# **Product Datasheet**

# DUOX2 Antibody - BSA Free NB110-61576

Unit Size: 0.1 ml

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

www.novusbio.com



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### NB110-61576

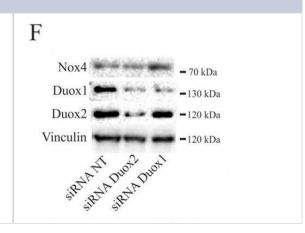
DUOX2 Antibody - BSA Free

Product Information	
Unit Size	0.1 ml
Concentration	1.0 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
Product Description	
Host	Rabbit
Gene ID	50506
Gene Symbol	DUOX2
Species	Human, Mouse, Rat, Porcine, Canine
Immunogen	Synthetic peptide made to an internal portion of human DUOX2 (within residues 400-500). [Swiss-Prot# Q9NRD8]
<b>Product Application Details</b>	
Applications	Western Blot, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin, Knockdown Validated
Recommended Dilutions	Western Blot 2 ug/ml, Immunohistochemistry, Immunocytochemistry/ Immunofluorescence 1:1000, Immunohistochemistry-Paraffin reported in scientific literature (PMID 24492313), Knockdown Validated
Application Notes	In Western blot a band is observed at ~150 kDa, and in Immunocytochemistry/Immunofluorescence membrane staining was seen in

## **Images**

Knockdown Validated: DUOX2 Antibody [NB110-61576] - Expression profile and silencing of NADPH-oxidases in mesenchymal cells.3T3 fibroblasts were transiently transfected by siRNAs to Duox1 or Duox2 and analyzed for Nox4 and Duox1/2 proteins in 2 experiments. Image collected and cropped by CiteAb from the following publication (//doi.org/10.1371/journal.pone.0154157), licensed under a CC-BY license.

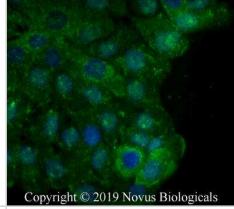
A431 cells.



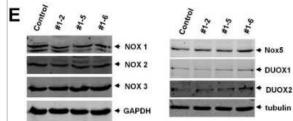
Western Blot: DUOX2 Antibody [NB110-61576] - Detection of DUOX2 in A549 in cell lysate.



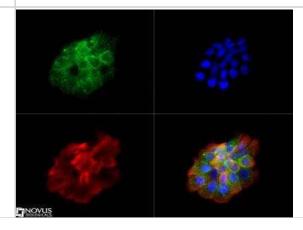
Immunocytochemistry/Immunofluorescence: DUOX2 Antibody [NB110-61576] - A431 cells were fixed for 10 minutes using 10% formalin and then permeabilized for 5 minutes using 1X PBS + 0.05% Triton-X100. The cells were incubated with anti-DUOX at 5 ug/ml overnight at 4C and detected with an anti-rabbit Dylight 488 (Green) at a 1:500 dilution. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 40X objective.



Western Blot: DUOX2 Antibody [NB110-61576] - Generation and validation of NOX4 knockout (KO) HeLa cell lines. NOX4 knockout did not influence NOX1, NOX2, NOX3, NOX5, DUOX1 and DUOX2 levels. Protein levels of NOX1, NOX2, NOX3, NOX5, DUOX1 and DUOX2 in parental HeLa cells and three clones of NOX4 knockout cells. Image collected and cropped by CiteAb from the following publication (https://dx.plos.org/10.1371/journal.pone.0170327), licensed under a CC-BY license.



Immunocytochemistry/Immunofluorescence: DUOX2 Antibody [NB110-61576] - DUOX2 antibody was tested in A431 cells with Dylight 488 (green). Nuclei and alpha-tubulin were counterstained with DAPI (blue) and Dylight 550 (red).



