


## Product datasheet

# Anti-U2AF65 antibody ab37530

★★★★★ [2 Abreviews](#) [20 References](#) [5 Images](#)

### Overview

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<b>Product name</b>	Anti-U2AF65 antibody
<b>Description</b>	Rabbit polyclonal to U2AF65
<b>Host species</b>	Rabbit
<b>Tested applications</b>	<b>Suitable for:</b> IP, IHC-P, ChIP, ICC/IF, WB
<b>Species reactivity</b>	<b>Reacts with:</b> Mouse, Human, Zebrafish <b>Predicted to work with:</b> Cow, Xenopus laevis 
<b>Immunogen</b>	Synthetic peptide corresponding to Human U2AF65 aa 200-300. (Peptide available as <a href="#">ab37529</a> )
<b>General notes</b>	<p>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.</p> <p>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&amp;As</p>

### Properties

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<b>Form</b>	Liquid
<b>Storage instructions</b>	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.
<b>Storage buffer</b>	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.
<b>Purity</b>	Immunogen affinity purified
<b>Clonality</b>	Polyclonal
<b>Isotype</b>	IgG

## Applications

**The Abpromise guarantee** Our **Abpromise guarantee** covers the use of ab37530 in the following tested applications.

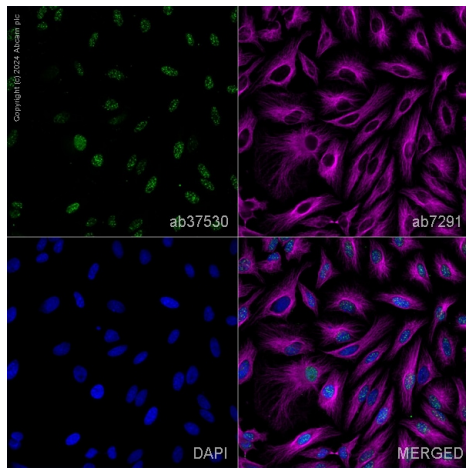
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IP		Use at an assay dependent concentration.
IHC-P		Use a concentration of 5 µg/ml.
ChIP		Use at an assay dependent concentration. PubMed: 27252533
ICC/IF	★★★★★ (1)	Use a concentration of 1 µg/ml.
WB	★★★★★ (1)	1/250. Detects a band of approximately 65 kDa (predicted molecular weight: 53 kDa).

## Target

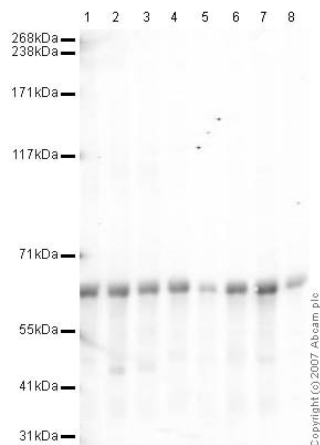
<b>Function</b>	Necessary for the splicing of pre-mRNA. Induces cardiac troponin-T (TNNT2) pre-mRNA exon inclusion in muscle. Regulates the TNNT2 exon 5 inclusion through competition with MBNL1. Binds preferentially to a single-stranded structure within the polypyrimidine tract of TNNT2 intron 4 during spliceosome assembly. Required for the export of mRNA out of the nucleus, even if the mRNA is encoded by an intron-less gene. Represses the splicing of MAPT/Tau exon 10.
<b>Sequence similarities</b>	Belongs to the splicing factor SR family. Contains 3 RRM (RNA recognition motif) domains.
<b>Post-translational modifications</b>	Lysyl-hydroxylation at Lys-15 and Lys-276 affects the mRNA splicing activity of the protein, leading to regulate some, but not all, alternative splicing events.
<b>Cellular localization</b>	Nucleus.

## Images



Immunocytochemistry/ Immunofluorescence - Anti-U2AF65 antibody (ab37530)

ab37530 staining U2AF65 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 ab37530 at 5µg/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 magenta). Nuclear DNA was labelled with DAPI (shown in blue). Also suitable in cells fixed with 4% paraformaldehyde (10 min). 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-U2AF65 antibody (ab37530)

**All lanes** : Anti-U2AF65 antibody (ab37530) at 1/250 dilution

**Lane 1** : HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate

**Lane 2** : Jurkat whole cell lysate (**ab7899**)

**Lane 3** : A-431 whole cell lysate (**ab7909**)

**Lane 4** : HEK-293 whole cell lysate (**ab7902**)

**Lane 5** : Hep G2 whole cell lysate (**ab7900**)

**Lane 6** : MCF-7 (Human breast adenocarcinoma cell line) Whole Cell Lysate

**Lane 7** : SHSY-5Y (Human neuroblastoma cell line) Whole Cell Lysate

**Lane 8** : U2OS (Human osteosarcoma cell line) Whole Cell Lysate

Lysates/proteins at 10 µg per lane.

