abcam

Product datasheet

Anti-BRG1 antibody [EPR3912] - BSA and Azide free ab222230

KO VALIDATED Recombinant RabMAb

1 References 19 Images

Overview

Product name	Anti-BRG1 antibody [EPR3912] - BSA and Azide free
Description	Rabbit monoclonal [EPR3912] to BRG1 - BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** ChIC/CUT&RUN-seq, Flow Cyt (Intra), WB, IHC-P, ICC/IF, IP
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: HAP1, HEK-293T, K562, RAW264.7, NIH/3T3, PC-12 and Molt-4 cell lysate. ICC/IF: HeLa cells; SMARCA4-HAP1 cells. IHC-P: Human colon, urinary bladder transitional carcinoma, breast carcinoma, bladder cancer, ovarian carcinoma and normal tonsil tissue, mouse and rat stomach tissue. Flow Cyt (intra): HeLa cells IP: K562 cell lysate, NIH/3T3 cell lysate. ChIC/CUT&RUN-Seq: HeLa cells.
General notes	ab222230 is the carrier-free version of **ab108318**. 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 cell-based 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. This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information **see here**.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	pH: 7.20 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR3912
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab222230 in the following tested applications.

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

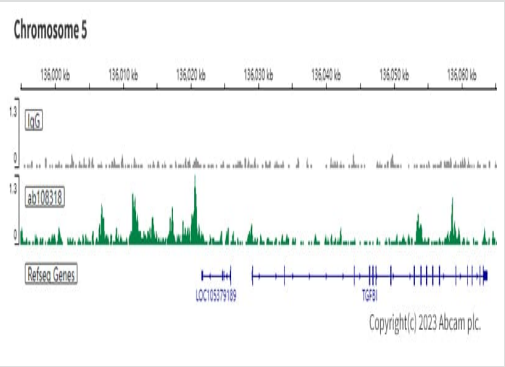
Application	Abreviews	Notes
ChIC/CUT&RUN-seq		Use at an assay dependent concentration.
Flow Cyt (Intra)		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Predicted molecular weight: 185 kDa.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See **IHC antigen retrieval protocols**. Heat up to 98 degrees C, below boiling, and then let cool for 10-20 min.
ICC/IF		Use at an assay dependent concentration.
IP		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



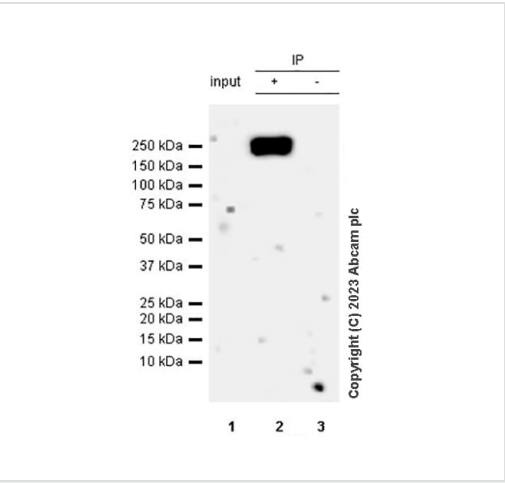
ChIC/CUT&RUN sequencing - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

ChIC/CUT&RUN was performed using a pAG-MNAse at a final concentration of 700 ng/mL, 2.5 x 10^5 HeLa (Human cervix adenocarcinoma epithelial cell line) cells and 5µg of **ab108318** [EPR3912]. The resulting DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 10 million reads. The negative IgG control **ab172730** is also shown.

Additional screenshots of mapped reads can be downloaded **here**.

The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab108318**).



Immunoprecipitation - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab108318**).

BRG1 was immunoprecipitated from NIH/3T3 (mouse embryonic fibroblast), whole cell lysate with **ab108318** at 1/30 dilution (2µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using **ab108318** at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP)(**ab131366**) was used as secondary antibody at 1/5000 dilution.

