Anti-Paxillin Antibody [SY23-02] - BSA and Azide free

HA750114

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, IP
Molecular Wt:	Predicted band size: 65 kDa
Clone number:	SY23-02

Description: Paxillin is a focal adhesion phosphoprotein that is localized to the cytoskeleton. Phosphorylation of paxillin has been shown to occur in response to PDGF treatment, v-Src transformation or cross-linking of integrins. FAK (focal adhesion kinase) and PYK2 have been shown to phosphorylate paxillin. FAK phosphorylates paxillin specifically on Tyr 118 in vitro. However, FAK phosphorylation does not seem to be required for the recruitment of paxillin to cell adhesion sites. Paxillin may play a role in signal transduction, regulation of cell morphology and the recruitment of structural and signaling molecules to focal adhesions. It has been shown that the amount of paxillin is reduced in mitotic cells by proteolytic downregulation and that paxillin is alternatively phosphorylated on serine rather than on tyrosine and serine during mitosis.

Immunogen: Synthetic peptide within Human Paxillin aa 1-59 / 591.

Positive control: HeLa cell lysate, A431 cell lysate, PANC-1 cell lysate, RAW264.7 cell lysate, C6 cell lysate, Rat testis tissue lysate, SK-OV-3, HeLa, C6, human kidney tissue, mouse testis tissue, human breast carcinoma tissue, mouse ovary tissue, rat testis tissue.

Subcellular location: Cytoplasm, cytoskeleton, Cell junction, focal adhesion, cell cortex.

Database links: SwissProt: P49023 Human | Q8VI36 Mouse | Q66H76 Rat

Recommended Dilutions:

WB	1:1,000-1:5,000
IF-Cell	1:50-1:200
IF-Tissue	1:50-1:200
IHC-P	1:50-1:200
IP	Use at an assay dependent concentration.

Storage Buffer: PBS (pH7.4).

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

Purity: Protein A affinity purified.

Images

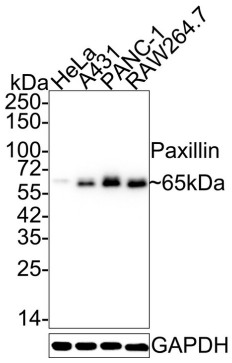

Fig1: Western blot analysis of Paxillin on different lysates with Rabbit anti-Paxillin antibody (HA750114) at 1/5,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: A431 cell lysate
Lane 3: PANC-1 cell lysate
Lane 4: RAW264.7 cell lysate

Lysates/proteins at 15 µg/Lane.

Predicted band size: 65 kDa
Observed band size: 65 kDa

Exposure time: 2 minutes; 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 (HA750114) at 1/5,000 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.

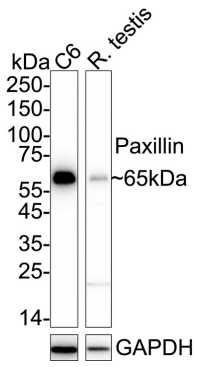

Fig2: Western blot analysis of Paxillin on different lysates with Rabbit anti-Paxillin antibody (HA750114) at 1/1,000 dilution.

Lane 1: C6 cell lysate (20 µg/Lane)
Lane 2: Rat testis tissue lysate (40 µg/Lane)

Predicted band size: 65 kDa
Observed band size: 65 kDa

Exposure time: 8 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 (HA750114) 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.

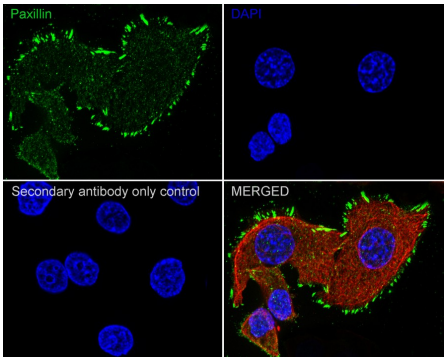

Fig3: Immunocytochemistry analysis of SK-OV-3 cells labeling Paxillin with Rabbit anti-Paxillin antibody (HA750114) at 1/100 dilution.

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-Paxillin antibody (HA750114) 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 (HA601187, 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.

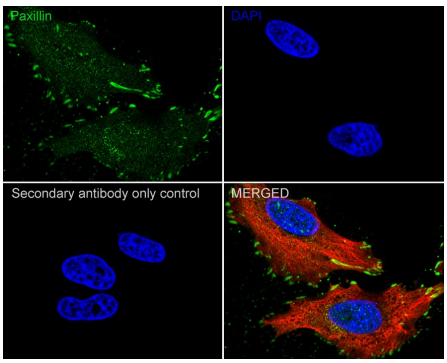

Fig4: Immunocytochemistry analysis of HeLa cells labeling Paxillin with Rabbit anti-Paxillin antibody (HA750114) at 1/100 dilution.

