10320 Camino Santa Fe, Suite G
San Diego, CA 92121
Tel: 858.875.1900 Fax: 858.875.1999

Anti-BRG1 SMARCA4 Antibody Picoband™ (monoclonal, 3F4)

Catalog # ABO14862

Specification

Anti-BRG1 SMARCA4 Antibody Picoband™ (monoclonal, 3F4) - Product Information

Application	**WB, IHC, FC**
Primary Accession	P51532
Host	**Mouse**
Isotype	**Mouse IgG1**
Reactivity	**Rat, Human, Mouse**
Clonality	**Monoclonal**
Format	**Lyophilized**

Description
Anti-BRG1 SMARCA4 Antibody Picoband™ (monoclonal, 3F4) . Tested in Flow Cytometry, IHC, WB applications. This antibody reacts with Human, Mouse, Rat.

Reconstitution
Add 0.2ml of distilled water will yield a concentration of 500 µg/ml.

Anti-BRG1 SMARCA4 Antibody Picoband™ (monoclonal, 3F4) - Additional Information

Gene ID 6597

Other Names
Transcription activator BRG1, 3.6.4.-, ATP-dependent helicase SMARCA4, BRG1-associated factor 190A, BAF190A, Mitotic growth and transcription activator, Protein BRG-1, Protein brahma homolog 1, SNF2-beta, SWI/SNF-related matrix-associated actin-dependent regulator of chromatin subfamily A member 4, SMARCA4, BAF190A, BRG1, SNF2B, SNF2L4

Calculated MW
181 kDa KDa

Application Details
Western blot, 0.1-0.5 µg/ml
 Immunohistochemistry (Paraffin-embedded Section), 0.5-1 µg/ml
 Flow Cytometry, 1-3 µg/1x10^6 cells

Subcellular Localization
Nucleus.

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).

Contents
Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.05mg NaN3.

Immunogen
E. coli-derived human BRG1 recombinant protein (Position: Q555-E763).

Cross Reactivity
No cross-reactivity with other proteins.

Storage

Store at -20˚C for one year from date of receipt. After reconstitution, at 4˚C for one month. It can also be aliquotted and stored frozen at -20˚C for six months. Avoid repeated freeze-thaw cycles.

Anti-BRG1 SMARCA4 Antibody Picoband™ (monoclonal, 3F4) - Protein Information

Name SMARCA4

Synonyms BAF190A, BRG1, SNF2B, SNF2L4

Function
Involved in transcriptional activation and repression of select genes by chromatin remodeling (alteration of DNA-nucleosome topology). Component of SWI/SNF chromatin remodeling complexes that carry out key enzymatic activities, changing chromatin structure by altering DNA-histone contacts within a nucleosome in an ATP-dependent manner. Component of the CREST-BRG1 complex, a multiprotein complex that regulates promoter activation by orchestrating the calcium- dependent release of a repressor complex and the recruitment of an activator complex. In resting neurons, transcription of the c-FOS promoter is inhibited by SMARCA4-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 the 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 postmitotic 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 postmitotic 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). Acts as a corepressor of ZEB1 to regulate E-cadherin transcription and is required for induction of epithelial-mesenchymal transition (EMT) by ZEB1. Binds via DLX1 to enhancers located in the intergenic region between DLX5 and DLX6 and this binding is stabilized by the long non-coding RNA (lncRNA) Evf2 (By similarity). Binds to RNA in a promiscuous manner (By similarity). Binding to RNAs including lncRNA Evf2 leads to inhibition of SMARCA4 ATPase and chromatin remodeling activities (By similarity). In brown adipose tissue, involved in the regulation of thermogenic genes expression (By similarity).

Cellular Location
Nucleus {ECO:0000255|PROSITE-ProRule:PRU00549, ECO:0000269|PubMed:20418909, ECO:0000269|PubMed:25593309} Note=Colocalizes with long non-coding RNA Evf2 in nuclear RNA clouds (By similarity). Localizes to sites of DNA damage (PubMed:25593309) {ECO:0000250|UniProtKB:Q3TKT4, ECO:0000269|PubMed:25593309}

Tissue Location

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).

