abcam

ab185620

Anti-CDKN2A/p14ARF antibody [EPR17878]

Recombinant | RabMAb | KO Validated | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 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	EPR17878
Purification technique	Affinity purification Protein A
Concentration	1.145 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Human
Dilution info	1/80
Notes	-

WB

Tested

Species	Human
Dilution info	1/1000
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/250
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/120
Notes	ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Target data

See full target information CDKN2A

Function

Capable of inducing cell cycle arrest in G1 and G2 phases. Acts as a tumor suppressor. Binds to MDM2 and blocks its nucleocytoplasmic shuttling by sequestering it in the nucleolus. This inhibits the oncogenic action of MDM2 by blocking MDM2-induced degradation of p53 and enhancing p53-dependent transactivation and apoptosis. Also induces G2 arrest and apoptosis in a p53-independent manner by preventing the activation of cyclin B1/CDC2 complexes. Binds to BCL6 and down-regulates BCL6-induced transcriptional repression. Binds to E2F1 and MYC and blocks their transcriptional activator activity but has no effect on MYC transcriptional repression. Binds to TOP1/TOPOI and stimulates its activity. This complex binds to rRNA gene promoters and may play a role in rRNA transcription and/or maturation. Interacts with NPM1/B23 and promotes its polyubiquitination and degradation, thus inhibiting rRNA processing. Plays a role in inhibiting ribosome biogenesis, perhaps by binding to the nucleolar localization sequence of transcription termination factor TTF1, and thereby preventing nucleolar localization of TTF1 (By similarity). Interacts with COMMD1 and promotes

its 'Lys63'-linked polyubiquitination. Interacts with UBE2I/UBC9 and enhances sumoylation of a number of its binding partners including MDM2 and E2F1. Binds to HUWE1 and represses its ubiquitin ligase activity. May play a role in controlling cell proliferation and apoptosis during mammary gland development. Isoform smARF. May be involved in regulation of autophagy and caspase-independent cell death; the short-lived mitochondrial isoform is stabilized by C1QBP.

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

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.

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.

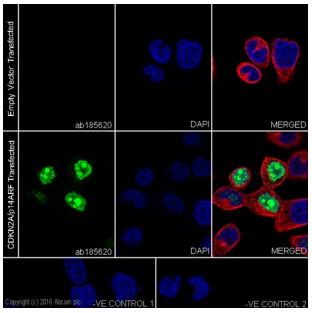
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

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.

---

6 product images



Immunocytochemistry/ Immunofluorescence - Anti-CDKN2A/p14ARF antibody [EPR17878] (ab185620)

CDKN2A/p14ARF Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-CDKN2A/p14ARF antibody

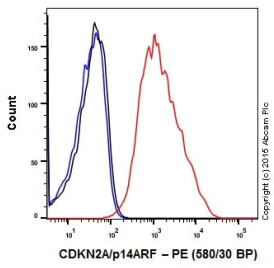
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (human cervix adenocarcinoma) cells transfected with CDKN2A/p14ARF or empty vector, labeling CDKN2A/p14ARF with ab185620 at 1/250 dilution, followed by Goat Anti-Rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution
The nuclear counterstain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) at 1/1000 dilution and Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) preadsorbed (ab150120) at 1/1000 dilution (red).

The negative controls are as follows:-

-ve control 1: ab185620 at 1/250 dilution followed by ab150120 at 1/1000 dilution.

-ve control 2: ab7291 at 1/1000 dilution followed by ab150077 at 1/1000 dilution.

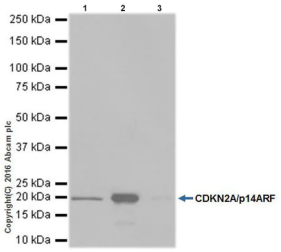


Flow Cytometry (Intracellular) - Anti-CDKN2A/p14ARF antibody [EPR17878] (ab185620)

CDKN2A/p14ARF Flow Cytometry (Intracellular) staining using rabbit Anti-CDKN2A/p14ARF antibody

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed HeLa (Human epithelial cell line from cervix adenocarcinoma) cells transfected with CDKN2A/p14ARF-GFP labeling

CDKN2A/p14ARF with ab185620 at 1/120 dilution (red) compared with a Rabbit IgG,monoclonal [EPR25A] - Isotype Control (ab172730) (black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti Rabbit IgG (PE) at 1/150 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-CDKN2A/p14ARF antibody [EPR17878] (ab185620)

CDKN2A/p14ARF was immunoprecipitated from 1mg of CDKN2A/p14ARF transfected HeLa (Human epithelial cells from cervix adenocarcinoma) whole cell lysate with ab185620 at 1/80 dilution. Western blot was performed from the immunoprecipitate using ab185620 at 1/1000 dilution.

VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/10000 dilution.

Lane 1: CDKN2A/p14ARF transfected HeLa whole cell lysate, 10µg (Input).

Lane 2: ab185620 IP in CDKN2A/p14ARF transfected HeLa whole cell lysate.

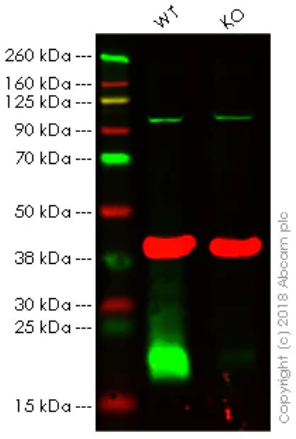
Lane 3: Rabbit IgG,monoclonal[EPR25A] - Isotype Control (ab172730) instead of ab185620 in CDKN2A/p14ARF transfected HeLa whole cell lysate.

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

Exposure time: 1 second.

All lanes:
Immunoprecipitation - Anti-CDKN2A/p14ARF antibody [EPR17878] (ab185620)

Predicted band size: 14 kDa



Western blot - Anti-CDKN2A/p14ARF antibody [EPR17878] (ab185620)

CDKN2A/p14ARF Western blot staining using rabbit Anti-CDKN2A/p14ARF antibody

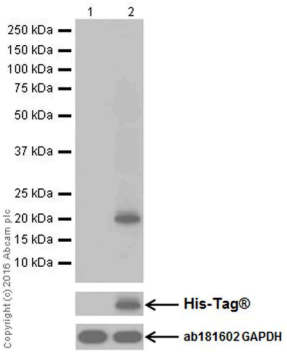
Lanes 1 - 2: Merged signal (red and green). Green - ab185620 observed at 14 kDa. Red - loading control, ab9484, observed at 37 kDa.

ab185620 was shown to specifically react with CDKN2A/p14ARF in wild-type HeLa cells as signal was lost in CDKN2A knockout cells. Wild-type and CDKN2A knockout samples were subjected to SDS-PAGE. ab185620 and ab9484 (Mouse anti-GAPDH 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.
All lanes:
Western blot - Anti-CDKN2A/p14ARF antibody [EPR17878] (ab185620) at 1/1000 dilution

Lane 1:
Wild-type HeLa whole cell lysate at 20 µg

Lane 2:
CDKN2A knockout HeLa whole cell lysate at 20 µg

Predicted band size: 14 kDa



Western blot - Anti-CDKN2A/p14ARF antibody [EPR17878] (ab185620)

Blocking/Dilution buffer: 5% NFDM/TBST.
All lanes:
Western blot - Anti-CDKN2A/p14ARF antibody [EPR17878] (ab185620) at 1/5000 dilution

Lane 1:
HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate transfected with empty vector (vector control) at 10 µg

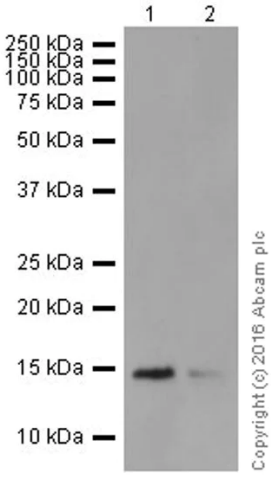
Lane 2:
HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate transfected with CDKN2A/p14ARF with His-tag at 10 µg

Secondary

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

Predicted band size: 14 kDa

Exposure time: 1s



Western blot - Anti-CDKN2A/p14ARF antibody [EPR17878] (ab185620)

Blocking/Dilution buffer: 5% NFDM/TBST.
The expression level of p14 ARF is relatively low in most cell types and will be induced by oncogene expression. (PMID: 9694807; PMID: 9694806).

All lanes:
Western blot - Anti-CDKN2A/p14ARF antibody [EPR17878] (ab185620) at 1/1000 dilution

Lane 1:
PC-3 (Human prostate adenocarcinoma cell line) whole cell lysate at 20 µg

Lane 2:
HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate at 20 µg

Secondary

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

Predicted band size: 14 kDa

Observed band size: 14 kDa

Exposure time: 3min

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.