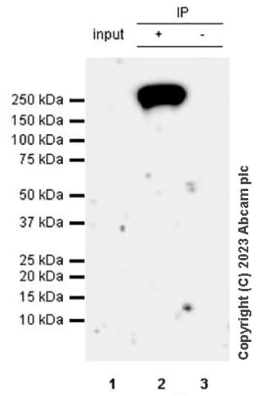
Lane 1 (Input): NIH/3T3 (mouse embryonic fibroblast), whole cell lysate, 10 µg

Lane 2 (+) : NIH/3T3 whole cell lysate

Lane 3 (-) : Rabbit monoclonal IgG (**ab172730**) instead of **ab108318** in NIH/3T3 whole cell lysate

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

Observed MW: 185 kDa.



Immunoprecipitation - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab108318**).

BRG1 was immunoprecipitated from K-562 (human chronic myelogenous leukemia lymphoblast), whole cell lysate with **ab108318** at 1/30 dilution (2µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using **ab108318** at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP) (**ab131366**) was used as secondary antibody at 1/5000 dilution.

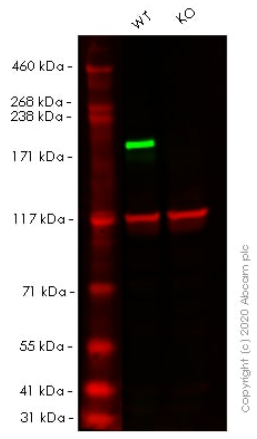
Lane 1 (Input): K-562 (human chronic myelogenous leukemia lymphoblast), whole cell lysate, 10 µg

Lane 2 (+) : K-562 whole cell lysate

Lane 3 (-) : Rabbit monoclonal IgG (**ab172730**) instead of **ab108318** in K-562 whole cell lysate

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

Observed MW: 185 kDa.



Western blot - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

All lanes : Anti-BRG1 antibody [EPR3912] (**ab108318**) at 1/10000 dilution

Lane 1 : Wild-type HEK-293T cell lysate
Lane 2 : SMARCA4 knockout HEK-293T cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

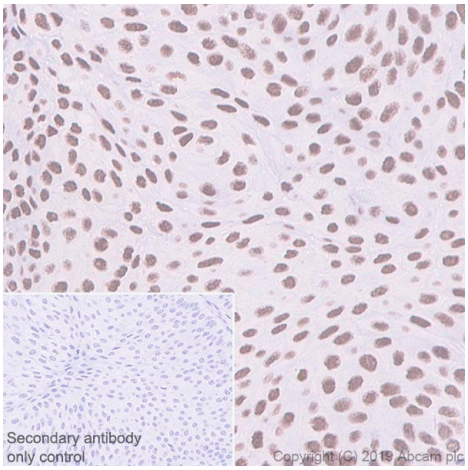
Predicted band size: 185 kDa
Observed band size: 185 kDa

This data was developed using the same antibody clone in a different buffer formulation (**ab108318**).

Lanes 1- 2: Merged signal (red and green). Green - **ab108318**

observed at 185 kDa. Red - Anti-Vinculin antibody [VIN-54] observed at 124 kDa.

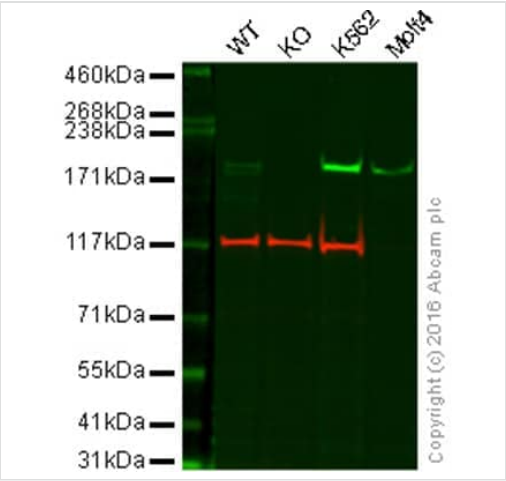
ab108318 was shown to react with BRG1 in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line **ab255432** (knockout cell lysate **ab263853**) was used. Wild-type HEK-293T and SMARCA4 knockout HEK-293T cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. **ab108318** and Anti-Vinculin antibody [VIN-54] overnight at 4°C at a 1 in 10000 dilution and a 1 in 20000 dilution respectively. 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 in 20000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human bladder cancer tissue sections labeling BRG1 with purified **ab108318** at 1/500 dilution (0.23 µg/ml). Heat mediated antigen retrieval was performed using **ab93684** (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab108318**).



