abcam

Product datasheet

# Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker ab133741

KO VALIDATED Recombinant RabMAb

14 Abreviews 161 References 17 Images

## Overview

| | |
|---|---|
| **Product name** | Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker |
| **Description** | Rabbit monoclonal [EPR8985(B)] to Lamin B1 - Nuclear Envelope Marker |
| **Host species** | Rabbit |
| **Tested applications** | **Suitable for:** IP, ICC/IF, WB, IHC-P |
| **Species reactivity** | **Reacts with:** Mouse, Rat, Human |
| **Immunogen** | Synthetic peptide. within Human Lamin B1 aa 500 to the C-terminus. The exact sequence is proprietary.<br>Database link: **P20700** |
| **Positive control** | WB: Hap1, HeLa, Jurkat, Molt4, Y79, Caco 2, C6, Raw264.7, PC-12 and NIH/3T3 cell lysates. Mouse brain,heart, kidney and spleen; and Rat brain, heart and spleen lysates. IHC-P: Human colon, liver and transitional cell carcinoma of the bladder tissues. ICC/IF: Ramos cells, HAP1-LMNB1 cells. |
| **General notes** | This product is a recombinant monoclonal antibody, which offers several advantages including:<br>- High batch-to-batch consistency and reproducibility<br>- Improved sensitivity and specificity<br>- Long-term security of supply<br>- Animal-free production<br>For more information **see here**.<br>Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**. |

## Properties

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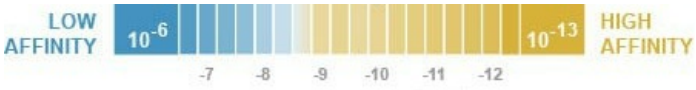
|---|---|
| **Form** | Liquid |
| **Storage instructions** | Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle. |
| **Dissociation constant ($K_D$)** | $K_D = 1.95 \times 10^{-10}$ M |




Learn more about $K_D$

| | |
|---|---|
| **Storage buffer** | pH: 7.20<br>Preservative: 0.01% Sodium azide<br>Constituents: 59% PBS, 40% Glycerol, 0.05% BSA |
| **Purity** | Protein A purified |
| **Clonality** | Monoclonal |
| **Clone number** | EPR8985(B) |
| **Isotype** | IgG |

## Applications

**The Abpromise guarantee** Our **Abpromise guarantee** covers the use of ab133741 in the following tested applications.

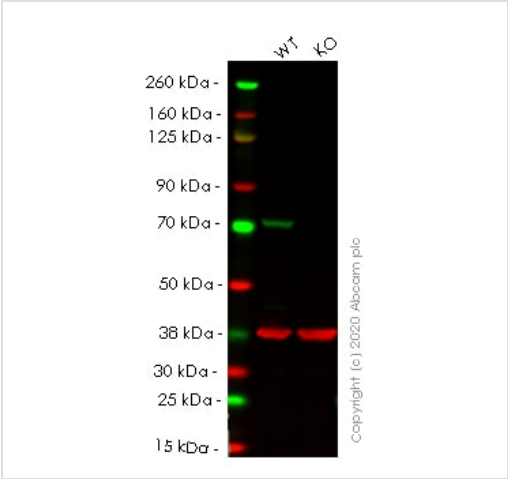
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

| Application | Abreviews | Notes |
|---|---|---|
| **IP** | | 1/20. |
| **ICC/IF** | ★★★★☆ **(4)** | Use a concentration of 1 µg/ml. |
| **WB** | ★★★★★ **(9)** | 1/1000 - 1/10000. Detects a band of approximately 70 kDa (predicted molecular weight: 66 kDa). |
| **IHC-P** | ★★★★★ **(1)** | 1/300. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See **IHC antigen retrieval protocols**. |

