**DESCRIPTION**

**Species Reactivity**: Human

**Specificity**: Detects human Nephrin in direct ELISAs and Western blots. In direct ELISAs, less than 20% cross-reactivity with recombinant mouse Nephrin is observed.

**Source**: Polyclonal Sheep IgG

**Purification**: Antigen Affinity-purified

**Immunogen**: Mouse myeloma cell line NS0-derived recombinant human Nephrin Gln23-Thr1029

**Accession #**: O60500

**Formulation**: Lyophilized from a 0.2 μm filtered solution in PBS with Trehalose. See Certificate of Analysis for details.

*Small pack size (-SP) is supplied either lyophilized or as a 0.2 μm filtered solution in PBS.*

**APPLICATIONS**

**Please Note**: Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Recommended Concentration</th>
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</thead>
<tbody>
<tr>
<td>Western Blot</td>
<td>1 µg/mL</td>
</tr>
<tr>
<td>Immunohistochemistry</td>
<td>1-15 µg/mL</td>
</tr>
<tr>
<td>Simple Western</td>
<td>20 µg/mL</td>
</tr>
</tbody>
</table>

**DATA**

**Western Blot**

Detection of Human Nephrin by Western Blot. Western blot shows lysates of human kidney tissue under reducing and non-reducing conditions. PVDF membrane was probed with 1 µg/mL Sheep Anti-Human Nephrin Antigen Affinity-purified Polyclonal Antibody (Catalog # AF4269) followed by HRP-conjugated Anti-Sheep IgG Secondary Antibody (Catalog # HAF016). A specific band for Nephrin was detected at approximately 150 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 2.

**Immunohistochemistry**

Nephrin in Human Kidney. Nephrin was detected in immersion fixed paraffin-embedded sections of human kidney using 1.7 µg/mL Sheep Anti-Human Nephrin Antigen Affinity-purified Polyclonal Antibody (Catalog # AF4269) overnight at 4 °C. Tissue was stained with the Anti-Sheep HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS019) and counterstained with hematoxylin (blue). Specific labeling was localized to podocytes in glomeruli. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

**PREPARATION AND STORAGE**

**Reconstitution**: Reconstitute at 0.2 mg/mL in sterile PBS.

**Shipping**: The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C.*

**Stability & Storage**: Use a manual defrost freezer and avoid repeated freeze-thaw cycles.

- 12 months from date of receipt, -20 to -70 °C as supplied.
- 1 month, 2 to 8 °C under sterile conditions after reconstitution.
- 6 months, -20 to -70 °C under sterile conditions after reconstitution.
Nephrin, also known as renal glomerulus-specific cell adhesion receptor and nephrosis 1 (NPHS1) is a 185-200 kDa type I transmembrane protein belonging to the immunoglobulin (Ig) superfamily. It is expressed on podocytes and is an essential component of the interpodocyte-spanning slit diaphragm complex. Nephrin forms cis-hetero-oligomeric complexes with Neph1, followed by trans-homophilic interaction with Nephrin on opposing cells. Mutations in the Nephrin gene is the pathogenic cause of congenital nephrotic syndrome. Mature human Nephrin contains a 1033 aa extracellular region and a 165 aa cytoplasmic tail. One potential soluble splice form is known where aa’s 1056-1095 are deleted, eliminating the transmembrane region. Over aa 23-1029, human Nephrin shares 84% and 89% aa sequence identity with mouse and canine Nephrin, respectively.