

# Product Datasheet

## EGLN3/PHD3 Antibody - BSA Free NB100-303SS

Unit Size: 0.025 ml

Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.

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Updated 4/13/2025 v.20.1

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**NB100-303SS**

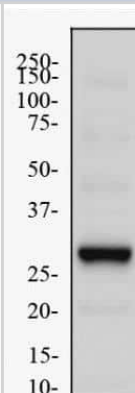
EGLN3/PHD3 Antibody - BSA Free

Product Information	
Unit Size	0.025 ml
Concentration	1 mg/ml
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	0.02% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	PBS
Target Molecular Weight	27 kDa
Product Description	
Host	Rabbit
Gene ID	112399
Gene Symbol	EGLN3
Species	Human, Mouse, Rat, Primate
Reactivity Notes	Primate reactivity reported in scientific literature (PMID: 23732909). Rat reactivity reported in scientific literature (PMID: 23545307).
Immunogen	A synthetic peptide made to the C-terminus of humanPHD3/HIF Prolyl Hydroxylase 3. [LocusLink ID 112399]
Product Application Details	
Applications	Western Blot, Simple Western, Chromatin Immunoprecipitation, Immunoblotting, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin, Immunoprecipitation, Chromatin Immunoprecipitation (ChIP)
Recommended Dilutions	Western Blot 1:500-1:2000, Simple Western 1:1000, Chromatin Immunoprecipitation reported in scientific literature (PMID 21620138), Immunohistochemistry, Immunocytochemistry/ Immunofluorescence reported in scientific literature (PMID 20801873), Immunoprecipitation 1:10-1:500. Use reported in scientific literature (PMID 15721254), Immunohistochemistry-Paraffin reported in scientific literature (PMID 25161887), Immunoblotting, Chromatin Immunoprecipitation (ChIP)
Application Notes	In Western blot, a band is seen at approx. 27 kDa. This antibody has been tested against HeLa and MEF cell lysates. In Simple Western only 10 - 15 uL of the recommended dilution is used per data point. See <a href="#">Simple Western Antibody Database</a> for Simple Western validation: Tested in Hypoxic HeLa lysate 0.5 mg/mL, separated by Size, antibody dilution of 1:1000, apparent MW was 31 kDa. Separated by Size-Wes, Sally Sue/Peggy Sue.



## Images

Western Blot: EGLN3/PHD3 Antibody [NB100-303] - Whole cell protein from MEF cells was separated on a 12% gel by SDS-PAGE, transferred to PVDF membrane and blocked in 5% non-fat milk in TBST. The membrane was probed with 2 ug/mL anti-EGLN3/PHD3 in 1% milk, and detected with an anti-rabbit HRP secondary antibody using chemiluminescence.



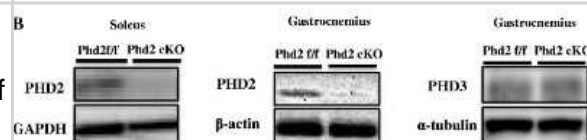
Simple Western: EGLN3/PHD3 Antibody [NB100-303] - Image shows a specific band for PHD3/HIF Prolyl Hydroxylase 3 in 0.5 mg/mL of Hypoxic HeLa cell lysate. This experiment was performed under reducing conditions using the 12-230 kDa separation system.



Immunohistochemistry-Paraffin: EGLN3/PHD3 Antibody [NB100-303] - LDL Receptor was detected in immersion fixed paraffin-embedded sections of human liver cancer using rabbit anti-human antibody (NB100-303) at 1:3000 overnight at 4C. Tissue was stained using the VisuCyte anti-rabbit HRP polymer detection reagent (VC003) with DAB chromogen (brown) and counterstained with hematoxylin (blue).



Western Blot: EGLN3/PHD3 Antibody [NB100-303] - Tamoxifen administration-induced PHD2 deletion in skeletal muscle in Phd2<sup>f/f</sup>/Rosa26CreERT2 mice. Anti-PHD2 and PHD3 Western blotting of gastrocnemius and soleus muscles at 6 weeks after tamoxifen administration. Image collected and cropped by CiteAb from the following publication (<https://www.skeletalmusclejournal.com/content/6/1/5>), licensed under a CC-BY license.



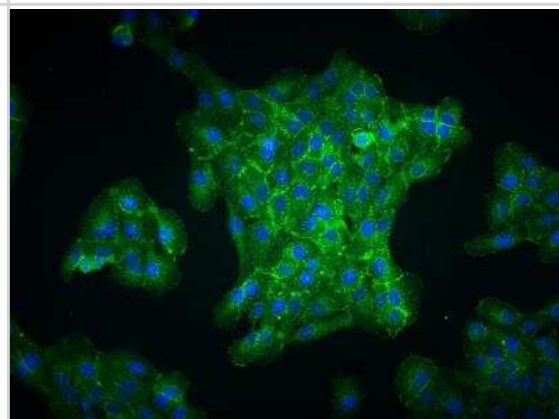
Immunocytochemistry/Immunofluorescence: EGLN3/PHD3 Antibody [NB100-303] - HeLa cells were fixed for 10 minutes using 10% formalin and then permeabilized for 5 minutes using 1X TBS + 0.5% Triton X-100. The cells were incubated with anti-HIF Prolyl Hydroxylase 3 (NB100-3030) at 1:200 overnight at 4C and detected with an anti-rabbit Dylight 488 (Green) at 1:500. Alpha tubulin, DM1A (NB100-690) was used as a co-stain at 1:1000 and detected with an anti-mouse DyLight 550 (Red) at 1:500. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 40X objective.



