abcam

Product datasheet

Anti-eNOS antibody [EPR19296] ab199956

Recombinant RabMAb

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Overview

Product name	Anti-eNOS antibody [EPR19296]
Description	Rabbit monoclonal [EPR19296] to eNOS
Host species	Rabbit
Tested applications	**Suitable for:** IP, WB
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: Human fetal heart, fetal kidney and fetal liver lysates; Mouse brain, heart, kidney and spleen lysates; Rat brain, heart and spleen lysates; whole cell lysate from HepG2 cells treated with 10 µg/ml LPS for 2 hours and 6 hours; DDDDK tagged human eNOS full length recombinant protein (aa2-1203) **ab198066**. IP: HepG2 whole cell lysate.
General notes	This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information **see here**. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol, 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR19296

Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab199956 in the following tested applications.

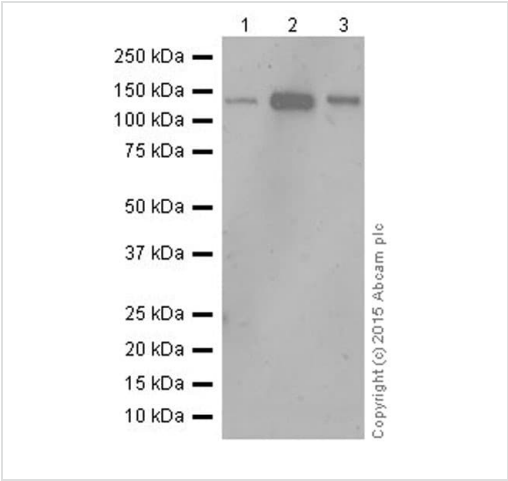
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IP	★★★★☆ **(1)**	1/50.
WB	★★★★☆ **(2)**	1/1000. Detects a band of approximately 140 kDa (predicted molecular weight: 133 kDa).

Target

Function	Produces nitric oxide (NO) which is implicated in vascular smooth muscle relaxation through a cGMP-mediated signal transduction pathway. NO mediates vascular endothelial growth factor (VEGF)-induced angiogenesis in coronary vessels and promotes blood clotting through the activation of platelets. Isoform eNOS13C: Lacks eNOS activity, dominant-negative form that may down-regulate eNOS activity by forming heterodimers with isoform 1.
Tissue specificity	Platelets, placenta, liver and kidney.
Involvement in disease	Variation in NOS3 seem to be associated with susceptibility to coronary spasm.
Sequence similarities	Belongs to the NOS family. Contains 1 FAD-binding FR-type domain. Contains 1 flavodoxin-like domain.
Post-translational modifications	Phosphorylation by AMPK at Ser-1177 in the presence of Ca(2+)-calmodulin (CaM) activates activity. In absence of Ca(2+)-calmodulin, AMPK also phosphorylates Thr-495, resulting in inhibition of activity (By similarity). Phosphorylation of Ser-114 by CDK5 reduces activity.
Cellular localization	Cell membrane. Membrane, caveola. Cytoplasm, cytoskeleton. Golgi apparatus. Specifically associates with actin cytoskeleton in the G2 phase of the cell cycle and which is favored by interaction with NOSIP and results in a reduced enzymatic activity.

Images



Western blot - Anti-eNOS antibody [EPR19296] (ab199956)

All lanes : Anti-eNOS antibody [EPR19296] (ab199956) at 1/1000 dilution

Lane 1 : Human fetal heart lysate
Lane 2 : Human fetal kidney lysate
Lane 3 : Human fetal liver lysate

Lysates/proteins at 20 µg per lane.

