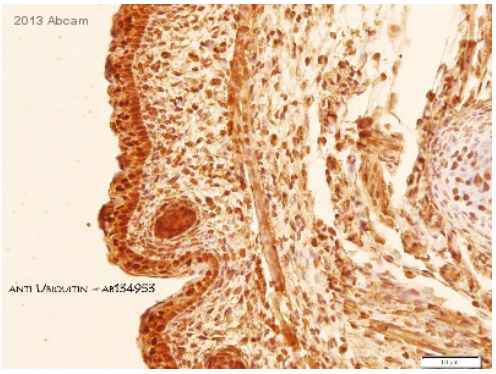
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab134953).




Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ubiquitin antibody [EPR8830] - BSA and Azide free (ab230145)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human bladder carcinoma tissue sections labeling Ubiquitin with Purified ab134953 at 1:800 dilution (0.9 µg/ml). Heat mediated antigen retrieval was performed using Perform heat mediated antigen retrieval using EDTA Buffer, PH9. Tissue was counterstained with Hematoxylin. ab97051 Goat Anti-Rabbit IgG H&L (HRP) secondary antibody was used at 1:500 dilution. PBS instead of the primary antibody was used as the negative control.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab134953).

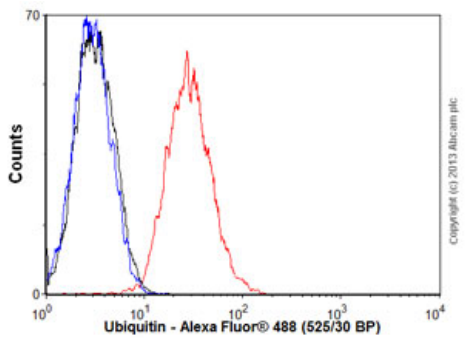



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ubiquitin antibody [EPR8830] - BSA and Azide free (ab230145)

This image is courtesy of an anonymous Abreview.

ab134953 staining Ubiquitin in Mouse embryo tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffin-embedded sections). Tissue was fixed with formaldehyde and blocked with 1% FBS/BSA for 1 hour at room temperature; antigen retrieval was by heat mediation in citrate buffer, pH6. Samples were incubated with primary antibody (1/100 in 1% FBS/BSA) for 16 hours at 4°C. An undiluted HRP-conjugated Goat anti-rabbit IgG polyclonal was used as the secondary antibody.

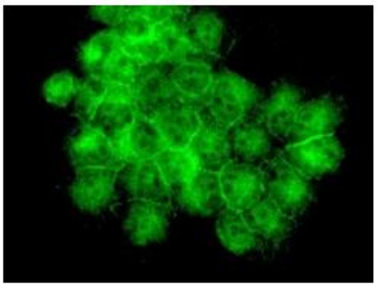
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab134953).




Flow Cytometry - Anti-Ubiquitin antibody [EPR8830] - BSA and Azide free (ab230145)

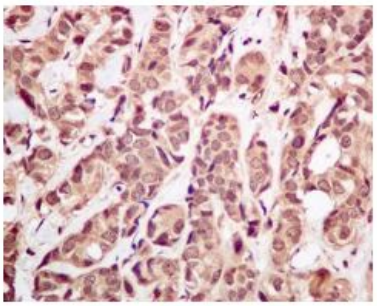
Overlay histogram showing HepG2 cells stained with ab134953 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab134953, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit IgG (H&L) (ab150077) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (0.1µg/1x$10^6$ cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This antibody gave a positive signal in HepG2 cells fixed with 4% paraformaldehyde (10

min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab134953).



Immunocytochemistry/ Immunofluorescence - Anti-Ubiquitin antibody [EPR8830] - BSA and Azide free (ab230145)

Immunofluorescent staining of JAR cells labelling Ubiquitin using ab134953 at 1/250 dilution
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab134953).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ubiquitin antibody [EPR8830] - BSA and Azide free (ab230145)

Immunohistochemical analysis of paraffin embedded Human breast carcinoma tissue labelling Ubiquitin with ab134953 at 1/250 dilution
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab134953).



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