abcam

Product datasheet

Anti-Cdk2 antibody [E304] ab32147



★★★★★ 8 Abreviews 214 References 画像数 14

製品の概要

製品名 Anti-Cdk2 antibody [E304]

製品の詳細 Rabbit monoclonal [E304] to Cdk2

由来種 Rabbit

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

種交差性 交差種: Mouse, Rat, Human

免疫原 Synthetic peptide within Human Cdk2 aa 250 to the C-terminus (C terminal). The exact sequence

is proprietary.

エピトープ The epitope is within the C-terminus of human Cdk2

ポジティブ・コントロール HeLa cells HeLa whole cell lysate (ab150035).

特記事項 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: 59% PBS, 40% Glycerol, 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 E304

アプリケーション

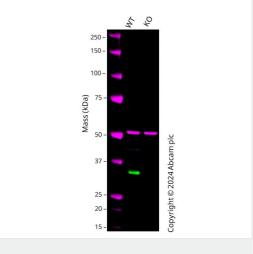
The Abpromise guarantee Abpromise保証は、次のテスト済みアプリケーションにおけるab32147の使用に適用されます アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
ICC/IF	****(3)	1/200. For unpurified use at 1/100.
IP		1/40.
WB	★★★★★ (5)	1/1000 - 1/10000. Detects a band of approximately 33 kDa (predicted molecular weight: 34 kDa).
IHC-P		1/50. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See IHC antigen retrieval protocols .
Flow Cyt (Intra)		1/80. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.

ターゲット情報

機能	Involved in the control of the cell cycle. Interacts with cyclins A, B1, B3, D, or E. Activity of CDK2 is maximal during S phase and G2.
配列類似性	Belongs to the protein kinase superfamily. CMGC Ser/Thr protein kinase family. CDC2/CDKX subfamily.
	Contains 1 protein kinase domain.

画像



Western blot - Anti-Cdk2 antibody [E304] (ab32147)

All lanes: Anti-Cdk2 antibody [E304] (ab32147) at 1/2000 dilution

Lane 1: Wild-type MCF7 cell lysate

Lane 2: CDK2 knockout MCF7 cell lysate

Lysates/proteins at 10 µg per lane.

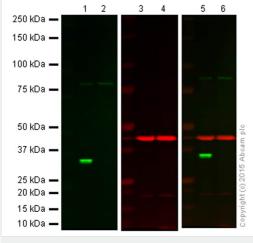
Secondary

All lanes: Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse

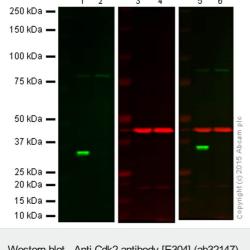
IgG H&L 680RD at 1/20000 dilution

Predicted band size: 34 kDa **Observed band size:** 34 kDa

Western blot: Anti-CDK2 antibody [E304] (ab32147) staining at 1/2000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] (ab7291) loading control staining at 1/20000 dilution, shown in magenta. In Western blot, ab32147 was shown to bind specifically to CDK2. A band was observed at 34 kDa in wild-type MCF7 cell lysates with no signal observed at this size in CDK2 knockout cell line ab282628. To generate this image, wild-type and CDK2 knockout MCF7 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.



Western blot - Anti-Cdk2 antibody [E304] (ab32147)



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Immunocytochemistry/ Immunofluorescence - Anti-Cdk2 antibody [E304] (ab32147)

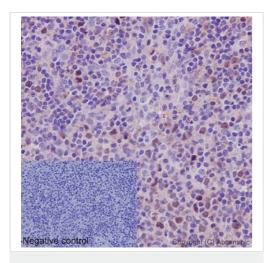
Lanes 1, 3 and 5: Wild-type HAP1 cell lysate (20 µg) Lanes 2, 4 and 6: CDK2 knockout HAP1 cell lysate (20 µg) Lanes 1 and 2: Green signal from target – ab32147 observed at 34 kDa

