

Anti-UBE3A antibody [EPR23077-14] ab272168

リコンビナント **RabMAb**

画像数 11

製品の概要

製品名	Anti-UBE3A antibody [EPR23077-14]
製品の詳細	Rabbit monoclonal [EPR23077-14] to UBE3A
由来種	Rabbit
アプリケーション	適用あり: Indirect ELISA, IP, WB, ICC/IF, Flow Cyt (Intra) 適用なし: IHC-P
種交差性	交差種: Mouse, Rat, Human
免疫原	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
ポジティブ・コントロール	WB: HeLa, K562, HepG2, A549, HEK-293T, PC-12, PC-12 (treated with 10 uM MG-132 for 4 hours), RAW 264.7 and RAW 264.7 (treated with 10 uM MG-132 for 4 hours) whole cell lysates; Mouse spleen tissue lysate, Rat brain tissue lysate. ICC/IF: HeLa and RAW 264.7 cells. Flow Cyt (intra): HeLa and RAW 264.7 cells. IP: K562 and RAW 264.7 whole cell lysates.
特記事項	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 .

製品の特性

製品の状態	Liquid
保存方法	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.
バッファー	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
精製度	Protein A purified
ポリ/モノ	モノクローナル

クローン名	EPR23077-14
アイソタイプ	IgG

アプリケーション

The Abpromise guarantee **Abpromise保証は、**次のテスト済みアプリケーションにおけるab272168の使用に適用されます
アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

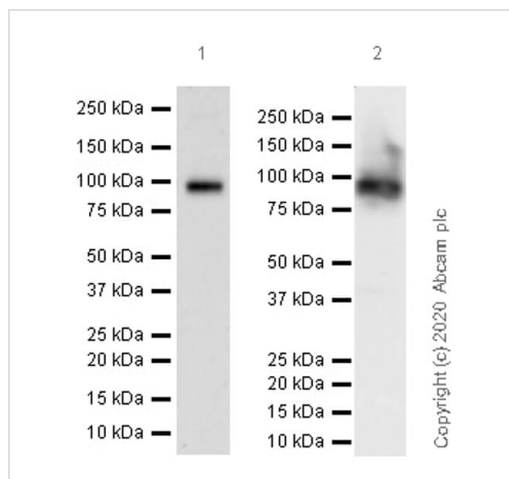
アプリケーション	Abreviews	特記事項
Indirect ELISA		Use a concentration of 1 µg/ml.
IP		1/30.
WB		1/1000. Detects a band of approximately 100, 37 kDa (predicted molecular weight: 100 kDa).
ICC/IF		1/100.
Flow Cyt (Intra)		1/50.

追加情報 Is unsuitable for IHC-P.

ターゲット情報

機能	E3 ubiquitin-protein ligase which accepts ubiquitin from an E2 ubiquitin-conjugating enzyme in the form of a thioester and transfers it to its substrates. Several substrates have been identified including the RAD23A and RAD23B, MCM7 (which is involved in DNA replication), annexin A1, the PML tumor suppressor, and the cell cycle regulator CDKN1B. Catalyzes the high-risk human papilloma virus E6-mediated ubiquitination of p53/TP53, contributing to the neoplastic progression of cells infected by these viruses. Additionally, may function as a cellular quality control ubiquitin ligase by helping the degradation of the cytoplasmic misfolded proteins. Finally, UBE3A also promotes its own degradation in vivo. Plays an important role in the regulation of the circadian clock: involved in the ubiquitination of the core clock component ARNTL/BMAL1, leading to its proteasomal degradation (PubMed:24728990).
パスウェイ	Protein modification; protein ubiquitination.
関連疾患	Angelman syndrome
配列類似性	Contains 1 HECT (E6AP-type E3 ubiquitin-protein ligase) domain.
翻訳後修飾	Phosphorylation at Tyr-659 by ABL1 impairs E3 ligase activity and protects p53/TP53 from degradation in (HPV)-infected cells.
細胞内局在	Nucleus. Cytoplasm.