## Target

| | |
|---|---|
| **Function** | Lamins are components of the nuclear lamina, a fibrous layer on the nucleoplasmic side of the inner nuclear membrane, which is thought to provide a framework for the nuclear envelope and may also interact with chromatin. |
| **Involvement in disease** | Defects in LMNB1 are the cause of leukodystrophy demyelinating autosomal dominant adult-onset (ADLD) [MIM:169500]. ADLD is a slowly progressive and fatal demyelinating leukodystrophy, presenting in the fourth or fifth decade of life. Clinically characterized by early autonomic abnormalities, pyramidal and cerebellar dysfunction, and symmetric demyelination of the CNS. It differs from multiple sclerosis and other demyelinating disorders in that neuropathology shows preservation of oligodendroglia in the presence of subtotal demyelination and lack of astrogliosis. |
| **Sequence similarities** | Belongs to the intermediate filament family. |
| **Post-translational modifications** | B-type lamins undergo a series of modifications, such as farnesylation and phosphorylation. Increased phosphorylation of the lamins occurs before envelope disintegration and probably plays a role in regulating lamin associations. |
| **Cellular localization** | Nucleus inner membrane. |

## Images




Western blot - Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

**All lanes :** Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741) at 1/1000 dilution

**Lane 1 :** Wild-type HeLa cell lysate
**Lane 2 :** LMNB1 knockout HeLa cell lysate

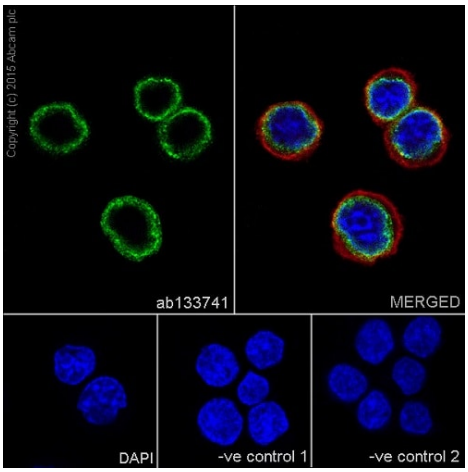
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**Predicted band size:** 66 kDa
**Observed band size:** 66-70 kDa

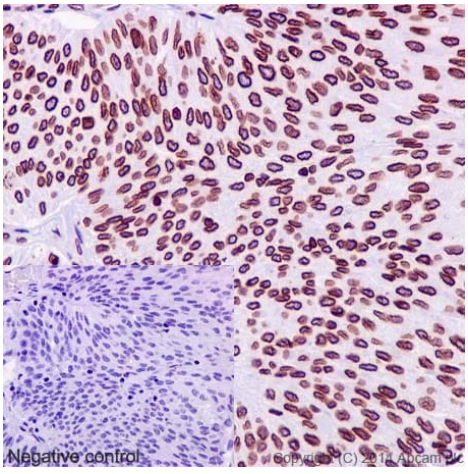
**Lanes 1- 2:** Merged signal (red and green). Green - ab133741 observed at 66-70 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

ab133741 was shown to react with LMNB1 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line **ab255404** (knockout cell lysate **ab263825**) was used. Wild-type HeLa and LMNB1 knockout HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab133741 and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye®800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye®680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



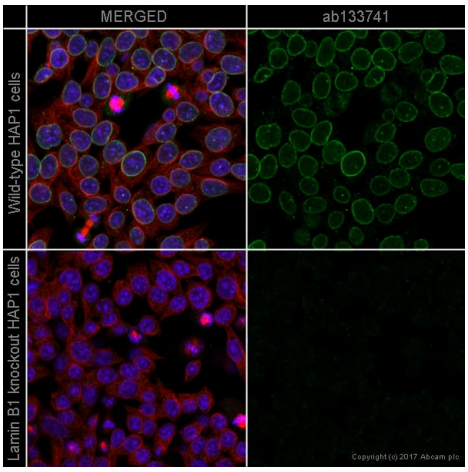

Immunocytochemistry/ Immunofluorescence - Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized Ramos (Human Burkitt's lymphoma cell line) cells labeling Lamin B1 with ab133741 at 1/100 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/200 dilution (green). Nuclear envelope staining on Ramos cell line is observed. The nuclear counter stain is DAPI (blue). Tubulin is detected with **ab7291** (anti-Tubulin mouse mAb) at 1/500 dilution and **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).
The negative controls are as follows:
-ve control 1: ab133741 at 1/100 dilution followed by **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
-ve control 2: **ab7291** (anti-Tubulin mouse mAb) at 1/500 dilution followed by **ab150077** (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/200 dilution.




Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

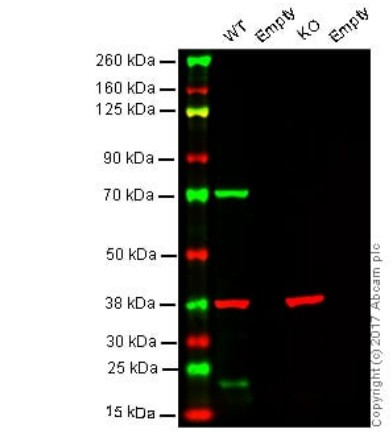
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human transitional cell carcinoma of the bladder tissue labeling Lamin B1 with purified ab133741 at 1/300. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. A prediluted HRP-polymer conjugated anti-rabbit IgG was used as the secondary antibody. Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.




Immunocytochemistry/ Immunofluorescence - Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

ab133741 staining Lamin B1 in wild-type HAP1 cells (top panel) and Lamin B1 knockout HAP1 cells (bottom panel). The cells were fixed with 100% methanol (5min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab133741 at 1µg/ml dilution and **ab195889** at 1/250 dilution (shown in pseudo-color red) overnight at +4°C. The cells were then incubated with **ab150081** (Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488)) at 1/1000 dilution for 1 hour. Nuclear DNA was labelled in blue with DAPI.

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).




Western blot - Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

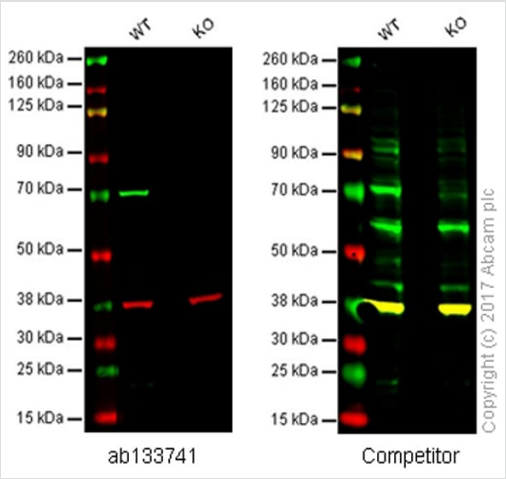
**Lanes 1 & 3 :** Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741) at 1/1000 dilution

**Lane 1 :** Wild-type HAP1 whole cell lysate at 20 µg
**Lanes 2 & 4 :** Empty
**Lane 3 :** LMNB1 knockout HAP1 whole cell lysate at 20 µg

**Predicted band size:** 66 kDa

**Lanes 1 - 4:** Merged signal (red and green). Green - ab133741 observed at 70 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab133741 was shown to specifically react with Lamin B1 in wild type HAP1 cells. No band was observed when knockout samples were used. Wild-type and Lamin B1 knockout samples were subjected to SDS-PAGE. Ab133741 and **ab8245** (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/10000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed **ab216773** and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed **ab216776** secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.




Western blot - Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

**Lanes 1 & 3 :** Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

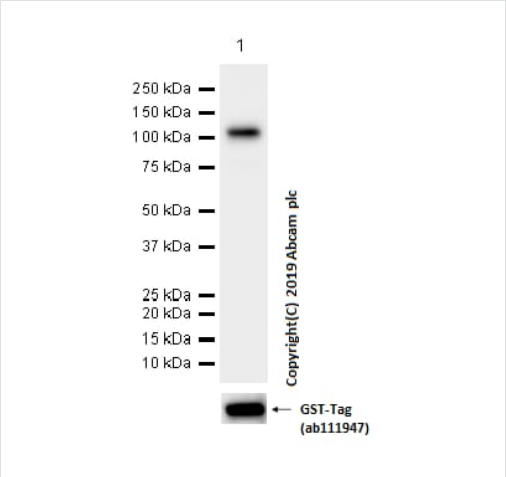
**Lane 1 :** Wild-type HAP1 whole cell lysate at 20 µg
**Lane 2 :** Empty
**Lane 3 :** Lamin B1 knockout HAP1 cell lysate at 20 µg

