abcam

Product datasheet

Anti-Nucleolin antibody ab22758

8 Abreviews 166 References 6 Images

Overview

Product name	Anti-Nucleolin antibody
Description	Rabbit polyclonal to Nucleolin
Host species	Rabbit
Specificity	Replenishment batches of our polyclonal antibody, ab22758 are tested in WB. Previous batches were additionally validated in ICC/IF, IHC-Fr, IHC-P and IP. These applications are still expected to work and are covered by our Abpromise guarantee. You may also be interested in our alternative recombinant antibody, **ab129200**.
Tested applications	**Suitable for:** WB, IP, IHC-Fr, IHC-P, ICC/IF
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide conjugated to KLH derived from within residues 1 - 100 of Human Nucleolin. Read Abcam's proprietary immunogen policy (Peptide available as **ab25315**.)
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 ab22758 in the following tested applications.

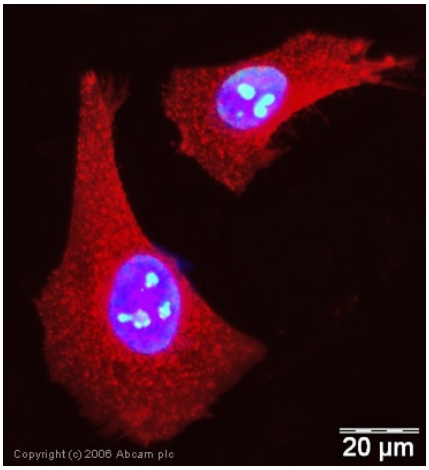
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB	★★★★★ (4)	Use a concentration of 1 µg/ml. Detects a band of approximately 76 kDa (predicted molecular weight: 76 kDa).
IP	★★★★★ (1)	Use at an assay dependent concentration.
IHC-Fr		Use at an assay dependent concentration.
IHC-P		Use a concentration of 1 µg/ml. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
ICC/IF	★★★★☆ (3)	Use a concentration of 1 µg/ml.

Target

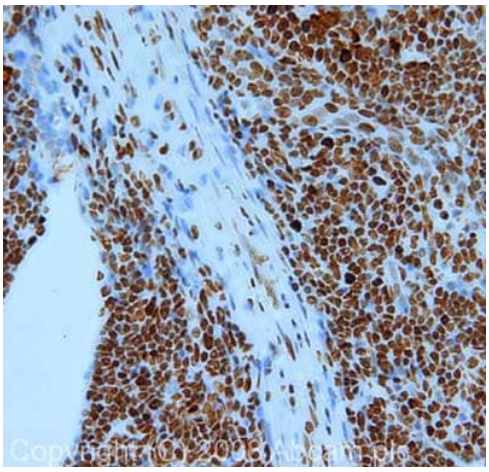
Function	Nucleolin is the major nucleolar protein of growing eukaryotic cells. It is found associated with intranucleolar chromatin and pre-ribosomal particles. It induces chromatin decondensation by binding to histone H1. It is thought to play a role in pre-rRNA transcription and ribosome assembly. May play a role in the process of transcriptional elongation. Binds RNA oligonucleotides with 5'-UUAGGG-3' repeats more tightly than the telomeric single-stranded DNA 5'-TTAGGG-3' repeats.
Sequence similarities	Contains 4 RRM (RNA recognition motif) domains.
Post-translational modifications	Some glutamate residues are glycylated by TTLL8. This modification occurs exclusively on glutamate residues and results in a glycine chain on the gamma-carboxyl group.
Cellular localization	Nucleus > nucleolus. Cytoplasm. Localized in cytoplasmic mRNP granules containing untranslated mRNAs.

Images



Immunocytochemistry/ Immunofluorescence - Anti-Nucleolin antibody (ab22758)

ICC/IF image of ab22758 stained human HeLa cells. The cells were methanol fixed (5 min) and incubated with the antibody (ab22758, 1µg/ml) for 1h at room temperature. The secondary antibody (green) was Alexa Fluor® 488 goat anti-rabbit IgG (H+L) used at a 1/1000 dilution for 1h. Image-iT™ FX Signal Enhancer was used as the primary blocking agent, 5% BSA (in TBS-T) was used for all other blocking steps. DAPI was used to stain the cell nuclei (blue). Alexa Fluor® 594 WGA was used to label plasma membranes (red).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Nucleolin antibody (ab22758)

IHC image of Nucleolin staining in human tonsil FFPE section, performed on a 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 ab22758, 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.

