

Anti-Cyclin D1 antibody [EPR2241] - C-terminal

Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175) is a rabbit monoclonal antibody detecting Cyclin D1 in **Western Blot, IP, IHC-P, ICC/IF**. Suitable for **Human, Mouse, Rat**.

- KO validated for confirmed specificity
- Biophysical QC for unrivalled batch-batch consistency
- Over 780 publications

Lab Essentials

Recombinant

RabMAb

KO Validated

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	EPR2241
Purification technique	Affinity purification Protein A
Specificity	The immunogen used for this product shares 66% homology with CCND2 (seven amino acid stretch with 100% homology). Based on internal testing in WB, this product shows a weak cross-reactivity to Cyclin D2. For IHC usage, this product shows a tissue localization specific to Cyclin D1 with no cross-reactivity to Cyclin D2.
Dissociation constant	2.4 x 10 ⁻¹¹ M
Concentration	0.428 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested	
Species	Rat
Dilution info	1/100 - 1/500
Notes	Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

Species	Human
Dilution info	1/100 - 1/500
Notes	Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

Expected

Species	Mouse
Dilution info	Use at an assay dependent concentration.
Notes	-

IP

Tested	
Species	Human
Dilution info	1/30
Notes	-

Expected

Species	Rat, Mouse
Dilution info	Use at an assay dependent concentration.
Notes	-

WB

Tested	
Species	Mouse
Dilution info	1/10000 - 1/50000
Notes	Compared to the working concentration with cyclin D1 recombinant protein, a 50 fold increase is required with cyclin D2 recombinant protein in western blot testing.

Species	Human
Dilution info	1/10000 - 1/50000
Notes	Compared to the working concentration with cyclin D1 recombinant protein, a 50 fold increase is required with cyclin D2 recombinant protein in western blot testing.

Expected

Species	Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

ICC/IF

Tested	
Species	Human
Dilution info	1/50
Notes	-

Expected	
Species	Rat, Mouse
Dilution info	Use at an assay dependent concentration.
Notes	-

Target data

Function	Regulatory component of the cyclin D1-CDK4 (DC) complex that phosphorylates and inhibits members of the retinoblastoma (RB) protein family including RB1 and regulates the cell-cycle during G(1)/S transition (PubMed:1827756, PubMed:1833066, PubMed:19412162, PubMed:33854235, PubMed:8114739, PubMed:8302605). Phosphorylation of RB1 allows dissociation of the transcription factor E2F from the RB/E2F complex and the subsequent transcription of E2F target genes which are responsible for the progression through the G(1) phase (PubMed:1827756, PubMed:1833066, PubMed:19412162, PubMed:8114739, PubMed:8302605). Hypophosphorylates RB1 in early G(1) phase (PubMed:1827756, PubMed:1833066, PubMed:19412162, PubMed:8114739, PubMed:8302605). Cyclin D-CDK4 complexes are major integrators of various mitogenic and antimitogenic signals (PubMed:1827756, PubMed:1833066, PubMed:19412162, PubMed:8302605). Also a substrate for SMAD3, phosphorylating SMAD3 in a cell-cycle-dependent manner and repressing its transcriptional activity (PubMed:15241418). Component of the ternary complex, cyclin D1/CDK4/CDKN1B, required for nuclear translocation and activity of the cyclin D-CDK4 complex (PubMed:9106657). Exhibits transcriptional corepressor activity with INSM1 on the NEUROD1 and INS promoters in a cell cycle-independent manner (PubMed:16569215, PubMed:18417529).
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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

Product Specifications

Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175) was developed by Abcam using patented rabbit monoclonal antibody technology and is validated for use in ICC/IF, IHC-P, IP, WB in human, mouse, rat samples. Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175) specifically detects Cyclin D1 (UniProt ID: P24385; Molecular weight: 34kDa) and is sold in 50 µL, 100 µL and 1 mL selling sizes.

Quality and Validation

Abcam's high quality manufacturing and validation processes ensure Anti-Cyclin D1 antibody [EPR2241] - C-terminal

(ab134175) has high sensitivity and specificity alongside high lot-to-lot consistency and reproducibility. The specificity of Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175) has been confirmed by testing in knockout samples.

Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175) has been cited over 783 times in peer reviewed journals and is trusted by the scientific community.

Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175) has 15 independent reviews from customers.

Related Products

Antibody clone EPR2241 is also available pre-conjugated to a variety of labels for your convenience - Alexa Fluor® 488, Alexa Fluor® 647, HRP, Alexa Fluor® 594, Alexa Fluor® 555 (ab190194, ab190563, ab190564, ab203446, ab203448).

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.

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

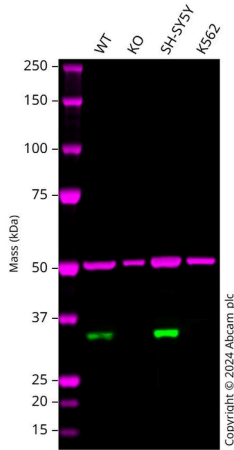
We do not recommend this combination. It is not covered by our product promise.

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](#):

17 product images



Western blot - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

Cyclin D1 Western blot staining using rabbit Anti-Cyclin D1 antibody

Western blot: Anti-CCND1 antibody [EPR2241] (ab134175) staining at 1/10000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] (ab7291) loading control staining at 1/20000 dilution, shown in magenta. In Western blot, ab134175 was shown to bind specifically to CCND1. A band was observed at 34 kDa in wild-type A549 cell lysates with no signal observed at this size in CCND1 knockout cell line. To generate this image, wild-type and CCND1 knockout A549 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.

All lanes:

Western blot - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175) at 1/10000 dilution

Lane 1:

Wild-type A549 cell lysate at 20 µg

Lane 2:

CCND1 knockout A549 cell lysate at 20 µg

Lane 2:

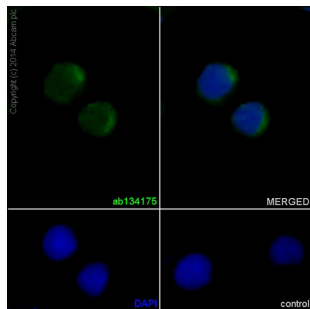
Western blot - Human CCND1 knockout A549 cell line (ab286759)

Lane 3:

SH-SY5Y cell lysate at 20 µg

Lane 4:

K562 cell lysate at 20 µg

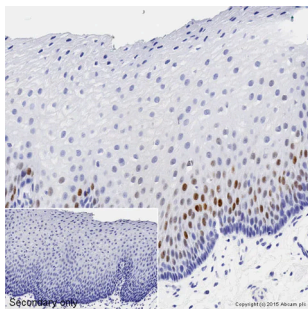


Immunocytochemistry/ Immunofluorescence - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

Cyclin D1 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-Cyclin D1 antibody

Immunofluorescent staining of Ramos (Human Burkitt's lymphoma cell line) cells (fixed in 4% PFA, permeabilized with 0.1% Triton X 100) using purified ab134175 at a dilution of 1/50. An Alexa Fluor® 488 goat anti-rabbit antibody was used as the secondary at a dilution of 1/500 and the cells were counterstained with DAPI.

The negative control is shown in the bottom right hand panel - for the negative control, Alex Fluor® 594 goat anti-mouse was used at a dilution of 1/500.



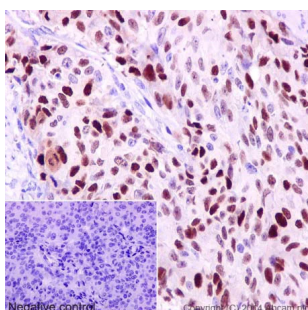
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

Cyclin D1 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Cyclin D1 antibody

IHC image of ab134175 staining Cyclin D1 in human esophagus formalin fixed paraffin embedded tissue sections*, performed on a Leica Bond. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab134175, 5µg/ml working concentration, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with hematoxylin and mounted with DPX. No primary antibody was used in the secondary only control (shown on the inset).

