**Qty:** 100 µg/200 µL

Mouse anti-nNOS

**Catalog No.** 37-2800

**Lot No.**

# Mouse anti-nNOS

## FORM

This monoclonal antibody is supplied as a 200 µL aliquot at a concentration of 0.5 mg/mL in PBS, pH 7.4, containing 0.1% sodium azide. This antibody is highly purified from mouse ascites by protein-A chromatography.

**CLONE:** 3G6B10 **ISOTYPE:** $IgG_1$-kappa

## IMMUNOGEN

Recombinant protein derived from the N-terminal region of rat nNOS protein.

## SPECIFICITY

This antibody is specific for the ~160 kDa nNOS.

## REACTIVITY

Reactivity has been confirmed with rat brain tissue lystaes. The nNOS target band at ~160 kDa was detected by immunoprecipitation and Western blots. Immunohistochemical tests using human, mouse, and rat brain tissue sections yielded positive and specific cytoplasmic staining in neurons and astrocytes. Based on amino acid sequence homology, the antibody is expected to recognize mouse (95%) and human (81%) nNOS.

| Sample | Immuno-precipitation[1] | Western Blotting | Immunohistochemistry (FFPE) | ELISA |
|---|---|---|---|---|
| Rat | +++ | +++ | +++ | N/A |
| Mouse | ND | ND | +++ | N/A |
| Human | ND | ND | +++ | N/A |
| Immunogen | N/A | N/A | N/A | +++ |

(Excellent +++, Good++, Poor +, No reactivity 0, Not applicable N/A, Not Determined ND)

## USAGE

Working concentrations for specific applications should be determined by the investigator. Appropriate concentrations will be affected by several factors, including secondary antibody affinity, antigen concentration, sensitivity of detection method, temperature and length of incubations, etc. The suitability of this antibody for applications other than those listed below has not been determined. The following concentration ranges are recommended starting points for this product.

**Western Blotting:** 1 µg/mL
**ELISA:** 0.1-1.0 µg/mL
**Immunoprecipitation:** 7 µg/each
**Immunohistochemistry (FFPE):** 2 µg/mL

## STORAGE

Store at 2-8°C for up to one month. Store at –20°C for long-term storage. Avoid repeated freezing and thawing.

## BACKGROUND

Nitric oxide (NO) is a potent bio-regulatory molecule which plays important roles in the regulation of a variety of normal developmental and physiological processes[1-3]. NO is generated by nitric oxide synthase (NOS) in a reaction which converts L-arginine and oxygen into citrulline and nitric oxide[1-3]. Three distinct mammalian genes have been identified which encode NOS isoforms: neuronal (nNOS or bNOS), macrophage or inducible (iNOS), and endothelial (eNOS)[1-4]. The NOS isoforms can be subdivided into two general categories, constitutive or inducible, based on differences in their regulation and activities[1]. The constitutive isoforms include eNOS and nNOS[1-3]. Although the nomenclature of the NOS family members suggests restricted isoform expression patterns, the160 kDa neuronal-type NO synthase (nNOS) has been detected in a variety of cell types including neurons, epithelial cells, mesangial cells, and skeletal muscle cells.

nNOS is a widely distributed calmodulin-regulated enzyme whose activity is coupled to several neurotransmitter systems both in the brain and in peripheral tissues. The physiological role of nNOS has been perhaps best characterized in the peripheral nervous system, where NO functions as a nonadrenergic-noncholinergic transmitter in several pathways in the gastrointestinal and urogenital tracts. In the CNS, deregulation of nNOS in the brain is associated with over activation of the glutamate receptor and appears to contribute to neuronal damage in animal models of stoke[5]. Mice that lack nNOS are more resistant to neuronal damage following focal cerebral ischemia[6]. Unlike eNOS, the nNOS protein lacks a consensus sequence for fatty acid modification. Nevertheless, nNOS is localized primarily to the particulate fraction via its interactions with specific membrane proteins[7]. In skeletal muscle, nNOS is enriched in fast-twitch muscle fibers where it associates with the sarcolemma[8]. This association with skeletal muscle membranes is due to the association of nNOS with dystrophin[9]. In particular, the dystrophin complex interacts with the N-terminus of nNOS which contains a GLGF motif present in other cytoskeletal-associated proteins[9]. In cases where dystrophin is absent such as in human Duchenne muscular dystrophy (DMD) or *mdx* mice, nNOS can no longer be detected in the sarcolemma and accumulates in the cytosol. This aberrant targeting of nNOS is thought to play a role in the preferential degeneration of fast-twitch muscle fibers seen in DMD[9]. The GLGF motif is also thought to be involved in anchoring nNOS to synaptic membranes complexes in the brain[10].

## REFERENCES

1. Sessa WC. *J Vasc Res* 31:131-143, 1994.
2. Lowenstein CL *et al. Annals of Int Med* 120:227-237, 1994.
3. Nathan C *et al. J Biol Chem* 269:13725-13728, 1994.
4. Snyder S. *Nature* 377:196-197, 1995.
5. Dawson TM et al. Ann Neurol 32:297-311, 1992.
6. Huang PL *et al. Science* 265:1883-1885, 1994.
7. Hecker M *et al. J Neurochem* 62:1524-1529, 1994
8. Kobzik L *et al. Nature* 372:546-548, 1994.
9. Brenman JE *et al. Cell* 82:743-752, 1995.
10. Bredt DS. *Proc Soc Exp Biol Med* 211:41-48, 1996.

## RELATED PRODUCTS

| ***Product*** | ***PAD*/clone*** | ***Cat. No.*** |
|---|---|---|
| Ms x Nitrotyrosine | HM11 | 32-2900 |
| Ms x eNOS | eNOS-6C6 | 33-4500 |
| Ms x eNOS | eNOS-9D10 | 33-4600 |
| Rb x phospho-eNOS (Ser1179) | ZMD.279 | 36-9100 |
| Rb x iNOS | Z-JL8 | 61-7700 |
| Rb x nNOS | Z-RNN3 | 61-7000 |
| Ms x Calmodulin | CaM85 | 13-6900 |
| Rb x Calmodulin | - | 61-8500 |
| Ms x α-CaM Kinase II | CBα-2 | 13-7300 |
| Ms x β-CaM Kinase II | CBβ-2 | 13-9800 |
| Rb x phospho-CaM Kinase II (Thr286) | PS286 | 36-9100 |
| Protein A | Sepharose® 4B | 10-1041 |
| rec-Protein G | Sepharose® 4B | 10-1241 |

*PAD: Polyclonal Antibody Designation

| **Conjugate** | **ZyMAX™ Goat x Rabbit IgG (H+L)** | **ZyMAX™ Goat x Mouse IgG (H+L)** |
|---|---|---|
| Purified | 81-6100 | 81-6500 |
| FITC | 81-6111 | 81-6511 |
| TRITC | 81-6114 | 81-6514 |
| Cy™3 | 81-6115 | 81-6515 |
| Cy™5 | 81-6116 | 81-6516 |
| HRP | 81-6120 | 81-6520 |
| AP | 81-6122 | 81-6522 |
| Biotin | 81-6140 | 81-6540 |



## For Research Use Only