abcam

Product datasheet

Anti-BRG1 antibody [EPNCIR111A] ab110641

KO VALIDATED Recombinant RabMAb

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Overview

Product name	Anti-BRG1 antibody [EPNCIR111A]
Description	Rabbit monoclonal [EPNCIR111A] to BRG1
Host species	Rabbit
Tested applications	**Suitable for:** ChIP, WB, IP, IHC-P, ICC/IF, Flow Cyt
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide within Mouse BRG1 aa 200-300. The exact sequence is proprietary. Database link: P51532
Positive control	K562, HeLa, MOLT4, NIH3T3, and PC12 cell lysates; Human kidney and testis tissues; HeLa cells. ICC/IF: HeLa cells, SMARCA4-HAP1 cells
General notes	This antibody was developed as part of a collaboration between Epitomics, the National Cancer Institute's Center for Cancer Research and the lab of Gordon Hager. 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	PBS 49%,Sodium azide 0.01%,Glycerol 50%,BSA 0.05%
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPNCIR111A

Isotype	IgG

Applications

Our Abpromise guarantee covers the use of **ab110641** 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
ChIP	★★★★☆	Use at an assay dependent concentration.
WB	★★★★★	1/10000 - 1/50000. Predicted molecular weight: 185 kDa.
IP	★★★★☆	1/10 - 1/100.
IHC-P		1/100 - 1/250. Antigen retrieval is recommended. Heat up to 98 °C, below boiling, and then let cool for 10-20 min.
ICC/IF		1/500.
Flow Cyt		Use at an assay dependent concentration.

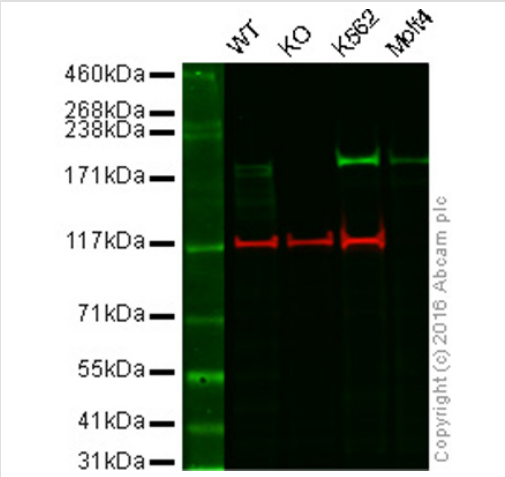
Target

Function

Transcriptional coactivator cooperating with nuclear hormone receptors to potentiate transcriptional activation. Component of the CREST-BRG1 complex, a multiprotein complex that regulates promoter activation by orchestrating a calcium-dependent release of a repressor complex and a recruitment of an activator complex. In resting neurons, transcription of the c-FOS promoter is inhibited by BRG1-dependent recruitment of a phospho-RB1-HDAC repressor complex. Upon calcium influx, RB1 is dephosphorylated by calcineurin, which leads to release of the repressor complex. At the same time, there is increased recruitment of CREBBP to the promoter by a CREST-dependent mechanism, which leads to transcriptional activation. The CREST-BRG1 complex also binds to the NR2B promoter, and activity-dependent induction of NR2B expression involves a release of HDAC1 and recruitment of CREBBP. Belongs to the neural progenitors-specific chromatin remodeling complex (npBAF complex) and the neuron-specific chromatin remodeling complex (nBAF complex). During neural development a switch from a stem/progenitor to a post-mitotic chromatin remodeling mechanism occurs as neurons exit the cell cycle and become committed to their adult state. The transition from proliferating neural stem/progenitor cells to post-mitotic neurons requires a switch in subunit composition of the npBAF and nBAF complexes. As neural progenitors exit mitosis and differentiate into neurons, npBAF complexes which contain ACTL6A/BAF53A and PHF10/BAF45A, are exchanged for homologous alternative ACTL6B/BAF53B and DPF1/BAF45B or DPF3/BAF45C subunits in neuron-specific complexes (nBAF). The npBAF complex is essential for the self-renewal/proliferative capacity of the multipotent neural stem cells. The nBAF complex along with CREST plays a role regulating the activity of genes essential for dendrite growth. SMARCA4/BAF190A may promote neural stem cell self-renewal/proliferation by enhancing Notch-dependent proliferative signals, while concurrently making the neural stem cell insensitive to SHH-dependent differentiating cues (By similarity). Also involved in vitamin D-coupled transcription regulation via its association with the WINAC complex, a chromatin-remodeling complex recruited by vitamin D receptor (VDR), which is required for the ligand-bound VDR-