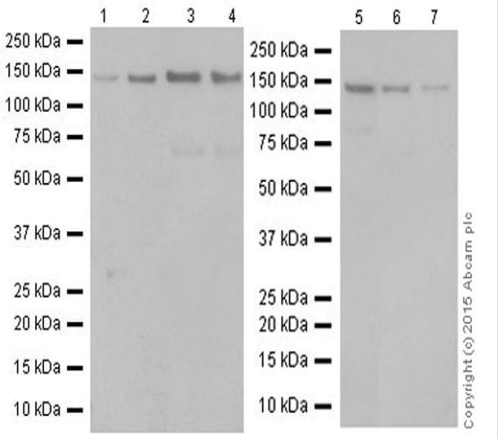
Secondary
All lanes : Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/10000 dilution

Predicted band size: 133 kDa
Observed band size: 140 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.

eNOS is shown to be expressed in platelets, placenta, liver and kidney (UniProt entry P29474). It also plays an important role in the cardiovascular system. (PMID: 15210449 & 15936740).



Western blot - Anti-eNOS antibody [EPR19296] (ab199956)

All lanes : Anti-eNOS antibody [EPR19296] (ab199956) at 1/1000 dilution

Lane 1 : Mouse brain lysate
Lane 2 : Mouse heart lysate
Lane 3 : Mouse kidney lysate
Lane 4 : Mouse spleen lysate
Lane 5 : Rat brain lysate
Lane 6 : Rat heart lysate
Lane 7 : Rat spleen lysate

Lysates/proteins at 10 µg per lane.

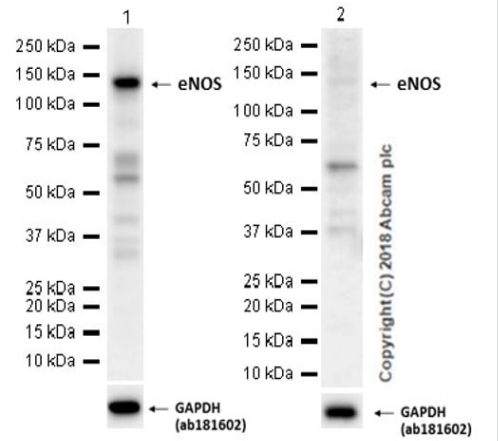
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

Predicted band size: 133 kDa
Observed band size: 140 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: Lane 1, 2, 3, 4, 6 and 7: 30 seconds; Lane 5: 2 minutes.

eNOS is shown to be expressed in platelets, placenta, liver and kidney (UniProt entry P29474). It also plays an important role in the cardiovascular system. (PMID: 15210449 & 15936740).



Western blot - Anti-eNOS antibody [EPR19296] (ab199956)

All lanes : Anti-eNOS antibody [EPR19296] (ab199956) at 1/1000 dilution

Lane 1 : HepG2 (Human hepatocellular carcinoma epithelial cell) whole cell lysate
Lane 2 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate

Lysates/proteins at 20 µg per lane.

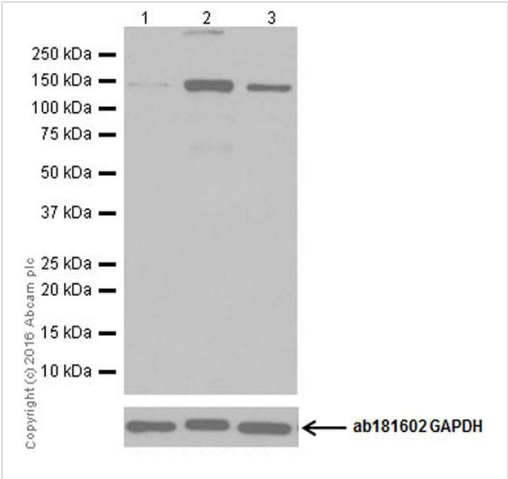
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 133 kDa

Observed band size: 140 kDa

Exposure Time: Lane1: 40 seconds; Lane2: 180 seconds

The expression profile observed in HeLa is consistent with the literature (PMID: 19925457).
Negative control: HeLa (PMID: 19925457)



Western blot - Anti-eNOS antibody [EPR19296] (ab199956)

All lanes : Anti-eNOS antibody [EPR19296] (ab199956) at 1/5000 dilution

Lane 1 : HepG2 (Human liver hepatocellular carcinoma cell line) whole cell lysate
Lane 2 : Whole cell lysate from HepG2 cells treated with 10 µg/ml LPS for 2 hours
Lane 3 : Whole cell lysate from HepG2 cells treated with 10 µg/ml LPS for 6 hours

Lysates/proteins at 20 µg per lane.