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre

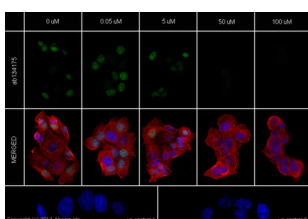


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

Cyclin D1 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Cyclin D1 antibody

Immunohistochemical staining of paraffin embedded human endometrial adenocarcinoma with purified ab134175 at a dilution of 1/100. An HRP goat anti-rabbit ([ab97051](#)) was used as the secondary antibody at a dilution of 1/500 and the sample was counterstained with hematoxylin. Antigen retrieval was performed using Tris-EDTA buffer, pH 9.0.

PBS was used instead of the primary antibody as the negative control (inset).



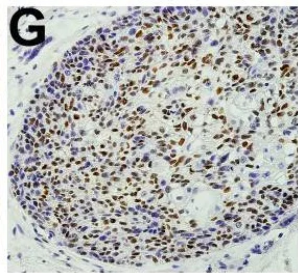
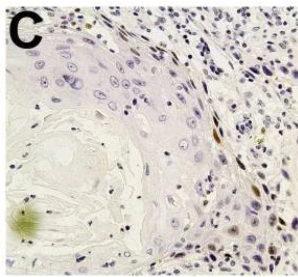
Immunocytochemistry/ Immunofluorescence - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

Cyclin D1 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-Cyclin D1 antibody

Unpurified ab134175 staining Cyclin D1 in MCF7 (Human breast adenocarcinoma cell line) cells treated with KN-93 ([ab120980](#)).

The cells were fixed with 100% methanol (5min) and then blocked in 1% BSA/10% normal goat serum/0.3M glycine in 0.1%PBS-Tween for 1h. The cells were then incubated with ab134175 at 10µg/ml and [ab7291](#) at 1µg/ml overnight at +4°C, followed by a further incubation at room temperature for 1h with an Goat anti-Rabbit Alexa 488 secondary ([ab150081](#)) at 2 µg/ml (shown in green) and Goat anti-Mouse Alexa 594 secondary ([ab150120](#)) at 2 µg/ml (shown in pseudo color red). Nuclear DNA was labeled in blue with DAPI.

Negative controls: 1– Rabbit primary and anti-mouse secondary antibody; 2 – Mouse primary antibody and anti-rabbit secondary antibody. Controls 1 and 2 indicate that there is no unspecific reaction between primary and secondary antibodies used.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

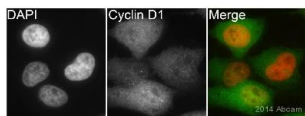
Immunohistochemistry was performed on human head and neck squamous cell carcinoma tissue using a rabbit monoclonal antibody against the CCND1 protein ab134175 on 3- μ m slides using 224 paraffin sections via the standard SP method.

Panel C: An example of low Cyclin D1 expression.

Panel G: An example of high Cyclin D1 expression.

For full image please see paper.

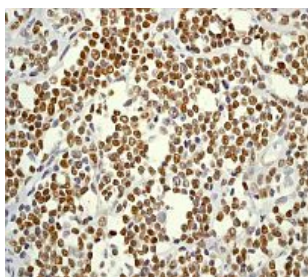
Feng et al PLoS One.
2011;6(10):e26399. doi:
10.1371/journal.pone.002
6399. Epub 2011 Oct 31.
Fig 1. Reproduced under
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This image is courtesy of a
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submitted by Kirk
Mcmanus.

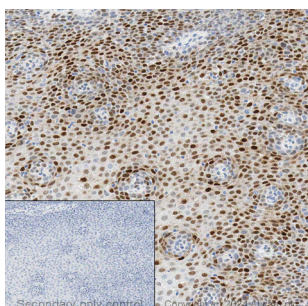
Immunocytochemistry/ Immunofluorescence - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

Immunocytochemical analysis of HeLa (Human epithelial cell line from cervix adenocarcinoma) cells, labeling Cyclin D1 with ab134175 at a dilution of 1/200. Cells were paraformaldehyde fixed and permeabilized with 0.5% Triton X-100 in PBS. Incubation with the primary antibody was for 1 hour at 22°C. Cells were counterstained with DAPI following immunostaining.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

