

Product Specification Sheet

NPT2 Antibodies

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| Cat # NPT25-P | Human NPT2 control/blocking peptide # 5 | SIZE: 100 ug |
| Cat # NPT25-S | Rabbit Anti-Human NPT2 antiserum #5 | SIZE: 100 ul |
| Cat # NPT25-A | Rabbit Anti-Human NPT2 IgG #5, aff pure | SIZE: 100 ug |

Inorganic phosphate (Pi) levels are hormonally regulated that affects the physiological activity of bone, kidney, and small intestine. Majority of the Pi is absorbed in the small intestine and reabsorbed in the proximal tubules in the kidney. At least 4 groups of structurally and functionally related proteins are involved in Pi transport: **Type I-related** NaPi transporters designated **NPT1**, **Npt1**, and **NaPi-1** respectively in humans, mouse, and rabbit are expressed in the kidney and liver. Its expression and activity are not regulated by Pi deprivation or parathyroid hormone (PTH) and its role in Pi-homeostasis is not clear. **Type IIa-related** cotransporters, designated **NaPi-2** in rat, **NaPi-3** or **NPT2** in humans, **NaPi-4** in opossum, **NaPi-5** in flounder vessel, **NaPi-6** or **Npt-2** in mouse, and **NaPi-7** in rabbit, is the primary target for Pi regulation by dietary, hormonal, and tubular Pi reabsorption. Deletion of Npt2 gene produces severe Pi wasting. Type II transporters are expressed in kidney, brain, lung, bone and small intestine. **Type IIb**, designated as **NaPi-IIb** or **NaPi-2b** in rat/mouse, and **NaPi-3b** in human, is closely related isoform of the NaPi-2 family. It is expressed in small intestine and lung. **Type III NaPi** transporters, originally described as a family of cell surface receptors for gibbon ape leukemia virus (**GALV**) and murine amphotropic retrovirus (A-MuLV), share very low (<20%) sequence homology with Type I and II proteins, and are found in most tissues. Human homologs are now designated as **PiT1** (also known as GALV receptors or GLVR1) and **PiT2** (A-MuLV receptors or GLVR2 or Ram1). Pit1 and Pit2 are ~62% related and predicted to contain 10 TM domains that is in contrast with Type 1/2 transporters (6-8 TM). The N and C-termini are predicted to be cytoplasmic.

Source of Antigen and Antibodies

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|------------------------|---|
| Antigen | Human Type-IIa, NPT2/Napi-3 is ~640 aa (chromosome 8) and mouse and rat 637 aa proteins. A 21 AA synthetic peptide (designated NPT25-P; control peptide) within the 1st extracellular loop (between TM1-2) of Human NPT2a (126-146 aa) was synthesized, conjugated to KLH |
| Ab Host/type | Rabbit, Polyclonal antiserum # NPT25-S and IgG, purified over antigen-agarose (Cat # NPT25-A). An antibody to this antigenic peptide is also produced in chickens (Cat # NPT26-S). |
| 2-Ab | Cat # 20320, goat anti-rabbit IgG-HRP (AP, biotin, FITC conjugates also available). |
| -ve control IgG | Cat # 20009-1, Rabbit (non-immune) Serum IgG, purified, suitable for ELISA, Western, IHC as -ve control |

Form & Storage of Antibodies/Peptide Control

Antiserum (unpurified)
100ul solution lyophilized powder
Supplied 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 -200C 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 1:1K-5K for antiserum and 1-10 ug/ml for affinity pure antibody using Chemiluminescence technique. NPT2a apparent mol wt is 80-90 kDa, and under reducing conditions 2 bands of 40-50 kDa are observed. It may also form multimeric structures (170 and 200 kDa). (see Murer et al 2000)

ELISA: Control peptide can be used to coat ELISA plates at 1 ug/ml and detected with antibodies (1:10-50K for neat serum and 0.5-1 ug/ml for affinity pure).

Histochemistry & Immunofluorescence: not tested.

Specificity & Cross-reactivity

The NPT25-P peptide is 100% conserved in rat, mouse, ovine, opossum, 95% in rabbit, 85% in Zebra fish, and 71% in frog NPT2a. The NPT25-P sequence also has significant sequence homology (~65%) with mouse, rat, and human Type-IIb transporters. Therefore, antibodies may show some crossreactivity with Type2b NPT2. No significant sequence homology of NPT25-P is observed with other Pi-transporters (Type-I, III, or IV) transporters. Antibody crossreactivity in various species or isoform is not established. 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: (1). Magnin S et al (1993) PNAS 90, 5979; Collins JF et al (1996) FASEB J 8, 862; Hartman CM et al (1996) PNAS 93, 7409.; (2) Murer H et al (2000) Physiol. Rev. 80, 1373 (review); Werner A et al (2001) Am. J. Physiol. Integ. Comp. Physiol. 280, R301

**This product is for In vitro research use only.*
NPT25-S-A-P 71213A

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