abcam

Product datasheet

Anti-Lin28A antibody ab63740

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Overview

Product name Anti-Lin28A antibody

Description Rabbit polyclonal to Lin28A

Host species Rabbit

Specificity From Jan 2024, QC testing of replenishment batches of this polyclonal changed. All tested and

expected application and reactive species combinations are still covered by our Abcam product promise. However, we no longer test all applications. For more information on a specific batch,

please contact our Scientific Support who will be happy to help.

Tested applications Suitable for: IP, Sandwich ELISA, ICC/IF, WB

Species reactivity Reacts with: Mouse, Human

Predicted to work with: Rat, Horse, Pig, Chimpanzee

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: HUES-7, MEL-1, F9. ES-E14TG2a, Mouse EG. ICC/IF: mES cells.

General notesThe 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

Clonality Polyclonal

Isotype IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab63740 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.
Sandwich ELISA		Use a concentration of 0.1 µg/ml. Can be paired for Sandwich ELISA with Mouse monoclonal [6D1F9] to Lin28A (ab76369). For sandwich ELISA, use this antibody as Detection at 0.1 µg/ml with Mouse monoclonal [6D1F9] to Lin28 (ab76369) as Capture.
ICC/IF	★★★★ <u>(2)</u>	Use a concentration of 1 µg/ml.
WB	**** <u>(2)</u>	Use a concentration of 1 µg/ml. Detects a band of approximately 30 kDa (predicted molecular weight: 23 kDa).

Target

Function

Acts as a 'translational enhancer', driving specific mRNAs to polysomes and thus increasing the efficiency of protein synthesis. Its association with the translational machinery and target mRNAs results in an increased number of initiation events per molecule of mRNA and, indirectly, in stabilizing the mRNAs. Binds IGF2 mRNA, MYOD1 mRNA, ARBP/36B4 ribosomal protein mRNA and its own mRNA. Essential for skeletal muscle differentiation program through the translational up-regulation of IGF2 expression (By similarity). Acts as a suppressor of microRNA (miRNA) biogenesis by specifically binding the precursor let-7 (pre-let-7), a miRNA precursor. Acts by binding pre-let-7 and recruiting ZCCHC11/TUT4 uridylytransferase, leading to the terminal uridylation of pre-let-7. Uridylated pre-let-7 miRNAs fail to be processed by Dicer and undergo degradation. Degradation of pre-let-7 in embryonic stem (ES) cells contributes to the maintenance of ES cells. In contrast, LIN28A down-regulation in neural stem cells by miR-125, allows the processing of pre-let-7. Specifically recognizes the 5'-GGAG-3' motif in the terminal loop of pre-let-7. Also recognizes and binds non pre-let-7 pre-miRNAs that contain the 5'-GGAG-3' motif in the terminal loop, leading to their terminal uridylation and subsequent degradation.

Tissue specificity

Expressed in embryonic stem cells (ES cells), placenta and testis.

Sequence similarities

Belongs to the lin-28 family.

Contains 2 CCHC-type zinc fingers.
Contains 1 CSD (cold-shock) domain.

Developmental stage

Expressed in fetal liver. Expression decreases during differentiation of ES cells or upon induction

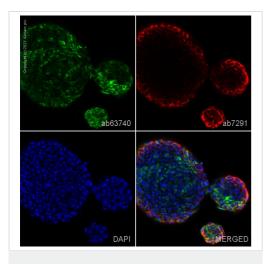
of neuronal differentiation by retinoic acid.

Domain

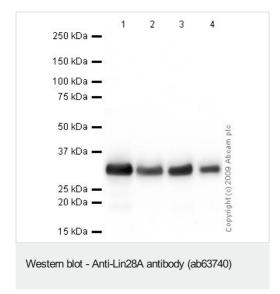
The CSD domain is required for function in muscle differentiation.

Cytoplasm. Nucleus > nucleolus. Nucleolar localization observed in 10-15% of the nuclei in differentiated myotubes (By similarity). Shuttles between the cytoplasm and the nucleus. Localizes to cytoplasmic processing bodies and stress granules.

