Anti-p16INK4a Antibody

ER2001-30

Product Type:	Rabbit polyclonal IgG, primary antibodies
Species reactivity:	Human, Mouse
Applications:	WB, IF-Cell, FC
Molecular Wt:	Predicted band size: 16 kDa

Description: p16 (also known as p16INK4a, cyclin-dependent kinase inhibitor 2A, CDKN2A, multiple tumor suppressor 1 and numerous other synonyms), is a protein that slows cell division by slowing the progression of the cell cycle from the G1 phase to the S phase, thereby acting as a tumor suppressor. It is encoded by the CDKN2A gene. A deletion (the omission of a part of the DNA sequence during replication) in this gene can result in insufficient or non-functional p16, accelerating the cell cycle and resulting in many types of cancer. p16 can be used as a biomarker to improve the histological diagnostic accuracy of grade 3 cervical intraepithelial neoplasia (CIN). p16 is also implicated in the prevention of melanoma, oropharyngeal squamous cell carcinoma, cervical cancer, vulvar cancer and esophageal cancer. p16 was discovered in 1993. It is a protein with 148 amino acids and a molecular weight of 16 kDa that comprises four ankyrin repeats. The name of p16 is derived from its molecular weight, and the alternative name p16INK4a refers to its role in inhibiting cyclin-dependent kinase CDK4.

Positive control: HeLa cell lysate, HepG2 cell lysate, HEK-293 cell lysate, NIH:OVCAR-3 cell lysate, A20 cell lysate, MEF cell lysate, HeLa, MEF.

Subcellular location: Cytoplasm, Nucleus.

Database links: SwissProt: P42771 Human | P51480 Mouse

Recommended Dilutions:

WB	1:5,000
IF-Cell	1:5,000
FC	1:1,000

Storage Buffer: PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20℃ long term.

Purity: Protein A affinity purified.

Images

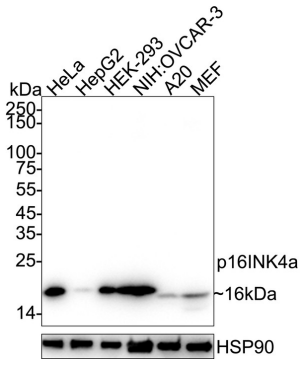

Fig1: Western blot analysis of p16INK4a on different lysates with Rabbit anti-p16INK4a antibody (ER2001-30) at 1/5,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: HepG2 cell lysate
Lane 3: HEK-293 cell lysate
Lane 4: NIH:OVCAR-3 cell lysate
Lane 5: A20 cell lysate
Lane 6: MEF cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 16 kDa
Observed band size: 16 kDa

Exposure time: 10 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 (ER2001-30) at 1/5,000 dilution was used in primary antibody dilution (K1803) at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

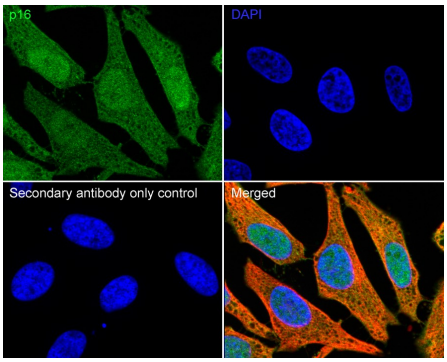

Fig2: Immunocytochemistry analysis of HeLa cells labeling p16INK4a with Rabbit anti-p16INK4a antibody (ER2001-30) at 1/5,000 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-p16INK4a antibody (ER2001-30) at 1/5,000 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (HA601187, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

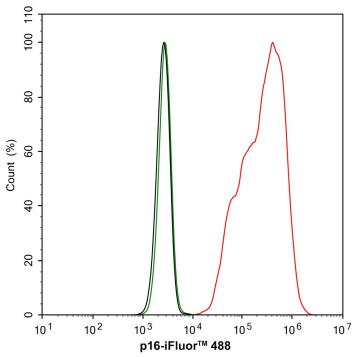

Fig3: Flow cytometric analysis of HeLa cells labeling p16INK4a.

Cells were fixed and permeabilized. Then stained with the primary antibody (ER2001-30, 1/1,000) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4℃ for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

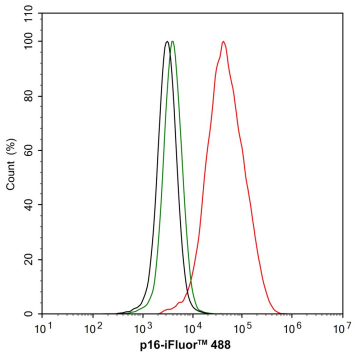

Fig4: Flow cytometric analysis of MEF cells labeling p16INK4a.

Cells were fixed and permeabilized. Then stained with the primary antibody (ER2001-30, 1/1,000) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4℃ for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Background References

1. Safwan-Zaiter H et al. P16INK4A-More Than a Senescence Marker. Life (Basel). 2022 Aug
2. Shi J et al. P16ink4a overexpression ameliorates cardiac remodeling of mouse following myocardial infarction via CDK4/pRb pathway. Biochem Biophys Res Commun. 2022 Mar