

Anti-p21 Antibody [PSH22-37]

HA724295



| | |
|----------------------------|---|
| Product Type: | Recombinant Rabbit monoclonal IgG, primary antibodies |
| Species reactivity: | Human |
| Applications: | WB, IF-Cell, IHC-P, IP |
| Molecular Wt: | Predicted band size: 18 kDa |
| Clone number: | PSH22-37 |

Description: p21Cip1 (alternatively p21Waf1), also known as cyclin-dependent kinase inhibitor 1 or CDK-interacting protein 1, is a cyclin-dependent kinase inhibitor (CKI) that is capable of inhibiting all cyclin/CDK complexes, though is primarily associated with inhibition of CDK2. p21 represents a major target of p53 activity and thus is associated with linking DNA damage to cell cycle arrest. This protein is encoded by the CDKN1A gene located on chromosome 6 (6p21.2) in humans.

Positive control: HCT 116 cell lysate, A549 cell lysate, MCF7 cell lysate, MCF7 treated with 10 μ M Nutlin 3a for 24 hours cell lysate, MCF7, human colon cancer tissue, human ovarian cancer tissue, human breast cancer tissue.

Subcellular location: Cytoplasm, Nucleus.

Database links: SwissProt: P38936 Human

Recommended Dilutions:

| | |
|----------------|--------------------|
| WB | 1:2,000 |
| IF-Cell | 1:5,000 |
| IHC-P | 1:200 |
| IP | 1-2 μ g/sample |

Storage Buffer: 1*PBS (pH7.4), 0.1% BSA, 40% Glycerol, 0.2% Proclean 950.

Storage Instruction: Shipped at 4 $^{\circ}$ C. Store at +4 $^{\circ}$ C short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20 $^{\circ}$ C long term.

Purity: Protein A affinity purified.

Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880

Technical:0086-571-89986345

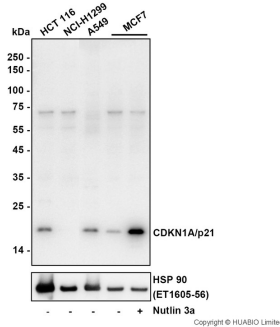
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Images

Fig1: Western blot analysis of p21 on different lysates with Rabbit anti-p21 antibody (HA724295) at 1/2,000 dilution.

Lane 1: HCT 116 (Human colon cancer cells)
 Lane 2: NCI-H1299 (Human non-small cell Lung carcinoma cells) (Negative)
 Lane 3: A549 (Human lung adenocarcinoma cells)
 Lane 4: MCF7 (Human breast cancer cells)
 Lane 5: MCF7 (Human breast cancer cells) treated with Nutlin 3a (10uM, 24h)



Lysates/proteins at 10 µg/Lane.
 Exposure time: 1 minutes; ECL: K1801

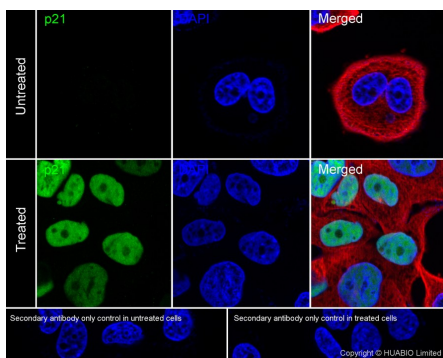
H1299 is a negative control (PMID: 23268708).

Blocking: 5% NFD/MTBST, 1 hour at room temperature
 Primary antibody: HA724295, 1/2,000 in primary antibody dilution buffer (K1803), overnight at 4 °C
 Secondary antibody: Goat anti-Rabbit IgG-HRP (HA1001), 1/50,000 in 5% NFD/MTBST, 1 hour at room temperature

Predicted band size: 21 kDa
 Observed band size: 21 kDa

Fig2: Application: Immunocytochemistry (IF-cell)

Species: Human
 Sample: MCF7 (Human cervix adenocarcinoma epithelial cell) treated or untreated with Nutlin 3a (10 µM, 24h)



Fixation: 4% Paraformaldehyde, 15 minutes at room temperature.
 Permeabilization: 0.1% Triton X-100, 15 minutes at room temperature.

Blocking: 1% BSA + 10% normal goat serum, 1 hour at room temperature.

Antibody dilution buffer: 1% BSA in PBST.

Primary antibody: HA724295, 1/5,000, overnight at 4 °C.

Secondary antibody: Goat Anti-Rabbit IgG (iFluor™ 488, HA1121), 45 minutes at room temperature.

Counterstain: Beta tubulin (M120505-2, red), 1/100, overnight at 4 °C. The Nuclear counterstain was DAPI (Blue).

