

Product datasheet

Anti-CDKN2A/p14ARF antibody [EPR17878] ab185620

KO VALIDATED

Recombinant

RabMAb

[4 References](#) [7 Images](#)

Overview

Product name	Anti-CDKN2A/p14ARF antibody [EPR17878]
Description	Rabbit monoclonal [EPR17878] to CDKN2A/p14ARF
Host species	Rabbit
Tested applications	Suitable for: Flow Cyt (Intra), ICC/IF, IP, WB
Species reactivity	Reacts with: Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: HeLa whole cell lysate transfected with CDKN2A/p14ARF with His-tag; PC-3 and HEK-293 whole cell lysates. ICC/IF: HeLa cells transfected with CDKN2A/p14ARF. Flow Cyt (intra): HeLa cells transfected with CDKN2A/p14ARF-GFP. IP: CDKN2A/p14ARF transfected HeLa whole cell lysate.
General notes	<p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <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 <p>For more information see here.</p> <p>Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents.</p>

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	<p>pH: 7.2</p> <p>Preservative: 0.01% Sodium azide</p> <p>Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA</p>
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR17878

Isotype

IgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab185620 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)		1/120. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
ICC/IF		1/250.
IP		1/80.
WB		1/1000. Detects a band of approximately 14 kDa (predicted molecular weight: 14 kDa).

Target

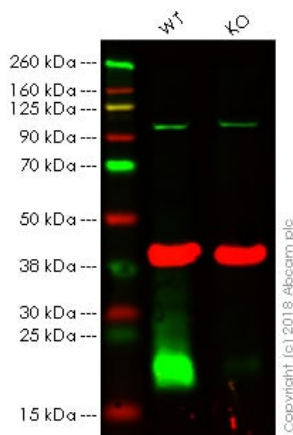
Relevance

The gene for CDK2NA generates several transcripts/proteins which differ from each other in their first exons. Three of these transcripts are generated by alternative splicing (isoform 1 a.k.a p16INK4A, isoform 2 and isoform 3 a.k.a p12), two of which are known to function as inhibitors of CDK4 kinase. One other transcript that is generated from this gene contains an alternate reading frame (ARF), with the first exon located 20kb upstream of the remainder of the gene (isoform 4 a.k.a. p14ARF, p19ARF, ARF). In spite of the structural and some functional differences, all the proteins encoded by the CDKN2A gene are involved in cell cycle G1 control.

Cellular localization

Cytoplasmic and Nuclear

Images



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

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

Lane 1 : Wild-type HeLa whole cell lysate

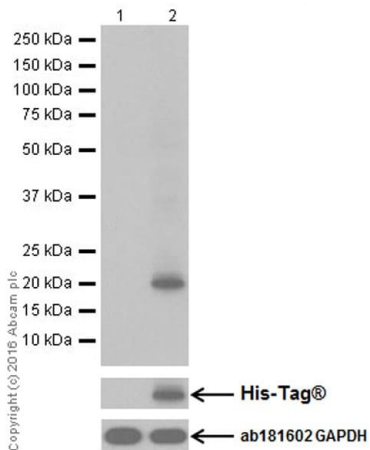
Lane 2 : CDKN2A knockout HeLa whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 14 kDa

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.



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

All lanes : 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)

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

Lysates/proteins at 10 µg per lane.

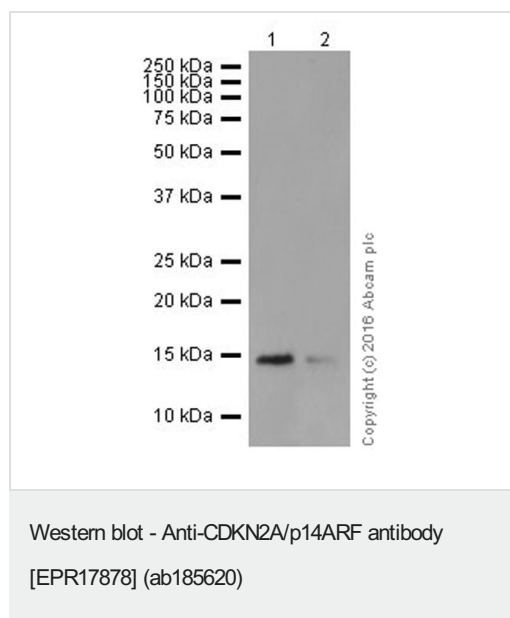
Secondary

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

Predicted band size: 14 kDa

Exposure time: 1 second

Blocking/Dilution buffer: 5% NFDM/TBST.



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

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

Lane 2 : HEK-293 (Human epithelial cell line from embryonic kidney) 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

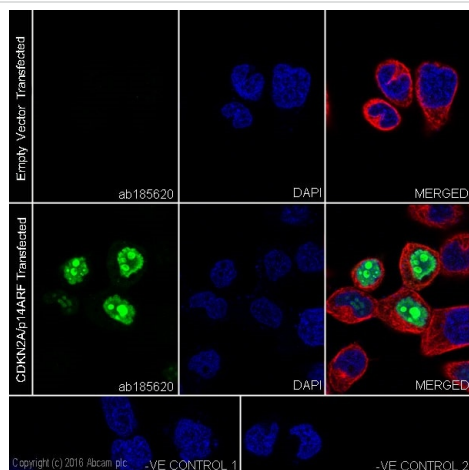
Predicted band size: 14 kDa

Observed band size: 14 kDa

Exposure time: 3 minutes

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).



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

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

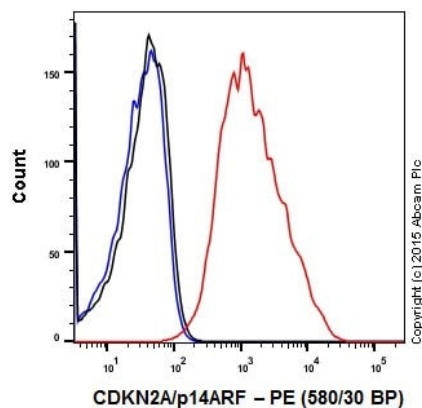
The nuclear counterstain is DAPI (blue).

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:-

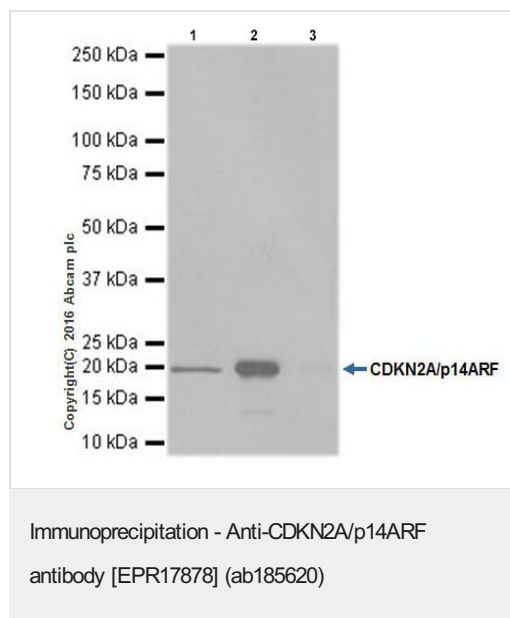
-ve control 1: ab185620 at 1/250 dilution followed by **ab150120** at 1/1000 dilution.

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



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

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.



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.

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-CDKN2A/p14ARF antibody [EPR17878] (ab185620)

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

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