**Predicted band size:** 66 kDa

**Lanes 1 - 3:** Merged signal (red and green).

Green - Target observed at 70 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

This western blot image is a comparison between ab133741 and a competitor's discontinued goat polyclonal antibody.




Western blot - Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

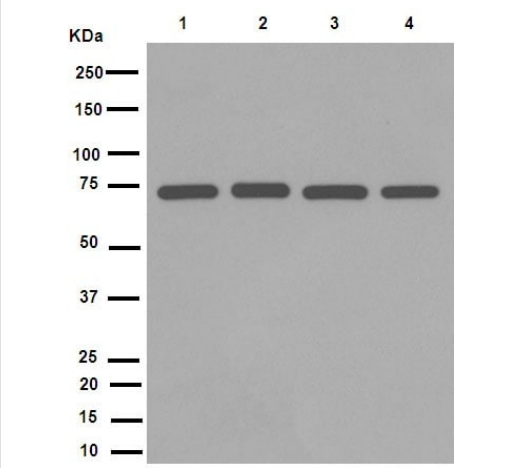
Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741) at 1/1000 dilution + GST-tagged Recombinant Human Lamin B1 protein (aa 1 to 586) at 0.015 µg with 5% NFDM/TBST

**Secondary**
Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

**Predicted band size:** 66 kDa
**Observed band size:** 100 kDa

**Exposure time:** 1 second

Recombinant Human Lamin B1 protein (**ab114163**)




Western blot - Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

**All lanes :** Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741) at 1/50000 dilution (purified)

**Lane 1 :** Jurkat (Human T cell leukemia cells from peripheral blood) cell lysate
**Lane 2 :** Molt-4 (Human lymphoblastic leukemia cell line) cell lysate
**Lane 3 :** Y79 (Human retinoblastoma cell line) cell lysate
**Lane 4 :** Caco-2 (Human colorectal adenocarcinoma cells) cell lysate

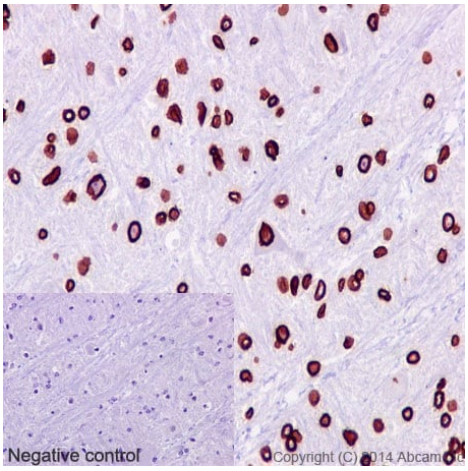
Lysates/proteins at 20 µg per lane.