	mediated transrepression of the CYP27B1 gene. Acts as a corepressor of ZEB1 to regulate E-cadherin transcription and is required for induction of epithelial-mesenchymal transition (EMT) by ZEB1.
Tissue specificity	Colocalizes with ZEB1 in E-cadherin-negative cells from established lines, and stroma of normal colon as well as in de-differentiated epithelial cells at the invasion front of colorectal carcinomas (at protein level).
Involvement in disease	Defects in SMARCA4 are the cause of rhabdoid tumor predisposition syndrome type 2 (RTPS2) [MIM:613325]. RTPS2 is a familial cancer syndrome predisposing to renal or extrarenal malignant rhabdoid tumors and to a variety of tumors of the central nervous system, including choroid plexus carcinoma, medulloblastoma, and central primitive neuroectodermal tumors. Rhabdoid tumors are the most aggressive and lethal malignancies occurring in early childhood.
Sequence similarities	Belongs to the SNF2/RAD54 helicase family. Contains 1 bromo domain. Contains 1 helicase ATP-binding domain. Contains 1 helicase C-terminal domain. Contains 1 HSA domain.
Post-translational modifications	Phosphorylated upon DNA damage, probably by ATM or ATR.
Cellular localization	Nucleus.

Images

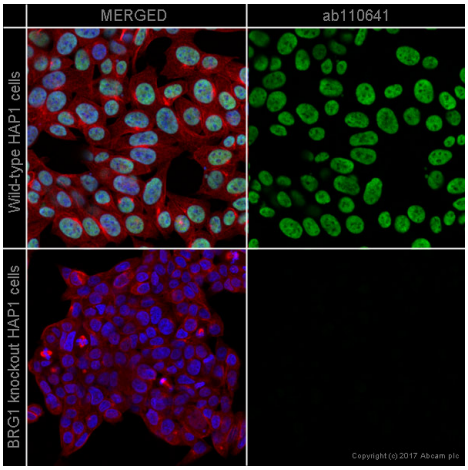


Western blot - Anti-BRG1 antibody [EPNCIR111A] (ab110641)

Lane 1: Wild-type HAP1 cell lysate (20 µg)
Lane 2: BRG1 knockout HAP1 cell lysate (20 µg)
Lane 3: K562 cell lysate (20 µg)
Lane 4: Molt-4 cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - ab110641 observed at 185 kDa. Red - loading control, ab18058, observed at 124 kDa.

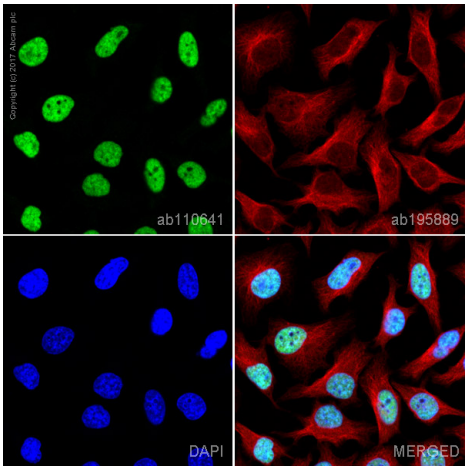
ab110641 was shown to specifically react with BRG1 in wild-type HAP1 cells. No band was observed when BRG1 knockout samples were used. Wild-type and BRG1 knockout samples were subjected to SDS-PAGE, ab110641 and ab18058 (loading control to Vinculin) were both diluted 1/10,000 and incubated overnight at 4°C. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1/10,000 dilution for 1hr at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-BRG1 antibody [EPNCIR111A] (ab110641)

ab110641 staining BRG1 in wild-type HAP1 cells (top panel) and BRG1 knockout HAP1 cells (bottom panel). The cells were fixed with 4% formaldehyde (10min), permeabilized with 0.1% 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 with ab110641 at 1/500 dilution and ab195889 at 1/250 dilution (shown in pseudocolour red) overnight at +4°C, followed by a further incubation at room temperature for 1h with a goat secondary antibody to Rabbit IgG (Alexa Fluor® 488) (ab150081) at 2 µg/ml (shown in green). Nuclear DNA was labelled in blue with DAPI.