画像



Western blot - Anti-UBE3A antibody [EPR23077-14] (ab272168)

All lanes : Anti-UBE3A antibody [EPR23077-14] (ab272168) at 1/1000 dilution

Lane 1 : HeLa (human cervix adenocarcinoma epithelial cell) treated with 10 μ M MG-132 for 24 hours, whole cell lysate

Lane 2 : HEK-293T (human embryonic kidney epithelial cell) whole cell lysate

Lysates/proteins at 20 μ g per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (**ab97051**) at 1/100000 dilution

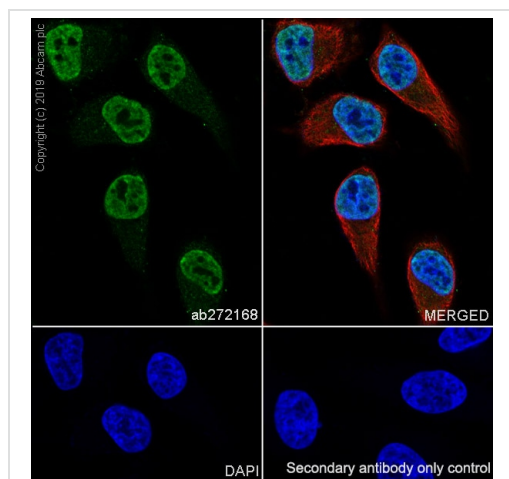
Predicted band size: 100 kDa

Observed band size: 100 kDa

Blocking and diluting buffer and concentration: 5% NFDM/TBST.

Fresh lysate was used in lane 2.

Exposure time: Lane 1: 3 minutes; Lane 2: 48 seconds.

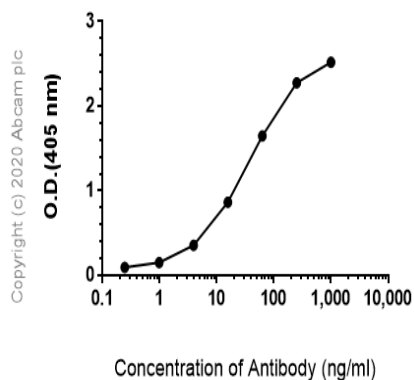


Immunocytochemistry/ Immunofluorescence - Anti-UBE3A antibody [EPR23077-14] (ab272168)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa cells labelling UBE3A with ab272168 at 1/100 dilution, followed by **ab150077** Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) antibody at 1/1000 dilution (Green). Confocal image showing nuclear and weak cytoplasmic staining in HeLa cell line **ab195889**. Anti-alpha Tubulin antibody (Alexa Fluor[®] 594) was used to counterstain tubulin at 1/200 dilution (Red). The Nuclear counterstain was DAPI (Blue).

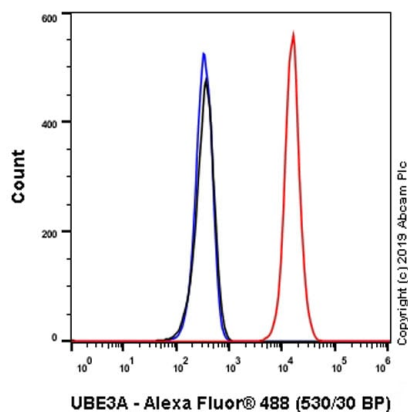
Secondary antibody only control: Secondary antibody is **ab150077** Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) at 1000 dilution.

Indirect ELISA antibody dose-response curve



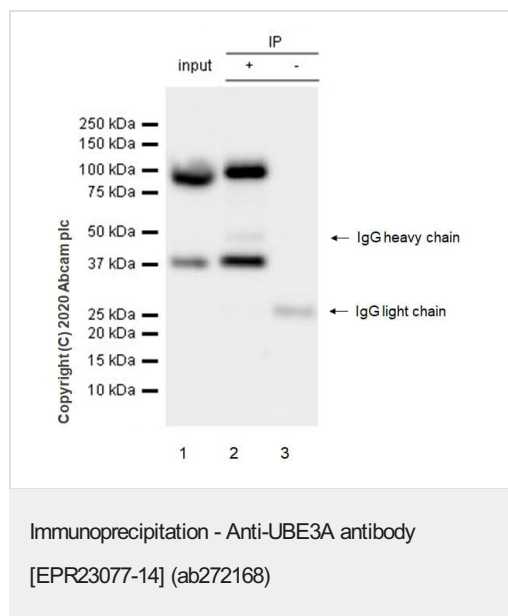
Indirect ELISA - Anti-UBE3A antibody [EPR23077-14] (ab272168)

Indirect ELISA using ab272168 at varying antibody concentrations (1000-0 ng/ml) and Human UBE3A antigen at 1000 ng/ml. Alkaline Phosphatase-conjugated AffiniPure Goat Anti-Rabbit IgG (H+L) at 1/2500 dilution was used as a secondary antibody.