Lanes 3 and 4: Red signal from loading control – ab8226 observed at 42 kDa

Lanes 5 and 6: Merged (red and green) signal

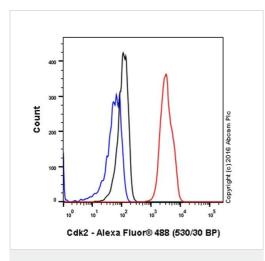
ab32147 was shown to specifically react with CDK2 when CDK2 knockout samples were used. Wild-type and CDK2 knockout samples were subjected to SDS-PAGE. ab32147 and ab8226 (loading control to beta actin) were both diluted 1/1000 and incubated overnight at 4°C. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1/10 000 dilution for 1 h at room temperature before imaging.

ab32147 staining Cdk2 in the HeLa cell line by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with 4% Paraformaldehyde permeabilized with 0.1% Triton X-100. Samples were incubated with primary antibody (1/200). ab150078 (1/500) an Alexa Fluor[®] 555-conjugated Goat anti-rabbit lgG was used as the secondary antibody. Nuclei were counterstained with DAPI.



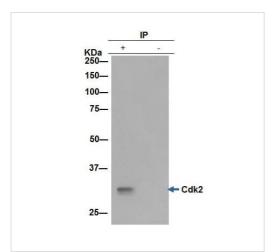
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cdk2 antibody [E304] (ab32147)

ab32147 staining Cdk2 in human tonsil tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffinembedded sections). Tissue was fixed and paraffin-embedded, antigen retrieval was by heat mediation in Tris/EDTA buffer pH9. Samples were incubated with primary antibody (1/50). An undiluted HRP-conjugated mouse anti-rabbit IgG was used as the secondary antibody. Tissue counterstained with Hematoxylin. PBS was used in the negative control rather than the Primary antibody.



Flow Cytometry (Intracellular) - Anti-Cdk2 antibody [E304] (ab32147)

Intracellular Flow Cytometry analysis of HeLa (human cervix adenocarcinoma) cells labelling Cdk2 with purified ab32147 at 1/80 (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. An Alexa Fluor[®] 488-conjugated goat anti-rabbit lgG (1/2000) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal lgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.

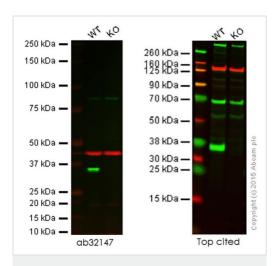


Immunoprecipitation - Anti-Cdk2 antibody [E304] (ab32147)

ab32147 (purified) at 1/40 immunoprecipitating Cdk2 from HeLa cells(Lane 1). Lane 2 - PBS. For western blotting, a HRP-conjugated anti-rabbit lgG, specific to the non-reduced form of lgG was used as the secondary antibody (1/1000).

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



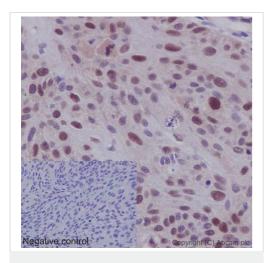
Western blot - Anti-Cdk2 antibody [E304] (ab32147)

Lanes 1: Wild-type HAP1 cell lysate (20 µg)

Lanes 2: CDK2 knockout HAP1 cell lysate (20 µg)

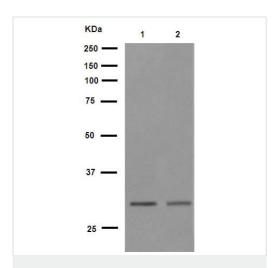
Lanes 1 - 2: Merged signal (red and green). Green - ab32147 observed at 34 kDa. Red - loading control, **ab8226**, observed at 42 kDa or **ab18058**, observed at 130 kDa.