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-Paxillin antibody (HA750114) 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 (HA601187, 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.

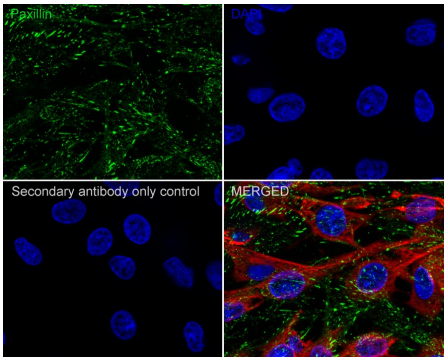

Fig5: Immunocytochemistry analysis of C6 cells labeling Paxillin with Rabbit anti-Paxillin antibody (HA750114) at 1/50 dilution.

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-Paxillin antibody (HA750114) at 1/50 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.

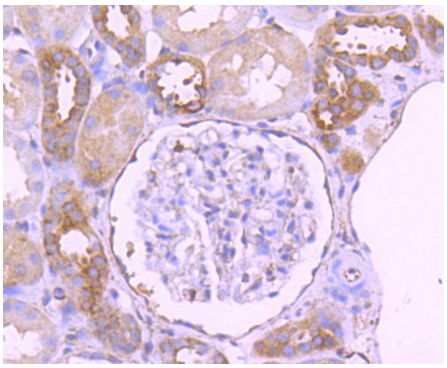

Fig6: Immunohistochemical analysis of paraffin-embedded human kidney tissue using anti-Paxillin antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750114, 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.

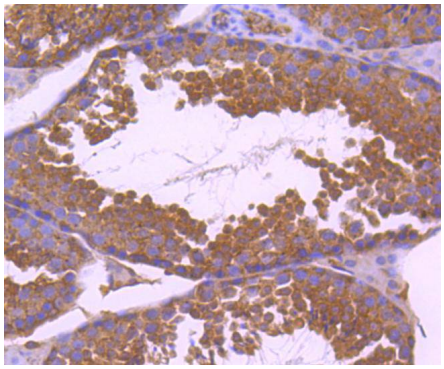

Fig7: Immunohistochemical analysis of paraffin-embedded mouse testis tissue using anti-Paxillin antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750114, 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.

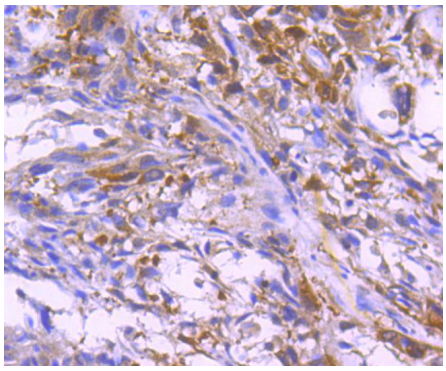

Fig8: Immunohistochemical analysis of paraffin-embedded human breast carcinoma tissue using anti-Paxillin antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750114, 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.

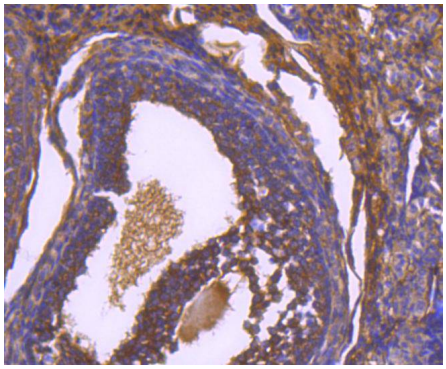

Fig9: Immunohistochemical analysis of paraffin-embedded mouse ovary tissue using anti-Paxillin antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750114, 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.

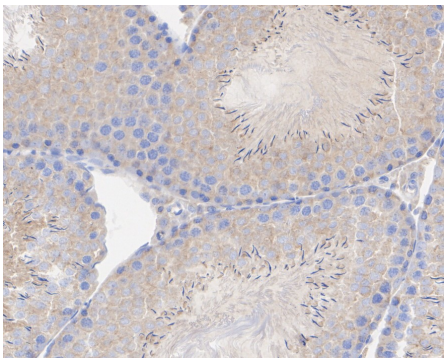

Fig10: Immunohistochemical analysis of paraffin-embedded rat testis tissue using anti-Paxillin antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750114, 1/200) 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.

Background References

1. Izumi D et al. CXCL12/CXCR4 activation by cancer-associated fibroblasts promotes integrin 1 clustering and invasiveness in gastric cancer. Int J Cancer 138:1207-19 (2016).
2. Yariswamy M et al. Cardiac-restricted Overexpression of TRAF3 Interacting Protein 2 (TRAF3IP2) Results in Spontaneous Development of Myocardial Hypertrophy, Fibrosis, and Dysfunction. J Biol Chem 291:19425-36 (2016).