abcam

Product datasheet

Anti-Cytokeratin 7 antibody [EPR1619Y] - Cytoskeleton Marker ab68459





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Overview

Product name Anti-Cytokeratin 7 antibody [EPR1619Y] - Cytoskeleton Marker

Description Rabbit monoclonal [EPR1619Y] to Cytokeratin 7 - Cytoskeleton Marker

Host species Rabbit

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

Species reactivity Reacts with: Human

Synthetic peptide within Human Cytokeratin 7 aa 1-100 (N terminal). The exact sequence is **Immunogen**

proprietary.

Positive control WB: HeLa, SK-OV-3 and T47D cell lysates. IHC-P: Human ovarian carcinoma and human

bladder carcinoma tissues. ICC/IF: HeLa and TD47 cells. Flow Cyt (intra): HeLa cells. IP: HeLa

whole cell lysate (ab150035).

General notes 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 monoclonal antibodies. For details on our patents, please refer to **RabMAb patents**.

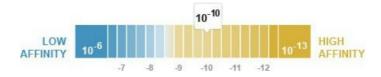
Properties

Form

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Avoid freeze / thaw cycle.

 $K_D = 2.10 \times 10^{-10} M$ Dissociation constant (K_D)



Learn more about K_D

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol, 0.05% BSA

Purity Protein A purified

ClonalityMonoclonalClone numberEPR1619Y

Isotype IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab68459 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)		1/20 - 1/30. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB		1/5000 - 1/10000. Detects a band of approximately 51 kDa (predicted molecular weight: 51 kDa).
IP		1/20 - 1/30.
IHC-P		1/1000. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See IHC antigen retrieval protocols. For unpurified use at 1/100 - 1/250.
ICC/IF	**** <u>(1)</u>	1/100 - 1/250.

Target

Function Blocks interferon-dependent interphase and stimulates DNA synthesis in cells. Involved in the

translational regulation of the human papillomavirus type 16 E7 mRNA (HPV16 E7).

Tissue specificity Expressed in cultured epidermal, bronchial and mesothelial cells but absent in colon, ectocervix

and liver. Observed throughout the glandular cells in the junction between stomach and esophagus

but is absent in the esophagus.

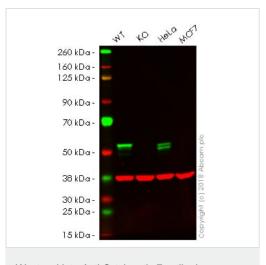
Sequence similaritiesBelongs to the intermediate filament family.

Post-translational modifications

Arg-20 is dimethylated, probably to asymmetric dimethylarginine.

Cellular localization Cytoplasm.

Images



Western blot - Anti-Cytokeratin 7 antibody [EPR1619Y] - Cytoskeleton Marker (ab68459)

All lanes : Anti-Cytokeratin 7 antibody [EPR1619Y] - Cytoskeleton Marker (ab68459) at 1/5000 dilution

Lane 1 : Wild-type A549 (Human lung carcinoma cell line) whole cell lysate

Lane 2 : KRT7 knockout A549 (Human lung carcinoma cell line) whole cell lysate

Lane 3: HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

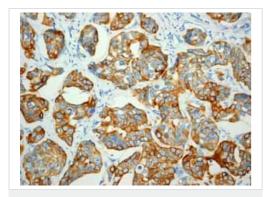
Lane 4 : MCF7 (Human breast adenocarcinoma cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 51 kDa **Observed band size:** 50 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab68459 observed at 50 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab68459 was shown to specifically react with KRT7 (Cytokeratin 7) in wild-type A549 cells as signal was lost in KRT7 knockout cells. Wild-type and KRT7 knockout samples were subjected to SDS-PAGE. Ab68459 and <u>ab8245</u> (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/5000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed <u>ab216773</u> and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed <u>ab216776</u> secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.

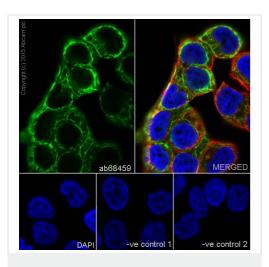


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cytokeratin 7 antibody

[EPR1619Y] - Cytoskeleton Marker (ab68459)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human breast carcinoma tissue labelling Cytokeratin 7 with unpurified ab68459.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-Cytokeratin 7 antibody [EPR1619Y] - Cytoskeleton Marker (ab68459)

Immunocytochemistry/Immunofluorescence analysis of T47D cells labelling Cytokeratin 7 with purified ab68459 at a dilution of 1/100. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, an Alexa Fluor[®] 488-conjugated goat anti-rabbit IgG (1/1000) 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 (1/1000) were also used.

Control 1: primary antibody (1/100) and secondary antibody, **ab150120**, an Alexa Fluor[®] 594-conjugated goat anti-mouse lgG (1/1000).

Control 2: <u>ab7291</u> (1/1000) and secondary antibody, <u>ab150077</u>, an Alexa Fluor[®] 488-conjugated goat anti-rabbit lgG (1/1000).

Alexa Fluor[®] 488 (<u>ab185048</u>) and Alexa Fluor[®] 647 (<u>ab192077</u>) conjugated versions are available for this clone.

