# abcam

## Product datasheet

## Anti-UBE2M/UBC12 antibody [EPR5333] - BSA and Azide free ab236056



### 5 Images

#### Overview

**Product name** Anti-UBE2M/UBC12 antibody [EPR5333] - BSA and Azide free

**Description** Rabbit monoclonal [EPR5333] to UBE2M/UBC12 - BSA and Azide free

**Host species** Rabbit

**Tested applications** Suitable for: Flow Cyt (Intra), IHC-P, WB, ICC/IF

**Species reactivity** Reacts with: Human

Predicted to work with: Mouse, Rat

**Immunogen** Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: 293T and Ramos cell lysates. IHC-P: Human colonic adenocarcinoma and lung carcinoma

tissues. ICC/IF: Ramos cells.

**General notes** ab236056 is the carrier-free version of ab109507.

> Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cellbased assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

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.

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar<sup>®</sup> is a trademark of Fluidigm Canada Inc.

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 production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit

#### **Properties**

Form Liquid

**Storage instructions** Shipped at 4°C. Store at +4°C. Do Not Freeze.

Storage buffer pH: 7.2

Constituent: PBS

Carrier free Yes

Purity Protein A purified

Clonality Monoclonal
Clone number EPR5333

**Isotype** IgG

#### **Applications**

The Abpromise guarantee Our Abpromise guarantee covers the use of ab236056 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
Flow Cyt (Intra)		Use at an assay dependent concentration. <b>ab172730</b> - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
WB		Use at an assay dependent concentration. Detects a band of approximately 20 kDa (predicted molecular weight: 21 kDa).
ICC/IF		Use at an assay dependent concentration.

## Target

Function Accepts the ubiquitin-like protein NEDD8 from the UBA3-NAE1 E1 complex and catalyzes its

covalent attachment to other proteins. The specific interaction with the E3 ubiquitin ligase RBX1, but not RBX2, suggests that the RBX1-UBE2M complex neddylates specific target proteins, such

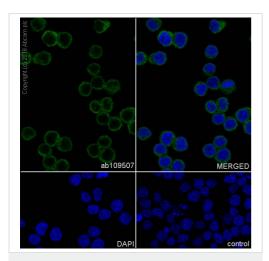
as CUL1, CUL2, CUL3 and CUL4. Involved in cell proliferation.

**Pathway** Protein modification; protein neddylation.

**Sequence similarities**Belongs to the ubiquitin-conjugating enzyme family. UBC12 subfamily.

**Domain**Both the N-terminal docking peptide and the catalytic core domain must bind the UBA3-NAE1

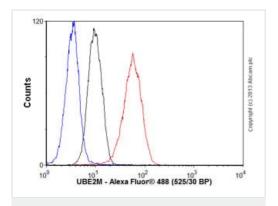
complex simultaneously for optimal transfer of NEDD8.



Immunocytochemistry/ Immunofluorescence - Anti-UBE2M/UBC12 antibody [EPR5333] - BSA and Azide free (ab236056)

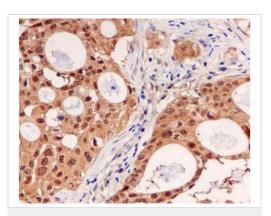
Immunocytochemistry/ Immunofluorescence analysis of Ramos (human Burkitt's lymphoma B lymphocyte) cells labeling UBE2M/UBC12 with purified  $\underline{ab109507}$  at 1/250 dilution (8  $\mu$ g/mL). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with None. Goat anti rabbit lgG (Alexa Fluor® 488,  $\underline{ab150077}$ ) was used as the secondary antibody at 1/1000 (2  $\mu$ g/mL) dilution. DAPI (blue) was used as nuclear counterstain.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab109507).



Flow Cytometry (Intracellular) - Anti-UBE2M/UBC12 antibody [EPR5333] - BSA and Azide free (ab236056)

Overlay histogram showing Ramos cells stained with ab109507 (red line). The cells were fixed with 4% paraformaldehyde (10 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 (ab109507, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit lgG (H&L) (ab150077) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit lgG (monoclonal) (0.1µg/1x10<sup>6</sup> 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 Ramos cells fixed with 80% methanol (5 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 (ab109507).

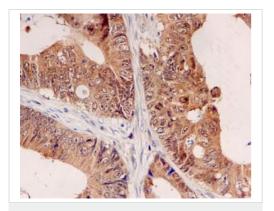


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-UBE2M/UBC12 antibody [EPR5333] - BSA and Azide free (ab236056)

Immunohistochemical analysis of paraffin-embedded Human lung carcinoma tissue using <u>ab109507</u> at a dilution of 1/100.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab109507</u>).

Heat mediated antigen retrieval was performed with citrate buffer pH 6 before commencing with IHC staining protocol.

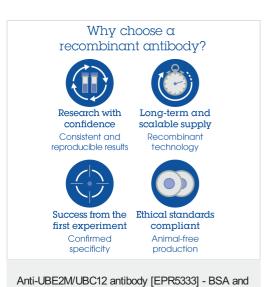


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-UBE2M/UBC12 antibody [EPR5333] - BSA and Azide free (ab236056)

Immunohistochemical analysis of paraffin-embedded human colonic adenocarcinoma tissue using **ab109507** at a dilution of 1/100.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab109507).

Heat mediated antigen retrieval was performed with citrate buffer pH 6 before commencing with IHC staining protocol.



Azide free (ab236056)

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