Western blot - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

All lanes : Anti-BRG1 antibody [EPR3912] (**ab108318**) at 1/1000 dilution

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

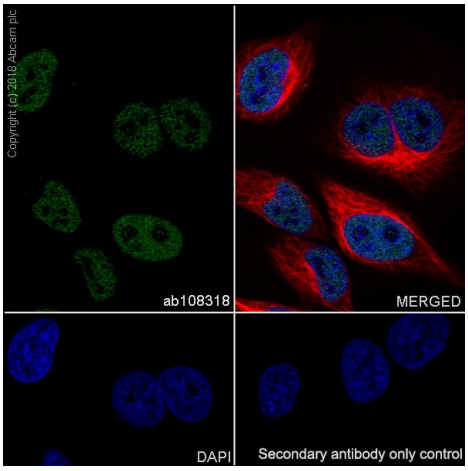
Lysates/proteins at 20 µg per lane.

Predicted band size: 185 kDa

This data was developed using the same antibody clone in a different buffer formulation (**ab108318**).

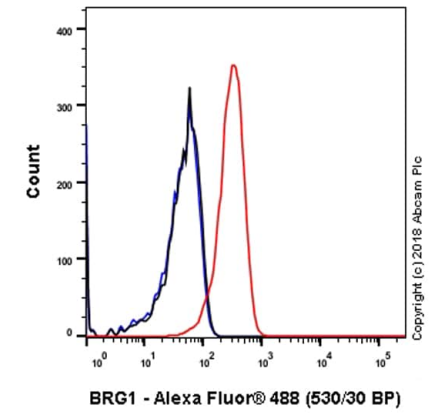
Lanes 1 - 4: Merged signal (red and green). Red - loading control, **ab18058** (unpurified), observed at 124 kDa.

ab108318 was shown to specifically react with BRG1 in wild-type HAP1 cells along with additional cross reactive bands. No band was observed when BRG1 knockout samples were used. Wild-type and BRG1 knockout samples were subjected to SDS-PAGE, **ab108318** and **ab18058** (loading control to Vinculin) were diluted 1/1000 and 1/10,000 respectively 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.



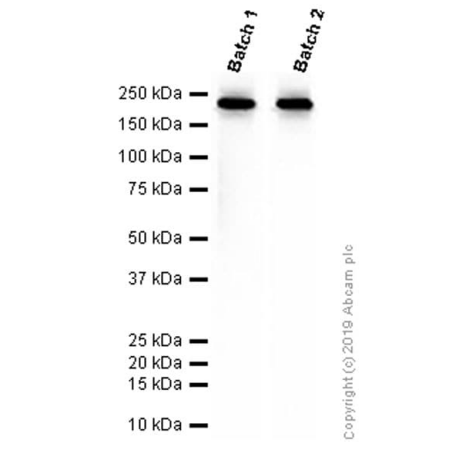
Immunocytochemistry/ Immunofluorescence - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

Immunocytochemistry/ Immunofluorescence analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling BRG1 with purified **ab108318** at 1/50 dilution (2.3 µg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) at 1/200 (2.5 µg/ml). Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1/1000 (2 µg/ml) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab108318**).



