abcam

Product datasheet

Anti-HuD antibody ab96474

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Overview

Product name	Anti-HuD antibody
Description	Rabbit polyclonal to HuD
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:** ICC/IF, IHC-P, WB
Species reactivity	**Reacts with:** Mouse, Rat, Human
	Predicted to work with: Rabbit, Chicken, Macaque monkey, Gorilla ⚠
Immunogen	Synthetic peptide corresponding to Mouse HuD aa 1-100 conjugated to keyhole limpet haemocyanin. (Peptide available as **ab151535**)
Positive control	This antibody gave a positive signal within Western Blot in the following tissue lysates: Mouse Brain P7; Rat Brain P7; Mouse Brain P0; Rat Brain P0. This antibody gave a positive result in IHC in the following FFPE tissue: Human hippocampus.
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 ab96474 in the following tested applications.
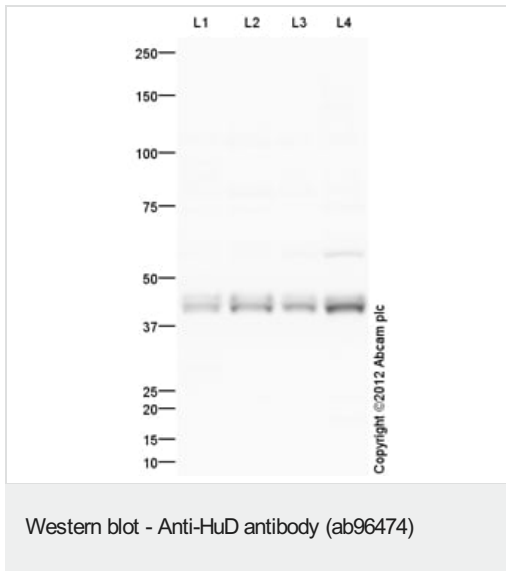
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
ICC/IF		Use a concentration of 5 µg/ml.
IHC-P		Use a concentration of 5 µg/ml.
WB		Use a concentration of 1 µg/ml. Detects a band of approximately 42 kDa (predicted molecular weight: 42 kDa). Abcam recommends using milk as the blocking agent (3%).

Target

Function	May play a role in neuron-specific RNA processing. Protects CDKN1A mRNA from decay by binding to its 3'-UTR (By similarity). Binds to AU-rich sequences (AREs) of target mRNAs, including VEGF and FOS mRNA.
Tissue specificity	Brain.
Sequence similarities	Belongs to the RRM elav family. Contains 3 RRM (RNA recognition motif) domains.
Post-translational modifications	Methylation at Arg-243 by CARM1 weakens protective binding to the 3'-UTR of CDKN1A mRNA and down-regulates CDKN1A protein expression, thereby maintaining cells in a proliferative state. Methylation is inhibited by NGF, which facilitates neurite outgrowth.

Images

Western blot - Anti-HuD antibody (ab96474)

All lanes : Anti-HuD antibody (ab96474) at 1 µg/ml (Milk blocking - 3%)

Lane 1 : P7 Mouse Brain Tissue Lysate Lab Lysate
Lane 2 : P7 Rat Brain Rat Tissue Lysate
Lane 3 : P0 Mouse Brain Mouse Tissue Lysate
Lane 4 : P0 Rat Brain Rat Tissue Lysate

Lysates/proteins at 25 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/10000 dilution
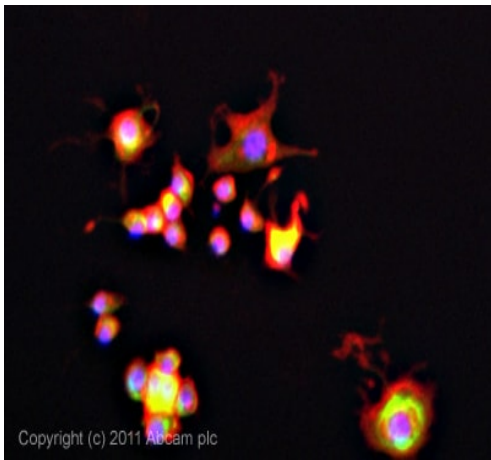
Developed using the ECL technique.

Performed under reducing conditions.

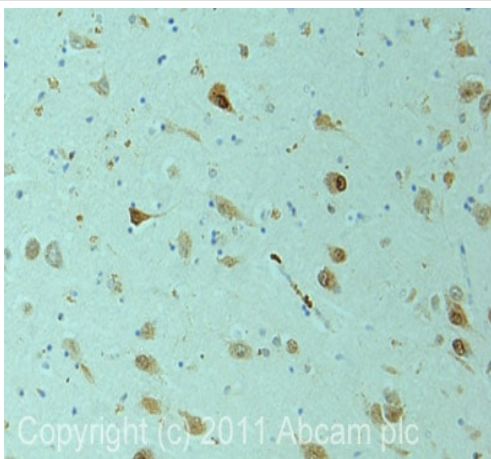**Predicted band size:** 42 kDa
Observed band size: 40,42 kDa

Exposure time: 8 minutes

This blot was produced using a 10% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 50 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 3% Milk before being incubated with ab96474 overnight at 4°C. Antibody binding was detected using an anti-rabbit antibody conjugated to HRP, and visualised using ECL development solution.

ICC/IF image of ab96474 stained PC12 cells. The cells were 4% formaldehyde 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 ab96474 at 1µg/ml overnight at +4°C. The secondary antibody (green) was DyLight® 488 goat anti- rabbit (**ab96899**) IgG (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 1.43µM.

Immunocytochemistry/ Immunofluorescence - Anti-HuD antibody (ab96474)

IHC image of HuD staining in Human hippocampus 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 ab96474, 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.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HuD antibody (ab96474)

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