Product Datasheet

Nucleolin Antibody
NB600-241

Unit Size: 0.1 ml

Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.

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Reviews: 2 Publications: 26

Protocols, Publications, Related Products, Reviews, Research Tools and Images at:
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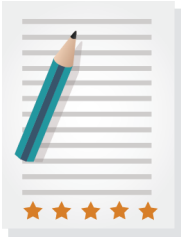
Updated 4/13/2025 v.20.1



NB600-241

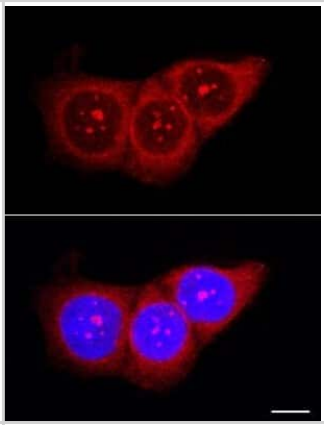
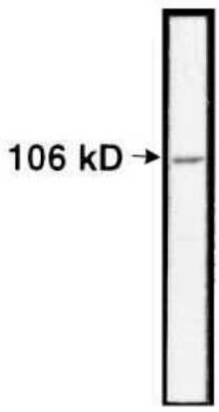
Nucleolin Antibody

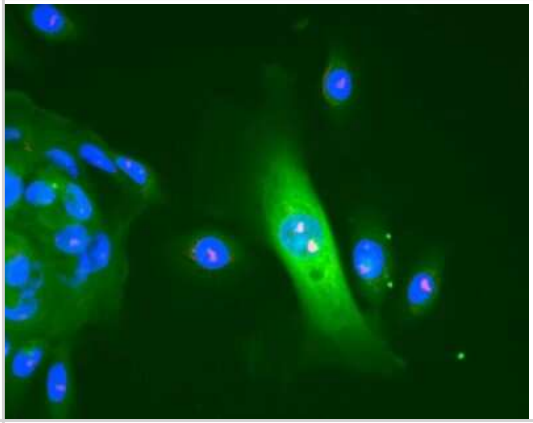
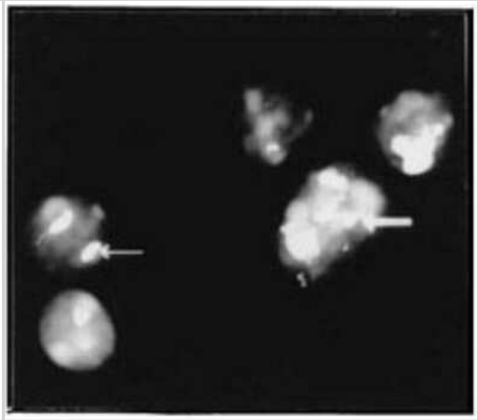
Product Information	
Unit Size	0.1 ml
Concentration	This product is unpurified. The exact concentration of antibody is not quantifiable.
Storage	Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	0.1% Sodium Azide
Isotype	IgG
Purity	Unpurified
Buffer	Whole antisera
Target Molecular Weight	106 kDa

Product Description	
Host	Rabbit
Gene ID	4691
Gene Symbol	NCL
Species	Human, Mouse, Rat, Plant
Immunogen	A fusion protein containing amino acids 284-709 of human Nucleolin. [UniProt# P19338]

