

Anti-Cdk6 Antibody [SD20-50]

ET1612-3



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human
Applications:	IF-Cell, IF-Tissue, IHC-P, FC, WB
Molecular Wt:	Predicted band size: 37 kDa
Clone number:	SD20-50

Description: Cell division protein kinase 6 (CDK6) is an enzyme encoded by the CDK6 gene.[5][6] It is regulated by cyclins, more specifically by Cyclin D proteins and Cyclin-dependent kinase inhibitor proteins. The protein encoded by this gene is a member of the cyclin-dependent kinase, (CDK) family, which includes CDK4. CDK family members are highly similar to the gene products of *Saccharomyces cerevisiae* cdc28, and *Schizosaccharomyces pombe* cdc2, and are known to be important regulators of cell cycle progression in the point of regulation named R or restriction point. This kinase is a catalytic subunit of the protein kinase complex, important for the G1 phase progression and G1/S transition of the cell cycle and the complex is composed also by an activating sub-unit; the cyclin D. The activity of this kinase first appears in mid-G1 phase, which is controlled by the regulatory subunits including D-type cyclins and members of INK4 family of CDK inhibitors. This kinase, as well as CDK4, has been shown to phosphorylate, and thus regulate the activity of, tumor suppressor Retinoblastoma protein making CDK6 an important protein in cancer development.

Immunogen: Synthetic peptide within Human Cdk6 aa 277-326 / 326.

Positive control: HeLa cell lysate, SK-OV-3 cell lysate, MCF7 cell lysate, K-562 cell lysate, HeLa, SW480, 293, human colon carcinoma tissue, human tonsil tissue.

Subcellular location: Cytoplasm, Nucleus, Cell projection.

Database links: SwissProt: Q00534 Human

Recommended Dilutions:

WB	1:500-1:2,000
IF-Cell	1:50-1:200
IF-Tissue	1:50-1:200
IHC-P	1:50-1:200
FC	1:50-1:100

Storage Buffer: 1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Shipped at 4°C. Store at +4°C short term (1-2 weeks). Store at -20°C long term.

Purity: Protein A affinity purified.

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Orders:0086-571-88062880

Technical:0086-571-89986345

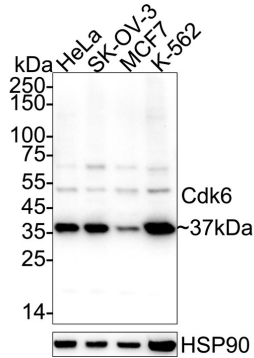
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Images

Fig1: Western blot analysis of Cdk6 on different lysates with Rabbit anti-Cdk6 antibody (ET1612-3) at 1/2,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: SK-OV-3 cell lysate
Lane 3: MCF7 cell lysate
Lane 4: K-562 cell lysate



Lysates/proteins at 20 µg/Lane.

Predicted band size: 37 kDa
Observed band size: 37 kDa

Exposure time: 59 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1612-3) at 1/2,000 dilution was used in primary antibody dilution (K1803) at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

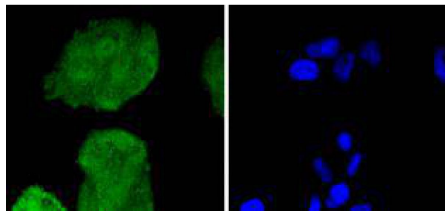


Fig2: ICC staining of Cdk6 in HeLa cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1612-3, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

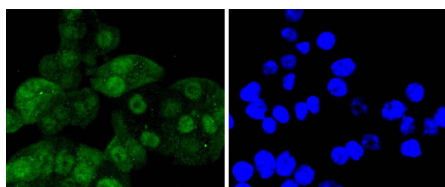


Fig3: ICC staining of Cdk6 in SW480 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1612-3, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

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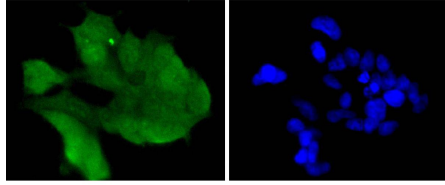


Fig4: ICC staining of Cdk6 in 293 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1612-3, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

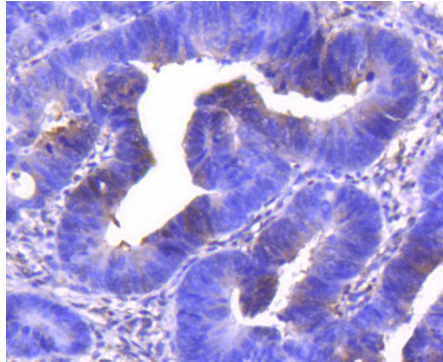


Fig5: Immunohistochemical analysis of paraffin-embedded human colon carcinoma tissue using anti-Cdk6 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1612-3, 1/50) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

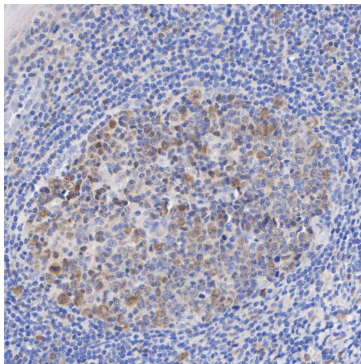


Fig6: Immunohistochemical analysis of paraffin-embedded human tonsil tissue with Rabbit anti-Cdk6 antibody (ET1612-3) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1612-3) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

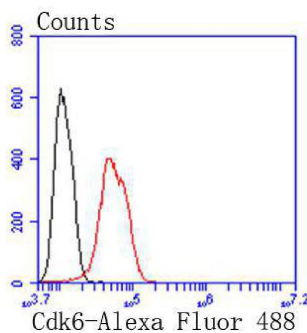


Fig7: Flow cytometric analysis of Cdk6 was done on HeLa cells. The cells were fixed, permeabilized and stained with the primary antibody (ET1612-3, 1/50) (red). After incubation of the primary antibody at room temperature for an hour, the cells were stained with a Alexa Fluor 488-conjugated Goat anti-Rabbit IgG Secondary antibody at 1/1000 dilution for 30 minutes. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

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Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

1. Yuan, W. et al. 2016. CDK6 mediates the effect of attenuation of miR-1 on provoking cardiomyocyte hypertrophy. *Mol. Cell. Biochem.* 412: 289-96.
2. Guo, B. et al. 2015. Naringin suppresses the metabolism of A375 cells by inhibiting the phosphorylation of c-Src. *Tumour biology : the journal of the International Society for Oncodevelopmental Biology and Medicine.*

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