Flow Cytometry (Intracellular) - Anti-UBE3A antibody [EPR23077-14] (ab272168)

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed, 90% methanol-permeabilized HeLa (Human cervix adenocarcinoma epithelial cell) cells labelling UBE3A with ab272168 at 1/500 dilution (Red) compared with a Rabbit monoclonal IgG (**ab172730**) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) at 1/2000 dilution was used as the secondary antibody.



UBE3A was immunoprecipitated from 0.35 mg K-562 (human chronic myelogenous leukemia lymphoblast) whole cell lysate with ab272168 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab272168 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) ([ab131366](#)) was used at 1/5000 dilution.

Lane 1: K-562 (human chronic myelogenous leukemia lymphoblast) whole cell lysate 10 ug

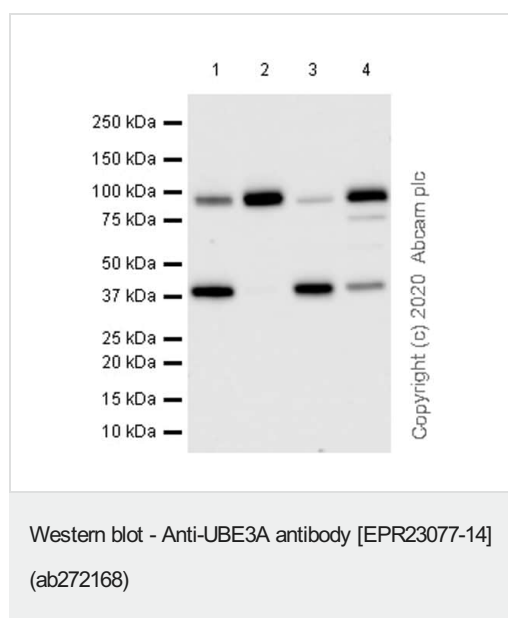
Lane 2: ab272168 IP in K-562 whole cell lysate

Lane 3: Rabbit monoclonal IgG ([ab172730](#)) instead of ab272168 in K-562 whole cell lysate

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 15 seconds

A 37 kDa degraded band is observed.



All lanes : Anti-UBE3A antibody [EPR23077-14] (ab272168) at 1/1000 dilution

Lane 1 : PC-12 (rat adrenal gland pheochromocytoma) whole cell lysate

Lane 2 : PC-12 treated with 10 μM MG-132 for 4 hours, whole cell lysate

Lane 3 : RAW 264.7 (mouse abelson murine leukemia virus-induced tumor macrophage) whole cell lysate

Lane 4 : RAW 264.7 treated with 10 μM MG-132 for 4 hours, whole cell lysate

Lysates/proteins at 20 μg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/100000 dilution

Predicted band size: 100 kDa

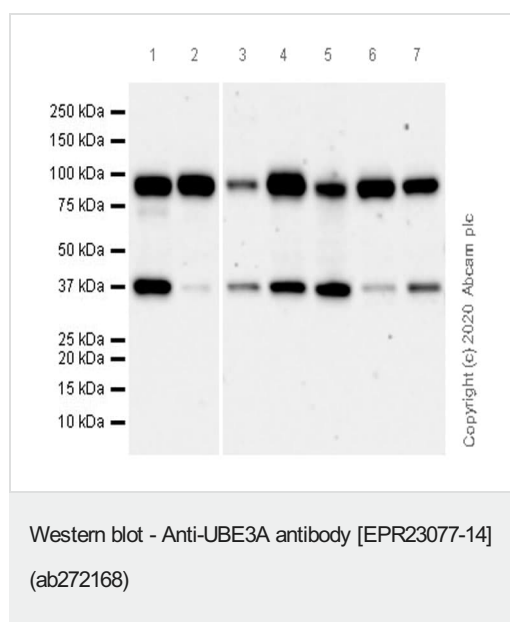
Observed band size: 100,37 kDa

Blocking and dilution buffer: 5% NFDM/TBST.

Exposure time: 3 minutes.

The 37-kDa band might be a degraded fragment (not verified), which can be inhibited/reduced by MG-132 treatment (lanes 2 and

4).



