# Anti-Nephrin Antibody [PSH07-70] - BSA and Azide free

# HA751182

| | |
|---|---|
| **Product Type:** | Recombinant Rabbit multiclonal IgG, primary antibodies |
| **Species reactivity:** | Human, Mouse, Rat |
| **Applications:** | WB, IHC-P, IF-Tissue, IHC-Fr |
| **Molecular Wt:** | Predicted band size: 135 kDa |
| **Clone number:** | PSH07-70 |

**Description:** Nephrin is a protein necessary for the proper functioning of the renal filtration barrier. The renal filtration barrier consists of fenestrated endothelial cells, the glomerular basement membrane, and the podocytes of epithelial cells. Nephrin is a transmembrane protein that is a structural component of the slit diaphragm. They are present on the tips of the podocytes as an intricate mesh and convey strong negative charges which repel protein from crossing into the Bowman's space. A defect in the gene for nephrin, NPHS1, is associated with congenital nephrotic syndrome of the Finnish type and causes massive amounts of protein to be leaked into the urine, or proteinuria. Nephrin is also required for cardiovascular development.

**Immunogen:** Recombinant protein within human Nephrin aa 1-1,050.

**Positive control:** Mouse kidney tissue lysate, Rat kidney tissue lysate, human kidney tissue, mouse kidney tissue, rat kidney tissue.

**Subcellular location:** Cell membrane.

**Database links:** SwissProt: O60500 Human | Q9QZS7 Mouse | Q9R044 Rat

**Recommended Dilutions:**

| | |
|---|---|
| **WB** | 1:1,000-1:5,000 |
| **IHC-P** | 1:3,000 |
| **IF-Tissue** | 1:200-1:1,000 |
| **IHC-Fr** | 1:500 |

**Storage Buffer:** PBS (pH7.4).

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

**Purity:** Protein A affinity purified.

## Images

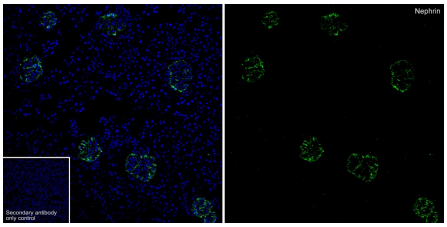


**Fig1:** Immunofluorescence analysis of frozen mouse kidney tissue with Rabbit anti-Nephrin antibody (HA751182) at 1/500 dilution.

The section was pre-treated using 1% SDS buffer (in PBS, pH 7.4) for 5 minutes at room temperature.

The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA751182, red) at 1/500 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

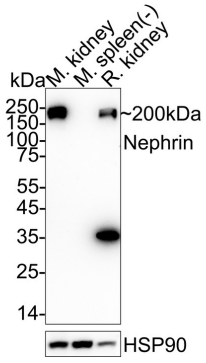


**Fig2:** Western blot analysis of Nephrin on different lysates with Rabbit anti-Nephrin antibody (HA751182) at 1/1,000 dilution.

Lane 1: Mouse kidney tissue lysate
Lane 2: Mouse spleen tissue lysate (negative)
Lane 3: Rat kidney tissue lysate

Lysates/proteins at 40 µg/Lane.

Predicted band size: 135 kDa
Observed band size: 200 kDa

Exposure time: 1 minute 30 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 (HA751182) 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.

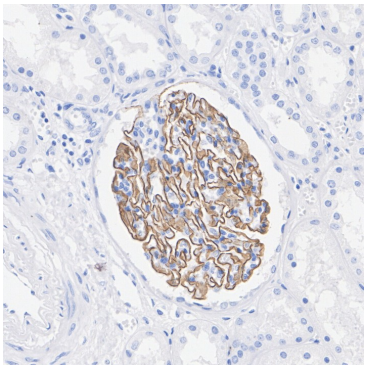

**Fig3:** Immunohistochemical analysis of paraffin-embedded human kidney tissue with Rabbit anti-Nephrin antibody (HA751182) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with dd$H_2O$ and PBS, and then probed with the primary antibody (HA751182) 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.

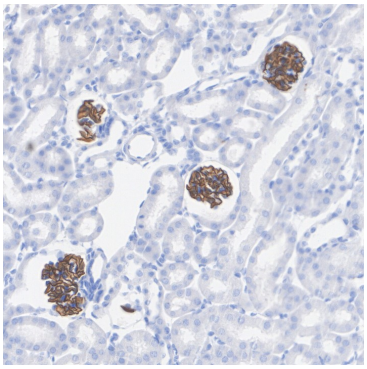

**Fig4:** Immunohistochemical analysis of paraffin-embedded mouse kidney tissue with Rabbit anti-Nephrin antibody (HA751182) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 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 (HA751182) 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.

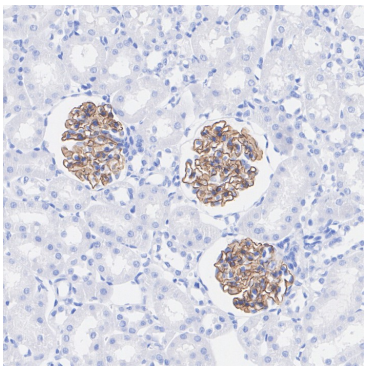

**Fig5:** Immunohistochemical analysis of paraffin-embedded rat kidney tissue with Rabbit anti-Nephrin antibody (HA751182) at 1/3,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 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 (HA751182) at 1/3,000 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.

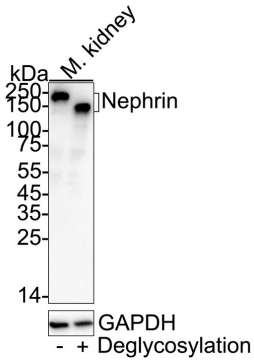


**Fig6:** Western blot analysis of Nephrin on different lysates with Rabbit anti-Nephrin antibody (HA751182) at 1/5,000 dilution.

Lane 1: Mouse kidney tissue lysate
Lane 2: Mouse kidney tissue lysate treated with deglycosylation

Lysates/proteins at 14.4 µg/Lane.

Predicted band size: 135 kDa
Observed band size: 200/135 kDa

Exposure time: 3 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 (HA751182) at 1/5,000 dilution was used in primary antibody dilution (K1803) at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

## Background References

1. Watts AJB et al. Discovery of Autoantibodies Targeting Nephrin in Minimal Change Disease Supports a Novel Autoimmune Etiology. J Am Soc Nephrol. 2022 Jan
2. Hengel FE et al. Autoantibodies Targeting Nephrin in Podocytopathies. N Engl J Med. 2024 May