Anti-PAX8 Antibody [JJ08-88]

ET1701-50

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P
Molecular Wt:	Predicted band size: 48 kDa
Clone number:	JJ08-88

Description: PAX8 is a transcription factor crucial to the organogenesis and development of the thyroid gland, urogenital tract, placenta and inner ear. In the thyroid, PAX8 is a master gene that regulates maintenance of the differentiated thyroid follicular cell phenotype, where it controls and activates the transcription of the main proteins responsible for the functional activity of follicular cells such as thyroglobulin, thyroperoxidase and sodium/iodide symporter. In the developing kidney PAX8 is important for renal vescicle formation. PAX8 regulates the expression of the WT1 gene. PAX8 appears to be currently the most sensitive and specific marker for renal cell carcinoma and ovarian non-mucinous carcinoma. Follicular and papillary thyroid carcinoma are virually always PAX8 positive (while anaplastic carcinoma is positive in most cases and medullary thyroid carcinoma negative). PAX8 is also found in almost all cases of ovarian serous, endometrioid, transitional and clear cell carcinoma (while mucinous carcinoma is positive in a minor number of cases), and endometrial carcinoma.

Immunogen: Recombinant protein within Human PAX8 aa 101-450 / 450.

Positive control: SKOV-3 cell lysates, NIH: OVCAR-3 cell lysates, SKOV-3, NIH:OVCAR-3, human ovarian carcinoma tissue, human thyroid cancer tissue, human thyroid tissue, human kidney tissue, mouse kidney tissue, rat kidney tissue.

Subcellular location: Nucleus.

Database links: SwissProt: Q06710 Human | Q00288 Mouse | P51974 Rat

Recommended Dilutions:

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

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

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃ or -80℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

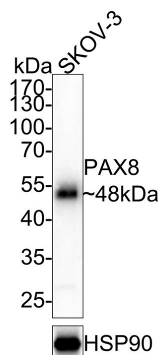

Fig1: Western blot analysis of PAX8 on SKOV-3 cell lysates with Rabbit anti-PAX8 antibody (ET1701-50) at 1/1,000 dilution.

Lysates/proteins at 10 µg/Lane.

Predicted band size: 48 kDa
Observed band size: 48 kDa

Exposure time: 20 seconds;

10% 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 (ET1701-50) 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 1:100,000 dilution was used for 1 hour at room temperature.

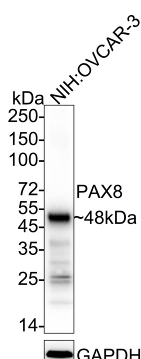

Fig2: Western blot analysis of PAX8 on NIH: OVCAR-3 cell lysates with Rabbit anti-PAX8 antibody (ET1701-50) at 1/1,000 dilution.

Lysates/proteins at 20 µg/Lane.

Predicted band size: 48 kDa
Observed band size: 48 kDa

Exposure time: 18 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 (ET1701-50) at 1/1,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

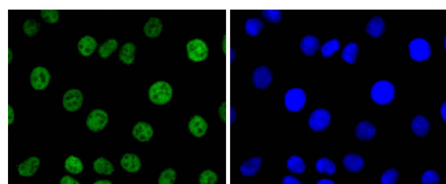

Fig3: ICC staining of PAX8 in SKOV-3 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 10% negative goat serum for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1701-50, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 conjugate-Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

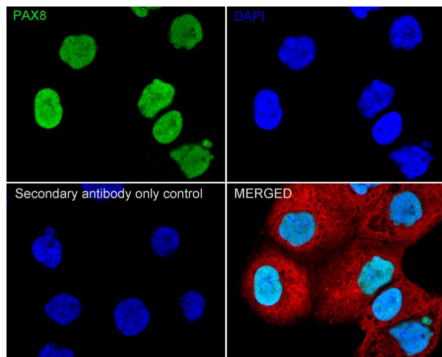

Fig4: Immunocytochemistry analysis of NIH:OVCAR-3 cells labeling PAX8 with Rabbit anti-PAX8 antibody (ET1701-50) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 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-PAX8 antibody (ET1701-50) at 1/100 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 (M1305-2, 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.

