

Product Specification Sheet

Inducible Nitric Oxide Synthase 2 (iNOS/NOS-2) Antibodies

Cat # INOS24-A

Rabbit Anti-Human iNOS/NOS-2 IgG # 4

SIZE: 100 ul

Nitric oxide (NO), a diffusible free radical gas, acts as a neurotransmitter in brain and peripheral nervous system. It accounts for the activity of endothelium-derived relaxing factors, which stimulate vasodilatation by releasing NO from the endothelium. NO is synthesized by L-arginine, oxygen, and NADPH by three known isoforms of heme-containing flavoproteins termed NO synthase (NOS, I-III, mol wt. ~130-160 kDa). One group of enzyme is constitutive, agonist-triggered, and dependent on Ca²⁺/Calmodulin and is inhibited by L-arginine analogues (L-NNA, L-NMMA). It is found in endothelium, adrenal glands, brain and platelets. The other principle group is inducible, Ca²⁺/Calmodulin-independent, and inhibited by NMMA and L-NNA. It has been found in macrophage, hepatocytes, tumor cells, vascular smooth muscle and endothelial cells. Analyses of cDNA clones have identified three distinct NOS genes in mammals: neuronal (nNOS/bNOS/NOS-I), endothelial (eNOS/NOS-III), and macrophage (mNOS/iNOS/NOS-II). Both nNOS and eNOS are constitutive and the mNOS/iNOS is inducible. Sequence homology among different cloned isoforms is ~ 50%. Human, rat, and mouse iNOS/NOS-2 are ~1145 aa proteins (1).

Source of Peptide Antigen and Antibodies

Antigen	Human NOS2 recombinant protein 1-175 fragment corresponding to human iNOS (protein accession # P35228) (1)
Ab Host/type	Rabbit, Polyclonal IgG # INOS24-A
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

IgG

100ul solution lyophilized powder
Supplied PBS, pH 7.4, 0.1% gelatin, 0.05% azide,
Reconstitute powder in 100 ul PBS

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. (1:1K-3K). It is suggested that users optimize actual dilution and conditions according their application. The antibody recognizes 130 kDa protein in Western blots from mouse macrophage RAW264.7 cells (see refs 2).

ELISA: Control peptide should be coated at 1 ug/ml.

Immunocytochemistry. We recommend the use of affinity pure antibody to reduce background (use at 5-10 ug/ml). Useful on tissue sections fixed with 3.5% paraformaldehyde. An overnight incubation with antibody at 4°C is recommended and detection by ABC (peroxidase) technique. Positive staining is seen in Glial, Purkinje and layer 5 cortical neurons.

Antibody specificity and Cross-reactivity

Antibody #INOS24-A reacts with human iNOS and it may also react with the mouse and rat iNOS due to the use of large fragment of iNOS used as antigen. No significant reactivity is seen with iNOS-1/NOS-3 or other proteins. Antibody crossreactivity in various other species is not established. Antibodies to mouse/rat iNOS (#INOS-A, iNOS22-S, and iNOS23-M) are also available.

General References: (1) Lowenstein CJ et al (1992) PNAS 89, 6711-6715; Lyons CR et al (1992) J Biol Chem. 267, 6370-6374; Zie Q-W et al (1992) Science 256, 225-228; Nunokawa Y et al (1993) BBRC 191, 89

Citations of for iNOS (see updated list at the web site)

XU, X, 2001, Eur. J. Pharmacol. 416, 1-9 (WB); Xu, X, 2001, Biochem. Pharmacol. 59, 509-516 (WB); Leiro JM, 2003, Int. J. Immunopharmacol. 4, 163-177; Leiro J, 2004, J. Leukoc. Biol., 75: 1156 – 1165;

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iNOS24-A 110119A