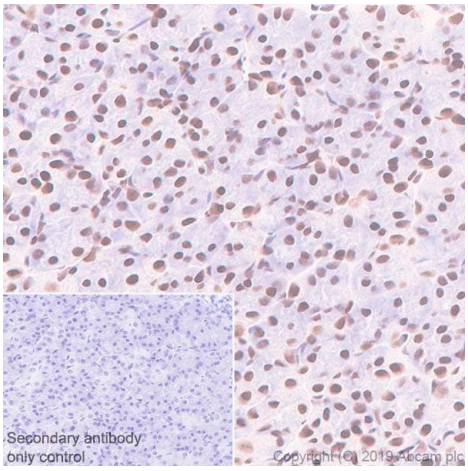
Flow Cytometry (Intracellular) - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

Intracellular Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling BRG1 with purified **ab108318** at 1/20 dilution (10µg/ml) (red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue). This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab108318**).



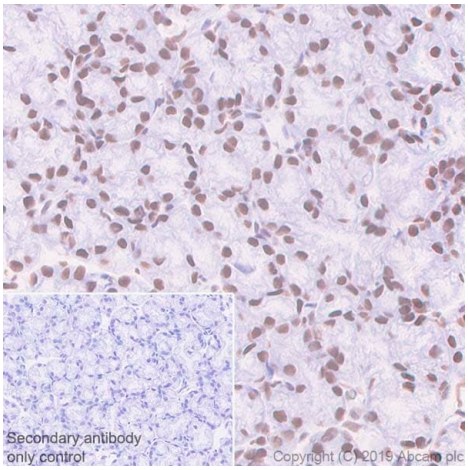
Western blot - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

This data was developed using **ab108318**, the same antibody clone in a different buffer formulation. Different batches of **ab108318** were tested on K-562 (Human chronic myelogenous leukemia lymphoblast) lysate at 0.05 µg/ml. 15 µg of lysate was loaded in each lane. Bands observed at 185 kDa.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

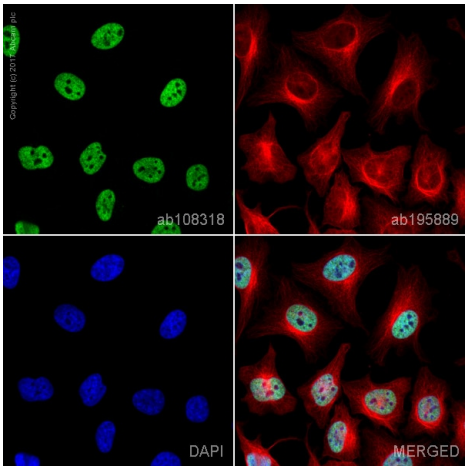
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Rat stomach tissue sections labeling BRG1 with purified **ab108318** at 1:500 dilution (0.23 µg/ml). Heat mediated antigen retrieval was performed using **ab93684** (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab108318**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Mouse stomach tissue sections labeling BRG1 with purified **ab108318** at 1/500 dilution (0.23 µg/ml). Heat mediated antigen retrieval was performed using **ab93684** (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab108318**).

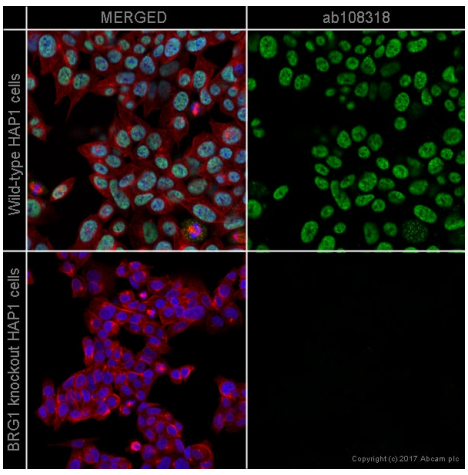


Immunocytochemistry/ Immunofluorescence - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

ab108318 (unpurified) 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 **ab108318** 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).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab108318**).

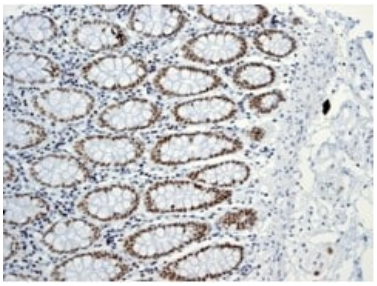


Immunocytochemistry/ Immunofluorescence - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

ab108318 (unpurified) 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 **ab108318** 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).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab108318**).