1 2 3

250 kDa —

150 kDa —

100 kDa —

75 kDa —

50 kDa —

37 kDa —

25 kDa —

20 kDa —

15 kDa —

10 kDa —

10 kDa —

Western blot - Anti-Cytokeratin 7 antibody [EPR1619Y] - Cytoskeleton Marker (ab68459)

All lanes : Anti-Cytokeratin 7 antibody [EPR1619Y] - Cytoskeleton Marker (ab68459) at 1/10000 dilution (purified)

Lane 1: HeLa whole cell lysate

Lane 2: SK-OV-3 whole cell lysate

Lane 3: T47D whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

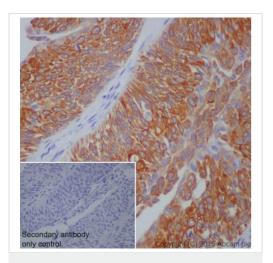
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 51 kDa

Observed band size: 51 kDa

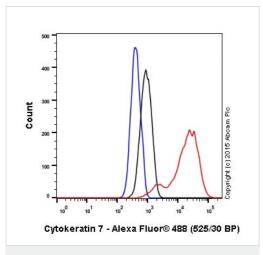
Blocking and dilution buffer: 5% NFDM/TBST.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human bladder carcinoma tissue labelling Cytokeratin 7 with purified ab68459 at a dilution of 1/1000. Heat mediated antigen retrieval was performed using EDTA buffer pH 9. ab97051, a HRP-conjugated goat anti-rabbit lgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cytokeratin 7 antibody

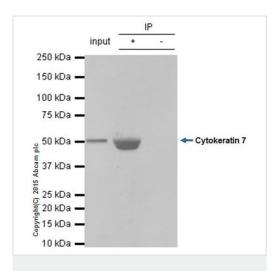
[EPR1619Y] - Cytoskeleton Marker (ab68459)



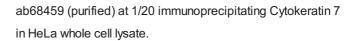
Flow Cytometry (Intracellular) - Anti-Cytokeratin 7 antibody [EPR1619Y] - Cytoskeleton Marker (ab68459)

Intracellular Flow Cytometry analysis of HeLa cells labelling Cytokeratin 7 with purified ab68459 at a dilution of 1/20 (red). Cells were fixed with 80% methanol. An Alexa Fluorr[®]488-conjugated goat anti-rabbit lgG (1/500) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal lgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.

Alexa Fluorr[®]488 (<u>ab185048</u>) and Alexa Fluorr[®]647 (<u>ab192077</u>) conjugated versions are available for this clone.



Immunoprecipitation - Anti-Cytokeratin 7 antibody [EPR1619Y] - Cytoskeleton Marker (ab68459)



Lane 1 (input): HeLa whole cell lysate (10µg)

Lane 2 (+): ab68459 + HeLa whole cell lysate.

Lane 3 (-): Rabbit monoclonal lgG (<u>ab172730</u>) instead of ab68459 in HeLa whole cell lysate.

For western blotting, VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/1000 dilution.

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



Western blot - Anti-Cytokeratin 7 antibody [EPR1619Y] - Cytoskeleton Marker (ab68459) **All lanes :** Anti-Cytokeratin 7 antibody [EPR1619Y] - Cytoskeleton Marker (ab68459) at 1/10000 dilution (unpurified)

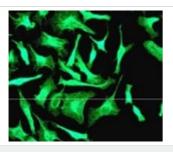
Lane 1 : HeLa cell lysate Lane 2 : T47D cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

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

Predicted band size: 51 kDa **Observed band size:** 51 kDa



Immunocytochemistry/ Immunofluorescence - Anti-Cytokeratin 7 antibody [EPR1619Y] - Cytoskeleton Marker (ab68459) Immunocytochemistry/Immunofluorescence analysis of HeLa cells labelling Cytokerain 7 with unpurified ab68459 at a dilution of 1/100.

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cytokeratin 7 antibody

[EPR1619Y] - Cytoskeleton Marker (ab68459)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human endometrial carcinoma tissue labelling Cytokeratin 7 with unpurified ab68459.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cytokeratin 7 antibody

[EPR1619Y] - Cytoskeleton Marker (ab68459)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human ovarian carcinoma tissue labelling Cytokeratin 7 with unpurified ab68459.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cytokeratin 7 antibody

[EPR1619Y] - Cytoskeleton Marker (ab68459)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human urinary bladder transitional carcinoma tissue labelling Cytokeratin 7 with unpurified ab68459.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cytokeratin 7 antibody

[EPR1619Y] - Cytoskeleton Marker (ab68459)

ab68459 showing negative staining in human sarcoma tissue.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cytokeratin 7 antibody

[EPR1619Y] - Cytoskeleton Marker (ab68459)

ab68459 showing negative staining in human colonic adenocarcinoma tissue.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

Flow Cytometry (Intracellular) - Anti-Cytokeratin 7 antibody [EPR1619Y] - Cytoskeleton Marker (ab68459) Overlay histogram showing HeLa cells stained with unpurified ab68459 (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 ab68459, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat antirabbit lgG (H+L) (ab96899) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit lgG (monoclonal) $(1\mu g/1x10^6 \text{ cells})$ used under the same conditions. Acquisition of >5,000 events was performed.

OI-RD Scanning - Anti-Cytokeratin 7 antibody [EPR1619Y] - Cytoskeleton Marker (ab68459) Equilibrium disassociation constant (K_D) Learn more about K_D

Click here to learn more about K_D



Cytoskeleton Marker (ab68459)

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