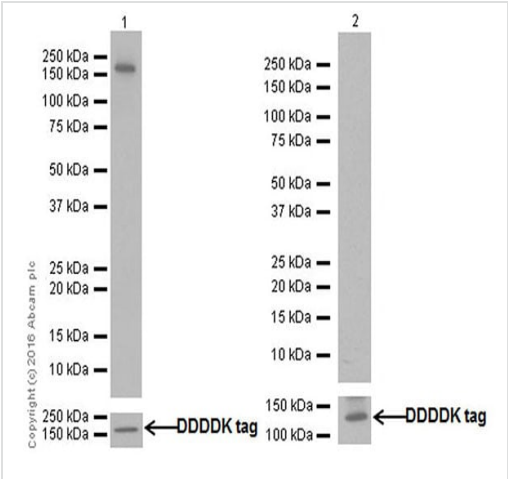
Secondary
All lanes : Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/10000 dilution

Predicted band size: 133 kDa
Observed band size: 140 kDa

Exposure time: 15 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

The expression profile observed is consistent with what has been described in the literature (PMID:17855661 & 11004215).



Western blot - Anti-eNOS antibody [EPR19296] (ab199956)

All lanes : Anti-eNOS antibody [EPR19296] (ab199956) at 1/1000 dilution

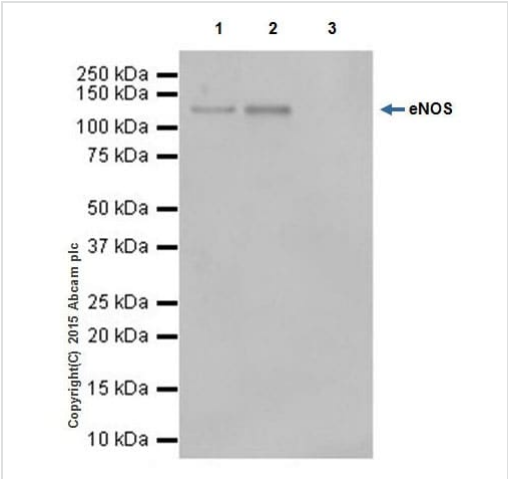
Lane 1 : Recombinant Human eNOS protein (Tagged) (**ab198066**) at 0.01 µg
Lane 2 : Recombinant Human iNOS protein (**ab135010**) at 0.05 µg

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

Predicted band size: 133 kDa
Observed band size: 140 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: Lane 1: 1 second; Lane 2: 3 minutes.



Immunoprecipitation - Anti-eNOS antibody [EPR19296] (ab199956)

eNOS was immunoprecipitated from 1 mg of HepG2 (Human liver hepatocellular carcinoma cell line) whole cell lysate with ab199956 at 1/50 dilution. Western blot was performed from the immunoprecipitate using ab199956 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/10000 dilution.

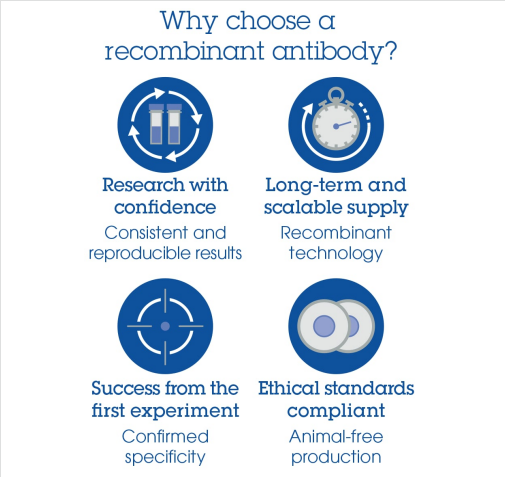
Lane 1: HepG2 whole cell lysate 10 µg (Input).

Lane 2: ab199956 IP in HepG2 whole cell lysate.

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of ab199956 in HepG2 whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 3 seconds.



Anti-eNOS antibody [EPR19296] (ab199956)

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