Product Specification Sheet

NEPH2 Antibodies

□ Cat. # NEPH21-P	Rat NEPH2 Control/blocking peptide # 1	**SIZE**: 100 ug
□ Cat. # NEPH21-A	**Rabbit** Anti-Rat NEPH2 IgG # 1 (aff pure)	**SIZE**: 100 ug

Nephrin is encoded by NPHS1 gene (chromosome 19q13.1), the core protein of the interpodocytes slit diaphragm of kidney glomerulus, these podocytes are highly differentiated with characteristic interdigitating foot processes covering the outer glomerular basement membrane, the space between these foot processes is spanned by a tight junction to provide the large surface area for filtration.

Nephrin interacts with podocin and with CD2AP C-terminal domain, It tends to be a signaling molecule that activates canonical protein kinase cascades, which is initiated by three closely related proteins called NEPH proteins (**NEPH1, NEPH2, & NEPH3**), The NEPH proteins share a common domain architecture consisting of 5 extracellular Ig domain followed by transmembrane domain, These proteins bind to C-terminal domain of podocin, which interacts with C-terminal of nephrin and greatly enhances nephrin-induced signaling.

NEPH2 (Human 778aa, chrm 11q24) has no N-terminal signal peptide; the C-terminus will be inside and do have transmembrane region. It has the common domain architecture of 5 Ig domains. NEPH2 id present in brain, heart, kidney and nervous system. At least 4 alternatively spliced have been reported in mice NEPH2 (Q8BR86, 778 aa).

Isoform 1, 778 aa, Q8BR86	1-778 aa
Isoform 2, 777 aa, Q8BR86-2	19-44 missing, 565 R changed
Isoform 3, 612 aa; Q8BR86-3	518-529 aa; 603-624 aa changed, 625-778 missing
Isoform 4, 766 aa, Q8BR86-4	518-529 aa

Source of Antigen and Antibodies

Antigen	14-aa peptide 727-740 aa from Rat NEPH2/KIRRE (protein accession # Q09GS6, refs 1) **Designation (NEPH21-P, control peptide**); **Epitope location**~ C-terminal, Cytoplasmic domain
Ab Host/type	Rabbit, Polyclonal Aff pure IgG (cat # NEPH21-A) purified over antigen-agarose column
2-ab	**Goat Anti-rabbit IgG-HRP** cat # 20320 (AP, biotin, FITC conjugates also available)
-ve control IgG	# 20009-1, Rabbit (non-immune) IgG, purified, suitable for ELISA, Western, IHC as –ve control

Form & Storage of Antibodies/Peptide Control

Affinity pure IgG

□ 100 ug/100ul □ solution □ lyophilized powder

Supplied in **Buffer**: PBS+0.1% BSA

Reconstitute powder in PBS at 1mg/ml

Control/blocking peptide

□ 100 ug/100 ul □ solution □ lyophilized powder

Supplied in Buffer: PBS pH 7.5,

Reconstitute powder in PBS at 1 mg/ml.

Storage

Short-term: unopened, undiluted liquid vials at -20OC and powder at 4oC or -20oC..

Long-term: at –20C or below in suitable aliquots after reconstitution. Do not freeze and thaw and store working, diluted solutions.

Stability: 6-12 months at –20oC or below.
Shipping: 4oC for solutions and room temp for powder.

Recommended Usage

Western Blotting (1-10 ug/ml for affinity pure antibody using ECL technique).

ELISA: Control peptide can be used to coat ELISA plates at 1 ug/ml and detected with antibodies (0.5-1 ug/ml for affinity pure).

Histochemistry & Immunofluorescence: Not tested. We recommend the use of aff pure IgG at 2-20 ug/ml.

Specificity & Cross-reactivity

The Rat NEPH21-P Control/blocking peptide is 100% conserved in human and mouse NEPH2/KIRR3. Based upon the sequences of various NEPH2 isoforms, the epitope of NEPH21-P is found in isoforms 1-2, and 4. No significant sequence homology is seen with other related family members Antibody cross-reactivity in various species has not been studied. Control peptide, because of its low mol. Wt (<3 kDa), is not suitable for Western. It should be used for ELISA or antibody blocking experiments (use 5-10 ug control peptide per 1 ug of aff pure IgG or 1 ul antiserum) to confirm antibody specificity (see detailed protocol see detailed protocol at the web site).

General References: Lorenz Sellin et al (2002) FASEB journal article 10.1096, published online; Heli Putaala et al (2001) Human Mol. Gen.10,1-8; Dorbit B.Donoviel et al(2001) Mol and Celluar Biol 21, 4829-4836, David B. Kershaw et al (1997) JBC 272, 15708-15714.

*This product is for In vitro research use only.

Related material available from ADI

Antibodies & Peptides: Nephrin related proteins (NEPH 1-3, Filtrin, Podocin, Podocalyxin).

NEPH21-A-P 71212A

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