**Secondary**
**All lanes :** HRP goat anti-rabbit (H+L) at 1/1000 dilution

**Predicted band size:** 66 kDa
**Observed band size:** 70 kDa

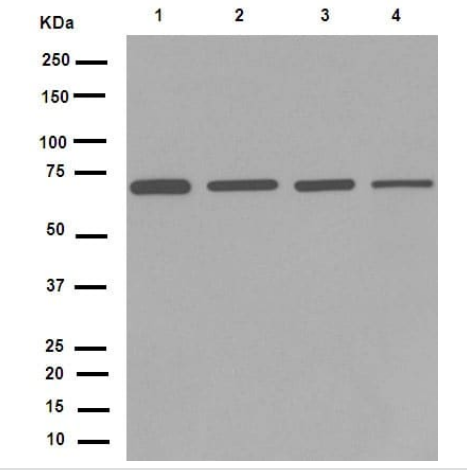
Blocking buffer: 5% NFDM/TBST

Dilution buffer: 5% NFDM/TBST




Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

Immunohistochemical staining of paraffin embedded Mouse Cerebral cortex with purified ab133741 at a working dilution of 1/300. The secondary antibody used is a HRP polymer for rabbit IgG. Nuclear envelope staining on neuron cells of Cerebral cortex tissue is observed. The sample is counter-stained with hematoxylin. Antigen retrieval was perfomed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.




Western blot - Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

**All lanes :** Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741) at 1/50000 dilution (purified)

**Lane 1 :** C6 (Rat glial tumor cells) cell lysate
**Lane 2 :** PC12 (Rat adrenal gland pheochromocytoma) cell lysate
**Lane 3 :** NIH/3T3 (Mouse embyro fibroblast cells) cell lysate
**Lane 4 :** RAW264.7 (Mouse macrophage cells transformed with Abelson murine leukemia virus) cell lysate

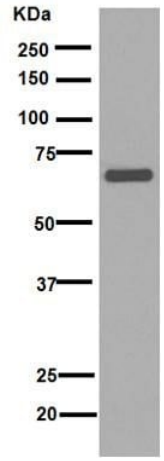
Lysates/proteins at 20 µg per lane.

**Secondary**
**All lanes :** HRP goat anti-rabbit (H+L) at 1/1000 dilution

**Predicted band size:** 66 kDa
**Observed band size:** 70 kDa

Blocking buffer: 5% NFDM/TBST

Dilution buffer: 5% NFDM/TBST




Western blot - Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741) at 1/10000 dilution (purified) + Jurkat (Human T cell leukemia cells from peripheral blood) cell lysate at 10 µg

**Secondary**

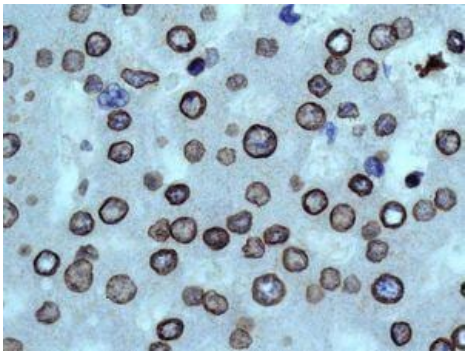
Peroxidase conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

**Predicted band size:** 66 kDa

**Observed band size:** 70 kDa

Blocking buffer and concentration: 5% NFDM/TBST.

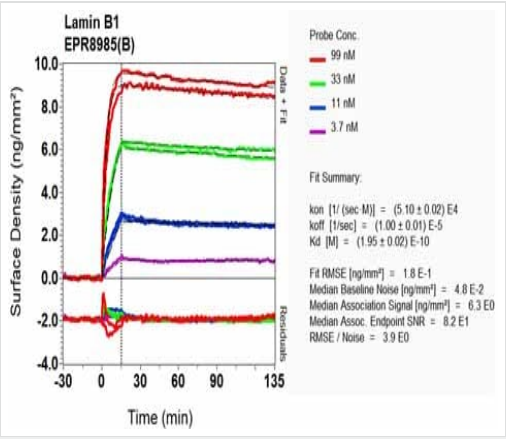
Diluting buffer and concentration: 5% NFDM /TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human liver tissue labelling Lamin B1 with unpurified ab133741 at 1/250.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

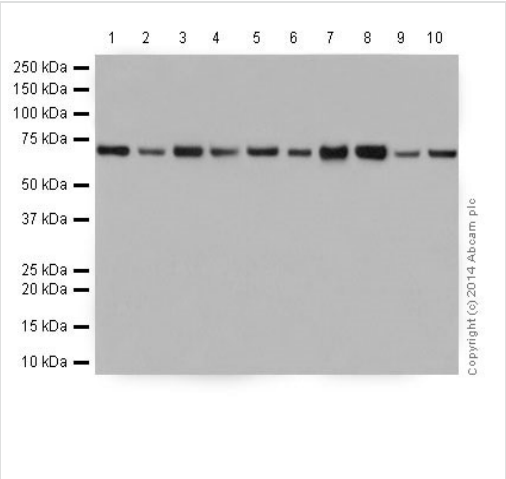



OI-RD Scanning - Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

Equilibrium disassociation constant ($K_D$)