Anti-BRG1 SMARCA4 Antibody Picoband™ (monoclonal, 3F4) - Protocols

Provided below are standard protocols that you may find useful for product applications.

- Western Blot
- Blocking Peptides
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- Cell Culture

Anti-BRG1 SMARCA4 Antibody Picoband™ (monoclonal, 3F4) - Images

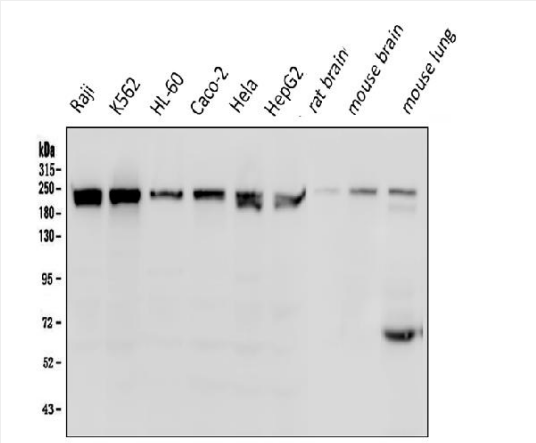

Figure 1. Western blot analysis of BRG1 using anti-BRG1 antibody (M00223-1).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.
Lane 1: human Raji whole cell lysates;
Lane 2: human K562 whole cell lysates;
Lane 3: human HL-60 whole cell lysates;
Lane 4: human Caco-2 whole cell lysates;
Lane 5: human Hela whole cell lysates;
Lane 6: human HepG2 whole cell lysates;
Lane 7: rat brain tissue lysates;
Lane 8: mouse brain tissue lysates;
Lane 9: mouse lung tissue lysates.
After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with mouse anti-BRG1 antigen affinity purified monoclonal antibody (Catalog # M00223-1) at 0.5 µg/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-mouse IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1001) with Tanon 5200 system. A specific band was detected for

BRG1 at approximately 181KD. The expected band size for BRG1 is at 181KD.

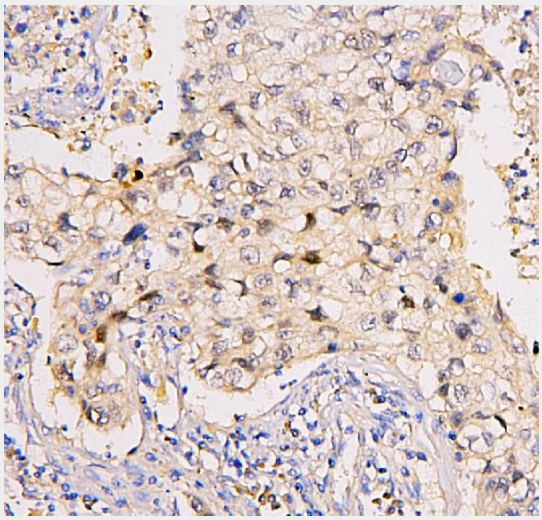

Figure 2. IHC analysis of BRG1 using anti-BRG1 antibody (M00223-1).
BRG1 was detected in paraffin-embedded section of human lung cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-BRG1 Antibody (M00223-1) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

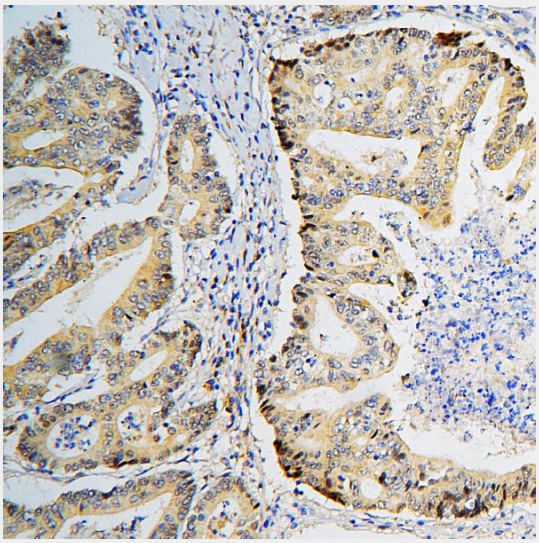

Figure 3. IHC analysis of BRG1 using anti-BRG1 antibody (M00223-1).
BRG1 was detected in paraffin-embedded section of human rectal cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-BRG1 Antibody (M00223-1) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