Immunohistochemical analysis of paraffin-embedded human mantle cell lymphoma tissue, labeling Cyclin D1 with unpurified ab134175 at 1/100 dilution. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

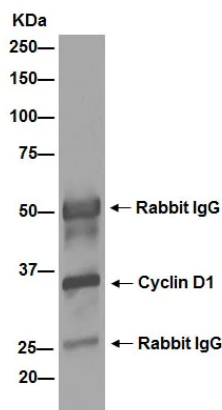


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

Immunohistochemical analysis of formalin fixed paraffin embedded human tonsil labelling Cyclin D1 with ab134175 at a concentration of 1 μ g/ml. The immunostaining was performed on a Ventana DISCOVERY ULTRA (Roche Tissue Diagnostics) instrument with a OptiView DAB IHC Detection Kit. Heat mediated antigen retrieval was performed with DISCOVERY cell conditioning solution (CC1) 100°C, pH8.5 for 32mins.

Anti-Cyclin D1 antibody [EPR2241] ab134175 was incubated for 16mins at 37°C. Sections were counterstained with Hematoxylin II. Image inset shows absence of staining in secondary antibody only control.

Customers are encouraged to optimise antigen retrieval conditions, antibody concentration, incubation times and temperature for best results in their own IHC assay workflow (automated and manual).



Immunoprecipitation - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

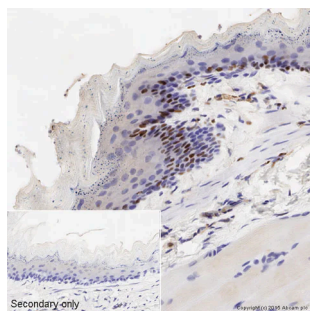
ab134175 (purified) at 1/30 immunoprecipitating cyclin D1 in A431 (Human epidermoid carcinoma cell line) cells. For western blotting, an HRP-conjugated goat anti-rabbit IgG, was used as the secondary antibody (1/1000).

Blocking/Dilution buffer and concentration: 5% NFDN/TBST.

All lanes:

Immunoprecipitation - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

Predicted band size: 34 kDa

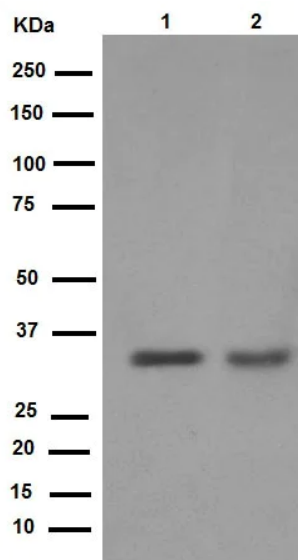


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

Cyclin D1 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Cyclin D1 antibody

IHC image of ab134175 staining Cyclin D1 in rat esophagus formalin fixed paraffin embedded tissue sections, performed on a Leica Bond. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab134175, 5µg/ml working concentration, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with hematoxylin and mounted with DPX. No primary antibody was used in the secondary only control (shown on the inset).

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.



Western blot - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

Blocking/Dilution buffer: 5% NFDm/TBST.

All lanes:

Western blot - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175) at 1/100000 dilution

Lane 1:

MCF7 (Human breast adenocarcinoma cell line) cell lysate at 20 µg

Lane 2:

LNCap (Human prostate cancer cell line) cell lysate at 20 µg

Secondary

All lanes:

HRP goat anti-rabbit (H+L) at 1/1000 dilution

Predicted band size: 34 kDa

Observed band size: 34 kDa



Western blot - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

Cyclin D1 Western blot staining using rabbit Anti-Cyclin D1 antibody

MG-63 cells were incubated at 37°C for 24h with vehicle control (0 µM) and different concentrations of 3-aminobenzamide (3-AB) ([ab141069](#)). Decreased expression of cyclin D1 (unpurified ab134175) in MG-63 cells correlates with an increase in 3-aminobenzamide (3-AB) concentration, as described in literature.

