

Anti-p40 Antibody [PDH0-02]

HA721222



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	IHC-P, WB, IF-Cell
Molecular Wt:	Predicted band size: 77 kDa
Clone number:	PDH0-02

Description: p63 protein (p63) is a nuclear protein, a transcription factor. The p40 is an isoform of p63, a transcription factor that regulates many cell activities, including cell proliferation, maintenance, and differentiation. It performs as a sensitive and specific tool for aiding in the identification of squamous cell carcinoma of the lung. In addition to its utility as a squamous differentiation marker, p40 has also been proven to be a valuable marker for highlighting myoepithelial and basal cell populations in prostate, breast, skin, and salivary gland. Strong p40 expression is frequently observed in esophageal cancerous squamous lesions. Immunohistochemical detection of p40 can also be helpful in identifying urothelial carcinoma. In cases of prostate carcinoma, p40 is almost always found to be negative for basal cell staining. Among carcinomas, p40 has approximately the same sensitivity as p63 but a higher specificity, as the TAp63 isoform is expressed more widespread in eg., adenocarcinomas. Moreover, p63 occur in lymphomas that are p40 negative. Placenta is recommended as positive tissue control for p40, where an at least weak to moderate, distinct nuclear staining reaction of cytotrophoblasts must be seen. The cytotrophoblasts should be visible even at low magnification (5x objective). Supportive to placenta, tonsil can be used as positive and negative tissue control. Virtually all squamous epithelial cells must show a moderate to strong, distinct nuclear staining reaction. No nuclear or cytoplasmic staining reaction should be seen in other cell types.

Immunogen: Synthetic peptide within human p40 aa 2-50 / 586 (Q9H3D4-2)

Positive control: Human tonsil tissue, human skin tissue, human lung squamous cell carcinoma tissue, human bladder carcinoma tissue, human prostate tissue, human breast tissue, rat skin tissue, mouse bladder tissue, A431 cell lysates, A431.

Subcellular location: Nucleus.

Database links: SwissProt: Q9H3D4 Human

Recommended Dilutions:

IHC-P	1:1,000-1:2,000
WB	1:1,000
IF-Cell	1:100

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

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

Purity: Protein A affinity purified.

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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

Images

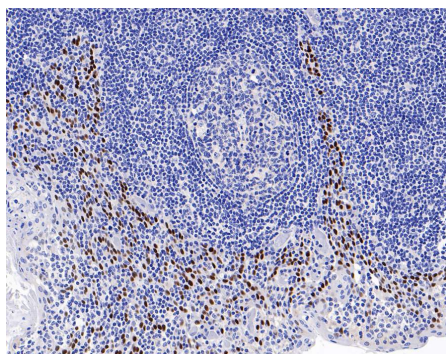


Fig1: Immunohistochemical analysis of paraffin-embedded human tonsil tissue with Rabbit anti-p40 antibody (HA721222) at 1/1,000 dilution.

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 (HA721222) at 1/1,000 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.

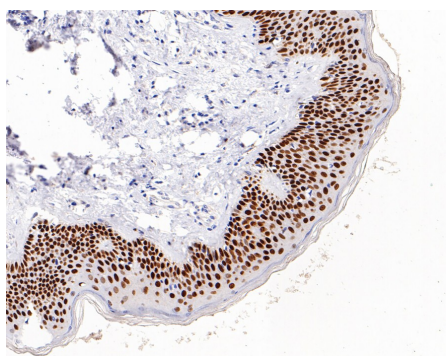


Fig2: Immunohistochemical analysis of paraffin-embedded human skin tissue with Rabbit anti-p40 antibody (HA721222) at 1/1,000 dilution.

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 (HA721222) at 1/1,000 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.

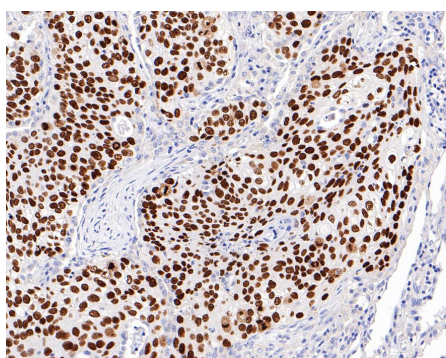


Fig3: Immunohistochemical analysis of paraffin-embedded human lung squamous cell carcinoma tissue with Rabbit anti-p40 antibody (HA721222) at 1/1,000 dilution.

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 (HA721222) at 1/1,000 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.

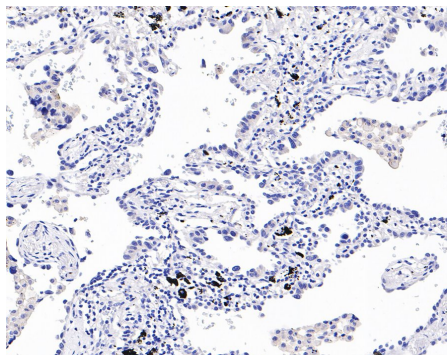


Fig4: Immunohistochemical analysis of paraffin-embedded human lung adenocarcinoma tissue (Negative control) with Rabbit anti-p40 antibody (HA721222) at 1/1,000 dilution.

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 (HA721222) at 1/1,000 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.

