abcam

ab79398

Anti-RPA70 antibody [EPR3472]

Anti-RPA70 antibody [EPR3472] (ab79398) is a rabbit monoclonal antibody detecting RPA70 in **Western Blot, Flow Cytometry (Intra), IP, IHC-P, ICC/IF**. Suitable for **Human**.

- Biophysical QC for unrivalled batch-batch consistency
- Over 70 publications
- Trusted since 2009

Recombinant | RabMAb | 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	EPR3472
Purification technique	Affinity purification Protein A
Concentration	0.29 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Human
Dilution info	1/100 - 1/250
Notes	See IHC antigen retrieval protocols. Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

IP

Tested

Species	Human
Dilution info	1/10 - 1/20
Notes	-

WB

Tested

Species	Human
Dilution info	1/1000
Notes	**For unpurified use at 1/2000 - 1/5000.**

ICC/IF

Tested

Species	Human
Dilution info	1/100 - 1/250
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/80
Notes	For unpurified use at 1/50.ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Target data

See full target information RPA1

Function	As part of the heterotrimeric replication protein A complex (RPA/RP-A), binds and stabilizes single-stranded DNA intermediates that form during DNA replication or upon DNA stress. It prevents their reannealing and in parallel, recruits and activates different proteins and complexes involved in DNA metabolism (PubMed:17596542, PubMed:27723717, PubMed:27723720). Thereby, it plays an essential role both in DNA replication and the cellular response to DNA damage (PubMed:9430682). In the cellular response to DNA damage, the RPA complex controls DNA repair and DNA damage checkpoint activation. Through recruitment of ATRIP activates the ATR kinase a master regulator of the DNA damage response (PubMed:24332808). It is required for the recruitment of the DNA double-strand break repair factors RAD51 and RAD52 to chromatin in response to DNA damage (PubMed:17765923). Also recruits to sites of DNA damage proteins like XPA and XPG that are involved in nucleotide excision repair and is required for this mechanism of DNA repair (PubMed:7697716). Also plays a role in base excision repair (BER) probably through interaction with UNG (PubMed:9765279). Also recruits SMARCAL1/HARP, which is involved in replication fork restart, to sites of DNA damage. Plays a role in telomere maintenance (PubMed:17959650, PubMed:34767620). As part of the alternative replication protein A complex, aRPA, binds single-stranded DNA and probably plays a role in DNA repair. Compared to the RPA2-containing, canonical RPA complex, may not support chromosomal DNA replication and cell cycle progression through S-phase. The aRPA may not promote efficient priming by DNA polymerase alpha but could support DNA synthesis by polymerase delta in presence of PCNA and replication factor C (RFC), the dual incision/excision reaction of nucleotide excision repair and RAD51-dependent strand exchange (PubMed:19996105). RPA stimulates 5'-3' helicase activity of the BRIP1/FANCJ (PubMed:17596542).

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	Stable for 12 months at -20°C

Notes

What is this antibody validated in?
Anti-RPA70 antibody [EPR3472] (ab79398) is a rabbit recombinant monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Intra), Immunoprecipitation (IP), Immunohistochemistry (IHC-P), Immunocytochemistry/immunofluorescence (ICC/IF) in Human samples.

What is the molecular weight of RPA70?
Anti-RPA70 [EPR3472] (ab79398) specifically detects a band for RPA70 (UniProt: P27694) at a molecular weight of 70kDa.

Trusted by the scientific community
Anti-RPA70 [EPR3472] (ab79398) was first used in a scientific publication in 2009 and has been cited over 70 times in peer-reviewed journals.

Reviewed by scientists
Anti-RPA70 [EPR3472] (ab79398) has over 5 independent reviews from customers.

Other related products
We have a range of other formats of antibody clone [EPR3472] also available for your convenience: ab79398, Alexa Fluor® 488 - ab199097, Alexa Fluor® 647 - ab199240, Carrier free - ab239890, PE - ab303077, APC - ab303078, HRP - ab303079, Alkaline Phosphatase - ab308863, Alexa Fluor® 594 - ab310589, Alexa Fluor® 555 - ab312119, Alexa Fluor® 568 - ab312603, Alexa Fluor® 750 - ab321735

Species reactivity
Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

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.

