

Anti-nNOS Antibody

R1510-28



Product Type:	Rabbit polyclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IHC-P, FC, IHC-Fr
Molecular Wt:	Predicted band size: 161 kDa

Description:	Nitric oxide (NO) has a broad range of biological activities and has been implicated in signaling pathways in phylogenetically diverse species. Nitric oxide synthases (NOSs), the enzymes responsible for synthesis of NO, contain an N-terminal oxygenase domain and a C-terminal reductase domain. NOS activity requires homodimerization as well as three cosubstrates (L-arginine, NADPH and O ₂) and five cofactors or prosthetic groups (FAD, FMN, calmodulin, tetrahydrobiopterin and heme). Several distinct NOS isoforms have been described and been shown to represent the products of three distinct genes. These include two constitutive Ca ²⁺ /CaM-dependent forms of NOS, including NOS1 (also designated ncNOS) whose activity was first identified in neurons, and NOS3 (also designated ecNOS), first identified in endothelial cells. The inducible form of NOS, NOS2 (also designated iNOS), is Ca ²⁺ -independent and is expressed in a broad range of cell types.
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Immunogen:	Synthetic peptide within C-terminal human nNOS.
Positive control:	Mouse brain tissue lysate, Rat brain tissue lysate, mouse colon tissue, rat colon tissue, SHG-44, SK-BR-3, HepG2, N2A.

Subcellular location:	Cell membrane, Cell projection
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Database links:	SwissProt: P29475 Human Q9Z0J4 Mouse P29476 Rat
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Recommended Dilutions:

WB	1:1,000
IF-Cell	1:50-1:200
IHC-P	1:10,000-1:50,000
FC	1:50-1:100
IHC-Fr	1:5,000

Storage Buffer:	1*PBS (pH7.4), 0.2% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
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Storage Instruction:	Shipped at 4°C. Store at +4°C short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20°C long term.
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Purity:	Immunogen affinity purified.
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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

Images

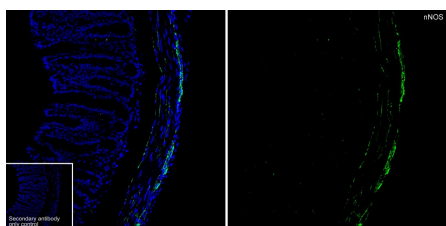


Fig1: Immunofluorescence analysis of frozen mouse colon tissue with Rabbit anti-nNOS antibody (R1510-28) at 1/5,000 dilution.

The section was not undergone antigen retrieval. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (R1510-28, green) at 1/5,000 dilution overnight at 4 °C, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

Fig2: Western blot analysis of nNOS on different lysates with Rabbit anti-nNOS antibody (R1510-28) at 1/1,000 dilution.

Lane 1: Mouse brain tissue lysate

Lane 2: Rat brain tissue lysate

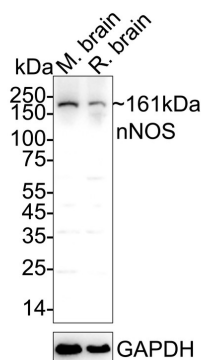
Lysates/proteins at 40 µg/Lane.

Predicted band size: 161 kDa

Observed band size: 161 kDa

Exposure time: 10 seconds; ECL: K1801;

4-20% SDS-PAGE gel.



Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (R1510-28) at 1/1,000 dilution was used in 5% NFDM/TBST at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

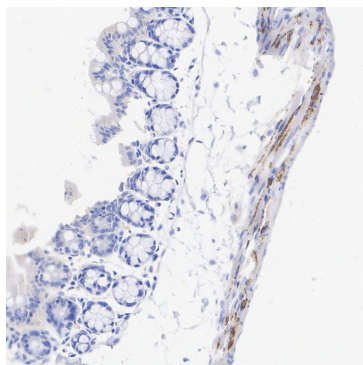


Fig3: Immunohistochemical analysis of paraffin-embedded mouse colon tissue with Rabbit anti-nNOS antibody (R1510-28) at 1/50,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (R1510-28) at 1/50,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

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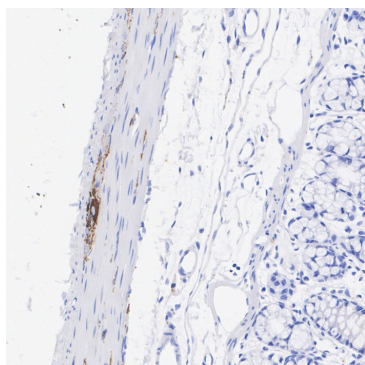


Fig4: Immunohistochemical analysis of paraffin-embedded rat colon tissue with Rabbit anti-nNOS antibody (R1510-28) at 1/10,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (R1510-28) at 1/10,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

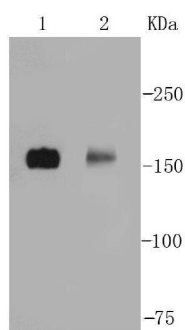


Fig5: Western blot analysis on SHG-44(1) and mouse brain(2) lysates using anti-nNOS rabbit polyclonal antibody.

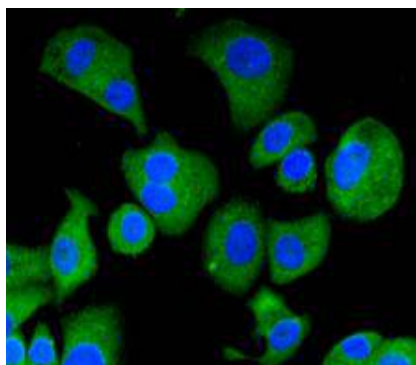


Fig6: Immunocytochemical staining of SK-BR-3 cells using anti-nNOS rabbit polyclonal antibody.

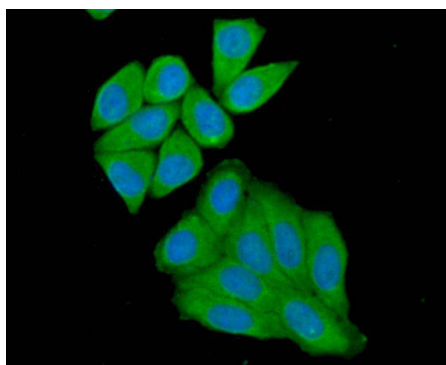


Fig7: Immunocytochemical staining of HepG2 cells using anti-nNOS rabbit polyclonal antibody.

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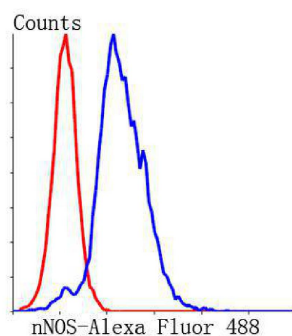


Fig8: Flow cytometric analysis of N2A cells with nNOS antibody at 1/50 dilution (blue) compared with an unlabelled control (cells without incubation with primary antibody; red). Alexa Fluor 488-conjugated Goat anti rabbit IgG was used as the secondary antibody.

Note: All products are “FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE”.

Background References

1. Kovanecz I et al. Oral Bisphenol A (BPA) given to rats at moderate doses is associated with erectile dysfunction, cavernosal lipofibrosis and alterations of global gene transcription. *Int J Impot Res* 26:67-75 (2014).
2. Pribiag H et al. Dystroglycan mediates homeostatic synaptic plasticity at GABAergic synapses. *Proc Natl Acad Sci U S A* 111:6810-5 (2014).

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