Images



Immunocytochemistry/ Immunofluorescence - Anti-Lin28A antibody (ab63740)



ab63740 staining Lin28A in mES cells. The cells were fixed with 100% methanol (5 min), permeabilized with 0.1% PBS-Tween 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 overnight at 4°C with ab63740 at 1µg/ml and ab7291, Mouse monoclonal [DM1A] to alpha Tubulin - Loading Control. Cells were then incubated with ab150081, Goat polyclonal Secondary Antibody to Rabbit lgG - H&L (Alexa Fluor® 488), pre-adsorbed at 1/1000 dilution (shown in green) and ab150120, Goat polyclonal Secondary Antibody to Mouse lgG - H&L (Alexa Fluor® 594), pre-adsorbed at 1/1000 dilution (shown in pseudocolour red). Nuclear DNA was labelled with DAPI (shown in blue).

Image was acquired with a confocal microscope (Leica-Microsystems TCS SP8) and a single confocal section is shown.

All lanes: Anti-Lin28A antibody (ab63740) at 1 µg/ml

Lane 1 : MEL-1 (Human embryonic stem cell, male cell line) Whole Cell Lysate (ab27198)

Lane 2: F9 (Mouse embryonic carcinoma cell line) Whole Cell Lysate (ab27193)

Lane 3 : E14Tg2a (Mouse embryonic stem cell line) Whole Cell Lysate

Lane 4 : Mouse EG (TMAS Embryonic Germ Cells) 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

Performed under reducing conditions.

Predicted band size: 23 kDa **Observed band size:** 30 kDa

The 30 kDa band observed is comparable to molecular weights seen with other commercially available antibodies to Lin28A.

1 2 3
250 kDa —
150 kDa —
150 kDa —
75 kDa —
50 kDa —
37 kDa —
25 kDa —
20 kDa —
15 kDa —
10 kDa —
15 kDa —
10 kDa —

10 kDa —

Western blot - Anti-Lin28A antibody (ab63740)

All lanes: Anti-Lin28A antibody (ab63740) at 1 µg/ml

Lane 1 : HUES7 (Human embryonic stem cell line) Whole Cell Lysate

Lane 2 : F9 (Mouse embryonic carcinoma cell line) Whole Cell

Lane 3 : E14Tg2a (Mouse embryonic stem 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 1/50000 dilution

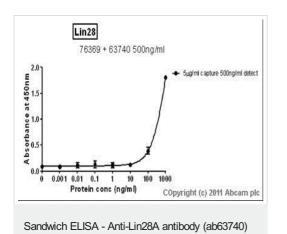
Developed using the ECL technique.

Performed under reducing conditions.

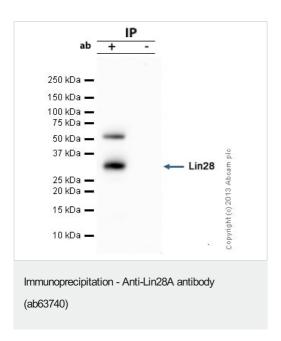
Predicted band size: 23 kDa
Observed band size: 30 kDa

Exposure time: 90 seconds

This blot was produced using a 4-12% Bis-tris gel under the MES buffer system. The gel was run at 200V for 35 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 2% Bovine Serum Albumin before being incubated with ab63740 overnight at 4°C. Antibody binding was detected using an anti-rabbit antibody conjugated to HRP, and visualised using ECL development solution ab133406.



Standard Curve for Lin28A (Analyte: (ab89225) Lin28 protein (His tag) (ab89225)); dilution range 1 pg/ml to 1 ug/ml using Capture Antibody (ab76369) Mouse monoclonal [6D1F9] to Lin28A (ab76369) at 0.2 ug/ml and Detector Antibody (ab63740) Rabbit polyclonal to Lin28A (ab63740) at 0.1 ug/ml.



Lin28A was immunoprecipitated using 0.5mg E14Tg2a whole cell extract, 5µg of Rabbit polyclonal to Lin28A 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, E14Tg2a 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 40μ I SDS loading buffer and incubated for 10min at 70oC; 10μ I of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab63740.

Secondary: Clean blot (HRP conjugate) at 1/1000 dilution.

Band: 30kDa: Lin28A.

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