Trial sizes available!
Test your antibody or perform pre-screening before committing to a larger quantity. Sold in 20µl. Discover our selection of trial-size 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.

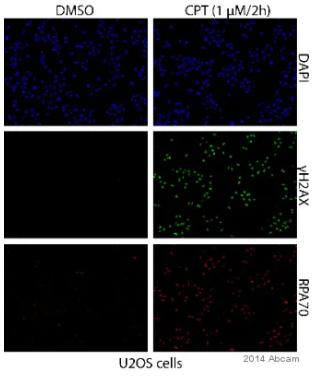
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

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.

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11 product images

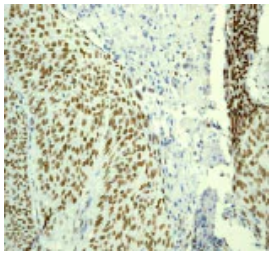


This image is courtesy of a customer review submitted by Remi Buisson.

Immunocytochemistry/ Immunofluorescence - Anti-RPA70 antibody [EPR3472] (ab79398)

RPA70 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-RPA70 antibody

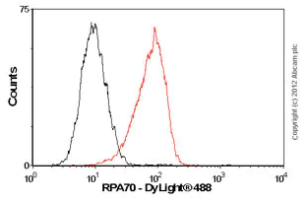
Unpurified ab79398 staining RPA70 in U-2 OS (Human bone osteosarcoma epithelial cell line) cells by ICC/IF (Immunocytochemistry/immunofluorescence).
Cells were fixed with paraformaldehyde, permeabilized with 0.5% Triton X-100 in PBS and blocked with 2% BSA for 1 hour at 25°C. Samples were incubated with primary antibody (1/500 in PBS + 0.5% Tween-20) for 2 hours at 25°C. A Cy3®-conjugated goat anti-rabbit IgG monoclonal (1/250) was used as the secondary antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-RPA70 antibody [EPR3472] (ab79398)

RPA70 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-RPA70 antibody

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human cervical squamous cell carcinoma labelling RPA70 with unpurified ab79398 at a dilution of 1/100. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



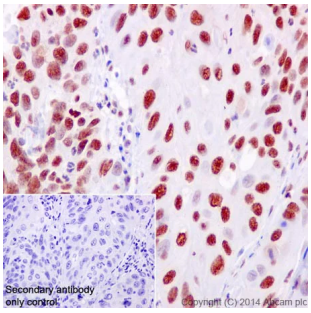
Flow Cytometry (Intracellular) - Anti-RPA70 antibody [EPR3472] (ab79398)

RPA70 Flow Cytometry (Intracellular) staining using rabbit Anti-RPA70 antibody

Overlay histogram showing HeLa (Human epithelial cell line from cervix adenocarcinoma) cells stained with unpurifiedab79398 (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 (unpurified ab79398, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit IgG (H+L) (ab96899) at 1/500 dilution for 30 min at 22°C.

Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10^6 cells) used under the same conditions.

Acquisition of >5,000 events was performed.

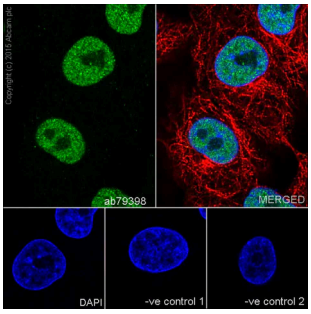


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-RPA70 antibody [EPR3472] (ab79398)

RPA70 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-RPA70 antibody

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human cervix carcinoma tissue labeling RPA70 with purified ab79398 at 1/100.
Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. ab97051, a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500).

Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.