Learn more about $K_D$

**Click here to learn more about $K_D$**




Western blot - Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

**All lanes :** Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741) at 1/10000 dilution

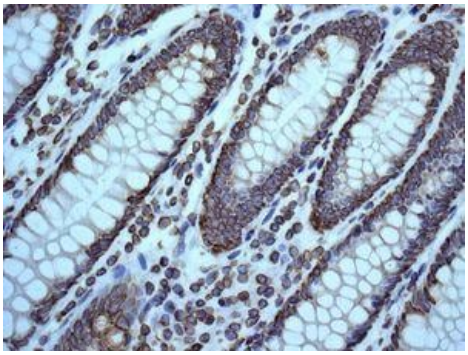
**Lane 1 :** Mouse brain lysates
**Lane 2 :** Mouse heart lysates
**Lane 3 :** Mouse kidney lysates
**Lane 4 :** Mouse spleen lysates
**Lane 5 :** Rat brain lysates
**Lane 6 :** Rat heart lysates
**Lane 7 :** Rat spleen lysates
**Lane 8 :** C6 (Rat glial tumor cells) whole cell lysates
**Lane 9 :** PC-12 (Rat adrenal gland pheochromocytoma) whole cell lysates
**Lane 10 :** NIH/3T3 (Mouse embryo fibroblast cells)

Lysates/proteins at 10 µg per lane.

**Secondary**
**All lanes :** Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

**Predicted band size:** 66 kDa
**Observed band size:** 70 kDa

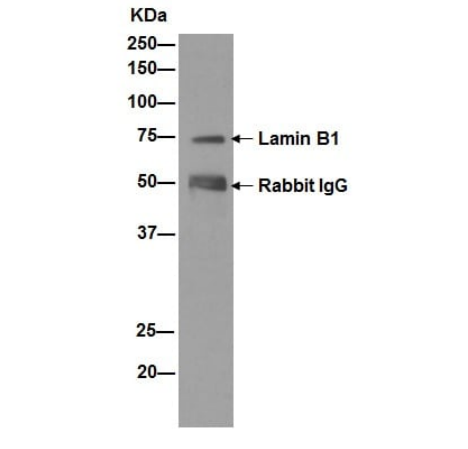
Blocking and Diluting buffer and concentration: 5% NFDM/TBST



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human colon tissue labelling Lamin B1 with unpurified ab133741 at 1/250.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

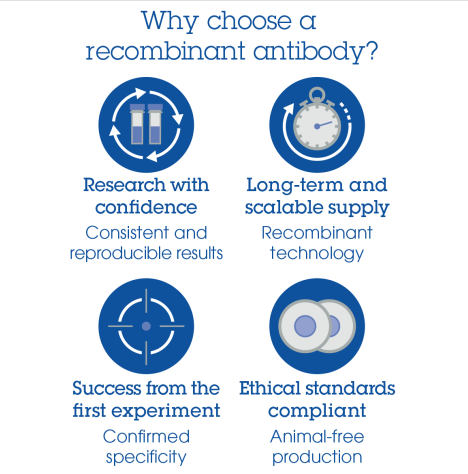



Immunoprecipitation - Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

ab133741 (purified) at 1/20 immunoprecipitating Lamin B1 in Jurkat cells (Lane 1). For western blotting, ab133741 was used at 1/1000 dilution and an HRP-conjugated goat anti-rabbit IgG was used as the secondary antibody (1/1500).

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.




Anti-Lamin B1 antibody [EPR8985(B)] - Nuclear Envelope Marker (ab133741)

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