



## Certificate of Analysis

**Anti-nNOS/NOS I**  
(rabbit antiserum)  
Catalog # 07-571  
Lot #DAM1604976

**Immunogen:** Synthetic peptide corresponding to N-terminus of rat nNOS (neuronal nitric oxide synthase).

**Specificity:** Recognizes nNOS, Mr 165kDa.

**Species Cross-Reactivity:** Human, mouse and rat. Other species cross-reactivity is unknown.

**Formulation:** 200µl of rabbit antiserum with 0.05% sodium azide. Frozen solution.

**Storage and Stability:** Stable for 2 years at -20°C from date of shipment. For maximum recovery of product, centrifuge the original vial after thawing and prior to removing the cap. Aliquot to avoid repeated freezing and thawing.

**FOR RESEARCH USE ONLY**  
**NOT FOR USE IN HUMANS**

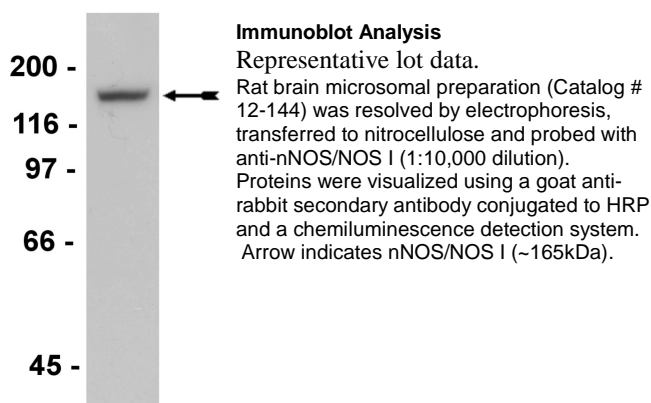
### Quality Control Testing

**Immunoblot Analysis:** A 1:4,000-1:10,000 dilution of this lot detected nNOS (Mr ~165kDa) in a rat brain microsomal preparation (Catalog # 12-144).

The use of fresh extract preparations is recommended.

### Additional Research Applications

**Immunocytochemistry:** This antibody has been reported by an independent laboratory to detect nNOS in C2C12 myotubes.<sup>1</sup>



### Application References:

1. Abdelmoity, A., *et al.*, *FEBS Lett.* **482**: 65-70, 2000.
2. Lau, K.S., *et al.*, *Physiol. Genomics* **2**: 21-27, 2000.
3. Sander, M., *et al.*, *Proc. Natl. Acad. Sci.* **97**: 13818-13823, 2000.

"Unless otherwise stated in our catalog or other company documentation accompanying the product(s), our products are intended for research use only and are not to be used for any other purpose, which includes but is not limited to, unauthorized commercial uses, in vitro diagnostic uses, ex vivo or in vivo therapeutic uses or any type of consumption or application to humans or animals."

### Immunoblot Protocol

1. Perform SDS-polyacrylamide gel electrophoresis (SDS-PAGE) on a freshly prepared neuronal extract and transfer the proteins to nitrocellulose. Wash the blotted nitrocellulose twice with water.
2. Block the blotted nitrocellulose in freshly prepared 3% nonfat dry milk (Catalog # 20-200) in PBS (PBS-MLK) for 20 minutes at room temperature with constant agitation.
3. Incubate the nitrocellulose with a **1:4,000-1:10,000 dilution of anti-nNOS/NOS I**, diluted in freshly prepared PBS-MLK overnight with agitation at 4°C.
4. Wash the nitrocellulose twice with water.
5. Incubate the nitrocellulose in the secondary reagent of choice (a goat anti-rabbit HRP conjugated IgG, Catalog # 12-348, 1:5000 dilution was used) in PBS-MLK for 1.5 hours with agitation at room temperature.
6. Wash the nitrocellulose twice with water.
7. Wash the nitrocellulose in PBS-0.05% Tween<sup>®</sup>-20 for 3-5 minutes.
8. Rinse the nitrocellulose in 4-5 changes of water.
9. Use detection method of choice (enhanced chemiluminescence was used).

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