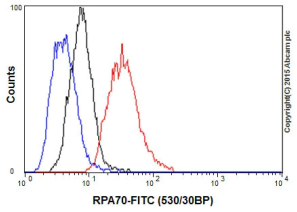
Immunocytochemistry/ Immunofluorescence - Anti-RPA70 antibody [EPR3472] (ab79398)

RPA70 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-RPA70 antibody

Immunocytochemistry/Immunofluorescence analysis of A549 (Human lung carcinoma cell line) cells labeling RPA70 with purified ab79398 at 1/200.
Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/500) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. ab7291, a mouse anti-tubulin (1/1000) and ab150120, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/500) were also used.

Control 1: primary antibody (1/200) and secondary antibody, ab150120, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/500).

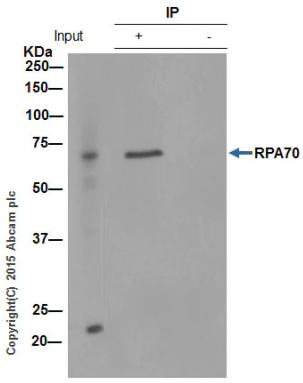
Control 2: ab7291 (1/1000) and secondary antibody, ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/500).



Flow Cytometry (Intracellular) - Anti-RPA70 antibody [EPR3472] (ab79398)

RPA70 Flow Cytometry (Intracellular) staining using rabbit Anti-RPA70 antibody

Intracellular Flow Cytometry analysis of HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling RPA70 with purified ab79398 at 1/80 (red).
Cells were fixed with 2% paraformaldehyde. A FITC-conjugated goat anti-rabbit IgG (1/150) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabeled control, cells without incubation with primary and secondary antibodies.



Immunoprecipitation - Anti-RPA70 antibody [EPR3472] (ab79398)

ab79398 (purified) at 1/20 immunoprecipitating RPA70 in HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate.
Lane 1 (input): HeLa whole cell lysate (10 µg)

Lane 2 (+): ab79398 + HeLa whole cell lysate (10 µg).

Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab79398 in HeLa whole cell lysate.

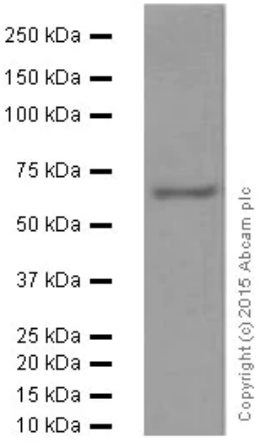
For western blotting, an HRP-conjugated anti-rabbit IgG, specific to the non-reduced form of IgG was used as the secondary antibody (1/1500).

Blocking/Dilution buffer: 5% NFDM/TBST.

All lanes:
Immunoprecipitation - Anti-RPA70 antibody [EPR3472] (ab79398)

Predicted band size: 68 kDa

Observed band size: 70 kDa



Western blot - Anti-RPA70 antibody [EPR3472] (ab79398)

Blocking/Dilution buffer: 5% NFDM/TBST.
All lanes:
Western blot - Anti-RPA70 antibody [EPR3472] (ab79398) at 1/1000 dilution

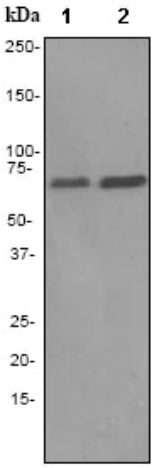
All lanes:
A549 (Human lung carcinoma cell line) cell lysate at 20 µg

Secondary

All lanes:
Peroxidase-conjugated goat anti-rabbit IgG, (H+L) at 1/1000 dilution

Predicted band size: 68 kDa

Observed band size: 70 kDa



Western blot - Anti-RPA70 antibody [EPR3472] (ab79398)

All lanes:
Western blot - Anti-RPA70 antibody [EPR3472] (ab79398) at 1/5000 dilution

