ALPHA DIAGNOSTIC INTERNATIONAL

Product Specification Sheet

## Natural Resistance-Associated Macrophage Protein (Nramp2) Antibodies

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| Cat # NRAMP22-P | Human NRAMP2 (with-IRE) Control/blocking peptide #2 | **SIZE**: 100 ug |
| Cat # NRAMP22-S | Rabbit Anti-Human NRAMP2 (with-IRE) antiserum # 2 | **SIZE**: 100 ul |
| Cat # NRAMP22-A | Rabbit Anti-Human NRAMP2 (with-IRE) IgG # 2 (aff pure) | **SIZE**: 100 ug |

Natural resistance to infection with unrelated intracellular parasite such as Mycobacteria, Salmonella, and Leishmania is controlled by a single gene that encodes a macrophage-specific membrane protein designated as Natural Resistance-Associated Macrophage Protein (**Nramp1**). Recently a second member of NRAMP family, termed **NRAMP2/DMT/DCT1 (Divalent Metal ion Transporter 1 or Divalent Cation Transporter 1)**, has been identified (human, rat and mouse 568 aa, ~65% identity with NRAMP1). Unlike NRAMP1, NRAMP2 expression is more ubiquitous and has been detected in most tissues. It is dramatically up-regulated by iron starvation in the intestine. **NRAMP2** gene produces two alternatively spliced transcripts generated by alternative use of two 3' exons encoding distinct C-termini of the protein as well as distinct 3' untranslated regions (UTRs). Interestingly, one Nramp2 mRNA contains an iron-responsive element (IRE) in its 3'UTR. The IRE is an RNA secondary structure present in the 5'- or the 3'-UTR of animal mRNAs encoding proteins involved in iron metabolism. The second Nramp2 splice isoform (**without-IRE, isoform II**) encodes a protein in which the C-terminal 18-aa of the IRE form (**with IRE**, **isoform I**) are replaced by a novel 25-aa segment and codes for a distinct 3' UTR lacking the IRE. The two isoforms are differentially localized and regulated in GI tract and kidney.

### Source of Antigen and Antibodies

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| **Antigen** | 17-aa peptide from **human NRAMP2 (1)**; **Designation (#NRAMP22-P, control/blocking peptide**) conjugated to KLH; **Epitope location**~ C-terminal, Cytoplasmic domain |
| **Ab Host/type** | Rabbit, Polyclonal unpurified antiserum (#**NRAMP22**-S) and IgG, purified over antigen-agarose (Cat # **NRAMP22**-A) |
| **2-Ab** | Cat # 20320, goat anti-rabbit IgG-HRP (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

**Antiserum (unpurified)**
□ 100 ul □ Solution □ Lyophilized
Supplied 0.1% azide, **Reconstitute** powder in 100 ul PBS

**Affinity pure IgG**
□ 100 ug/100 ul □ Solution □ Lyophilized
Supplied in **Buffer**: PBS + 0.1% BSA
**Reconstitute powder** in PBS at 1 mg/ml

**Control/blocking peptide**
□ 100 ug/100 ul □Solution Lyophilized
Supplied in Buffer: PBS pH 7.5,
**Reconstitute powder in PBS at 1 mg/ml.**

**Storage**
**Short-term**: unopened, undiluted liquid vials at -20°C and powder at 4°C or -20°C..
**Long-term**: at –20°C or below in suitable aliquots after reconstitution. Do not freeze and thaw and store working, diluted solutions.

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

### Recommended Usage

**Western Blotting** We recommend the use of (1:1K-5K for neat serum and 1-10 ug/ml for affinity pure antibody using ECL technique) (not tested)

**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. Affinity purified antibody at 5-20 ug/ml in paraformaldehyde fixed sections of tissues may be tested.

### Specificity & Cross-reactivity

The human NRAMP22-P control peptide sequence is 88% conserved in monkey NRAMP2 with IRE. This sequence is absent in NRAMP2 without IRE. No significant sequence homology exist with NRAMP1 or NRAMP2 from mouse or rat.. Antibody cross-reactivity in various species has not been 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 at the web site).

**General References**: Gunshin H et al (1997) Nature 388, 482; Fleming MD (1998) PNAS 95, 1148; Kishi F (1997) Mol Immunol. 6, 224; Gruenshield S (1995) Genomics 25, 514; Fleming MD et al (1997) Nature Genet. 16, 383; Canonne-Hergaux F (1999) Blood 93, 4406 (review).

**2. Citations for ADI Antibodies (**see updates at the web site)

Roth JA, 2000, J. Neurosci. 2000 20: 7595-7601
Moos T 2004, Journal of Neurochemistry 88, 233-245,WB, IHC
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*This product is for In vitro research use only.

**Related material available from ADI**

Antibodies NRAMP1/2, MTP1, Tf, Tfr,. And Defensins

NRAMP22-S-A-P 200109AC

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