abcam

Product datasheet

Anti-HMGA1 antibody [EPR7839] ab129153

Recombinant RabMAb

★★★★★ 3 Abreviews 21 References 10 Images

Overview

Product name Anti-HMGA1 antibody [EPR7839]

Description Rabbit monoclonal [EPR7839] to HMGA1

Host species Rabbit

Tested applications Suitable for: WB, IHC-P, Flow Cyt, ICC/IF

Species reactivity Reacts with: Mouse, Rat, Human

Immunogen Synthetic peptide within Human HMGA1 (N terminal). The exact sequence is proprietary.

Database link: P17096

Positive control WB: SK-OV-3, Caco-2, BxPC-3 and HepG2 whole cell lysate (ab7900), rat kidney and mouse

brain tissue lysates. IHC-P: Human testis and transitional cell carcinoma of bladder tissues.

ICC/IF: HeLa and BxPC-3 cells. Flow Cyt: HepG2 cells.

General notes Our RabMAb® technology is a patented hybridoma-based technology for making rabbit

monoclonal antibodies. For details on our patents, please refer to RabMab® patents

We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified

format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this

update, please contact our Scientific Support team.

This product is a recombinant rabbit monoclonal antibody.

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.

Stable for 12 months at -20°C.

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol, 0.05% BSA

Purity Protein A purified

Clonality Monoclonal

Clone number EPR7839

Isotype IgG

Applications

Our Abpromise guarantee covers the use of ab129153 in the following tested applications.

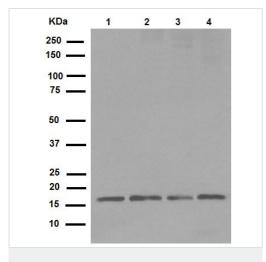
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		1/10000 - 1/50000. Detects a band of approximately 17 kDa (predicted molecular weight: 12 kDa).
IHC-P	****	1/250 - 1/500. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol. See IHC antigen retrieval protocols.
Flow Cyt		1/60. For unpurified use at 1/100 - 1/500. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
ICC/IF	****	1/500.

Target

Function	HMG-I/Y bind preferentially to the minor groove of A+T rich regions in double stranded DNA. It is suggested that these proteins could function in nucleosome phasing and in the 3'-end processing of mRNA transcripts. They are also involved in the transcription regulation of genes containing, or in close proximity to A+T-rich regions.
Involvement in disease	Note=A chromosomal aberration involving HMGA1 is found in pulmonary chondroid hamartoma. Translocation t(6;14)(p21;q23-24) with RAD51L1.
Sequence similarities	Belongs to the HMGA family. Contains 3 A.T hook DNA-binding domains.
Post-translational modifications	Constitutively phosphorylated on two or three sites. Phosphorylated upon DNA damage, probably by ATM or ATR. Hyperphosphorylated at early stages of apoptosis, followed by dephosphorylation and methylation, which coincides with chromatin condensation. Isoform HMG-Y can be phosphorylated by HIPK2. HMG-Y is not methylated. Methylation at Arg-58 is mutually exclusive with methylation at Arg-60.
Cellular localization	Nucleus. Chromosome.

Images



Western blot - Anti-HMGA1 antibody [EPR7839] (ab129153)

All lanes: Anti-HMGA1 antibody [EPR7839] (ab129153) (purified)

Lane 1 : HepG2 cell lysate
Lane 2 : SK-OV-3 cell lysate
Lane 3 : Caco-2 cell lysate
Lane 4 : BxPC-3 cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Peroxidase-conjugated goat anti-rabbit lgG (H+L) at 1/1000 dilution

Predicted band size: 12 kDa **Observed band size:** 17 kDa

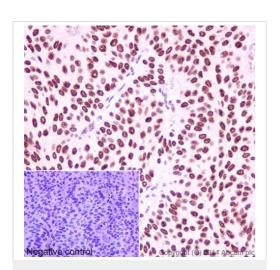
why is the actual band size different from the predicted?

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.

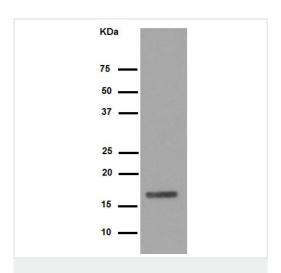
Immunocytochemistry/ Immunofluorescence - Anti-HMGA1 antibody [EPR7839] (ab129153)

Immunocytochemistry/Immunofluorescence analysis of BxPC-3 cells labelling HMGA1 with unpurified ab129153 at 1/250 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-HMGA1 antibody
[EPR7839] (ab129153)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human transitional cell carcinoma of bladder tissue labelling HMGA1 with purified ab129153 at 1/500. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. A prediluted HRP-polymer conjugated anti-rabbit lgG was used as the secondary antibody. Negative control using PBS instead of primary antibody. Counterstained with Hematoxylin.