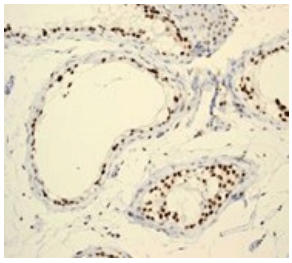
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



Immunocytochemistry/ Immunofluorescence - Anti-BRG1 antibody [EPNCIR111A] (ab110641)

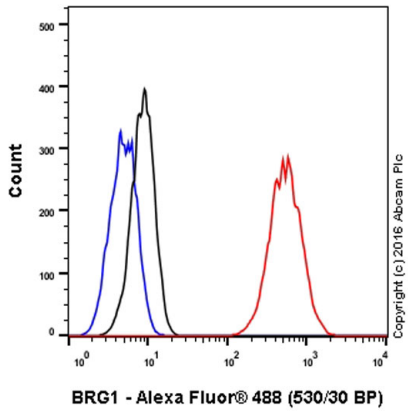
ab110641 staining BRG1 in HeLa cells. The cells were fixed with 4% formaldehyde (10min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab110641 at 1/500 dilution and ab195889 (Mouse monoclonal [DM1A] to alpha Tubulin - Microtubule Marker (Alexa Fluor® 594)) at 1/250 dilution overnight at +4°C, followed by a further incubation at room temperature for 1h with ab150081 (Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488)) at 2 µg/ml (shown in green). Nuclear DNA was labelled in blue with DAPI.

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



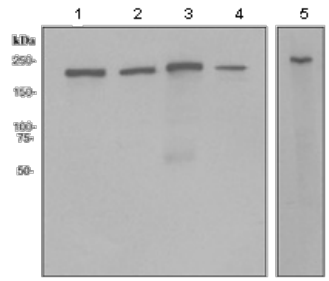
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BRG1 antibody [EPNCIR111A] (ab110641)

ab110641 at 1/100 dilution staining BRG1 in Human testis tissue by Immunohistochemistry, Paraffin-embedded tissue.



Flow Cytometry - Anti-BRG1 antibody [EPNCIR111A] (ab110641)

Flow Cytometry analysis of HeLa (human cervix adenocarcinoma) cells labeling BRG1 with purified ab110641 at 1/200 dilution(10µg/ml) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit IgG (Alexa Fluor® 488)(1/2000 dilution) was used as the secondary antibody. Rabbit monoclonal IgG (Black) was used as the isotype control, cells without incubation with primary antibody and secondary antibody (Blue) were used as the unlabeled control.



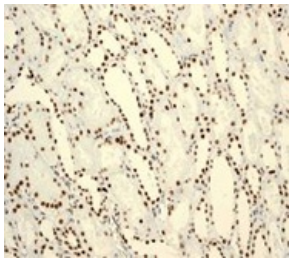
Western blot - Anti-BRG1 antibody [EPNCIR111A] (ab110641)

All lanes : Anti-BRG1 antibody [EPNCIR111A] (ab110641) at 1/10000 dilution

Lane 1 : K562 cell lysate
Lane 2 : HeLa cell lysate
Lane 3 : MOLT4 cell lysate
Lane 4 : NIH3T3 cell lysate
Lane 5 : PC12 cell lysate

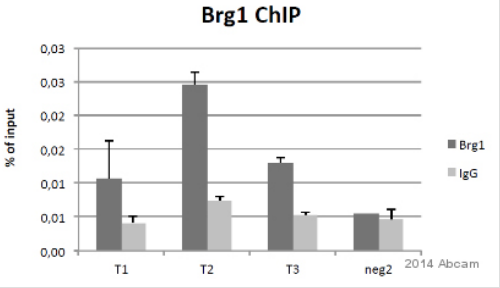
Lysates/proteins at 10 µg per lane.

Predicted band size: 185 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BRG1 antibody [EPNCIR111A] (ab110641)

ab110641 at 1/100 dilution staining BRG1 in Human kidney tissue by Immunohistochemistry, Paraffin-embedded tissue.



ChIP - Anti-BRG1 antibody [EPNCIR111A] (ab110641)

This image is courtesy of an Abreview submitted by Silvia Bonifacio

ChIP analysis using ab110641 binding BRG1 in mouse bone marrow derived macrophages. Cells were cross-linked for 10 minutes with formaldehyde. Samples were incubated with primary antibody for 16 hours at 4°C. Protein binding was detected using real-time PCR.
Positive control: PU.1 antibody.
Negative Control: rabbit IgG.

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