### **Publications**

Poinsignon L, Chissey A, Ajjaji A et al. Placental cartography of NADPH oxidase (NOX) family proteins: Involvement in the pathophysiology of preeclampsia Archives of biochemistry and biophysics 2023-10-20 [PMID: 37866451] (Immunohistochemistry, Western Blot, Human)

Yang M, Zhao Y, Ding Y et al. A truncated protein product of the germline variant of the DUOX2 gene leads to adenomatous polyposis Cancer Biology and Medicine 2021-02-15 [PMID: 33628596]

Mota M, Metge BJ, Hinshaw DC et al. Merlin deficiency alters the redox management program in breast cancer Molecular Oncology 2021-04-01 [PMID: 33410252] (Western Blot)

Zhou YJ, Lu XF, Chen H et al. Single-cell transcriptomics reveals early molecular and immune alterations underlying the serrated neoplasia pathway toward colorectal cancer Cellular and molecular gastroenterology and hepatology 2022-10-07 [PMID: 36216310] (IHC-P)

Lee H, Ryu H, Park H et al. Dual Oxidase 2 (DUOX2) as a Proteomic Biomarker for Predicting Treatment Response to Chemoradiation Therapy for Locally Advanced Rectal Cancer: Using High-Throughput Proteomic Analysis and Machine Learning Algorithm International Journal of Molecular Sciences 2022-10-26 [PMID: 36361712] (IHC-P, Human)

#### Details:

Dilution used for IHC 1:100

Liang T, Zhang N, Ju H et al. Telocytes Reduce Oxidative Stress by Downregulating DUOX2 Expression in Inflamed Lungs of Mice Acta Biochim Biophys Sin (Shanghai) 2022-05-24 [PMID: 35607956]

Dyikanov D, Vasiluev P, Rysenkova K et al. Optimization of CRISPR/Cas9 technology to knock-out genes of interest in aneuploid cell lines Tissue Eng Part C Methods 2019-02-12 [PMID: 30747044] (WB)

Jafari N, Kim H, Park R et al. CRISPR-Cas9 Mediated NOX4 Knockout Inhibits Cell Proliferation and Invasion in HeLa Cells PLoS ONE 2017-01-18 [PMID: 28099519] (WB, Human)

Tyurin-Kuzmin PA, Zhdanovskaya ND, Sukhova AA et al. Nox4 and Duox1/2 Mediate Redox Activation of Mesenchymal Cell Migration by PDGF. PLoS ONE. 2016-04-26 [PMID: 27110716] (WB, Mouse)

Wakatsuki S, Furuno A, Ohshima M, Araki T. Oxidative stress-dependent phosphorylation activates ZNRF1 to induce neuronal/axonal degeneration. J. Cell Biol. 2015-11-23 [PMID: 26572622] (ICC/IF, Mouse)

Macfie TS, Poulsom R, Parker A et al. DUOX2 and DUOXA2 Form the Predominant Enzyme System Capable of Producing the Reactive Oxygen Species H2O2 in Active Ulcerative Colitis and are Modulated by 5-Aminosalicylic Acid. Inflamm. Bowel Dis. 2014-02-14 [PMID: 24492313] (IHC-P, Human)

Morand, S et al. Effect of iodide on nicotinamide adenine dinucleotide phosphate oxidase activity and Duox2 protein expression in isolated porcine thyroid follicles. Endocrinology;144(4):1241-8. 2003-04-01 [PMID: 12639906] (WB, Porcine)



### **Procedures**

### Western Blot protocol for DUOX2 Antibody (NB110-61576)

DUOX2 Antibody:

