abcam

Product datasheet

Anti-DGCR8 antibody [EPR18757] - BSA and Azide free ab240325



5 Images

Overview

Product name Anti-DGCR8 antibody [EPR18757] - BSA and Azide free

Description Rabbit monoclonal [EPR18757] to DGCR8 - BSA and Azide free

Host species Rabbit

Tested applications Suitable for: WB, Flow Cyt (Intra), ICC/IF, IP

Species reactivity Reacts with: Mouse, Rat, Human

Immunogen Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

General notes ab240325 is the carrier-free version of ab191875.

> Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for

increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cellbased assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the

need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C. Do Not Freeze.

Storage buffer pH: 7.2

Constituent: PBS

Carrier free Yes

Purity Protein A purified

Clonality Monoclonal
Clone number EPR18757

Isotype IgG

Applications

The Abpromise guarantee

Our Abpromise guarantee covers the use of ab240325 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		Use at an assay dependent concentration. Detects a band of approximately 100 kDa (predicted molecular weight: 86 kDa).
Flow Cyt (Intra)		Use at an assay dependent concentration.
ICC/IF		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.

Target

Function

Component of the microprocessor complex that acts as a RNA- and heme-binding protein that is involved in the initial step of microRNA (miRNA) biogenesis. Component of the microprocessor complex that is required to process primary miRNA transcripts (pri-miRNAs) to release precursor miRNA (pre-miRNA) in the nucleus. Within the microprocessor complex, DGCR8 function as a molecular anchor necessary for the recognition of pri-miRNA at dsRNA-ssRNA junction and directs DROSHA to cleave 11 bp away form the junction to release hairpin-shaped pre-miRNAs that are subsequently cut by the cytoplasmic DICER to generate mature miRNAs. The hemebound DGCR8 dimer binds pri-miRNAs as a cooperative trimer (of dimers) and is active in triggering pri-miRNA cleavage, whereas the heme-free DGCR8 monomer binds pri-miRNAs as a dimer and is much less active. Both double-stranded and single-stranded regions of a pri-miRNA are required for its binding. Involved in the silencing of embryonic stem cells self-renewal.

Tissue specificity

Ubiquitously expressed.

Sequence similarities

Contains 2 DRBM (double-stranded RNA-binding) domains.

Contains 1 WW domain.

Domain

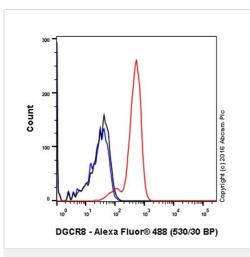
Both DRBM domains are required for efficient binding to pri-miRNA. The region between residues 276 and 498 has an autoinhibitory function on pri-miRNA processing activity.

Cellular localization

Nucleus. Nucleus > nucleolus. Colocalizes with nucleolin and DROSHA in the nucleolus. Mostly detected in the nucleolus as electron-dense granular patches around the fibrillar center (FC) and granular component (GC). Also detected in the nucleoplasm as small foci adjacent to splicing speckles near the chromatin structure. Localized with DROSHA in GW bodies (GWBs), also

known as P-bodies.

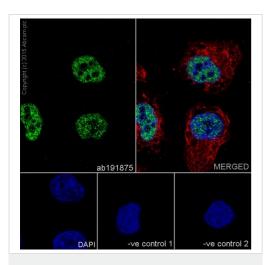
Images



Flow Cytometry (Intracellular) - Anti-DGCR8 antibody [EPR18757] - BSA and Azide free (ab240325)

Intracellular Flow Cytometry analysis of Jurkat (human acute T cell leukemia) labelling DGCR8 with purified <u>ab191875</u> at 1/1000 (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. Alexa Fluor[®] 488 goat anti-rabbit lgG (1/2000) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal lgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab191875).



Immunocytochemistry/ Immunofluorescence - Anti-DGCR8 antibody [EPR18757] - BSA and Azide free (ab240325)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cells from cervix adenocarcinoma) cells labeling DGCR8 with <u>ab191875</u> at 1/500 dilution, followed by Goat anti-rabbit lgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green).

Confocal image showing nuclear staining on HeLa cell line.

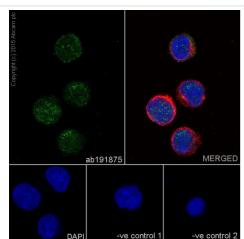
The nuclear counterstain is DAPI (blue).

Tubulin is detected with <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution and <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/1000 dilution (red).

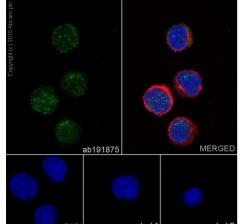
The negative controls are as follows:-

-ve control 1: <u>ab191875</u> at 1/500 dilution followed by <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/1000 dilution. -ve control 2: <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution followed by <u>ab150077</u> (Alexa Fluor®488 Goat Anti-Rabbit lgG H&L) at 1/1000 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab191875</u>).



Immunocytochemistry/ Immunofluorescence - Anti-DGCR8 antibody [EPR18757] - BSA and Azide free (ab240325)



The nuclear counterstain is DAPI (blue). Tubulin is detected with ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution and ab150120 (AlexaFluor®594 Goat anti-Mouse

Confocal image showing nuclear and weakly cytoplasmic staining

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized Jurkat (Human T cell leukemia cells from peripheral blood) cells labeling DGCR8 with ab191875 at 1/500 dilution, followed by Goat anti-rabbit lgG (Alexa Fluor® 488)

(ab150077) secondary antibody at 1/1000 dilution (green).

The negative controls are as follows:-

secondary) at 1/1000 dilution (red).

on Jurkat cell line.

-ve control 1: ab191875 at 1/500 dilution followed by ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/1000 dilution. -ve control 2: ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution followed by ab150077 (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/1000 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab191875).

250 kDa -150 kDa ← DGCR8 100 kDa 75 kDa Copyright(C) 2015 Abcam plc 50 kDa 37 kDa 25 kDa 20 kDa

Immunoprecipitation - Anti-DGCR8 antibody [EPR18757] - BSA and Azide free (ab240325)

DGCR8 was immunoprecipitated from 1mg of HEK-293 (Human epithelial cells from embryonic kidney) whole cell lysate with ab191875 at 1/60 dilution.

Western blot was performed from the immunoprecipitate using ab191875 at 1/1000 dilution.

Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG, was used as secondary antibody at 1/1500 dilution.

Lane 1: HEK-293 whole cell lysate 10ug (Input).

Lane 2: ab191875 IP in HEK-293 whole cell lysate.

Lane 3: Rabbit monoclonal lgG (ab172730) instead of ab191875 in HEK-293 whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 30 seconds.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab191875</u>).



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