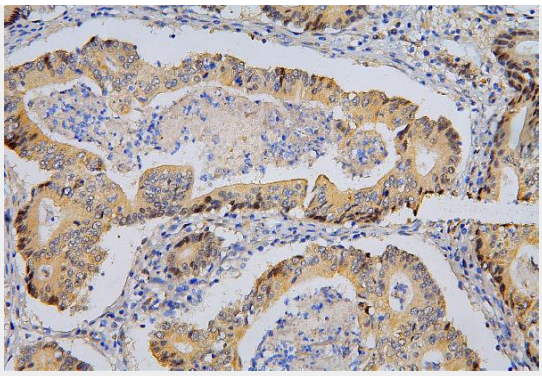

Figure 4. IHC analysis of BRG1 using anti-BRG1 antibody (M00223-1).
BRG1 was detected in paraffin-embedded section of human rectal cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-BRG1 Antibody (M00223-1) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

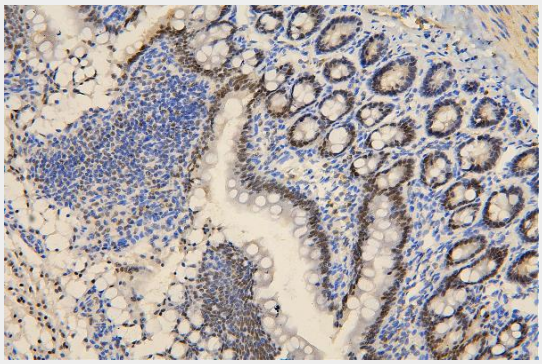

Figure 5. IHC analysis of BRG1 using anti-BRG1 antibody (M00223-1).
BRG1 was detected in paraffin-embedded section of rat intestine tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-BRG1 Antibody (M00223-1) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

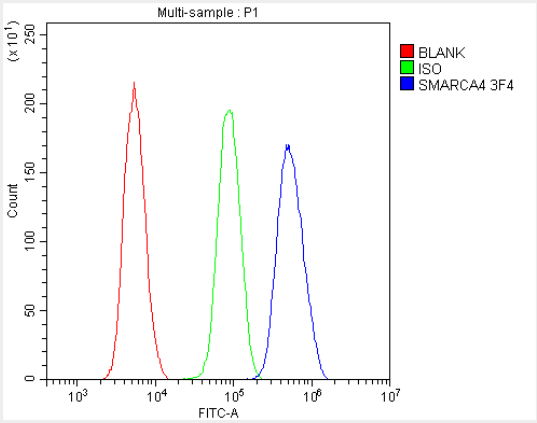

Figure 6. Flow Cytometry analysis of U20S cells using anti-BRG1 antibody (M00223-1).
Overlay histogram showing U20S cells stained with M00223-1 (Blue line).The cells were blocked

with 10% normal goat serum. And then incubated with mouse anti-BRG1 Antibody (M00223-1, 1 µg/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

Anti-BRG1 SMARCA4 Antibody Picoband™ (monoclonal, 3F4) - Background

Transcription activator BRG1 also known as ATP-dependent helicase SMARCA4 is a protein that in humans is encoded by the SMARCA4 gene. The protein encoded by this gene is a member of the SWI/SNF family of proteins and is similar to the brahma protein of Drosophila. Members of this family have helicase and ATPase activities and are thought to regulate transcription of certain genes by altering the chromatin structure around those genes. The encoded protein is part of the large ATP-dependent chromatin remodeling complex SNF/SWI, which is required for transcriptional activation of genes normally repressed by chromatin. In addition, this protein can bind BRCA1, as well as regulate the expression of the tumorigenic protein CD44. Mutations in this gene cause rhabdoid tumor predisposition syndrome type 2. Multiple transcript variants encoding different isoforms have been found for this gene.