abcam

Product datasheet

Anti-CDKN2A/p16INK4a antibody [EPR1473] - C-terminal ab108349

יעלאעבע RabMAb

★★★★★ 7 Abreviews 162 References

画像数 13

製品の概要

製品名 Anti-CDKN2A/p16lNK4a antibody [EPR1473] - C-terminal

製品の詳細 Rabbit monoclonal [EPR1473] to CDKN2A/p16INK4a - C-terminal

由来種 Rabbit

特異性 Expression levels of the CDKN2A/p16INK4a protein may vary with sample type. It is barely

> expressed in normal tissue, and mostly expressed in some tumour tissues, such as cervical cancer, breast cancer and so on. Moreover, only expressed in some cell lines. Please see

images for recommended positive controls.

アプリケーション 適用あり: WB, IP, IHC-P, Flow Cyt (Intra)

適用なし: ICC/IF

種交差性 交差種: Human

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

ポジティブ・コントロール WB: HeLa, HEK-293, HEK-293T, and Saos-2 cell lysates, His-tagged human CDKN2A

> recombinant protein lysates, human CDKN2A full-length recombinant protein with His-tag, human CDKN2B full-length recombinant protein with GST-tag. IHC-P: Human cervical carcinoma tissue.

ICC/IF: HeLa cells. Flow Cyt (intra): HEK-293 and HeLa cells. IP: HeLa cell lysate.

特記事項 Abcam recommended secondaries - Goat Anti-Rabbit HRP (ab205718) and Goat Anti-Rabbit

Alexa Fluor® 488 (ab150077). Or search our wide range of secondary antibodies for use with

your experiment.

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**.

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.

Avoid freeze / thaw cycle.

バッファー pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 40% Glycerol, PBS, 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 EPR1473

アイソタイプ IgG

アプリケーション

The Abpromise guarantee <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab108349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
WB	★★★★★ (3)	1/2000. Predicted molecular weight: 17 kDa. For unpurified use at 1/1000 - 1/10000.
IP		1/30. For unpurified use at 1/10 - 1/100.
IHC-P	★★★★ (2)	1/100. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See IHC antigen retrieval protocols. For unpurified use at 1/250 - 1/500.
Flow Cyt (Intra)		1/270 - 1/500. For unpurified use at 1/100 - 1/500. <u>ab172730</u> - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.

追加情報 Is unsuitable for ICC/IF.

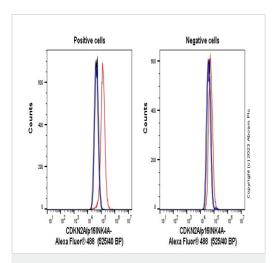
ターゲット情報

細胞内局在 Cytoplasmic and Nuclear

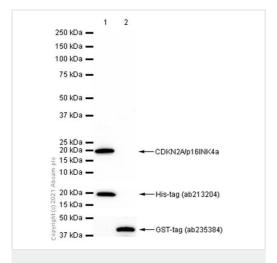
製品の状態 There are 4 isoforms produced by alternative splicing. Isoform 1 also known as: p16INK4a;

lsoform 3 also known as: p12; lsoform 4 also known as: p14ARF; p19ARF; ARF.

画像



Flow Cytometry (Intracellular) - Anti-CDKN2A/p16lNK4a antibody [EPR1473] - C-terminal (ab108349)



Western blot - Anti-CDKN2A/p16lNK4a antibody [EPR1473] - C-terminal (ab108349)

Flow cytometry overlay histogram showing left HeLa positive cells and right negative MCF7 stained with ab108349 (red line). The cells were fixed with 4% formaldehyde (10 min) and then permeabilised with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS containing 10% normal goat serum to block non-specific protein-protein interaction followed by the antibody (ab108349) (1x 10^6 in 100μ l at 0.04μ g/ml (1/52500)) for 30min at 22° C.

The secondary antibody Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min at 22°C

Isotype control antibody (black line) was Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control used at the same concentration and conditions as the primary antibody. Unlabelled sample (blue line) was also used as a control.

Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter.

