abcam

Product datasheet

Anti-eNOS (phospho S1177) antibody ab184154

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Overview

Species reactivity

Product name Anti-eNOS (phospho S1177) antibody

Description Rabbit polyclonal to eNOS (phospho S1177)

Host species Rabbit

Tested applications Suitable for: WB, ICC/IF

Reacts with: Human

Predicted to work with: Guinea pig, Pig ...

Immunogen Synthetic peptide within Human eNOS (phospho S1177). The exact sequence is proprietary.

Database link: P29474

Positive control THP1 whole cell lysate (untreated and treated with 100 nM PMA treatment for 6, 24 or 48 hr),

Jurkat, Raji and NCI-H929 whole cell lysate; HeLa cells.

General notesThe Life Science industry has been in the grips of a reproducibility crisis for a number of years.

Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

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: 7.00

Preservative: 0.025% Proclin 300

Constituents: 78% PBS, 1% BSA, 20% Glycerol (glycerin, glycerine)

Purity Immunogen affinity purified

Clonality Polyclonal

Isotype IgG

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Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab184154 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	
WB		1/500 - 1/3000. Predicted molecular weight: 133 kDa.	
ICC/IF	★★★★★ (1)	1/100 - 1/1000.	

Target

H	u	n	ct	10	n

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

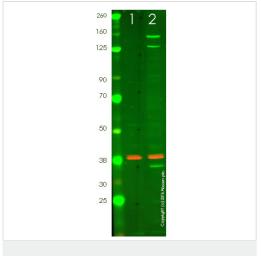
Phosphorylation by AMPK at Ser-1177 in the presence of Ca(2+)-calmodulin (CaM) activates activity. In absence of Ca(2+)-calmodulin, AMPK also phosphorylates Thr-495, resulting in inhibition of activity (By similarity). Phosphorylation of Ser-114 by CDK5 reduces activity.

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 (phospho S1177) antibody (ab184154)

All lanes : Anti-eNOS (phospho S1177) antibody (ab184154) at 1/1000 dilution

Lane 1: HUVEC whole cell lysate

Lane 2: VEGF (ab9571) treated HUVEC whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes: goat anti-rabbit antibody (green) at 1/20000 dilution

Performed under reducing conditions.

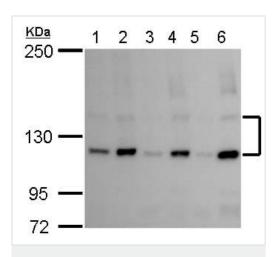
Predicted band size: 133 kDa

Additional bands at: 150 kDa (possible post-translational

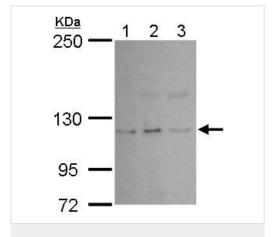
modification)

2e7 HUVEC, grown in serum free medium for 18 hours, were harvested with 0.5 mM EDTA. Cells, collected and resuspended in 10 mL cell culture medium, were stimulated for 2 minutes with 50 ng per mL VEGF (ab9571), after which 30 mL of ice-cold PBS was added. The tubes were spun for 2 min at 1.5k RPM and cell pellets were snap frozen. Control cells were treated in parallel, but without the addition of VEGF.

This blot was produced using a 4-12% Bis-Tris gel under the MOPS buffer system. The gel was run at 200V for 60 minutes before being transferred onto a nitrocellulose membrane at 30V for 70 minutes. The membrane was blocked for an hour before being incubated with anti-eNOS (phospho S1177) antibody (ab184154; diluted 1:1000) and loading control (anti-GAPDH antibody [6C5] ab8245; diluted 1:20000) at 4°C overnight. Antibody binding was detected using infrared labelled goat anti-rabbit (green) or anti-mouse (red) antibodies (diluted 1:20000) for 1 hour at room temperature before imaging.



Western blot - Anti-eNOS (phospho S1177) antibody (ab184154)



Western blot - Anti-eNOS (phospho S1177) antibody (ab184154)

All lanes : Anti-eNOS (phospho S1177) antibody (ab184154) at 1/500 dilution

Lanes 1 & 3 & 5: THP1 whole cell lysate

Lane 2: THP1 whole cell lysate (100 nM PMA treatment for 6 hr)

Lane 4: THP1 whole cell lysate (100 nM PMA treatment for 24 hr)

Lane 6: THP1 whole cell lysate (100 nM PMA treatment for 48 hr)

Lysates/proteins at 30 µg per lane.

Predicted band size: 133 kDa

5 % SDS-PAGE

All lanes : Anti-eNOS (phospho S1177) antibody (ab184154) at 1/500 dilution

Lane 1: Jurkat whole cell lysate

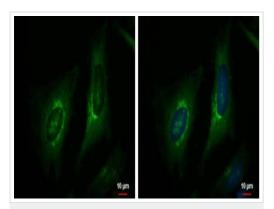
Lane 2: Raji whole cell lysate

Lane 3: NCI-H929 whole cell lysate

Lysates/proteins at 30 µg per lane.

Predicted band size: 133 kDa

5 % SDS-PAGE



Immunocytochemistry/ Immunofluorescence - AntieNOS (phospho S1177) antibody (ab184154)

Immunofluorescent analysis of methanol fixed HeLa cells labeling eNOS using ab184154 at 1/500 dilution. Image at the right costained with Hoechst 33342.

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