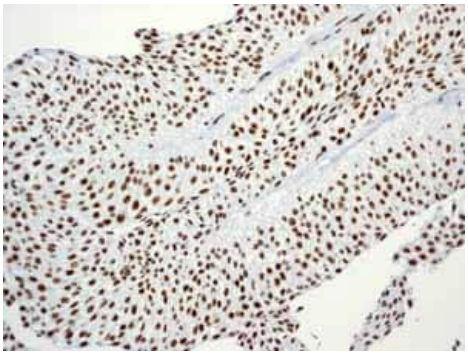


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

ab108318 (unpurified) at 1/100 staining BRG1 in paraffin-embedded Human colon tissue.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab108318**).

Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.

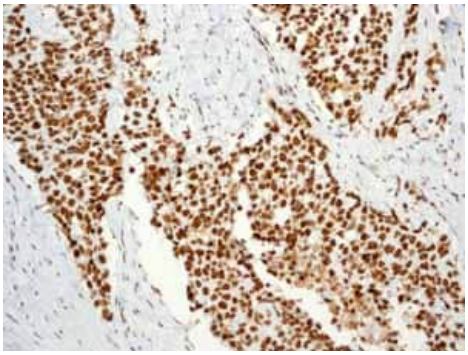


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

ab108318 (unpurified) showing positive staining in Urinary bladder transitional carcinoma tissue.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab108318**).

Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.

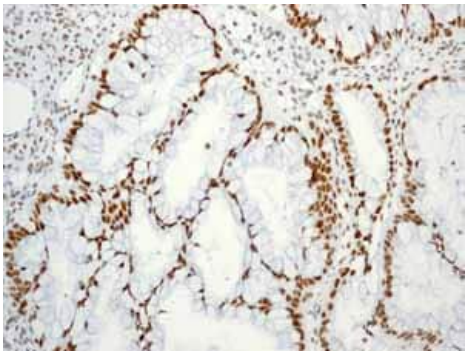


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

ab108318 (unpurified) showing positive staining in Breast carcinoma tissue.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab108318**).

Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.

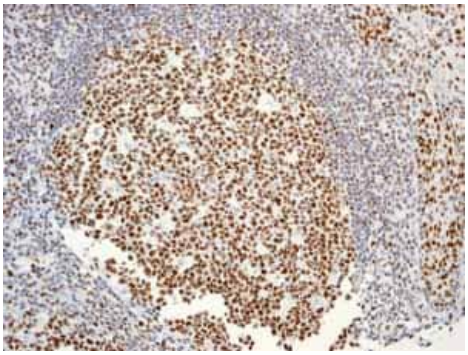


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

ab108318 (unpurified) showing positive staining in Ovarian carcinoma tissue.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab108318**).

Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.

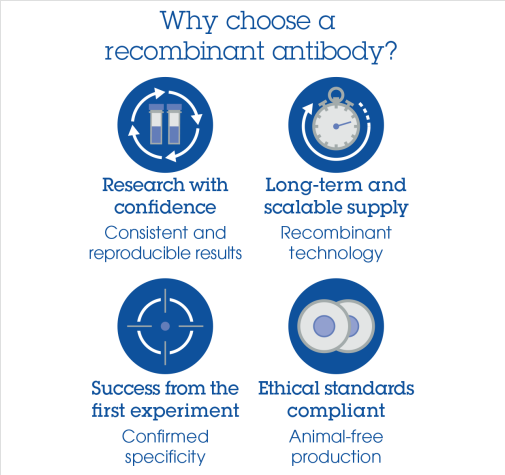


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

ab108318 (unpurified) showing positive staining in Normal tonsil tissue.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab108318**).

Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



Anti-BRG1 antibody [EPR3912] - BSA and Azide free (ab222230)

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit **https://www.abcam.com/abpromise** or contact our technical team.

Terms and conditions

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors