abcam

Product datasheet

Anti-hnRNP A2B1 antibody ab31645

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Overview

Product name	Anti-hnRNP A2B1 antibody
Description	Rabbit polyclonal to hnRNP A2B1
Host species	Rabbit
Specificity	The peptide used as the immunogen for this antibody is found within isoform A2 and B1 of the protein and should thus recognize a doublet around 35/37 kDa. 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:** IHC-P, WB, ICC, IP
Species reactivity	**Reacts with:** Human **Predicted to work with:** Mouse, Rat, Chicken, Dog, Xenopus laevis
Immunogen	Synthetic peptide corresponding to Human hnRNP A2B1 aa 50-150. (Peptide available as **ab31644**)
Positive control	WB: A549, HEK293T, HeLa, NIH 3T3, and MEF1 cell lysates, and mouse testis tissue lysate. IHC-P: Human colon tissue. ICC: HeLa cells.
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
Clonality	Polyclonal
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab31645 in the following tested applications.

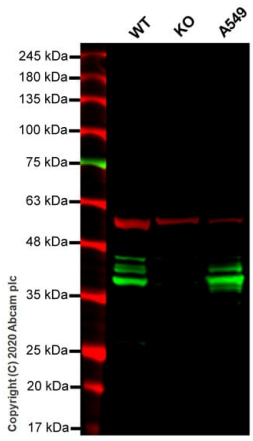
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IHC-P		Use a concentration of 1 µg/ml. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
WB	★★★★★ (2)	Use a concentration of 1 - 5 µg/ml. Detects a band of approximately 35, 38 kDa (predicted molecular weight: 35 , 37 kDa).
ICC		Use a concentration of 5 µg/ml.
IP		Use at an assay dependent concentration.

Target

Function	Involved with pre-mRNA processing. Forms complexes (ribonucleosomes) with at least 20 other different hnRNP and heterogeneous nuclear RNA in the nucleous.
Sequence similarities	Contains 2 RRM (RNA recognition motif) domains.
Cellular localization	Nucleus > nucleoplasm. Cytoplasm. Localized in cytoplasmic mRNP granules containing untranslated mRNAs. Component of ribonucleosomes. Predominantly nucleoplasmic, however isoform A2 is also found in the cytoplasm of cells in some tissues. Not found in the nucleolus.

Images



Western blot - Anti-hnRNP A2B1 antibody (ab31645)

All lanes : Anti-hnRNP A2B1 antibody (ab31645) at 1/500 dilution

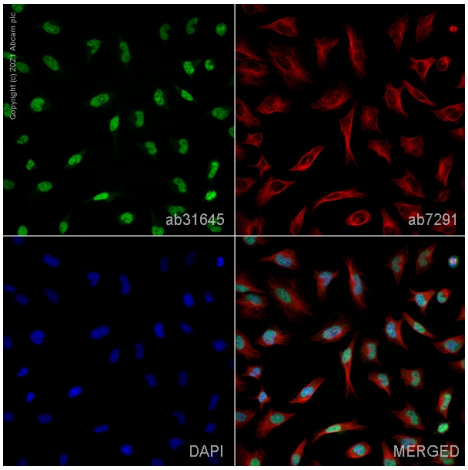
Lane 1 : Wild-type HEK293T cell lysate
Lane 2 : HNRNPA2B1 knockout HEK293T cell lysate
Lane 3 : A549 cell lysate at 20 µg

Performed under reducing conditions.

Predicted band size: 35 , 37 kDa
Observed band size: 37 kDa

Lanes 1-3: Merged signal (red and green). Green - ab31645 observed at 37 kDa. Red - loading control, **ab7291** observed at 50 kDa.

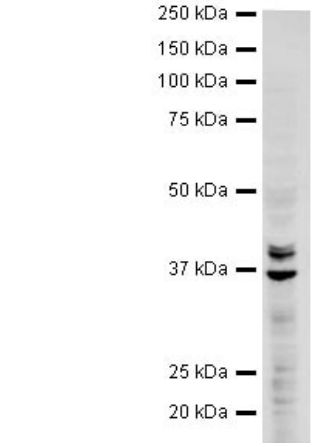
ab31645 Anti-hnRNP A2B1 antibody was shown to specifically react with hnRNP A2B1 in wild-type HEK293T cells. Loss of signal was observed when knockout cell line **ab266404** (knockout cell lysate **ab257224**) was used. Wild-type and hnRNP A2B1 knockout samples were subjected to SDS-PAGE. ab31645 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (**ab7291**) were incubated overnight at 4°C at 1 in 500 dilution and 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 10000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry - Anti-hnRNP A2B1 antibody (ab31645)

ab31645 staining hnRNP A2B1 in HeLa cells. The cells were fixed with 100% methanol (5 min), permeabilized with 0.1% PBS-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 overnight at 4°C with ab31645 at 5 µg/ml and **ab7291**, Mouse monoclonal [DM1A] to alpha Tubulin - Loading Control. Cells were then incubated with **ab150081**, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488), pre-adsorbed at 1/1000 dilution (shown in green) and **ab150120**, Goat polyclonal Secondary Antibody to Mouse IgG - 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 high-content analyser (Operetta CLS, Perkin Elmer) and a maximum intensity projection of confocal sections is shown.



Western blot - Anti-hnRNP A2B1 antibody (ab31645)

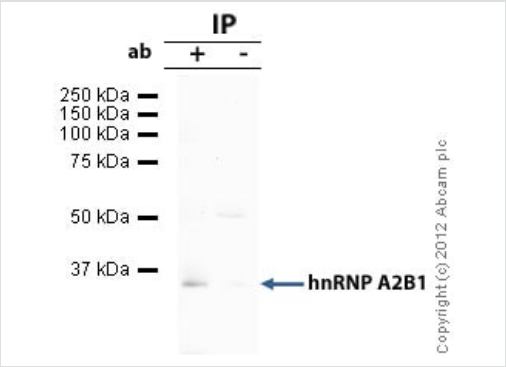
Anti-hnRNP A2B1 antibody (ab31645) at 1 µg/ml + HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate at 20 µg

Secondary
Goat polyclonal to Rabbit IgG (Alexa Fluor® 680) at 1/10000 dilution

Performed under reducing conditions.

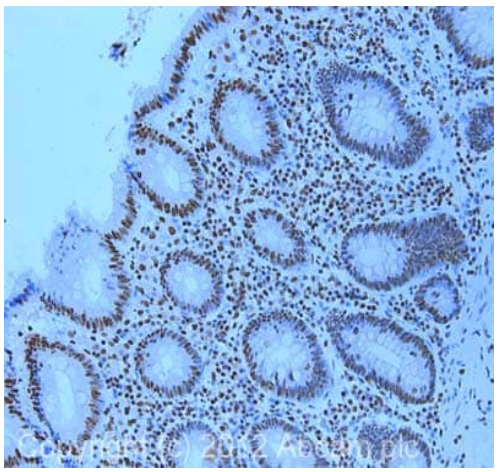
Predicted band size: 35 , 37 kDa
Observed band size: 35,38 kDa

ab31645 is expected to recognize isoform hnrnp A2 and hnrnp B1. The antibody detects bands at approximately 35 and 38 kDa which are of the correct size to correspond to the predicted molecular weight of these isoforms.



Immunoprecipitation - Anti-hnRNP A2B1 antibody (ab31645)

hnRNP A2B1 was immunoprecipitated using 0.5mg Hela whole cell extract, 5µg of Rabbit polyclonal to hnRNP A2B1 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, Hela 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 40µl SDS loading buffer and incubated for 10min at 70^{o}C; 10µl of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab31645.
Secondary: Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (**ab99697**).
Band: 35kDa: hnRNP A2B1.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-hnRNP A2B1 antibody (ab31645)

IHC image of ab31645 staining in human colon formalin fixed paraffin embedded tissue section, performed on a Leica Bond^{TM} 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 ab31645, 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.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

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