

Product Datasheet

eNOS/NOS3 Rabbit Polyclonal Antibody (orb546315)

Catalog Number	orb546315
Category	Antibodies
Description	eNOS/NOS3 Rabbit Polyclonal Antibody
Target	Nitric oxide synthase 3
Clonality	Polyclonal
Species/Host	Rabbit
Isotype	Rabbit IgG
Conjugation	Unconjugated
Reactivity	Human, Mouse, Rat
Form/Appearance	Lyophilized
Concentration	500 µg/ml
Buffer/Preservatives	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na ₂ HPO ₄ , 0.05mg NaN ₃ .
Reconstitution	Add 0.2ml of distilled water will yield a concentration of 500ug/ml.
Purification	Immunogen affinity purified.
Immunogen	E.coli-derived human eNOS/NOS3 recombinant protein (Position: Q1052-P1203).
UniProt ID	P29474
MW	133 kDa

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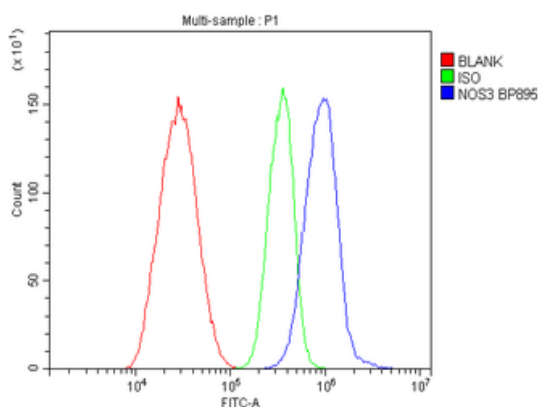
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Tested applications	ELISA, FC, IHC, WB
Dilution range	Western blot, 0.1-0.5µg/ml, Human, Mouse, Rat Immunohistochemistry (Paraffin-embedded Section), 0.5-1µg/ml, Human, Mouse, Rat Flow Cytometry (Fixed), 1-3µg/1x10 ⁶ cells, Human ELISA, 0.1-0.5µg/ml, Huma
Specificity	No cross reactivity with other proteins.
Cross Reactivity	No cross-reactivity with other proteins.
Antibody Type	Primary Antibody
Storage	Maintain refrigerated at 2-8°C for up to 2 weeks. For long term storage store at -20°C in small aliquots to prevent freeze-thaw cycles.
Note	For research use only
Expiration Date	12 months from date of receipt.



Flow Cytometry analysis of HepG2 cells using anti-NOS3 antibody. Overlay histogram showing HepG2 cells (Blue line). To facilitate intracellular staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-NOS3 Antibody (1 µg/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (5-10 µg/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 µg/1x10⁶) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.

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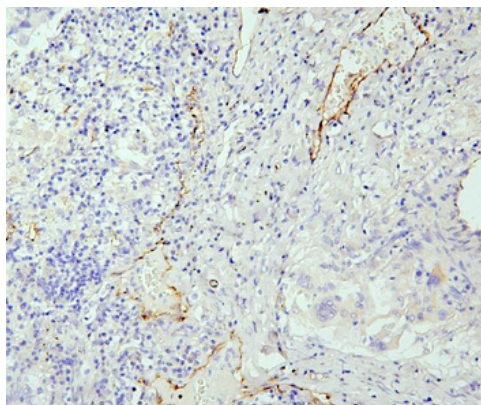
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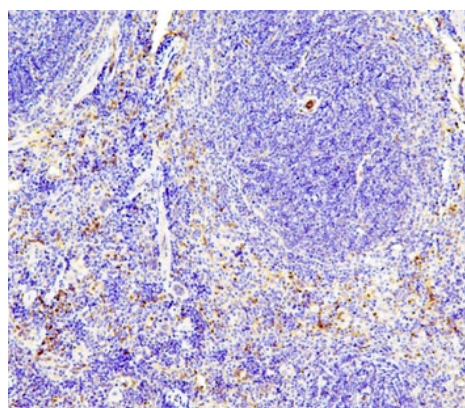
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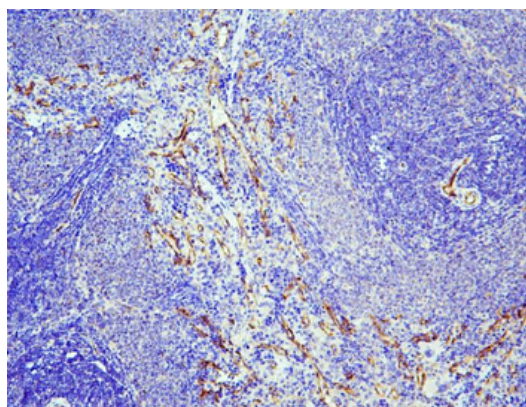
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IHC analysis of NOS3 using anti-NOS3 antibody. NOS3 was detected in paraffin-embedded section of human lung cancer tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 μ g/ml rabbit anti-NOS3 Antibody overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) with DAB as the chromogen.



IHC analysis of NOS3 using anti-NOS3 antibody. NOS3 was detected in paraffin-embedded section of mouse spleen tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 μ g/ml rabbit anti-NOS3 Antibody overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) with DAB as the chromogen.



IHC analysis of NOS3 using anti-NOS3 antibody. NOS3 was detected in paraffin-embedded section of rat spleen tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 μ g/ml rabbit anti-NOS3 Antibody overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) with DAB as the chromogen.

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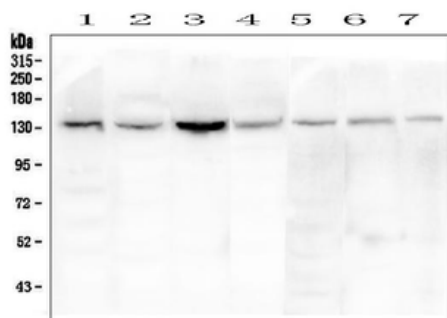
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Western blot analysis of NOS3 using anti-NOS3 antibody. Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50 ug of sample under reducing conditions. Lane 1: human placenta tissue lysates Lane 2: human K562 whole cell lysates Lane 3: human HepG2 whole cell lysates Lane 4: human THP-1 whole cell lysates Lane 5: rat kidney tissue lysates Lane 6: mouse kidney tissue lysates Lane 7: mouse small intestine tissue lysates. After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-NOS3 antigen affinity purified polyclonal antibody at 0.5 $\mu\text{g}/\text{mL}$ overnight at 4°C, then washed with TBS-0.1% Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit with Tanon 5200 system. A specific band was detected for NOS3 at approximately 133KD. The expected band size for NOS3 is at 133KD.

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