

# Datasheet for ABIN487479 **anti-Cyclin D2 antibody**

# 2 Images



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#### Overview

Quantity:	0.1 mg
Target:	Cyclin D2 (CCND2)
Reactivity:	Human, Mouse
Host:	Mouse
Clonality:	Monoclonal
Conjugate:	This Cyclin D2 antibody is un-conjugated
Application:	Western Blotting (WB), Immunoprecipitation (IP)
Product Details	
Clone:	DCS-5
Isotype:	lgG2b
Specificity:	This antibody reacts with Human and Mouse cyclin D2 on Western blotting and Immunoprecipitation.
Cross-Reactivity (Details):	Species reactivity (tested):Human and Mouse.
Characteristics:	Synonyms: Cyclin-D2, CCND2
Purification:	Protein-A Sepharose Chromatography.
Target Details	
Target:	Cyclin D2 (CCND2)
Alternative Name:	Cyclin D2 (CCND2 Products)

#### Target Details

Background:

Cyclin D2 (34 kDa) is one of three D-type cyclins that are synthesized in G1 phase and induced in response to agents that promote re-entry into the cell cycle. D-type cyclins assemble with cyclin-dependent kinases Cdk4 and Cdk6 to form complexes that phosphorylate key substrates, including the retinoblastoma protein Rb, involved in proliferation. Generally, D cyclins appear late in the G1 phase of the cell cycle, however, cyclin D2 accumulates in Go and at the G1/S interface. A role for cyclin D2 has been suggested in differentiation and oncogenic transformation. Cyclin D2 is expressed in macrophages in response to CSF-1 stimulation, and overexpression of cyclin D2 is associated with increased in vivo invasiveness of human squamous carcinoma cells. Synonyms: CCND2, Cyclin-D2

Gene ID:

894

UniProt:

P30279

Pathways:

Cell Division Cycle, Mitotic G1-G1/S Phases

#### **Application Details**

Application Notes:

Western blott: 1-5  $\mu$ g/mL for chemiluminescence detection system. Immunoprecipitation: 2  $\mu$  g/200-300  $\mu$ L of 5x10e cells. Positive Control: NIH/3T3 cells. It is reported that this clone is reactive with several Human cells such as U-2-OS, Bristol-8and Lovo36 (See Reference 10). This antibody is reported to work in Immunocytochemistry (See Reference 10)Detailed procedure is provided in Protocols.

Other applications not tested.

Optimal dilutions are dependent on conditions and should be determined by the user.

Protocol:

SDS-PAGE & Western Blotting1) Wash the cells 3 times with PBS and suspend with 10 volume of cold Lysis buffer (50 mMTris-HCl, pH 7. 2, 250 mM NaCl, 0. 1% NP-40, 2 mM EDTA, 10% glycerol) containing appropriate protease inhibitors. Incubate it at 4°C with rotating for 30 minutes, then sonicate briefly (up to 10 seconds). 2) Centrifuge the tube at 12,000 x g for 10 minutes at 4°C and transfer the supernatant to another tube. Measure the protein concentration of the supernatant and add the Lysis buffer to make 8 mg/mL solution. 3) Mix the sample with equal volume of Laemmli's sample buffer. 4) Boil the samples for 2 minutes and centrifuge. Load 10  $\mu$ L of the sample per lane in a 1mm thick SDS-polyacrylamide gel for electrophoresis. 5) Blot the protein to a polyvinylidene difluoride (PVDF) membrane at 1 mA/cm2 for 1 hourin a semi-dry transfer system. (Transfer Buffer: 25 mM Tris, 190 mM glycine, 20% MeOH). See the manufacture's manual for precise transfer procedure. 6) To reduce nonspecific binding, soak the membrane in 10% skimmed milk (in PBS, pH7. 2) for 1 hour at RT, or overnight at 4°C. 7) Incubate the membrane with primary antibody diluted with PBS, pH 7. 2 containing 1%skimmed milk as suggest in the APPLICATIONS for 1 hour at room temperature. (Theconcentration of antibody will depend on condition. 8) Wash the membrane with PBS-T (5 min x 6 times). 9) Incubate the membrane with the 1: 10,000 HRP-conjugated anti-mouse IgG diluted with1% skimmed milk (in PBS, pH 7. 2) for 1 hour at room temperature10) Wash the membrane with PBS (10 minutes x 3 times)11) Wipe excess buffer on the membrane, then incubate it with appropriatechemiluminescence reagent for 1 minute. 12)Remove extra reagent from the membrane by dabbing with paper towel, and seal it inplastic wrap. 13) Expose to an X-ray film in a dark room for 3 minutes. Positive Control for Western blotting:

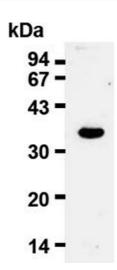
NIH/3T3Immunoprecipitation1) Wash the cells 3 times with PBS and suspend with 10 volume of cold Lysis buffer (50 mMTris-HCl pH 7. 2, 250 mM NaCl, 0. 1% NP-40, 2 mM EDTA, 10% glycerol) containingappropriate protease inhibitors. Incubate it at 4°C with rotating for 30 minutes, thensonicate briefly (up to 10 seconds)2) Centrifuge the tube at 12,000 x g for 10 min at 4°C and transfer the supernatant toanother tube. 3) Add primary antibody as suggest in the APPLICATIONS into 300 ìL of the supernatant. Mix well and incubate with gentle agitation for 30-120 minutes at 4°C.

Restrictions:

For Research Use only

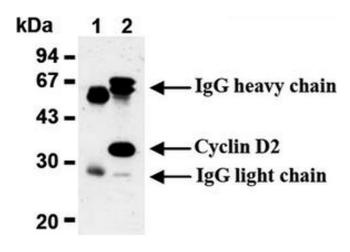
## Handling

Concentration:	1.0 mg/mL
Buffer:	PBS, pH 7.2 containing 50 % Glycerol without preservatives.
Preservative:	Without preservative
Storage:	-20 °C
Storage Comment:	Store the antibody undiluted at -20 °C. Shelf life: one year from despatch.
Expiry Date:	12 months



## **Western Blotting**

Image 1.



#### **Western Blotting**

Image 2.