

Datasheet for 600-401-P62**Lamin B1 Antibody****Overview**

Description:	Anti-Lamin B1 (RABBIT) Antibody - 600-401-P62
Item No.:	600-401-P62
Size:	100 µg
Applications:	IHC, WB
Reactivity:	Human, Rat
Host Species:	Rabbit

Product Details

Background:	Lamin-B1 is a protein that in humans is encoded by the LMNB1 gene. The nuclear lamina consists of a two-dimensional matrix of proteins located next to the inner nuclear membrane. The lamin family of proteins make up the matrix and are highly conserved in evolution. During mitosis, the lamina matrix is reversibly disassembled as the lamin proteins are phosphorylated. Lamin proteins are thought to be involved in nuclear stability, chromatin structure and gene expression. Vertebrate lamins consist of two types, A and B. This gene encodes one of the two B type proteins, B1. This antibody is suitable for researchers interested in chromatin antibody research.
Synonyms:	LMNB1, LMN2, LMNB, Lamin B1 antibody
Host Species:	Rabbit
Clonality:	Polyclonal
Format:	IgG

Target Details

Gene Name:	LMNB1
Reactivity:	Human, Rat
Immunogen Type:	Conjugated Peptide
Immunogen:	Lamin B1 affinity purified antibody was prepared from whole rabbit serum produced by repeated immunizations with a synthetic peptide corresponding to a sequence at the C-terminal of human Lamin B1.

Purity/Specificity:	Anti-Lamin B1 antibody is directed against human Lamin B1 protein. The product was affinity purified from monospecific antiserum by immunoaffinity purification. A BLAST analysis was used to suggest reactivity with this protein from human and rat based on homology for the immunogen sequence. Cross reactivity with Lamin B1 from other sources has not been determined.
Relevant Links:	• UniProtKB - P20700 • GeneID - 4001 • NCBI - NP_001172028.11

Application Details

Tested Applications:	IHC, WB
Application Note:	Anti-Lamin B1 is tested for Immunohistochemistry-P, Immunohistochemistry-F, Immunocytochemistry, and Western Blotting. Specific conditions for reactivity should be optimized by the end user. Expect a band approximately ~66.4 kDa corresponding to the appropriate cell lysate or extract. WB tested in Human, Mouse, Rat, IHC-P (heat) and IHC-F in human and rat, ICC in human, and ICC/IF in human.
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
IHC:	0.5-1µg/ml
WB:	0.1-0.5µg/ml

Formulation

Physical State:	Lyophilized
Concentration:	0.5 mg/mL by UV absorbance at 280 nm
Buffer:	0.9mg NaCl, 0.2mg Na ₂ HPO ₄ , 0.05mg NaN ₃
Preservative:	0.05mg Thimerosal
Stabilizer:	5mg BSA
Reconstitution Volume:	100 µL
Reconstitution Buffer:	Restore with deionized water (or equivalent)

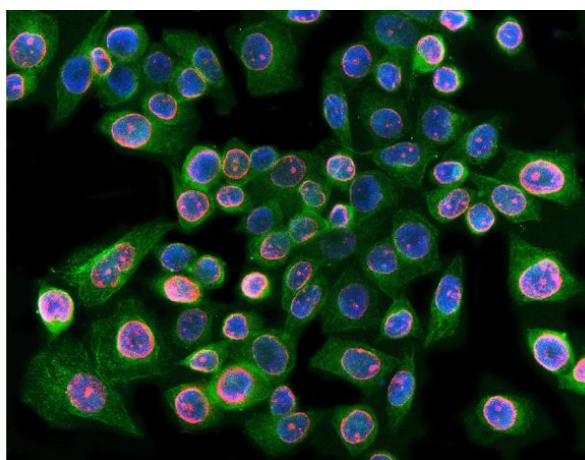
Shipping & Handling

Shipping Condition:	Ambient
----------------------------	---------

Storage Condition: Store vial at 4° C prior to opening. Aliquot contents and freeze at -20° C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use.

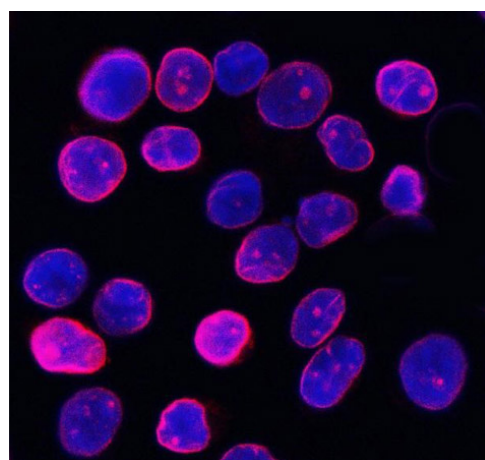
Expiration: Expiration date is one (1) year from date of receipt.

Images



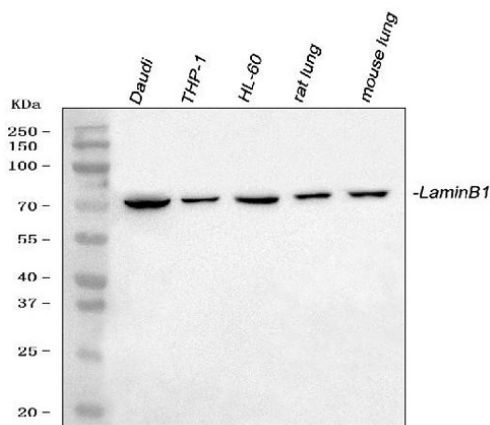
Immunofluorescence Microscopy

Immunofluorescence analysis of Lamin B1 and Tubulin beta using anti-Lamin B1 antibody (p/n 600-401-P62) and an anti-Tubulin beta antibody. Lamin B1 and Tubulin beta were detected in immunocytochemical section of SiHa cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5µg/mL rabbit anti-Lamin B1 antibody and mouse anti-Tubulin beta antibody overnight at 4°C. Cy3 Conjugated Goat Anti-Rabbit IgG and DyLight®488 Conjugated Goat Anti-Mouse IgG were used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.



Immunofluorescence Microscopy

Immunofluorescence analysis of Lamin B1 using anti-Lamin B1 antibody. Lamin B1 was detected in an immunocytochemical section of SiHa cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 µg/mL rabbit anti-Lamin B1 Antibody overnight at 4°C. Cy3 Conjugated Goat Anti-Rabbit IgG was used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.



Western Blot

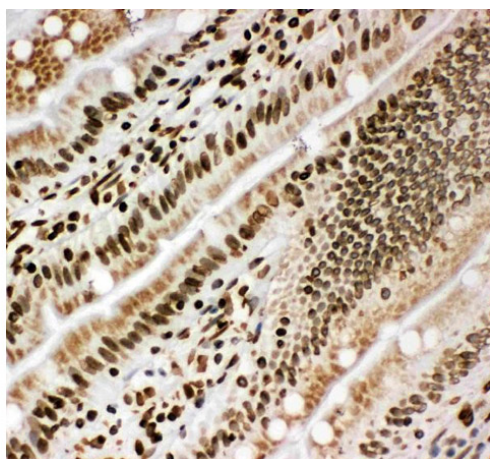
Western Blot analysis of Lamin B1 using anti-Lamin B1 antibody.

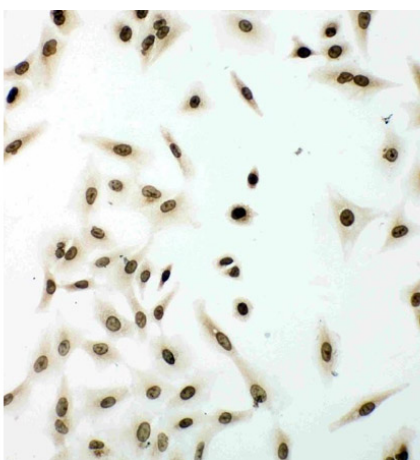
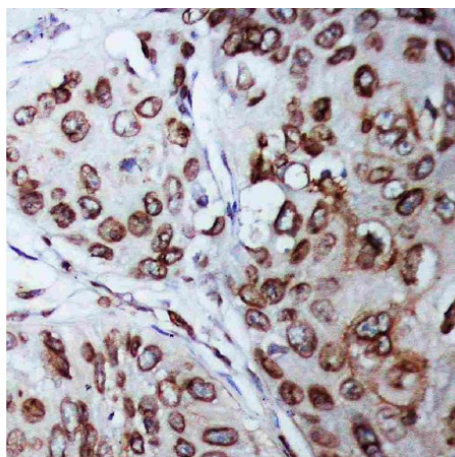
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30µg of sample under reducing conditions.

Lane 1: human Daudi whole cell lysates, Lane 2: human THP-1 whole cell lysates, Lane 3: human HL-60 whole cell lysates, Lane 4: rat lung tissue lysates, Lane 5: mouse lung tissue lysates. After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with affinity purified rabbit anti-Lamin B1 polyclonal antibody at 0.5 µg/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 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for Lamin B1 at approximately 70 kDa. The expected band size for Lamin B1 is at 70 kDa.

Immunohistochemistry

Immunohistochemistry analysis of Lamin B1 using anti-Lamin B1 antibody. Lamin B1 was detected in a frozen section of Rat Intestine tissue. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml rabbit anti-Lamin B1 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.





