Anti-PAX5 Antibody [JJ08-87]

ET1701-49



Product Type: Species reactivity: Applications: Molecular Wt: Clone number:	Recombinant Rabbit monoclonal IgG, primary antibodies Human, Mouse, Rat WB, IF-Cell, IF-Tissue, IHC-P, FC Predicted band size: 42 kDa JJ08-87
Description:	Paired box protein Pax-5 is a protein that in humans is encoded by the PAX5 gene. The PAX5 gene is a member of the paired box (PAX) family of transcription factors. The central feature of this gene family is a novel, highly conserved DNA-binding domain, known as the paired box. The PAX proteins are important regulators in early development, and alterations in the expression of their genes are thought to contribute to neoplastic transformation. The PAX5 gene encodes the B-cell lineage specific activator protein (BSAP) that is expressed at early, but not late stages of B-cell differentiation. Its expression has also been detected in developing CNS and testis, therefore, PAX5 gene product may not only play an important role in B-cell differentiation, but also in neural development and spermatogenesis.
lmmunogen:	Synthetic peptide within human PAX5 aa 240-280.
Positive control:	Daudi cell lysate, Raji cell lysate, A20 cell lysate, human lymph nodes tissue, human tonsil tissue, human spleen tissue, mouse spleen tissue, rat spleen tissue, human appendix tissue, human B cell lymphoma tissue, Raji.
Subcellular location:	Nucleus.
Database links:	SwissProt: Q02548 Human Q02650 Mouse Entrez Gene: 500453 Rat
Recommended Dilutions: WB IF-Cell IF-Tissue IHC-P FC	1:10,000-1:20,000 1:500 1:500-1:1,000 1:1,000-1:2,000 1:1,000-1:2,000
Storage Buffer:	1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Shipped at 4 $^\circ\!{\rm C}$. Store at +4 $^\circ\!{\rm C}$ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20 $^\circ\!{\rm C}$ long term.
Purity:	Protein A affinity purified.

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Technical:0086-571-89986345

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Images



Fig1: Western blot analysis of PAX5 on different lysates with Rabbit anti-PAX5 antibody (ET1701-49) at 1/10,000 dilution.

Lane 1: Daudi cell lysate (20 µg/Lane) Lane 2: Raji cell lysate (20 µg/Lane) Lane 3: A20 cell lysate (20 µg/Lane) Lane 4: CTLL-2 cell lysate (negative)

Predicted band size: 42 kDa Observed band size: 45 kDa

Exposure time: 15 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 (ET1701-49) at 1/10,000 dilution was used in 5% NFDM/TBST 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: Immunocytochemistry analysis of Ramos cells labeling PAX5 with Rabbit anti-PAX5 antibody (ET1701-49) at 1/500 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-PAX5 antibody (ET1701-49) at 1/500 dilution in 1% BSA in PBST 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.

Beta tubulin (HA601187, red) was stained at 1/100 dilution overnight at $+4^{\circ}$ C. Goat Anti-Mouse IgG H&L (iFluor \pm 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

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Fig3: Immunohistochemical analysis of paraffin-embedded human lymph nodes tissue with Rabbit anti-PAX5 antibody (ET1701-49) at 1/1,000 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 ddH₂O and PBS, and then probed with the primary antibody (ET1701-49) 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 tonsil tissue with Rabbit anti-PAX5 antibody (ET1701-49) at 1/1,000 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 ddH₂O and PBS, and then probed with the primary antibody (ET1701-49) 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 spleen tissue with Rabbit anti-PAX5 antibody (ET1701-49) at 1/1,000 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 ddH₂O and PBS, and then probed with the primary antibody (ET1701-49) 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.

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Fig6: Immunohistochemical analysis of paraffin-embedded mouse spleen tissue with Rabbit anti-PAX5 antibody (ET1701-49) at 1/2,000 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 ddH₂O and PBS, and then probed with the primary antibody (ET1701-49) 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 rat spleen tissue with Rabbit anti-PAX5 antibody (ET1701-49) at 1/2,000 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 ddH₂O and PBS, and then probed with the primary antibody (ET1701-49) 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 human appendix tissue with Rabbit anti-PAX5 antibody (ET1701-49) at 1/2,000 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 ddH₂O and PBS, and then probed with the primary antibody (ET1701-49) 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.

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Fig9: Immunohistochemical analysis of paraffin-embedded human B cell lymphoma tissue with Rabbit anti-PAX5 antibody (ET1701-49) at 1/2,000 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 ddH₂O and PBS, and then probed with the primary antibody (ET1701-49) 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: Immunohistochemical analysis of paraffin-embedded human kidney tissue (negative) with Rabbit anti-PAX5 antibody (ET1701-49) at 1/2,000 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 ddH₂O and PBS, and then probed with the primary antibody (ET1701-49) 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.



Fig11: Flow cytometric analysis of PAX5 was done on Raji cells. The cells were fixed, permeabilized and stained with the primary antibody (ET1701-49, 1/50) (red). After incubation of the primary antibody at room temperature for an hour, the cells were stained with a Alexa Fluor®488 conjugate-Goat anti-Rabbit IgG Secondary antibody at 1/1,000 dilution for 30 minutes.Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

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

Background References

- 1. Nasri Nasrabadi P et al. The Pleiotropy of PAX5 Gene Products and Function. Int J Mol Sci. 2022 Sep
- 2. Jia Z et al. PAX5 alterations in B-cell acute lymphoblastic leukemia. Front Oncol. 2022 Oct





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