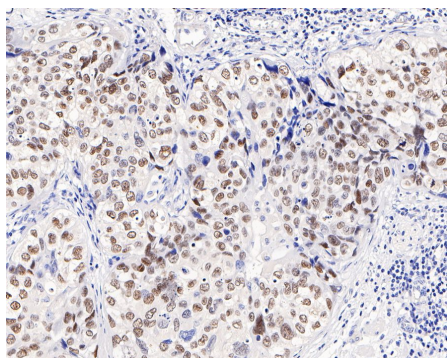


Fig5: Immunohistochemical analysis of paraffin-embedded human bladder carcinoma tissue with Rabbit anti-p40 antibody (HA721222) at 1/2,000 dilution.

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 (HA721222) at 1/2,000 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.

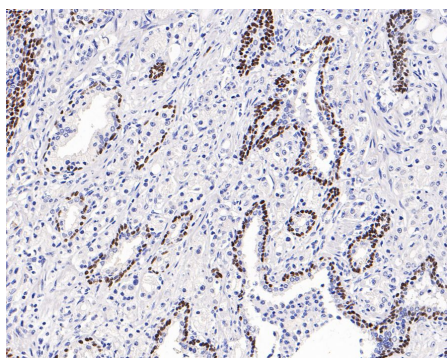


Fig6: Immunohistochemical analysis of paraffin-embedded human prostate tissue with Rabbit anti-p40 antibody (HA721222) at 1/2,000 dilution.

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 (HA721222) at 1/2,000 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.

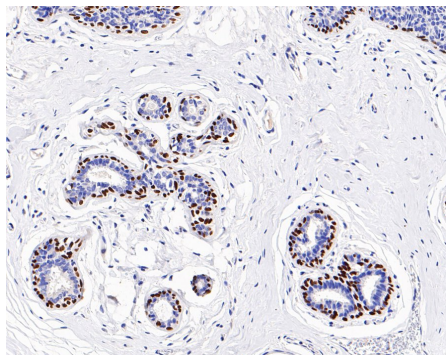


Fig7: Immunohistochemical analysis of paraffin-embedded human breast tissue with Rabbit anti-p40 antibody (HA721222) at 1/2,000 dilution.

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 (HA721222) at 1/2,000 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.

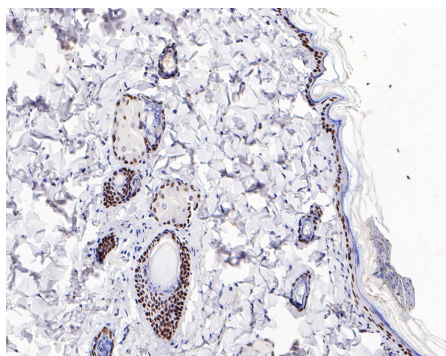


Fig8: Immunohistochemical analysis of paraffin-embedded rat skin tissue with Rabbit anti-p40 antibody (HA721222) at 1/2,000 dilution.

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 (HA721222) at 1/2,000 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.

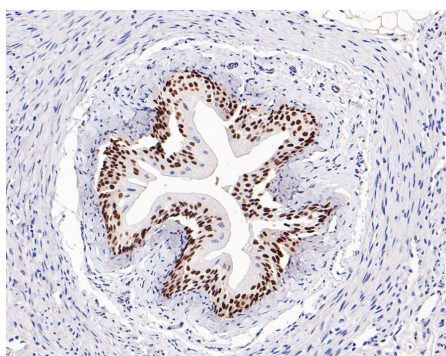


Fig9: Immunohistochemical analysis of paraffin-embedded mouse bladder tissue with Rabbit anti-p40 antibody (HA721222) at 1/2,000 dilution.

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 (HA721222) at 1/2,000 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.

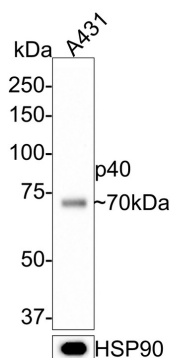


Fig10: Western blot analysis of p40 on A431 cell lysates with Rabbit anti-p40 antibody (HA721222) at 1/1,000 dilution.

Lysates/proteins at 10 µg/Lane.

Predicted band size: 77 kDa

Observed band size: 70 kDa

Exposure time: 3 minutes;

8% 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 (HA721222) at 1/1,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:300,000 dilution was used for 1 hour at room temperature.

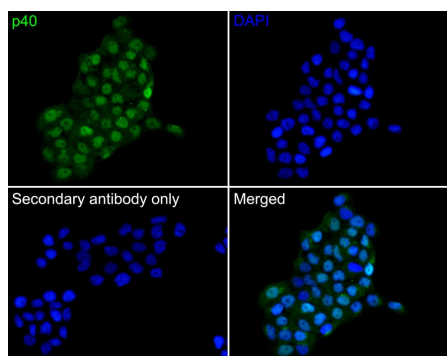


Fig11: Immunocytochemistry analysis of A431 cells labeling p40 with Rabbit anti-p40 antibody (HA721222) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 10 minutes at 37 °C, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-p40 antibody (HA721222) at 1/100 dilution in 2% negative goat serum overnight at 4 °C. 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.

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

Background References

1. Brustmann, Hermann. "p40 as a Basal Cell Marker in the Diagnosis of Prostate Glandular Proliferations: A Comparative Immunohistochemical Study with 34betaE12." Pathology research international vol. 2015 (2015): 897927.
2. Geddert, Helene et al. "The role of p63 and deltaNp63 (p40) protein expression and gene amplification in esophageal carcinogenesis." Human Pathology. 34,9 (2003): 850-856.

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