Immunohistochemistry

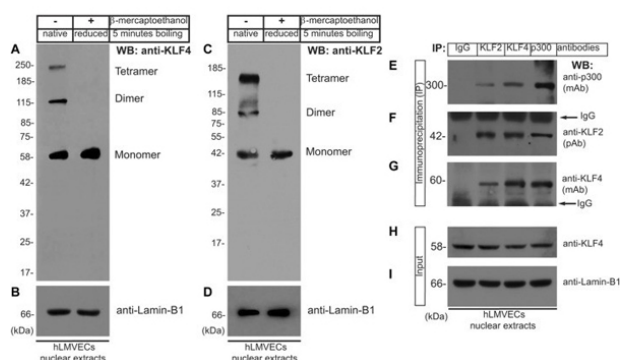
Immunohistochemistry analysis of Lamin B1 using anti-Lamin B1 antibody. Lamin B1 was detected in a paraffin-embedded section of Human Mammary Cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml rabbit anti-Lamin B1 Antibody overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit with DAB as the chromogen.

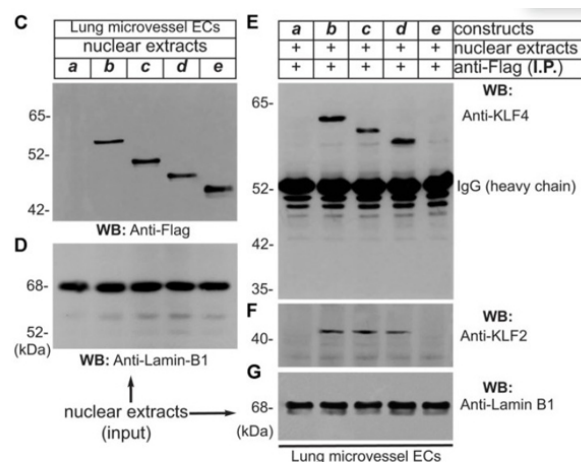
Immunocytochemistry

Immunocytochemistry analysis of Lamin B1 using anti-Lamin B1 antibody. Lamin B1 was detected in an immunocytochemical section of A549 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 1 µg/ml rabbit anti-Lamin B1 Antibody overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The section was developed using Streptavidin-Biotin-Complex (SABC) with DAB as the chromogen.

Western Blot

Oligomeric KLF4 and KLF2 protein complexes in quiescent ECs. (A,C) Nuclear extracts prepared from human lung microvascular endothelial cells (hLMVECs) were boiled in native or reducing sample buffers, resolved by 9% SDS-PAGE. The presence of mono-, di-, and tetrameric anti-KLF4 and anti-KLF2 polypeptide species in native SDS-PAGE were analyzed by indicated antibodies. (B,D) Equal loading were analyzed by anti-Lamin-B1 antibody. (E–G) hLMVECs extracts were subjected to immunoprecipitation (IP) with IgG (control), anti-KLF2, anti-KLF4, and anti-p300 antibodies, and analyzed by WB with indicated antibodies. (H,I) Nuclear lysates (input) were analyzed by WB with anti-KLF4 and anti-Lamin-B1 antibodies. Molecular weights are given in kiloDalton (kDa). This experiment was repeated at least three times. Fig 1. PMID: 36425528





Western Blot

(c) KLF4-Δ86 construct (lacking 85 amino acid residues from the N-terminus) fused in-frame with Flag-epitope tag; (d) KLF4-Δ96 construct (lacking 95 amino acid residues from the N-terminus) fused in-frame with Flag-epitope tag; (e) KLF4-Δ116 construct (lacking 115 amino acid residues from the N-terminus) fused in-frame with Flag-epitope tag as shown. (B) Strategy and timeline of human lung microvessel endothelial cell (hLMVECs) infection/transfection experiments. (C) Efficiency of retrovirus infection of hLMVECs was determined by WB with anti-Flag antibody. (D) Equal loading across the lanes were judged by anti-Lamin-B1 antibody. (E) Nuclear cell extracts were immunoprecipitated with an anti-Flag monoclonal antibody (mAb) and analyzed by WB with anti-Flag mAb. (F) The KLF4-wild-type, KLF4-Δ86, and KLF4-Δ96 immunoprecipitated KLF2 polypeptide species, while KLF4-Δ116 did not. (G) Equal levels checked with anti-Lamin-B1. These experiments were repeated at least 3 times. Fig 2. PMID: 36425528

References

- Mastej V et al. A requirement for Krüppel Like Factor-4 in the maintenance of endothelial cell quiescence. *Front Cell Dev Biol.* (2022)

Disclaimer

This product is for research use only and is not intended for therapeutic or diagnostic applications. Please contact a technical service representative for more information. All products of animal origin manufactured by Rockland Immunochemicals are derived from starting materials of North American origin. Collection was performed in United States Department of Agriculture (USDA) inspected facilities and all materials have been inspected and certified to be free of disease and suitable for exportation. All properties listed are typical characteristics and are not specifications. All suggestions and data are offered in good faith but without guarantee as conditions and methods of use of our products are beyond our control. All claims must be made within 30 days following the date of delivery. The prospective user must determine the suitability of our materials before adopting them on a commercial scale. Suggested uses of our products are not recommendations to use our products in violation of any patent or as a license under any patent of Rockland Immunochemicals, Inc. If you require a commercial license to use this material and do not have one, then return this material, unopened to: Rockland Inc., P.O. BOX 5199, Limerick, Pennsylvania, USA.