All lanes : Anti-UBE3A antibody [EPR23077-14] (ab272168) at 1/1000 dilution

Lane 1 : Mouse spleen tissue lysate

Lane 2 : Rat brain tissue lysate

Lane 3 : HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate

Lane 4 : K-562 (human chronic myelogenous leukemia lymphoblast) whole cell lysate

Lane 5 : HepG2 (human hepatocellular carcinoma epithelial cell) whole cell lysate

Lane 6 : HEK-293T (human embryonic kidney epithelial cell) whole cell lysate

Lane 7 : A549 (human lung carcinoma epithelial cell) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated ([ab97051](#)) at 1/100000 dilution

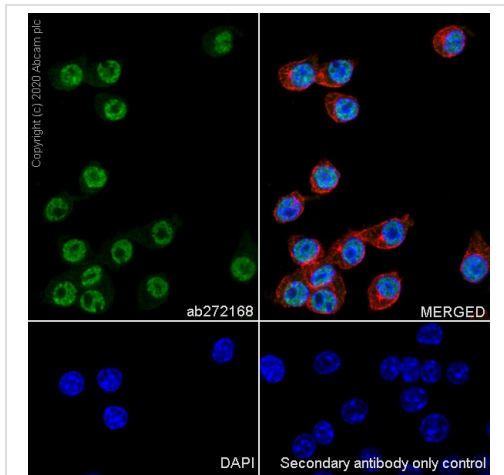
Predicted band size: 100 kDa

Observed band size: 100,37 kDa

Exposure time: 3 minutes

Blocking and diluting buffer and concentration: 5% NFDm/TBST.

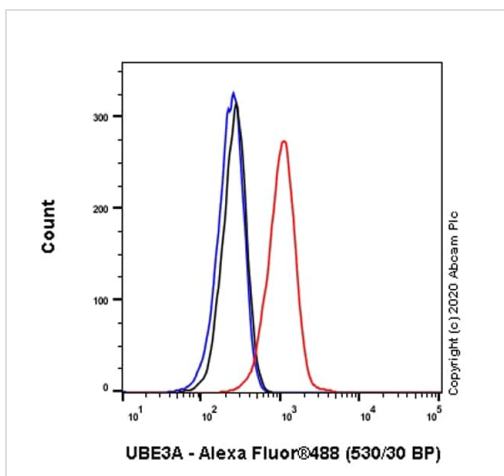
The 37 kDa band might be a degraded band (not verified). MG132 treatment or freshly made lysates can decrease the degradation.



Immunocytochemistry/ Immunofluorescence - Anti-UBE3A antibody [EPR23077-14] (ab272168)

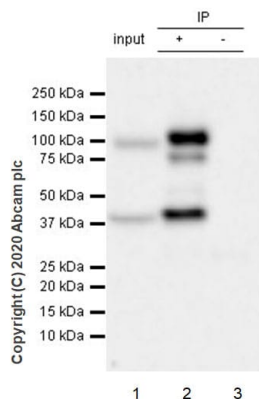
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized RAW 264.7 cells labelling UBE3A with ab272168 at 1/100 dilution, followed by **ab150077** Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) antibody at 1/1000 dilution (Green). Confocal image showing nuclear and weak cytoplasmic staining in RAW 264.7 cell line. **ab195889** Anti-alpha Tubulin antibody (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is **ab150077** Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1000 dilution.



Flow Cytometry (Intracellular) - Anti-UBE3A antibody [EPR23077-14] (ab272168)

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed, 90% methanol-permeabilized RAW 264.7 (mouse abelson murine leukemia virus-induced tumor macrophage) cells labelling UBE3A with ab272168 at 1/50 dilution (Red) compared with a Rabbit monoclonal IgG (**ab172730**) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) at 1/2000 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-UBE3A antibody
[EPR23077-14] (ab272168)

UBE3A was immunoprecipitated from 0.35 mg RAW 264.7 (mouse abelson murine leukemia virus-induced tumor macrophage) (treated with 10 μ M MG-132 for 4 hours) whole cell lysate with ab272168 at 1/30 dilution (2 μ g in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab272168 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) ([ab131366](#)) was used at 1/5000 dilution.

Lane 1: RAW 264.7 (mouse abelson murine leukemia virus-induced tumor macrophage) treated with 10 μ M MG-132 for 4 hours, whole cell lysate 10 μ g

Lane 2: ab272168 IP in RAW 264.7 treated with 10 μ M MG-132 for 4 hours, whole cell lysate

Lane 3: Rabbit monoclonal IgG ([ab172730](#)) instead of ab272168 in RAW 264.7 treated with 10 μ M MG-132 for 4 hours, whole cell lysate

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 10 seconds

A 37 kDa degraded band is observed.

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-UBE3A antibody [EPR23077-14] (ab272168)

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

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