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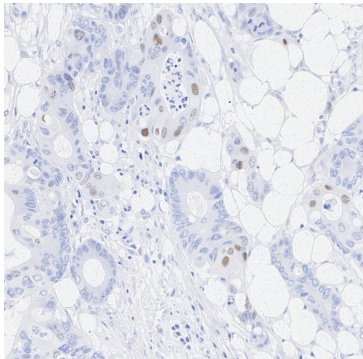


Fig3: Application: Immunohistochemistry (IHC-P)

Species: Human
Tissue: Colon cancer
Sample: Paraffin-embedded section

Antigen retrieval: Heat-mediated, Tris-EDTA buffer (pH 9.0), 20 minutes at 95°C.

Wash buffer: 1× PBST
Endogenous peroxidase blocking: 3% H₂O₂, 10 minutes at room temperature.
Blocking: 1% BSA + 10% normal goat serum, 10 minutes at room temperature.
Primary antibody: HA724295, 1/200, 1 hour at room temperature.
Secondary antibody: HA1119, 20 minutes at room temperature.

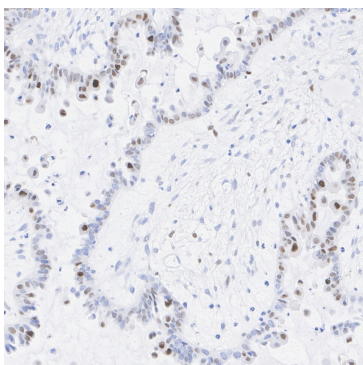
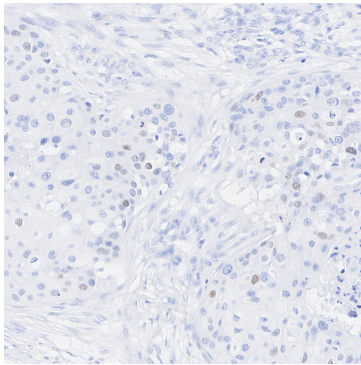


Fig4: Application: Immunohistochemistry (IHC-P)

Species: Human
Tissue: Ovarian Carcinoma
Sample: Paraffin-embedded section

Antigen retrieval: Heat-mediated, Tris-EDTA buffer (pH 9.0), 20 minutes at 95°C.

Wash buffer: 1× PBST
Endogenous peroxidase blocking: 3% H₂O₂, 10 minutes at room temperature.
Blocking: 1% BSA + 10% normal goat serum, 10 minutes at room temperature.
Primary antibody: HA724295, 1/200, 1 hour at room temperature.
Secondary antibody: HA1119, 20 minutes at room temperature.

**Fig5:** Application: Immunohistochemistry (IHC-P)

Species: Human

Tissue: Invasive Ductal Carcinoma of the Breast

Sample: Paraffin-embedded section

Antigen retrieval: Heat-mediated, Tris-EDTA buffer (pH 9.0), 20 minutes at 95°C.

Wash buffer: 1× PBST

Endogenous peroxidase blocking: 3% H₂O₂, 10 minutes at room temperature.

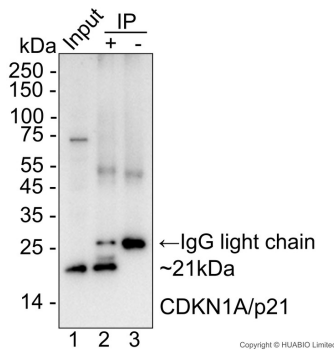
Blocking: 1% BSA + 10% normal goat serum, 10 minutes at room temperature.

Primary antibody: HA724295, 1/200, 1 hour at room temperature.

Secondary antibody: HA1119, 20 minutes at room temperature.

Fig6: Application: Immunoprecipitation (IP)

p21 was immunoprecipitated in 0.2 mg HCT 116 (Human colon cancer cell) cell lysate with HA724295 at 2 µg/10 µl beads. Western blot was performed from the immunoprecipitate using HA724295 at 1/2,000 dilution. Anti-Rabbit IgG for IP Nano-secondary antibody (NBI01H) at 1/5,000 dilution was used for 1 hour at room temperature.



Lane 1: HCT 116 cell lysate (input)

Lane 2: Rabbit IgG instead of HA724295 in HCT 116 cell lysate

Lane 3: HA724295 IP in HCT 116 cell lysate

Exposure time: 1 minute

Blocking: 5% NFDM/TBST, 1 hour at room temperature

Primary dilution: HA724295, 1/2,000 in primary antibody dilution buffer (K1803), 2 hours at room temperature

Predicted band size: 21 kDa

Observed band size: 21 kDa

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

Background References

1. Engeland K. Cell cycle regulation: p53-p21-RB signaling. Cell Death Differ. 2022 May
2. Bautista L et al . p21-Activated Kinases in Thyroid Cancer. Endocrinology. 2020 Aug

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