Western Blot Protocol

- 1. Perform SDS-PAGE (4-12% MOPS) on samples to be analyzed, loading 38 ug of total protein per lane.
- 2. Transfer proteins to Nitrocellulose according to the instructions provided by the manufacturer of the transfer apparatus.
- 3. Rinse membrane with dH2O and then stain the blot using Ponceau S for 1-2 minutes to access the transfer of proteins onto the nitrocellulose membrane. Rinse the blot in water to remove excess stain and mark the lane locations and locations of molecular weight markers using a pencil.
- 4. Rinse the blot in TBS for approximately 5 minutes.
- 5. Block the membrane using 5% NFDM + 1% BSA in TBS + Tween, 1 hour at RT.
- 6. Rinse the membrane in dH2O and then wash the membrane in wash buffer [TBS + 0.1% Tween] 3 times for 10 minutes each.
- 7. Dilute the rabbit anti-DUOX2 primary antibody (NB 110-61576) in blocking buffer and incubate 1 hour at room temperature.
- 8. Rinse the membrane in dH2O and then wash the membrane in wash buffer [TBS + 0.1% Tween] 3 times for 10 minutes each.
- 9. Apply the diluted rabbit-IgG 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 [TBS + 0.1% Tween] 3 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 (Pierce ECL).

Note: Tween-20 can be added to the blocking or antibody dilution buffer at a final concentration of 0.05-0.2%, provided it does not interfere with antibody-antigen binding.

# Immunocytochemistry/Immunofluorescence protocol for DUOX2 Antibody (NB110-61576) DUOX2 Antibody:

Immunocytochemistry Protocol

Culture cells to appropriate density in 35 mm culture dishes or 6-well plates.

- 1. Remove culture medium and add 10% formalin to the dish. Fix at room temperature for 30 minutes.
- 2. Remove the formalin and add ice cold methanol. Incubate for 5-10 minutes.
- 3. Remove methanol and add washing solution (i.e. PBS). Be sure to not let the specimen dry out. Wash three times for 10 minutes.
- 4. To block nonspecific antibody binding incubate in 10% normal goat serum from 1 hour to overnight at room temperature.
- 5. Add primary antibody at appropriate dilution and incubate at room temperature from 2 hours to overnight at room temperature.
- 6. Remove primary antibody and replace with washing solution. Wash three times for 10 minutes.
- 7. Add secondary antibody at appropriate dilution. Incubate for 1 hour at room temperature.
- 8. Remove antibody and replace with wash solution, then wash for 10 minutes. Add Hoechst 33258 to wash solution at 1:25,0000 and incubate for 10 minutes. Wash a third time for 10 minutes.
- 9. Cells can be viewed directly after washing. The plates can also be stored in PBS containing Azide covered in Parafilm (TM). Cells can also be cover-slipped using Fluoromount, with appropriate sealing.
- \*The above information is only intended as a guide. The researcher should determine what protocol best meets their needs. Please follow safe laboratory procedures.





## **Novus Biologicals USA**

10730 E. Briarwood Avenue Centennial, CO 80112

USA

Phone: 303.730.1950 Toll Free: 1.888.506.6887

Fax: 303.730.1966

nb-customerservice@bio-techne.com

### **Bio-Techne Canada**

21 Canmotor Ave Toronto, ON M8Z 4E6

Canada

Phone: 905.827.6400 Toll Free: 855.668.8722 Fax: 905.827.6402

canada.inquires@bio-techne.com

### **Bio-Techne Ltd**

19 Barton Lane Abingdon Science Park Abingdon, OX14 3NB, United Kingdom Phone: (44) (0) 1235 529449

Free Phone: 0800 37 34 15 Fax: (44) (0) 1235 533420 info.EMEA@bio-techne.com

## **General Contact Information**

www.novusbio.com

Technical Support: nb-technical@bio-

techne.com

Orders: nb-customerservice@bio-techne.com

General: novus@novusbio.com

# **Products Related to NB110-61576**

HAF008 Goat anti-Rabbit IgG Secondary Antibody [HRP]

NB7160 Goat anti-Rabbit IgG (H+L) Secondary Antibody [HRP]

NBP2-24891 Rabbit IgG Isotype Control

NB110-61576G DUOX2 Antibody [DyLight 488]

#### 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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