All lanes : Anti-CDKN2A/p16lNK4a antibody [EPR1473] - Cterminal (ab108349) at 1/1000 dilution

Lane 1 : Human CDKN2A full-length recombinant protein with Histag

Lane 2: Human CDKN2B full-length recombinant protein with GST-tag

Lysates/proteins at 0.01 µg per lane.

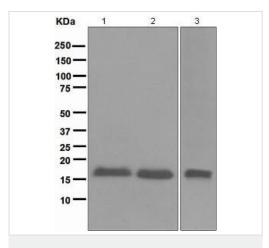
Secondary

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

Predicted band size: 17 kDa **Observed band size:** 17 kDa

Exposure time: 10 seconds

5% NFDM/TBST was used as a blocking and diluting buffer.



Western blot - Anti-CDKN2A/p16lNK4a antibody [EPR1473] - C-terminal (ab108349)

All lanes : Anti-CDKN2A/p16lNK4a antibody [EPR1473] - Cterminal (ab108349) at 1/1000 dilution (unpurified)

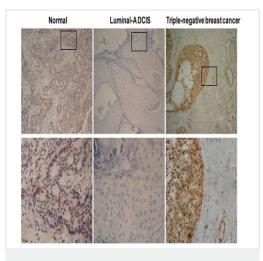
Lane 1 : HeLa (Human epithelial cell line from cervix adenocarcinoma) cell lysate

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

Lane 3: Saos-2 (Human osteosarcoma cell line) cell lysate

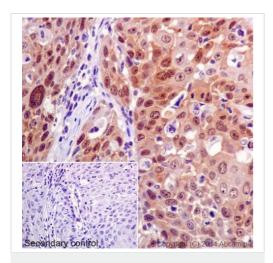
Lysates/proteins at 10 µg per lane.

Predicted band size: 17 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CDKN2A/p16INK4a antibody [EPR1473] - C-terminal (ab108349)

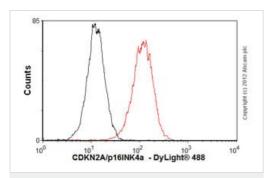
Image from Shan Met al., PLoS One. 2013;8(10):e76408. Fig 1.; doi: 10.1371/journal.pone.0076408. eCollection 2013. Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/ Formalin-fixed, paraffin-embedded human normal breast, luminal-A DCIS (ductal carcinoma *in situ*) and triple negative breast cancer tissues stained for CDKN2A/p16INK4a using ab108349 in immunohistochemical analysis.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CDKN2A/p16INK4a antibody [EPR1473] - C-terminal (ab108349)

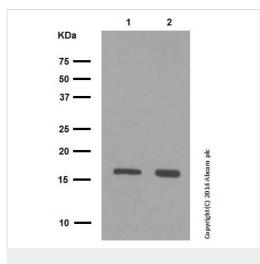
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human cervix carcinoma tissue labeling CDKN2A/p16INK4a with purified ab108349 at 1/100. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. ab97051, an HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500).

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



Flow Cytometry (Intracellular) - Anti-CDKN2A/p16INK4a antibody [EPR1473] - C-terminal (ab108349)

Overlay histogram showing HEK-293 (Human epithelial cell line from embryonic kidney) cells stained with unpurified ab108349 (red line). The cells were fixed with 4% paraformaldehyde (10 minutes) and then permeabilized with 0.1% PBS-Tween for 20 minutes. 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 (ab108349, 1/100) for 30 minutes at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit lgG (H+L) (ab96899) at 1/500 dilution for 30 minutes at 22°C. Isotype control antibody (black line) was rabbit lgG (monoclonal) (1µg/1x106 cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in HEK-293 cells fixed with 80% methanol (5 minutes)/permeabilized with 0.1% PBS-Tween for 20 minutes used under the same conditions.