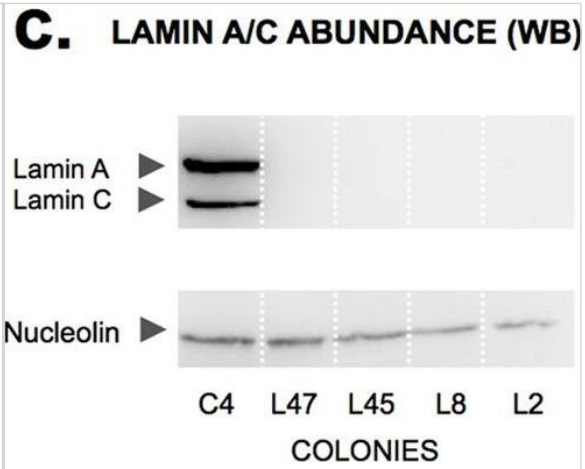
Product Application Details	
Applications	Western Blot, Simple Western, Chromatin Immunoprecipitation, Electron Microscopy, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin, Immunomicroscopy, Chromatin Immunoprecipitation (ChIP), Immunohistochemistry Whole-Mount, Knockdown Validated
Recommended Dilutions	Western Blot 1:4000, Simple Western 1:400, Chromatin Immunoprecipitation reported in scientific literature, Immunohistochemistry 1:50-1:100, Immunocytochemistry/ Immunofluorescence 1:50-1:100, Immunohistochemistry-Paraffin 1:50-1:100, Immunohistochemistry-Frozen reported in scientific literature (PMID 30411850), Immunomicroscopy, Electron Microscopy 1:10-1:500, Immunohistochemistry Whole-Mount reported in scientific literature (PMID 22483616), Chromatin Immunoprecipitation (ChIP), Knockdown Validated
Application Notes	In Western blot, a band at 1~106 kDa represents human Nucleolin. Heat mediated antigen retrieval is recommended for IHC-P. In Simple Western only 10 - 15 uL of the recommended dilution is used per data point. See Simple Western Antibody Database for Simple Western validation: Tested in HeLa lysate 0.5 mg/mL, separated by Size, antibody dilution of 1:400, apparent MW was 134 kDa. Separated by Size-Wes, Sally Sue/Peggy Sue.

Images

Immunocytochemistry/Immunofluorescence: Nucleolin Antibody [NB600-241] - Analysis of Nucleolin MCF-7 breast cancer cells. Image courtsey of product review by Lacey Litchfield.	
Simple Western: Nucleolin Antibody [NB600-241] - Image shows a specific band for Nucleolin in 0.5 mg/mL of HeLa lysate. This experiment was performed under reducing conditions using the 12-230 kDa separation system.	
Immunocytochemistry/Immunofluorescence: Nucleolin Antibody [NB600-241] - Endogenous nuclear nucleolin-COUP-TFII interaction in MCF-7 and T47D cells.NE (200A ug protein) from MCF-7 cells. Immunofluorescent staining of endogenous COUP-TFII (green) and nucleolin (red) in the nuclei (Hoechst, blue) of MCF-7 cells. Merged images are shown at the right. Bar is 10A um. Image collected and cropped by Citeab from the following publication (Identification and characterization of nucleolin as a COUP-TFII coactivator of retinoic acid receptor transcription in breast cancer cells. PLoS One (2012) licensed under a CC-BY license.	
Western Blot: Nucleolin Antibody [NB600-241] - Detection of nucleolin in crude PD31 nuclear extracts.	

Immunocytochemistry/Immunofluorescence: Nucleolin Antibody [NB600-241] - Human prostate epithelial cells stained with anti-nucleolin (red), CFSE (green) retaining cell has increased nucleolin in nucleolus. DAPI (blue). Image from verified customer review.	
Immunohistochemistry: Nucleolin Antibody [NB600-241] - Detection of Nucleolin in human tonsil germinal center.	
Immunocytochemistry/Immunofluorescence: Nucleolin Antibody [NB600-241] - In-situ immunofluorescent staining of PD31 murine pre-B cells.	

Western Blot: Nucleolin Antibody [NB600-241] - HT-LKO cells recapitulate the hallmarks of lamin A/C deficiency.(a) Quantitative PCR shows a >6-fold reduction (expressed as log2 fold change of ddCt value) of LMNA transcripts in different LKO colonies as compared to HT-WT clones; (b) Quantitative immunofluorescence shows a dramatic reduction of A-type lamin levels in HT-LKO colonies, approximating background levels (dotted white line); (c) Western blot for lamin A/C reveals the absence of both proteins in HT-LKO cells; (d) Immunostaining & nuclear counterstaining of HT-LKO cells reveals their aberrant nuclear morphology, virtual absence of lamin A (green) & local depletion of lamin B (red) (arrowheads) as opposed to HT-WT cells; (e) Time-lapse recordings after H2B-GFP transfection illustrate increased nuclear plasticity of HT-LKO vs. HT-WT cells, as evidenced by their larger projected area (Σ Area) & contour changes (Σ Outline) across time; (f) HT-LKO nuclei have a lower average nuclear circularity (dotted white line indicates a circularity of 0.85) & higher nuclear circularity fluctuations (average + 95% confidence interval) than HT-WT cells; The Y-axis has been cropped for clarity; (g) This translates into a significantly larger coefficient of variation (CoV) for the circularity across time ($p < 0.001$). (h) Nuclear ruptures occur more frequently in HT-LKO cells than in HT-WT cells ($p < 0.001$); (i) Representative montages of the NLS channel of a C4 HT-WT & L2 HT-LKO cell, with corresponding NLS/H2B ratio measurements (grey-coded bar plots). The moments of rupture events are indicated as red dots. Bar graphs reflect mean ± standard error (n = number of tracks). Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/27461848), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Publications

Freya Molenberghs, Marlies Verschuuren, Lauran Vandeweyer, Sarah Peeters, Johannes J Bogers, Claudina Perez Novo, Wim Vanden Berghe, Hans De Reu, Nathalie Cools, Mario Schelhaas, Winnok H De Vos Lamin B1 curtails early human papillomavirus infection by safeguarding nuclear compartmentalization and autophagic capacity. Cellular and molecular life sciences : CMLS 2024-03-18 [PMID: 38485766]

Godet AC, Roussel E, David F et al. Long non-coding RNA Neat1 and paraspeckle components are translational regulators in hypoxia eLife 2022-12-22 [PMID: 36546462] (Simple Western, Mouse)

Details:
Dilution used in Simple Western 1:50

Molenberghs F, Verschuuren M, Vandeweyer L et al. Lamin B1 curtails early human papillomavirus infection by safeguarding nuclear compartmentalization and autophagic capacity Research Square 2022-08-05 (WB, Human)

Zhang K, Miorin L, Makio T et al. Nsp1 protein of SARS-CoV-2 disrupts the mRNA export machinery to inhibit host gene expression Science advances 2021-02-01 [PMID: 33547084]

Wu R, Li L, Bai Y et al. The long noncoding RNA LUCAT1 promotes colorectal cancer cell proliferation by antagonizing Nucleolin to regulate MYC expression Cell Death Dis 2020-10-23 [PMID: 33097685] (Chemotaxis)

Podratz JL, Tang JJ, Polzin MJ et al. Mechano growth factor interacts with nucleolin to protect against cisplatin-induced neurotoxicity Exp. Neurol. 2020-06-05 [PMID: 32511954] (ICC/IF, Mouse)

Okur MN, Lee JH, Osmani W et al. Cockayne syndrome group A and B proteins function in rRNA transcription through nucleolin regulation Nucleic Acids Res. 2020-01-23 [PMID: 31970402] (Chemotaxis, Human)

Banerjee S, Aponte-Diaz D, Yeager C et al. Hijacking of Multiple Phospholipid Biosynthetic Pathways and Induction of Membrane Biogenesis by a Picornaviral 3CD Protein bioRxiv 2017-10-29 [PMID: 29782554] (ICC/IF, Human)

Hoffmann F, Umbreit C, Kruger T et al. Identification of Proteomic Markers in Head and Neck Cancer Using MALDI-MS Imaging, LC-MS/MS, and Immunohistochemistry. Prot. Clin. Appl. 2018-11-26 [PMID: 30411850] (IHC-Fr, Human)

Park OH, Park J, Yu M et al. Identification and molecular characterization of cellular factors required for glucocorticoid receptor-mediated mRNA decay. Genes Dev. 2016-09-15 [PMID: 27798850] (WB, Human)

Corne TD, Sieprath T, Vandenbussche J et al. Uncoordinated regulation of focal adhesions and cytoskeletal tension due to the loss of a-type lamins. Cell Adh Migr 2016-10-28 [PMID: 27791462] (WB, Human)

Sieprath T, Corne TD, Nooteboom M et al. Sustained accumulation of prelamin A and depletion of lamin A/C both cause oxidative stress and mitochondrial dysfunction but induce different cell fates. Nucleus 2015-05-21 [PMID: 25996284] (WB)

More publications at http://www.novusbio.com/NB600-241

Procedures

Serum protocol for Nucleolin Antibody (NB600-241)

IHC-FFPE sections

I. Deparaffinization:

A. Treat slides with Xylene: 3 changes for 5 minutes each. Drain slides for 10 seconds between changes.

B. Treat slides with 100% Reagent Alcohol: 3 changes for 5 minutes each. Drain slides for 10 seconds between changes.

II. Quench Endogenous Peroxidase:

A. Place slides in peroxidase quenching solution: 15-30 minutes. To Prepare 200 ml of Quenching Solution: Add 3 ml of 30% Hydrogen Peroxide to 200 ml of Methanol.
Use within 4 hours of preparation

B. Place slides in distilled water: 2 changes for 2 minutes each.

III. Retrieve Epitopes:

A. Preheat Citrate Buffer. Place 200 ml of Citrate Buffer Working Solution into container, cover and place into steamer. Heat to 90-96 degrees Celsius.