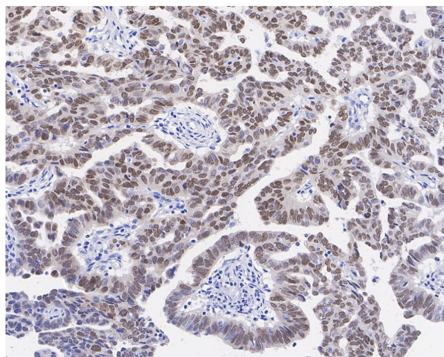

Fig5: Immunohistochemical analysis of paraffin-embedded human ovarian carcinoma tissue with Rabbit anti-PAX8 antibody (ET1701-50) at 1/500 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1701-50) at 1/500 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.

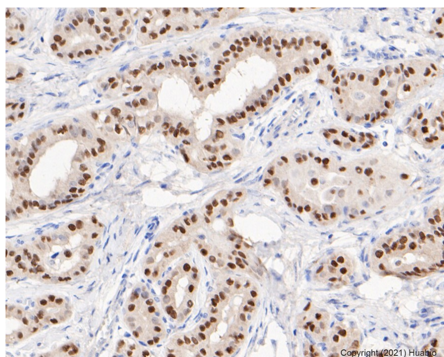

Fig6: Immunohistochemical analysis of paraffin-embedded human thyroid cancer tissue using anti-PAX8 antibody. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1701-50, 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.

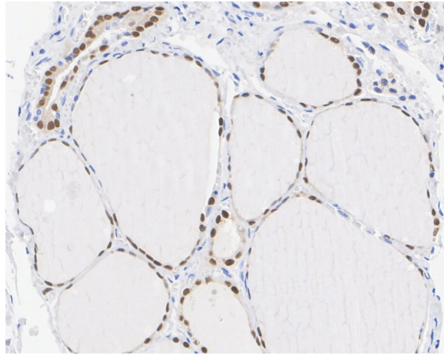

Fig7: Immunohistochemical analysis of paraffin-embedded human thyroid tissue with Rabbit anti-PAX8 antibody (ET1701-50) at 1/500 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1701-50) at 1/500 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.

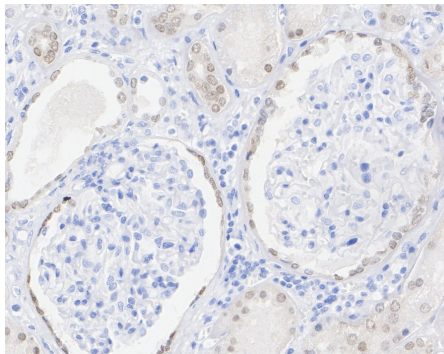

Fig8: Immunohistochemical analysis of paraffin-embedded human kidney tissue with Rabbit anti-PAX8 antibody (ET1701-50) at 1/500 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1701-50) at 1/500 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.

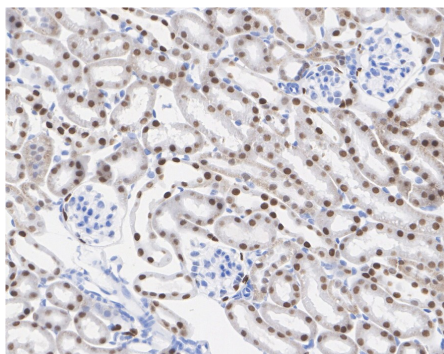

Fig9: Immunohistochemical analysis of paraffin-embedded mouse kidney tissue with Rabbit anti-PAX8 antibody (ET1701-50) at 1/500 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1701-50) at 1/500 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.

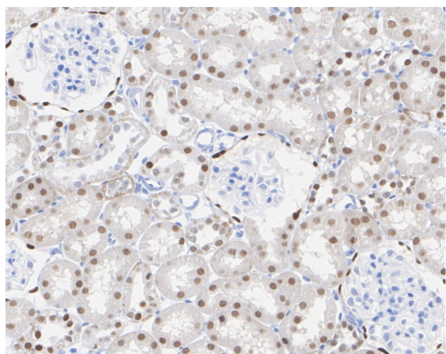

Fig10: Immunohistochemical analysis of paraffin-embedded rat kidney tissue with Rabbit anti-PAX8 antibody (ET1701-50) at 1/500 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1701-50) at 1/500 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.

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

Background References

1. Wang M et al. PAX2 and PAX8 reliably distinguishes ovarian serous tumors from mucinous tumors. Appl Immunohistochem Mol Morphol 23:280-7 (2015).
2. Sicking EM et al. Subtotal ablation of parietal epithelial cells induces crescent formation. J Am Soc Nephrol 23:629-40 (2012).