Western blot - Anti-HMGA1 antibody [EPR7839] (ab129153)

Anti-HMGA1 antibody [EPR7839] (ab129153) at 1/10000 dilution (purified) + Mouse brain tissue lysate at 10 μ g

Secondary

Peroxidase-conjugated goat anti-rabbit lgG (H+L) at 1/1000 dilution

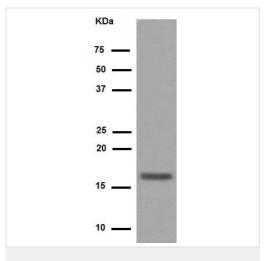
Predicted band size: 12 kDa

Observed band size: 17 kDa why is the actual band size different

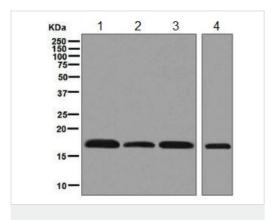
from the predicted?

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



Western blot - Anti-HMGA1 antibody [EPR7839] (ab129153)



Western blot - Anti-HMGA1 antibody [EPR7839] (ab129153)

Anti-HMGA1 antibody [EPR7839] (ab129153) at 1/50000 dilution (purified) + Rat kidney tissue lysate at 10 μg

Secondary

Peroxidase-conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

Predicted band size: 12 kDa

Observed band size: 17 kDa why is the actual band size different

from the predicted?

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.

All lanes : Anti-HMGA1 antibody [EPR7839] (ab129153) at 1/10000 dilution (unpurified)

Lane 1 : SK-OV-3 cell lysate
Lane 2 : Caco-2 cell lysate
Lane 3 : BxPC-3 cell lysate
Lane 4 : HepG2 cell lysate

Lysates/proteins at 10 µg per lane.

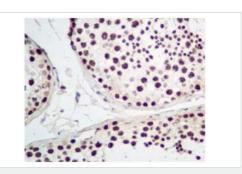
Secondary

All lanes: Goat anti-rabbit HRP at 1/2000 dilution

Predicted band size: 12 kDa

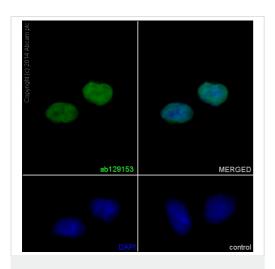
Observed band size: 17 kDa why is the actual band size different

from the predicted?



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-HMGA1 antibody
[EPR7839] (ab129153)

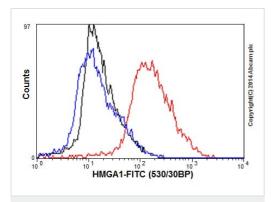
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human testis tissue labelling HMGA1 with unpurified ab129153 at 1/250.



Immunocytochemistry/ Immunofluorescence - Anti-HMGA1 antibody [EPR7839] (ab129153)

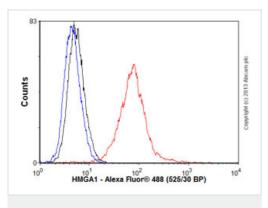
Immunocytochemistry/Immunofluorescence analysis of HeLa cells labelling HMGA1 with purified ab129153 at 1/500. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, an Alexa Fluor[®] 488-conjugated goat anti-rabbit lgG (1/500) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain.

Control: primary antibody (1/500) and secondary antibody, ab150120, an Alexa Fluor[®] 594-conjugated goat anti-mouse IgG (1/500).



Flow Cytometry - Anti-HMGA1 antibody [EPR7839] (ab129153)

Flow cytometry analysis of HepG2 cells labelling HMGA1 with purified ab129153 at 1/60 (red). Cells were fixed with 2% paraformaldehyde. A FITC-conjugated goat anti-rabbit lgG (1/150) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal lgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.



Flow Cytometry - Anti-HMGA1 antibody [EPR7839] (ab129153)

Overlay histogram showing HepG2 cells stained with unpurified ab129153 (red line). The cells were fixed with 4% paraformaldehyde (10 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific proteinprotein interactions followed by the antibody (ab129153, 1/870 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit lgG (H&L) (ab150077) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit lgG (monoclonal) (0.1µg/1x10⁶ cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This antibody gave a positive signal in HepG2 cells fixed with 80% methanol (5 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.

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