Whole cell lysates were prepared with RIPA buffer (containing protease inhibitors and sodium orthovanadate), 10 µg of each were loaded on the gel and the WB was run under reducing conditions. After transfer the membrane was blocked for an hour using 5% BSA before being incubated with unpurified ab134175 at 1/500 dilution and [ab8226](#) at 1 µg/ml overnight at 4°C. Antibody binding was detected using an anti-rabbit HRP secondary antibody ([ab97051](#)) at 1/10000 dilution and visualised using ECL development solution.

All lanes:

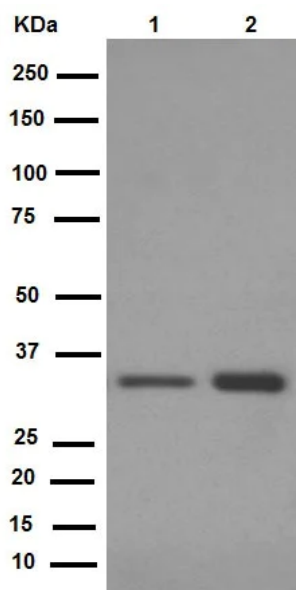
Western blot - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 34 kDa

Exposure time: 3min



Western blot - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

Blocking/Dilution buffer: 5% NFDM/TBST.

All lanes:

Western blot - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175) at 1/20000 dilution

Lane 1:

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

Lane 2:

A431 (Human epidermoid carcinoma cell line) cell lysate at 20 µg

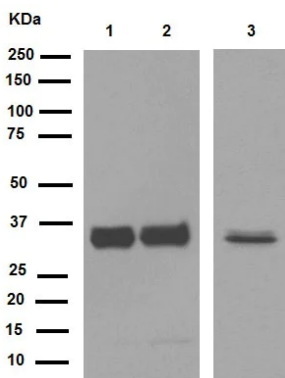
Secondary

All lanes:

HRP goat anti-rabbit (H+L) at 1/1000 dilution

Predicted band size: 34 kDa

Observed band size: 34 kDa



Western blot - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

Blocking/Dilution buffer: 5% NFDM/TBST.

All lanes:

Western blot - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175) at 1/10000 dilution

Lane 1:

Mouse kidney at 20 µg

Lane 2:

Mouse spleen at 20 µg

Lane 3:

Rat heart at 20 µg

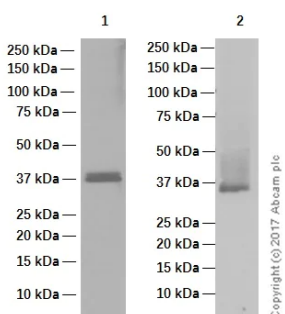
Secondary

All lanes:

HRP goat anti-rabbit (H+L) at 1/1000 dilution

Predicted band size: 34 kDa

Observed band size: 34 kDa



Western blot - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

Blocking and dilution buffer: 5% NFDM/TBST.

Lane 1:

Western blot - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175) at 0.04 µg/mL

Lane 2:

Western blot - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175) at 2 µg/mL

Lane 1:
His-Tagged full-length human Cyclin D1 (aa 1 to 295) recombinant protein at 0.015 µg

Lane 2:
His-Tagged full-length human Cyclin D2 (aa 1 to 289) recombinant protein at 0.015 µg

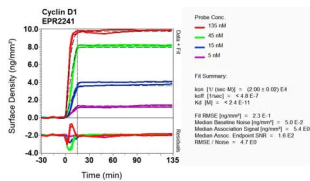
Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/20000 dilution

Predicted band size: 34 kDa

Observed band size: 37 kDa

Exposure time: 3min



OI-RD Scanning - Anti-Cyclin D1 antibody [EPR2241] - C-terminal (ab134175)

We have systematically measured KD (the equilibrium dissociation constant between the antibody and its antigen), of more than 840 recombinant antibodies to assess not only their individual KD values but also to see the average affinity of antibody. Based on the comparison with published literature values for mouse monoclonal antibodies, Recombinant antibodies appear to be on average 1-2 order of magnitude higher affinity.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.