Immunohistochemistry: EGLN3/PHD3 Antibody [NB100-303] - PHD3 was detected in immersion fixed paraffin-embedded sections of human heart using Rabbit Anti-Human EGLN3 polyclonal Antibody (NB100-303) at 5 ug/mL for 1 hour at room temperature followed by incubation with the Anti-Rabbit IgG VisUCyte(TM) HRP Polymer Antibody (VC003). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to nuclei.



Immunocytochemistry/Immunofluorescence: EGLN3/PHD3 Antibody [NB100-303] - Analysis of PHD3 in ARPE-19 cells using anti-PHD3 antibody. Image from verified customer review.



## Publications

Soni C, Perez OA, Voss WN, Pucella JN et Al. Plasmacytoid Dendritic Cells and Type I Interferon Promote Extrafollicular B Cell Responses to Extracellular Self-DNA Immunity 2020-05-27 [PMID: 32454024]

Anisyah Ridiandrie CC-chemokine class inhibition attenuates pathological angiogenesis while preserving physiological angiogenesis FASEB J, 2016-12-20;0(0):. 2016-12-20 [PMID: 27998907]

Casciello F, Al-Ejeh F, Kelly G et al. G9a drives hypoxia-mediated gene repression for breast cancer cell survival and tumorigenesis. Proc. Natl. Acad. Sci. U.S.A. 2017-06-19 [PMID: 28630300]

Nathan K. P. Wong, Emma L. Solly, Richard Le, Victoria A. Nankivell, Jocelyne Mulangala, Peter J. Psaltis, Stephen J. Nicholls, Martin K. C. Ng, Christina A. Bursill, Joanne T. M. Tan, Riccardo Alessandro TRIM2 Selectively Regulates Inflammation-Driven Pathological Angiogenesis without Affecting Physiological Hypoxia-Mediated Angiogenesis International Journal of Molecular Sciences 2024-03-15 [PMID: 38542330]

Meng X, Lin Z, Cao S et al. Estrogen-mediated downregulation of HIF-1 alpha signaling in B lymphocytes influences postmenopausal bone loss Bone research 2022-02-17 [PMID: 35177582] (Western Blot)

Kenneth K. Y. Ting, Pei Yu, Riley Dow, Hisham Ibrahim, Saraf Karim, Chanele K. Polenz, Daniel A. Winer, Minna Woo, Jenny Jongstra-Bilen, Myron I. Cybulsky Cholesterol accumulation impairs HIF-1 $\alpha$ -dependent immunometabolic reprogramming of LPS-stimulated macrophages by upregulating the NRF2 pathway Scientific Reports 2024-05-15 [PMID: 38750095]

Noonan ML, Ni P, Solis E et al. Osteocyte EglN1/Phd2 links oxygen sensing and biomineralization via FGF23 Bone Research 2023-01-18 [PMID: 36650133] (Western Blot)

Woods PS, Kimmig LM, Sun KA et al. HIF-1 alpha induces glycolytic reprogramming in tissue-resident alveolar macrophages to promote cell survival during acute lung injury eLife 2022-07-13 [PMID: 35822617] (WB, Mouse)

Chen T, Zhou Q, et al. miR-17/20 Controls Prolyl Hydroxylase 2 (PHD2)/Hypoxia-Inducible Factor 1 (HIF1) to Regulate Pulmonary Artery Smooth Muscle Cell Proliferation. J Am Heart Assoc 2016-12-05 [PMID: 27919930] (WB, Mouse)

Angelini A, Saha PK, Jain A Et al. PHDs/CPT1B/VDAC1 axis regulates long-chain fatty acid oxidation in cardiomyocytes Cell reports 2021-10-05 [PMID: 34610308] (WB, IP, Rat)

Xu YM, Gao Q, Zhang JZ et al. Prolyl hydroxylase 3 controls the intestine goblet cell generation through stabilizing ATOH1 Cell Death Differ. 2020-01-20 [PMID: 31959916] (WB, IP, Mouse, Human)

Pastor-Soler NM, Sutton TA, Mang HE et al. Muc1 is protective during kidney ischemia-reperfusion injury Am J Physiol Renal Physiol. 2015-04-28 [PMID: 25925251] (WB, Mouse)

More publications at <http://www.novusbio.com/NB100-303>



## Procedures

### Western Blot Protocol for PHD3/HIF Prolyl Hydroxylase 3 Antibody (NB100-303)

#### Western Blot Protocol

1. Perform SDS-PAGE on samples to be analyzed, loading 35 ug of sample lysate per lane.
2. Transfer proteins to membrane according to the instructions provided by the manufacturer of the membrane and transfer apparatus.
3. Stain according to standard Ponceau S procedure (or similar product) to assess transfer success, and mark molecular weight standards where appropriate.
4. Rinse the blot.
5. Block the membrane using standard blocking buffer for at least 1 hour.
6. Wash the membrane in wash buffer three times for 10 minutes each.
7. Dilute primary antibody in blocking buffer and incubate 1 hour at room temperature.
8. Wash the membrane in wash buffer three times for 10 minutes each.
9. Apply the diluted HRP conjugated secondary antibody in blocking buffer (as per manufacturers instructions) and incubate 1 hour at room temperature.
10. Wash the blot in wash buffer three times for 10 minutes each (this step can be repeated as required to reduce background).
11. Apply the detection reagent of choice in accordance with the manufacturers instructions.

Note: Tween-20 can be added to the blocking or antibody dilution buffer at a final concentration of 0.05-0.2%.





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### **Limitations**

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

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