This western blot image is a comparison between ab32147 and a competitor's top cited rabbit polyclonal antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Cdk2 antibody [E304] (ab32147)

ab32147 staining Cdk2 in human squamous cell carcinoma of cervix tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffin-embedded sections). Tissue was fixed and paraffin-embedded, antigen retrieval was by heat mediation in Tris/EDTA buffer pH9. Samples were incubated with primary antibody (1/50). An undiluted HRP-conjugated mouse antirabbit IgG was used as the secondary antibody. Tissue counterstained with Hematoxylin. PBS was used in the negative control rather than the Primary antibody.



Western blot - Anti-Cdk2 antibody [E304] (ab32147)

All lanes: Anti-Cdk2 antibody [E304] (ab32147) at 1/5000 dilution

Lane 1: C6 cell lysate

Lane 2: PC-12 cell lysate

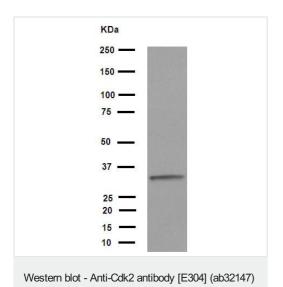
Lysates/proteins at 10 µg per lane.

Secondary

All lanes: Goat Anti-Rabbit lgG, (H+L), HRP-conjugated at 1/1000

dilution

Predicted band size: 34 kDa

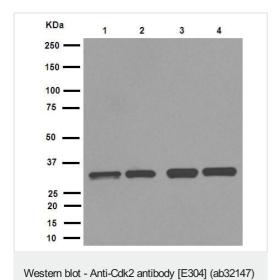


Anti-Cdk2 antibody [E304] (ab32147) at 1/1000 dilution + NIH/3T3 cell lysate at 20 μg

Secondary

Goat Anti-Rabbit IgG, (H+L), HRP-conjugated at 1/1000 dilution

Predicted band size: 34 kDa



All lanes: Anti-Cdk2 antibody [E304] (ab32147) at 1/1000 dilution

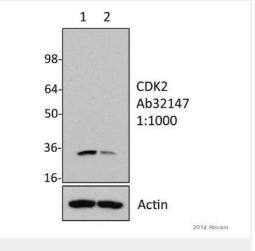
Lane 1 : Jurkat cell lysate
Lane 2 : Hela cell lysate
Lane 3 : K562 cell lysate
Lane 4 : 293 cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG, (H+L), HRP-conjugated at 1/1000 dilution

Predicted band size: 34 kDa



Western blot - Anti-Cdk2 antibody [E304] (ab32147)

This image is courtesy of an Abreview submitted by Sonia Rocha

All lanes : Anti-Cdk2 antibody [E304] (ab32147) at 1/1000 dilution (unpurified)

Lane 1 : Human osteosarcoma whole cell lysate - control, non-targeting siRNA

Lane 2: Human osteosarcoma whole cell lysate - siRNA for CDK2

Lysates/proteins at 20 µg per lane.

Secondary

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

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 34 kDa **Observed band size:** 34 kDa

Exposure time: 2 seconds

All lanes : Anti-Cdk2 antibody [E304] (ab32147) at 1/1000 dilution (unpurified)

Lane 1 : HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate (ab27252) at 10 µg

Lane 2 : HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate (ab27252) at 20 µg

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/10000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 34 kDa **Observed band size:** 34 kDa

Exposure time: 4 minutes

This blot was produced using a 10% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 50 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 5% Bovine Serum Albumin before being incubated with ab32147 overnight at 4°C. Antibody binding was detected using an anti-rabbit antibody conjugated to HRP, and visualised using ECL development solution.

1 2

250 kDa —

150 kDa —

150 kDa —

75 kDa —

50 kDa —

37 kDa —

25 kDa —

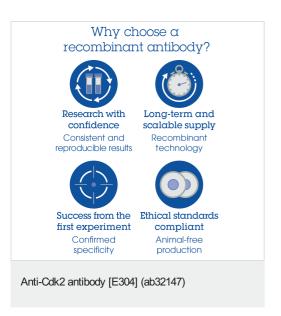
20 kDa —

10 kDa —

10 kDa —

11 kDa —

10 kDa —



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