B. Place rack of slides into hot Citrate Buffer for 20 minutes. Cover.

C. Carefully remove container with slides from steamer and cool on bench, uncovered, for 20 minutes.

D. Slowly add distilled water to further cool for 5 minutes.

E. Rinse slides with distilled water. 2 changes for 2 minutes each.

IV. Immunostaining Procedure:

A. Remove each slide from rack and circle tissue section with a hydrophobic barrier pen (e.g. Liquid Blocker-Super Pap Pen).

B. Flood slide with Wash Solution. Do not allow tissue sections to dry for the rest of the procedure.

C. Drain wash solution and apply 4 drops of Blocking Reagent to each slide and incubate for 15 minutes.

D. Drain Blocking Reagent (do not wash off the Blocking Reagent), apply 200 ul of Primary Antibody solution to each slide, and incubate for 1 hour.

E. Wash slides with Wash Solution: 3 changes for 5 minutes each.

F. Drain wash solution, apply 4 drops of Secondary antibody to each slide and incubate for 1 hour.

G. Wash slides with Wash Solution: 3 changes for 5 minutes each.

H. Drain wash solution, apply 4 drops of DAB Substrate to each slide and develop for 5-10 minutes. Check development with microscope.

I. Wash slides with Wash Solution: 3 changes for 5 minutes each.

J. Drain wash solution, apply 4 drops of Hematoxylin to each slide and stain for 1-3 minutes. Increase time if darker counterstaining is desired.

K. Wash slides with Wash Solution: 2-3 changes for 2 minutes each.

L. Drain wash solution and apply 4 drops of Bluing Solution to each slide for 1-2 minutes.

M. Rinse slides in distilled water.

N. Soak slides in 70% reagent alcohol: 3 minutes with intermittent agitation.

O. Soak slides in 95% reagent alcohol: 2 changes for 3 minutes each with intermittent agitation.

P. Soak slides in 100% reagent alcohol: 3 changes for 3 minutes each with intermittent agitation. Drain slides for 10 seconds between each change.

Q. Soak slides in Xylene: 3 changes for 3 minutes each with intermittent agitation. Drain slides for 10 seconds between each change.

R. Apply 2-3 drops of non-aqueous mounting media to each slide and mount coverslip.

S. Lay slides on a flat surface to dry prior to viewing under microscope.

NOTES:

-Use treated slides (e.g. HistoBond) to assure adherence of FFPE sections to slide.

-Prior to deparaffinization, heat slides overnight in a 60 degrees Celsius oven.

-All steps in which Xylene is used should be performed in a fume hood.

-For Epitope Retrieval, a microwave or pressure cooker may be substituted for the steamer method. Adjust times as necessary depending on conditions.

-For the initial IHC run with a new primary antibody, test tissues with and without Epitope Retrieval. In some instances, Epitope Retrieval may not be necessary.

-200 ul is the recommended maximum volume to apply to a slide for full coverage. Using more than 200 ul may allow solutions to wick off the slide and create drying artifacts. For small tissue sections less than 200 ul may be used.

-5 minutes of development with DAB Substrate should be sufficient. Do not develop for more than 10 minutes. If 5 minutes of development causes background staining, further dilution of the primary antibody may be necessary.

-Hematoxylin should produce a light nuclear counterstain so as not to obscure the DAB staining. Counterstain for 1-1.5 minutes for nuclear antigens. Counterstain for 2-3 minutes for cytoplasmic and membranous antigens. If darker counterstaining is desired increase time (up to 10 minutes).

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Products Related to NB600-241

HAF008	Goat anti-Rabbit IgG Secondary Antibody [HRP]
NB7160	Goat anti-Rabbit IgG (H+L) Secondary Antibody [HRP]
NBP2-24891	Rabbit IgG Isotype Control
NBP3-21345PEP	Nucleolin Recombinant Protein Antigen

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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