

Basic Information

Product Name	Anti-Cyclin D1/CCND1 Antibody
Gene Name	CCND1
Source	Rabbit
Isotype	IgG
Species Reactivity	human, rat
Tested Application	WB, FCM
Contents	500 ug/ml antibody with PBS , 0.02% NaN ₃ , 1 mg BSA and 50% glycerol.
Immunogen	A synthetic peptide corresponding to a sequence at the N-terminus of human Cyclin D1(19-37aa DANLLNDRVLRAMLKAEET), different from the related mouse and rat sequences by two amino acids.
concentration	500 ug/ml
Purification	Immunogen affinity purified.
Observed MW	34KD
Dilution Ratios	Western blot(WB): 1:500-2000 Flow cytometry (FCM):1-3 µg/1x10 ⁶ cells

Storage

12 months from date of receipt, -20°C as supplied. 6 months 2 to 8°C after reconstitution. Avoid repeated freezing and thawing.

Background Information

Cyclin D1, also known as CCND1, is a human gene. The protein encoded by this gene belongs to the highly conserved cyclin family, whose members are characterized by a dramatic periodicity in protein abundance throughout the cell cycle. Cyclin D1 encodes the regulatory subunit of a holoenzyme that phosphorylates and inactivates the retinoblastoma protein and promotes progression through the G1-S phase of the cell cycle. Amplification or overexpression of cyclin D1 plays pivotal roles in the development of a subset of human cancers including parathyroid adenoma, breast cancer, colon cancer, lymphoma, melanoma, and prostate cancer. The cyclin D1 gene is overexpressed in human breast cancers and is required for oncogene-induced tumorigenesis. Briskin et al. (2003) found that prolactin (PRL; 176760) induced IGF2 (147470) mRNA and IGF2 induced cyclin D1 protein expression in mouse mammary epithelial cultures. And they also concluded that IGF2 is a mediator of prolactin-induced alveologenesis and that prolactin, IGF2, and cyclin D1 are components of a developmental pathway in mammary gland.

Selected Validation Data

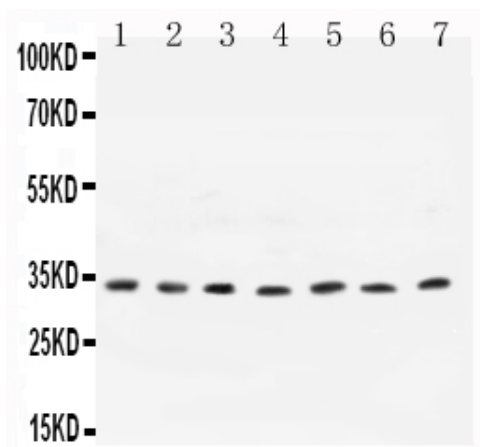


Figure 1. Western blot analysis of Cyclin D1 using anti-Cyclin D1 antibody (BA0770-2).

Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.

Lane 1: Rat Testis Tissue Lysate,

Lane 2: Human Placenta Tissue Lysate,

Lane 3: Rat Brain Tissue Lysate,

Lane 4: MCF-7 Whole Cell Lysate,

Lane 5: COLO320 Whole Cell Lysate,

Lane 6: SW620 Whole Cell Lysate,

Lane 7: MM231 Whole Cell Lysate.

After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-Cyclin D1 antigen affinity purified polyclonal antibody (Catalog # BA0770-2) at 0.5 µg/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for Cyclin D1 at approximately 34KD. The expected band size for Cyclin D1 is at 33KD.

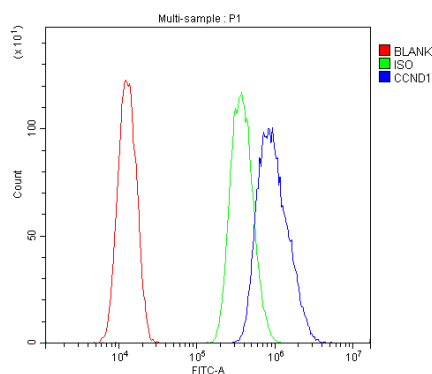


Figure 2. Flow Cytometry analysis of U-87MG cells using anti- Cyclin D1 antibody (BA0770-2).

Overlay histogram showing U-87MG cells stained with BA0770-2 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-Cyclin D1 Antibody (BA0770-2,1µg/1x10⁶ cells) for 30 min at 20°C. DyLight488 conjugated goat anti-rabbit IgG (BA1127, 5-10µg/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1µg/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.