# **Anti-PAX5 Antibody [JJ08-87]**

### ET1701-49



Product Type: Recombinant Rabbit monoclonal IgG, primary antibodies

Species reactivity: Human, Mouse, Rat

Applications: WB, IF-Cell, IF-Tissue, IHC-P, FC

Molecular Wt: Predicted band size: 42 kDa

Clone number: JJ08-87

**Description:** The Pax family of nuclear transcription factors is comprised of nine members that function

during embryogenesis to regulate the temporal and position-dependent differentiation of cells. Pax family genes are also involved in a variety of signal transduction pathways in the adult organism. Mutations in Pax proteins have been linked to disease and cancer in humans. For example, the human PAX5 gene encodes a B cell lineage-specific protein, Pax-5, also designated B cell specific activator protein or BSAP, which is expressed in pro-B, pre-B and mature B lymphocytes but not in plasma cells. Pax-5 functions to regulate not only B cell development, but also influences the balance between immunoglobulin secretion and B cell proliferation. Overexpression of Pax-5 has been implicated in cellular transformation, and in the case of small lymphocytic lymphomas with plasmacytoid differentiation, a t(9;14) (p13;q32) translocation resulting in the deregulation of PAX5 gene expression has been

detected.

**Immunogen:** Synthetic peptide within human PAX5 aa 240-280.

Positive control: Ramos cell lysate, Raji cell lysate, Daudi cell lysates, Hela, MCF-7, HepG2, human lymph

nodes tissue, human tonsil tissue, human spleen tissue, mouse spleen tissue, rat spleen

tissue, Raji.

Subcellular location: Nucleus.

Database links: SwissProt: Q02548 Human | Q02650 Mouse

Entrez Gene: 500453 Rat

**Recommended Dilutions:** 

 WB
 1:5,000

 IF-Cell
 1:50-1:200

 IF-Tissue
 1:50-1:200

 IHC-P
 1:400-1:1,000

 FC
 1:50-1:100

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.

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#### **Images**

kDa 250-150-100-72-55-42-35-25-14**Fig1:** Western blot analysis of PAX5 on different lysates with Rabbit anti-PAX5 antibody (ET1701-49) at 1/5,000 dilution.

Lane 1: Ramos cell lysate Lane 2: Raji cell lysate Lane 3: Daudi cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 42 kDa Observed band size: 50 kDa

Exposure time: 3 minutes 10 seconds;

4-20% SDS-PAGE gel.

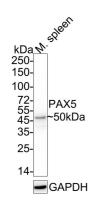
**Fig2:** Western blot analysis of PAX5 on Mouse spleen tissue lysates with Rabbit anti-PAX5 antibody (ET1701-49) at 1/5,000 dilution.

Lysates/proteins at 40 µg/Lane.

Predicted band size: 42 kDa Observed band size: 50 kDa

Exposure time: 30 seconds; ECL: K1801;

4-20% SDS-PAGE gel.



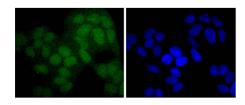
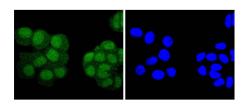
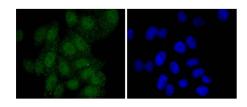


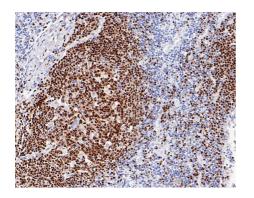
Fig3: ICC staining of PAX5 in Hela cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 10% negative goat serum for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1701-49, 1/50) for 1 hour 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:** ICC staining of PAX5 in MCF-7 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 (ET1701-49, 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).



**Fig5:** ICC staining of PAX5 in HepG2 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 (ET1701-49, 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).

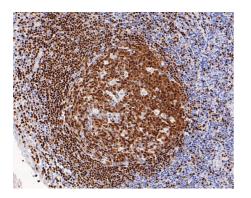


**Fig6:** Immunohistochemical analysis of paraffin-embedded human lymph nodes tissue with Rabbit anti-PAX5 antibody (ET1701-49) at 1/1,000 dilution.

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 ddH<sub>2</sub>O 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.

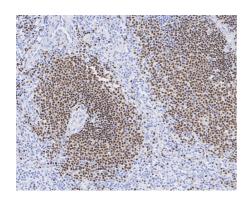
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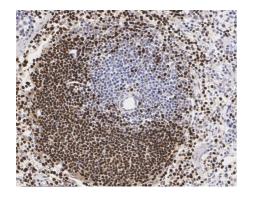
**Fig7:** Immunohistochemical analysis of paraffin-embedded human tonsil tissue with Rabbit anti-PAX5 antibody (ET1701-49) at 1/1,000 dilution.

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 ddH<sub>2</sub>O 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.



**Fig8:** 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 ddH<sub>2</sub>O 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.

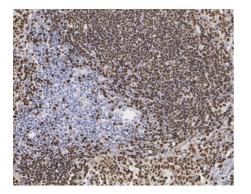


**Fig9:** Immunohistochemical analysis of paraffin-embedded mouse spleen tissue with Rabbit anti-PAX5 antibody (ET1701-49) at 1/1,000 dilution.

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 ddH<sub>2</sub>O 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.

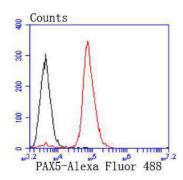
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**Fig10:** Immunohistochemical analysis of paraffin-embedded rat spleen tissue with Rabbit anti-PAX5 antibody (ET1701-49) at 1/1,000 dilution.

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 ddH<sub>2</sub>O 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.



**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. Zhao L et al. Paired box 5 is a frequently methylated lung cancer tumour suppressor gene interfering -catenin signalling and GADD45G expression. J Cell Mol Med 20:842-54 (2016).
- 2. Ren Y et al. Diagnostic utility of PAX2 and PAX5 in distinguishing non-small cell lung cancer from small cell lung cancer. Int J Clin Exp Pathol 8:14709-16 (2015).