Lane 1:
A549 (Human lung carcinoma cell line) cell lysate at 10 µg

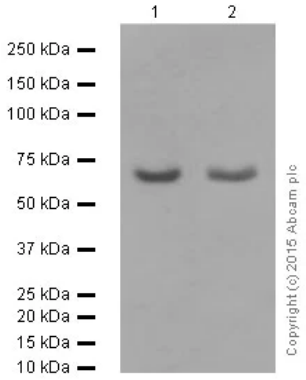
Lane 2:
HeLa (Human epithelial cell line from cervix adenocarcinoma) cell lysate at 10 µg

Secondary

All lanes:
HRP-conjugated goat anti-rabbit IgG at 1/2000 dilution

Predicted band size: 68 kDa

Observed band size: 70 kDa



Western blot - Anti-RPA70 antibody [EPR3472] (ab79398)

RPA70 Western blot staining using rabbit Anti-RPA70 antibody

Blocking/Dilution buffer: 5% NFDM/TBST.
All lanes:
Western blot - Anti-RPA70 antibody [EPR3472] (ab79398) at 1/4000 dilution

Lane 1:
HEK-293 (Human epithelial cell line from embryonic kidney) cell lysate at 20 µg

Lane 2:
HeLa (Human epithelial cell line from cervix adenocarcinoma) cell lysate at 20 µg

Secondary

All lanes:
Peroxidase-conjugated goat anti-rabbit IgG, (H+L) at 1/1000 dilution

Predicted band size: 68 kDa

Observed band size: 70 kDa

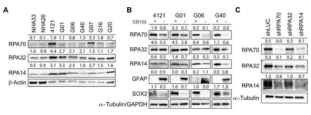


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Western blot - Anti-RPA70 antibody [EPR3472] (ab79398)

RPA70 Western blot staining using rabbit Anti-RPA70 antibody

RPA70 western blot using anti-RPA70 antibody [EPR3472] ab79398. Publication image and figure legend from Pedersen, H., Anne Adanma Obara, E., et al., 2020, Int J Mol Sci, PubMed 32111042.

ab79398 was used in this publication in western blot. This may not be the same as the application(s) guaranteed by Abcam. For a full list of applications guaranteed by Abcam for ab79398 please see the product overview.

RPA expression is crucial for the maintenance of glioblastoma cancer stem-like cells. (A) Immunoblot analysis of RPA70, RPA32 and RPA14 in a panel of GBM primary cell lines (4121, G01, G06, G40, G07, G16 and G20) and two normal human astrocyte cell lines (NHA33, NHA26). β-Actin

was used as a loading control. (B) Immunoblot analysis of RPA70, RPA32 and RPA14 expression in matched pairs of glioblastoma cancer stem-like cells (GSCs) and differentiated GBM cells (DGCs) isolated from 4121, G01, G06 and G40 primary GBM cell lines. α-Tubulin or Glyceraldehyde 3-phosphate dehydrogenase (GAPDH) was used as a loading control. (C) Immunoblot analysis validating knockdown efficiencies of lentiviral shRNAs targeting RPA70, RPA32 and RPA14 in G01-GSCs. α-Tubulin was used as a loading control. (D) CellTiter-Glo luminescent viability assay (Promega) of G01-GSCs transduced with lentiviral shRNA targeting RPA70, RPA32 or RPA14 assessed 72 h after virus wash. Data are presented as mean ± SD. Statistical significance was tested using a two-way ANOVA with post-hoc Sidak's multiple comparisons test, where *** $p < 0.001$. N = 3. (E) CellTiter-Glo luminescent viability assay (Promega) of G01-GSCs transduced with lentiviral shRNA targeting RPA70, RPA32 or RPA14 alone or 24 h after irradiation (3 Gy; COMBO) or sham-irradiated and assessed for viability 72 h later. (E) Extreme limiting dilution assay (ELDA) of G01-GSCs transduced with lentiviral shRNA targeting of RPA70 (2 independent shRNAs) shows significantly impaired self-renewal of G01-GSCs after 10 days. Data are presented as mean ± SD. Statistical significance was tested using a two-way ANOVA with post-hoc Sidak's multiple comparisons test, where * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$. N = 3.

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