### Secondary

**All lanes** : Rabbit IgG secondary antibody (**ab28446**) at 1/10000 dilution

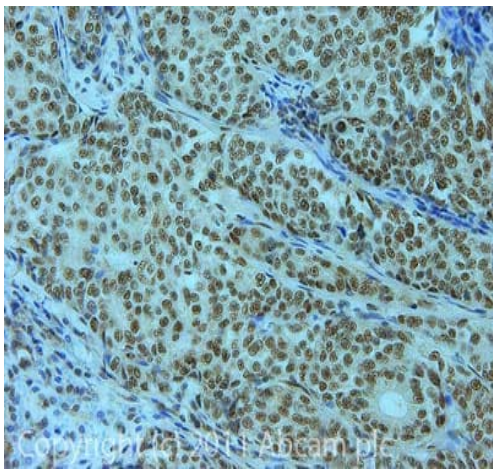
Performed under reducing conditions.

**Predicted band size:** 53 kDa

**Observed band size:** 65 kDa

Although the predicted band size is 53kDa based on Swiss-prot

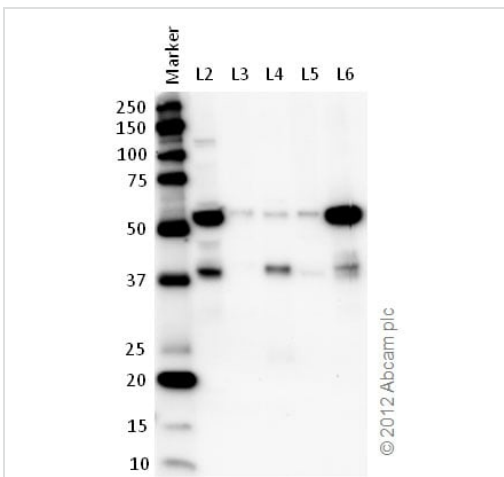
data, a band of 65kDa has been previously observed. **J Biol Chem.** 2004 Nov 26;279(48):49773-9 (PMID: 15377657)



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-U2AF65 antibody (ab37530)

IHC image of ab37530 staining in Human breast adenocarcinoma formalin fixed paraffin embedded tissue section, performed on a Leica Bond™ 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 ab37530, 5µg/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.



Western blot - Anti-U2AF65 antibody (ab37530)

**All lanes :** Anti-U2AF65 antibody (ab37530) at 1/250 dilution

**Lane 1 :** Marker

**Lane 2 :** Zebrafish brain homogenate at 20 µg

**Lane 3 :** Zebrafish heart homogenate at 10 µg

**Lane 4 :** Zebrafish liver homogenate at 10 µg

**Lane 5 :** Zebrafish skeletal muscle homogenate at 10 µg

**Lane 6 :** HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate at 10 µg

### Secondary

**All lanes :** Goat polyclonal to Rabbit IgG – H&L – Pre-Adsorbed (HRP) at 1/6000 dilution

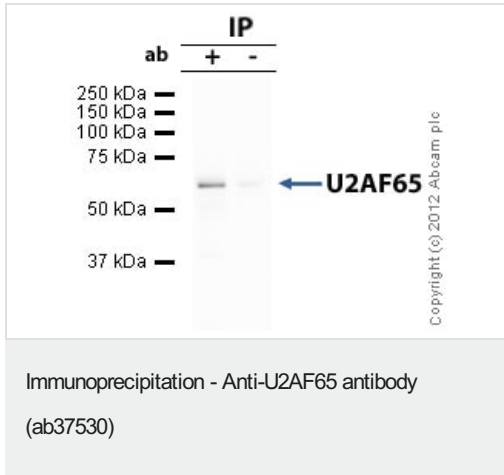
Developed using the ECL technique.

Performed under reducing conditions.

**Predicted band size:** 53 kDa

**Observed band size:** 53 kDa

**Exposure time:** 5 minutes



U2AF65 was immunoprecipitated using 0.5mg SHSY5Y whole cell extract, 5µg of Rabbit polyclonal to U2AF65 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, SHSY5Y 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°C; 10µl of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab37530.

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

Band: 65kDa: U2AF65.

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