Western blot - Anti-Nucleolin antibody (ab22758)

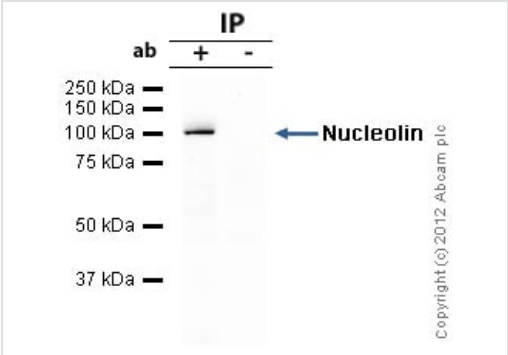
All lanes : Anti-Nucleolin antibody (ab22758) at 1 µg/ml

Lane 1 : MEF1 (Mouse embryonic fibroblast cell line) Whole Cell Lysate
Lane 2 : Brain (Mouse) Tissue Lysate
Lane 3 : Pancreas (Mouse) Tissue Lysate
Lane 4 : PC12 (Rat adrenal pheochromocytoma cell line) Whole Cell Lysate

Lysates/proteins at 10 µg per lane.

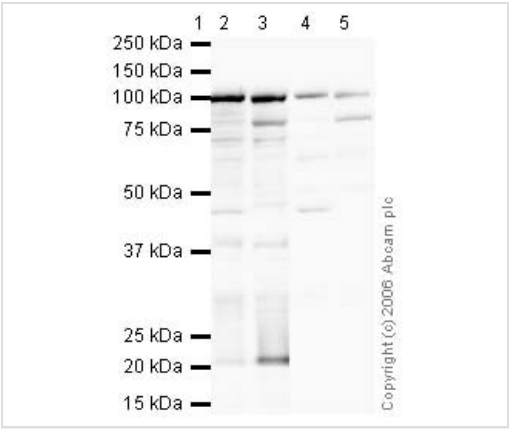
Secondary
All lanes : IRDye 680 Conjugated Goat Anti-Rabbit IgG (H+L) at 1/10000 dilution

Predicted band size: 76 kDa
Observed band size: 100 kDa
Additional bands at: 60 kDa. We are unsure as to the identity of these extra bands.



Immunoprecipitation - Anti-Nucleolin antibody (ab22758)

Nucleolin was immunoprecipitated using 0.5mg Jurkat whole cell extract, 5µg of Rabbit polyclonal to Nucleolin 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, Jurkat 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 ab22758.
Secondary: Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (**ab99697**).
Band: 100kDa: Nucleolin.



Western blot - Anti-Nucleolin antibody (ab22758)

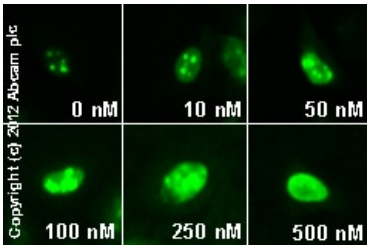
Lane 1 : Marker
Lanes 2-5 : Anti-Nucleolin antibody (ab22758) at 1 µg/ml

Lane 2 : Jurkat whole cell lysate (**ab7899**) at 20 µg
Lane 3 : A-431 whole cell lysate (**ab7909**) at 20 µg
Lane 4 : Jurkat whole cell lysate (**ab7899**) at 20 µg with Human Nucleolin peptide (**ab25315**) at 1 µg/ml
Lane 5 : A-431 whole cell lysate (**ab7909**) at 20 µg with Human Nucleolin peptide (**ab25315**) at 1 µg/ml

Secondary
Lanes 2-5 : Goat polyclonal to Rabbit IgG H&L (HRP) Pre-Adsorbed at 1/10000 dilution

Performed under reducing conditions.

Predicted band size: 76 kDa
Observed band size: 100 kDa



Immunocytochemistry/ Immunofluorescence - Anti-Nucleolin antibody (ab22758)

ab22758 staining nucleolin in HeLa cells treated with Triptolide from *Tripterygium wilfordii* (**ab120720**), by ICC/IF. Changes in nuclear localization of nucleolin (from nuclelous to whole nucleous) correlates with increased concentration of Triptolide from *Tripterygium wilfordii*, as described in literature.
The cells were incubated at 37°C for 1h in media containing different concentrations of **ab120720** (Triptolide from *Tripterygium wilfordii*) in DMSO, fixed with 4% formaldehyde for 10 minutes at room temperature and blocked with PBS containing 10% goat serum, 0.3 M glycine, 1% BSA and 0.1% tween for 2h at room temperature. Staining of the treated cells with ab22758 (1 µg/ml) was performed overnight at 4°C in PBS containing 1% BSA and 0.1% tween. A DyLight 488 goat anti-rabbit polyclonal antibody (**ab96899**) at 1/250 dilution was used as the secondary antibody.

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