abcam

Product datasheet

Anti-RPL5 antibody ab86863

11 References 4 Images

Overview

Product name Anti-RPL5 antibody

Description Rabbit polyclonal to RPL5

Host species Rabbit

Tested applications Suitable for: IP, IHC-P, WB, ICC/IF

Species reactivity Reacts with: Human

Immunogen Synthetic peptide corresponding to Human RPL5 aa 200 to the C-terminus conjugated to keyhole

limpet haemocyanin.

(Peptide available as ab97494)

Positive control This antibody gave a positive signal Human fetal liver tissue lysate as well as the following whole

cell lysates: HepG2; MEL-1; Jurkat.

General notes

The Life Science industry has been in the grips of a reproducibility crisis for a number of years.

Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -

80°C. Avoid freeze / thaw cycle.

Storage buffer pH: 7.40

Preservative: 0.02% Sodium azide

Constituent: PBS

Batches of this product that have a concentration < 1mg/ml may have BSA added as a stabilising

agent. If you would like information about the formulation of a specific lot, please contact our

scientific support team who will be happy to help.

Purity Immunogen affinity purified

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Clonality Polyclonal

Isotype IgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab86863 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		Use a concentration of 5 µg/ml.
IHC-P		Use a concentration of 1 µg/ml. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
WB		Use a concentration of 1 µg/ml. Detects a band of approximately 34 kDa (predicted molecular weight: 34 kDa).
ICC/IF		Use a concentration of 5 µg/ml.

Target

Function Required for rRNA maturation and formation of the 60S ribosomal subunits. This protein binds 5S

RNA.

Involvement in disease Defects in RPL5 are the cause of Diamond-Blackfan anemia type 6 (DBA6) [MIM:612561]. DBA6

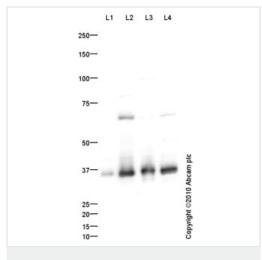
is a form of Diamond-Blackfan anemia, a congenital non-regenerative hypoplastic anemia that usually presents early in infancy. Diamond-Blackfan anemia is characterized by a moderate to severe macrocytic anemia, erythroblastopenia, and an increased risk of malignancy. 30 to 40% of Diamond-Blackfan anemia patients present with short stature and congenital anomalies, the most frequent being craniofacial (Pierre-Robin syndrome and cleft palate), thumb and urogenital

anomalies.

Sequence similaritiesBelongs to the ribosomal protein L18P family.

Cellular localization Cytoplasm. Nucleus > nucleolus.

Images



Western blot - Anti-RPL5 antibody (ab86863)

All lanes: Anti-RPL5 antibody (ab86863) at 1 µg/ml

Lane 1: Liver (Human)Tissue Lysate - Fetal tissue

Lane 2 : HepG2 (Human hepatocellular liver carcinoma cell line)

Whole Cell Lysate

Lane 3: MEL-1 (Human embryonic stem cell, male cell line) Whole

Cell Lysate (<u>ab27198</u>)

Lane 4: Jurkat (Human T cell lymphoblast-like cell line) Whole Cell

Lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat polyclonal to Rabbit lgG - H&L - Pre-Adsorbed (HRP) at 1/3000 dilution

Developed using the ECL technique.

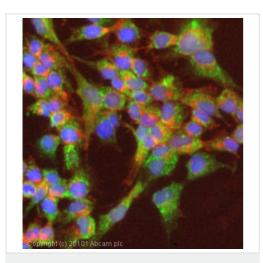
Performed under reducing conditions.

Predicted band size: 34 kDa **Observed band size:** 34 kDa

Additional bands at: 68 kDa. We are unsure as to the identity of

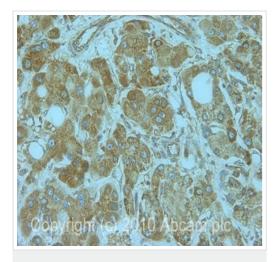
these extra bands.

Exposure time: 1 minute



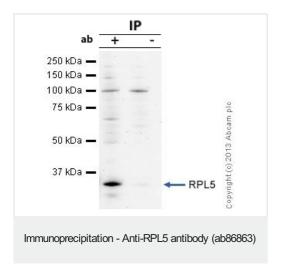
Immunocytochemistry/ Immunofluorescence - Anti-RPL5 antibody (ab86863)

ICC/IF image of ab86863 stained HepG2 cells. The cells were 4% PFA fixed (10 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab86863, 5 μ g/ml) overnight at +4°C. The secondary antibody (green) was Alexa Fluor® 488 goat anti-rabbit lgG (H+L) used at a 1/1000 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43 μ M. This antibody also gave a positive result in 4% PFA fixed (10 min) HeLa cells at 5 μ g/ml, and in 100% methanol fixed (5 min) HeLa, HepG2 and MCF7 cells at 5 μ g/ml



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RPL5 antibody (ab86863)

IHC image of RPL5 staining in human liver carcinoma formalin fixed paraffin embedded tissue section, performed on a Leica BondTM system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab86863, 1µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.



RPL5 was immunoprecipitated using 0.5mg HepG2 whole cell extract, 5µg of Rabbit polyclonal to RPL5 and 50µl of protein G magnetic beads (+). No antibody was added to the control (-). The antibody was incubated under agitation with Protein G beads for 10min, HepG2 whole cell extract lysate diluted in RIPA buffer was added to each sample and incubated for a further 10min under agitation.

Proteins were eluted by addition of $40\mu l$ SDS loading buffer and incubated for 10min at $70^{o}C$; $10\mu l$ of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab86863.

Secondary: Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (ab99697).

Band: 34kDa, non specific band - 68kDa as present in test (lane 1) and control (lane 2); 68kDa: We are unsure as to the identity of this extra band; RPL5

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