

### Nepriylsin-Like Protease- $\beta$ (NEPL- $\beta$ ) Antibodies

Cat. # NEPLB21-S	Rabbit Anti-Mouse NEPL- $\beta$ antiserum	<b>SIZE:</b> 100 ul
Cat. # NEPLB21-A	Rabbit Anti-Mouse NEPL- $\beta$ IgG (Affinity pure)	<b>SIZE:</b> 100 ug
Cat. # NEPLB21-P	Mouse NEPL- $\beta$ Control/blocking peptide	<b>SIZE:</b> 100 ug

The amyloid  $\beta$ -peptide (**A $\beta$** ) of 39 to 43 amino acids is constitutively produced in brain upon proteolysis of the  $\beta$ -amyloid precursor protein (**APP**) and exists as fragments of 40, 42 and 43 amino acids (A $\beta$ <sub>40</sub>, A $\beta$ <sub>42</sub>, A $\beta$ <sub>43</sub>). In the young and healthy humans, the secreted A $\beta$  is rapidly catabolized before it can be deposited in the brain. However, upon aging or the onset of familial **Alzheimer's disease**, alterations in either synthesis or degradation/clearance of A $\beta$  may contribute to amyloid depositions in the brain. A $\beta$  carries cleavage sites for a number of *in vivo* and *in vitro* proteases like cathepsin D and M-13 metalloproteases. The M-13 family comprises several zinc-dependent metalloproteases like **DINE**, **PHEX**, **KELL**, **ECE**, **XCE**, **nepriylsin (NEP)** and **nepriylsin-like proteases (NEPLs)**. The NEPLs (**NEPL- $\alpha$** , **NEPL- $\beta$** , **NEPL- $\gamma$** ) arise from the alternative splicing of a single NEPL gene and are zinc dependent metalloproteases with ~54 % homology to NEP.

**NEPL- $\beta$**  (variously called SEP/NL1/NEPII) is type II transmembrane enzyme containing a single polypeptide chain of 765aa (~110 KDa) with cytosolic and transmembrane domains and a large extracellular C-terminal core containing the peptidase active site. The aa sequence is 65.1 % identical to mouse NEP. Like NEP, NEPL- $\beta$  also contains 12 cysteine residues, 10 of which are conserved in all members of the family. There are two splice variants of NEPL- $\beta$ ; a secreted isoform of 126 KDa containing a 23 aa secretion signal sequence and a membrane associated isoform of 110 kDa. The secreted isoform is much more glycosylated than the membrane isoform. Testis is the only tissue where the soluble/secreted isoform is predominant. Unlike NEP, NEPL- $\beta$  has no proteolytic activity to A $\beta$ ; however both enzymes can cleave Leu<sub>5</sub>-enkephalin.

**FUNCTION:** Metalloprotease involved in sperm function, possibly by modulating the processes of fertilization and early embryonic development.

**SUBCELLULAR LOCATION:** Membrane; Single-pass type II membrane protein. Secreted. Note=A secreted form produced by proteolytic cleavage also exists.

**SIMILARITY:** Belongs to the peptidase M13 family [view classification].

**Protein name** Membrane metallo-endopeptidase-like 1  
**Synonyms** EC 3.4.24.11, Nepriylsin-2, Nepriylsin II, NL2 NEPII, NEP2(m), Nepriylsin-like peptidase, NEPLP, Nepriylsin-like 1 NL-1, Soluble secreted endopeptidase  
**Gene name** Name: Mmel1 ; Synonyms: Nep2, NI1, Sep

#### Source of Antigen and Antibodies

<b>Antigen</b>	A 13-aa peptide sequence ( <b>gene accession#Q9JLI3</b> ) ( <b>designated NEPLB21-P or control peptide</b> ), conjugated to KLH Epitope location ~ N-terminus
<b>Ab Host/type</b>	Rabbit, polyclonal Aff pure IgG1 ( <b>cat #NADC31-A</b> ) Antibodies have been <b>affinity purified</b> over the control-peptide Sepharose.
<b>2-ab</b>	Cat # 20320, goat anti-rabbit IgG-HRP (AP, biotin, FITC conjugates also

	available
<b>-ve control</b>	# 20009-1, Rabbit (non-immune) IgG, purified, suitable for ELISA, Western, IHC as -ve control

#### Form & Storage of Antibodies/Peptide Control

##### Antiserum (unpurified)

100ul solution lyophilized powder  
 Supplied in Buffer: 0.05% azide  
**Reconstitute powder in 100 ul PBS**

##### 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.

##### Recommended Usage

**Western Blotting** (1:1K-5K for neat serum and 1-10 ug/ml for affinity pure using Chemiluminescence technique).

**ELISA** (1:10K-1:100K; using 50-100 ng of control peptide/well).

**Histochemistry & Immunofluorescence:** not tested. We recommend the use of affinity pure antibody at 2-20 ug/ml.

##### Specificity & Cross-reactivity

Mouse NEPLB21-P antigenic/control peptide sequence is 76% conserved in rat NEPL- $\beta$ . No significant sequence homology of NEPLB21-P is seen with NEP or other NEPLs. Antibody reactivity in various species is not known. 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 at: [www.4adi.com/data/abblock.html](http://www.4adi.com/data/abblock.html)).

**General References:** (1) Shirovani K et al (2001) JBC 276, 21895-21901; Iwata et al. (2001) Science 292, 1550; Ikeda et al. (1999) J. Biol. Chem. 274, 32469; Boileau et al. (2001) Biochem. J 355, 107; Kiryu-Seo et al. (2000) PNAS 97, 4345.

*\*This product is for In vitro research use only.*

##### Related materials available from ADI

Antibodies: NEP, NEP-alpha, -beta, -gamma, DINE, PHEX.

NEPLB21-S-A-P 70904J