Western blot - Anti-CDKN2A/p16lNK4a antibody [EPR1473] - C-terminal (ab108349)

All lanes : Anti-CDKN2A/p16lNK4a antibody [EPR1473] - Cterminal (ab108349) at 1/2000 dilution (purified)

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

Lane 2: Saos-2 (Human osteosarcoma cell line) cell lysate

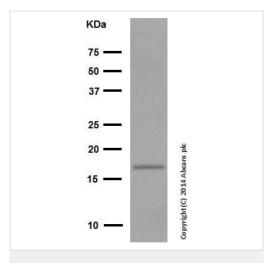
Lysates/proteins at 10 µg per lane.

Secondary

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

Predicted band size: 17 kDa **Observed band size:** 17 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-CDKN2A/p16lNK4a antibody [EPR1473] - C-terminal (ab108349)

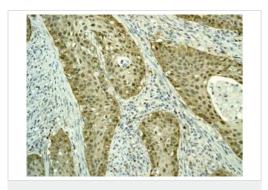
Anti-CDKN2A/p16INK4a antibody [EPR1473] - C-terminal (ab108349) at 1/2000 dilution (purified) + HeLa (Human epithelial cell line from cervix adenocarcinoma) cell lysate at 10 μ g

Secondary

Peroxidase-conjugated goat anti-rabbit lgG (H+L) at 1/1000 dilution

Predicted band size: 17 kDa **Observed band size:** 17 kDa

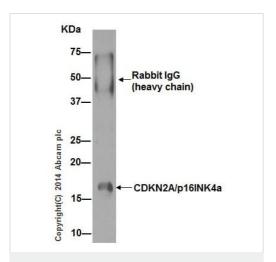
Blocking/Dilution buffer: 5% NFDM/TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CDKN2A/p16lNK4a antibody [EPR1473] - C-terminal (ab108349)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human cervical carcinoma tissue labeling CDKN2A/p16INK4a with unpurified ab108349 at a dilution of 1/250

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

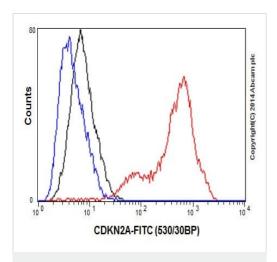


Immunoprecipitation - Anti-CDKN2A/p16lNK4a antibody [EPR1473] - C-terminal (ab108349)

ab108349 (purified) at 1/30

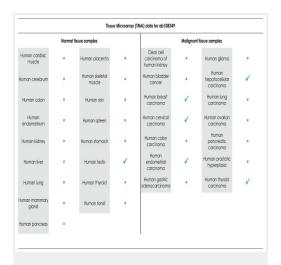
immunoprecipitating CDKN2A/p16INK4a in HeLa (Human epithelial cell line from cervix adenocarcinoma) cell lysate. For western blotting, a peroxidase-conjugated goat anti-rabbit lgG (H+L) was used as the secondary antibody (1/1000).

Blocking/Dilution buffer: 5% NFDM/TBST.

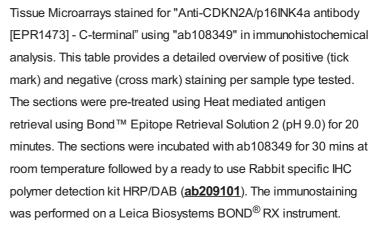


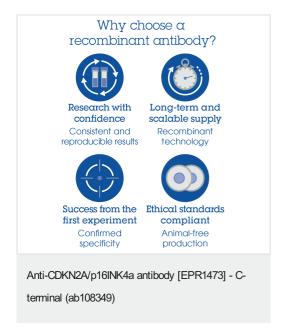
Flow Cytometry (Intracellular) - Anti-CDKN2A/p16INK4a antibody [EPR1473] - C-terminal (ab108349)

Intracellular Flow Cytometry analysis of HEK-293 (Human epithelial cell line from embryonic kidney) cells labeling CDKN2A/p16lNK4a with purified ab108349 at 1/270 (red). Cells were fixed with 2% paraformaldehyde. A FITC-conjugated goat anti-rabbit lgG (1/150) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal lgG. Blue - Unlabeled control, cells without incubation with primary and secondary antibodies.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CDKN2A/p16lNK4a antibody [EPR1473] - C-terminal (ab108349)





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