Technical Data Sheet

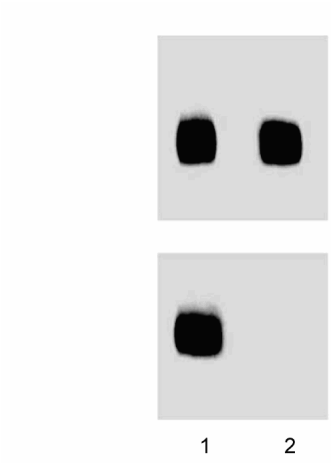
Purified Mouse Anti-eNOS (pS1177)

Product Information

Material Number:	**612392**
Size:	50 µg
Concentration:	250 µg/ml
Clone:	19/eNOS/S1177
Immunogen:	Human eNOS (pS1177)
Isotype:	Mouse IgG1
Reactivity:	QC Testing: Human
Target MW:	140 kDa
Storage Buffer:	Aqueous buffered solution containing BSA, glycerol, and ≤0.09% sodium azide.

Description

Nitric oxide synthase (NOS), a cell-type specific enzyme, catalyzes the synthesis of nitric oxide (NO). NO is a short-lived radical that transmits signals involved in vasorelaxation, neurotransmission, and cytotoxity. In neurons and endothelial cells, constitutive NOS (cNOS) is activated by agonists that increase intracellular Ca2+ levels and enhance calmodulin binding. Neuronal NOS (nNOS) and endothelial NOS (eNOS) have recognition sites for NADPH, FAD, FMN, and calmodulin. eNOS has a unique N-myristylation consensus sequence that may explain its membrane localization. Various protein kinases have been implicated in regulation of eNOS activity, including AMPK, PKA, PKB/Akt, PKC, and CaM Kinase II. During VEGF stimulation, eNOS is transiently phosphorylated at Ser-1177 by PKB/akt and dephosphorylated at Thr-495. At later time points, VEGF stimulation leads to an increase in Thr-495 phosphorylation mediated by PKC and a decrease in Ser-1177 phosphorylation. In addition, Ser-633 and Ser-1177 are phosphorylated by PKA and PKG in vitro. Thus, eNOS activity may be regulated through complex phosphorylated events mediated by multiple kinases at various phosphorylation sites. Human endothelial cells are routinely tested as a positive control for eNOS (pS1177) mAb. 100% homology is detected for immunogen sequence in human, mouse, rat, dog and bovine. Cross-reactivity with other species is expected but not confirmed.



Human Endothelial lysate was either left untreated (left column) or treated (right column) with 150 U/ml) of lambda phosphatase for 1 hour at 37°C. *The top panel was probed with eNOS (Cat. No. 610296). The bottom panel was probed with eNOS (pS1177) (Cat. No. 612392).*

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.
Store undiluted at -20° C.

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Application Notes

Application

Western blot	Routinely Tested
Flow cytometry	Tested During Development

Suggested Companion Products

Catalog Number	Name	Size	Clone
611450	Human Endothelial Cell Lysate	500 µg	(none)
554002	HRP Goat Anti-Mouse Ig	1.0 ml	(none)
610296	Mouse Anti-eNOS/NOS Type III	50 µg	3/eNOS/NOS Type III

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
4. Source of all serum proteins is from USDA inspected abattoirs located in the United States.

References

Dimmeler S, Fleming I, Fisslthaler B, Hermann C, Busse R, Zeiher AM. Activation of nitric oxide synthase in endothelial cells by Akt-dependent phosphorylation.. *Nature.* 1999; 399(6736):601-605.(Biology)

Gallis B, Corthals GL, Goodlett DR, et al. Identification of flow-dependent endothelial nitric-oxide synthase phosphorylation sites by mass spectrometry and regulation of phosphorylation and nitric oxide production by the phosphatidylinositol 3-kinase inhibitor LY294002. *J Biol Chem.* 1999; 274(42):30101-30108. (Biology)

Michell BJ, Chen Zp, Tiganis T, et al. Coordinated control of endothelial nitric-oxide synthase phosphorylation by protein kinase C and the cAMP-dependent protein kinase. *J Biol Chem.* 2001; 276(21):17625-17628.(Biology)