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Anti-RbAp48 RBBP4 Antibody Picoband™ (monoclonal, 9F3)

Catalog # ABO14782

Product Information

Application	WB, IHC
Primary Accession	Q09028
Host	Mouse
Isotype	Mouse IgG1
Reactivity	Rat, Human, Mouse
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-RbAp48 RBBP4 Antibody Picoband™ (monoclonal, 9F3) . Tested in IHC, WB applications. This antibody reacts with Human, Mouse, Rat.
Reconstitution	Add 0.2ml of distilled water will yield a concentration of 500ug/ml.

Additional Information

Gene ID	5928
Other Names	Histone-binding protein RBBP4, Chromatin assembly factor 1 subunit C, CAF-1 subunit C, Chromatin assembly factor I p48 subunit, CAF-I 48 kDa subunit, CAF-I p48, Nucleosome-remodeling factor subunit RBAP48, Retinoblastoma-binding protein 4, RBBP-4, Retinoblastoma-binding protein p48, RBBP4, RBAP48
Calculated MW	47656
Application Details	Western blot, 0.1-0.5 □g/ml Immunohistochemistry (Paraffin-embedded Section), 0.5-1 □g/ml
Subcellular Localization	Nucleus.
Source	Eukaryota
Contents	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.05mg NaN3.
Clone Names	Clone: 9F3
Immunogen	A synthetic peptide corresponding to a sequence at the C-terminus of human RbAp48, identical to the related mouse sequence.
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.

Protein Information

Name	RBBP4
Synonyms	RBAP48
Function	Core histone-binding subunit that may target chromatin assembly factors, chromatin remodeling factors and histone deacetylases to their histone substrates in a manner that is regulated by nucleosomal DNA (PubMed:10866654). Component of the chromatin assembly factor 1 (CAF-1) complex, which is required for chromatin assembly following DNA replication and DNA repair (PubMed:8858152). Component of the core histone deacetylase (HDAC) complex, which promotes histone deacetylation and consequent transcriptional repression (PubMed:9150135). Component of the nucleosome remodeling and histone deacetylase complex (the NuRD complex), which promotes transcriptional repression by histone deacetylation and nucleosome remodeling (PubMed:16428440, PubMed:28977666, PubMed:39460621). Component of the PRC2 complex, which promotes repression of homeotic genes during development (PubMed:29499137, PubMed:31959557). Component of the NURF (nucleosome remodeling factor) complex (PubMed:14609955, PubMed:15310751).
Cellular Location	Nucleus. Chromosome, telomere. Note=Localizes to chromatin as part of the PRC2 complex.
Tissue Location	Expressed in neuroblastoma cells.

Background

Histone-binding protein RBBP4 (also known as RbAp48, or NURF55) is a protein that in humans is encoded by the RBBP4 gene. This gene encodes a ubiquitously expressed nuclear protein which belongs to a highly conserved subfamily of WD-repeat proteins. It is present in protein complexes involved in histone acetylation and chromatin assembly. And it is part of the Mi-2 complex which has been implicated in chromatin remodeling and transcriptional repression associated with histone deacetylation. This encoded protein is also part of co-repressor complexes, which is an integral component of transcriptional silencing. It is found among several cellular proteins that bind directly to retinoblastoma protein to regulate cell proliferation. This protein also seems to be involved in transcriptional repression of E2F-responsive genes. Three transcript variants encoding different isoforms have been found for this gene.

Images

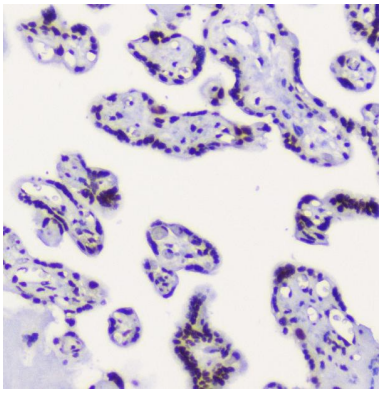


Figure 2. IHC analysis of RbAp48 using anti-RbAp48 antibody (M02702-1).
RbAp48 was detected in paraffin-embedded section of human placenta tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-RbAp48 Antibody (M02702-1) 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.

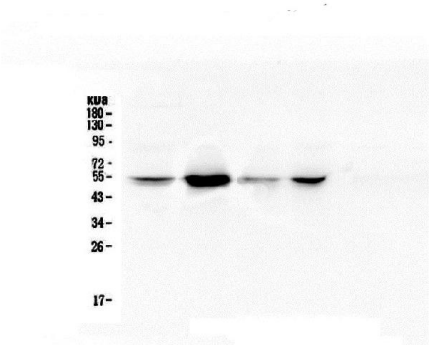


Figure 3. Western blot analysis of RbAp48 using anti-RbAp48 antibody (M02702-1).
Electrophoresis was performed on a 10% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.
Lane 1: human A549 whole cell lysate,
Lane 2: human Jurkat whole cell lysate,
Lane 3: human Hela whole cell lysate,
Lane 4: human PANC-1 whole cell lysate.
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-RbAp48 antigen affinity purified monoclonal antibody (Catalog # M02702-1) 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.

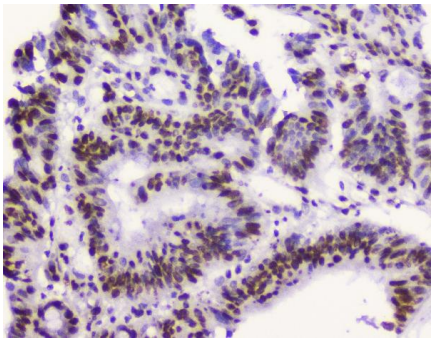


Figure 1. IHC analysis of RbAp48 using anti-RbAp48 antibody (M02702-1).
RbAp48 was detected in paraffin-embedded section of human intestinal cancer tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-RbAp48 Antibody (M02702-1) 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.

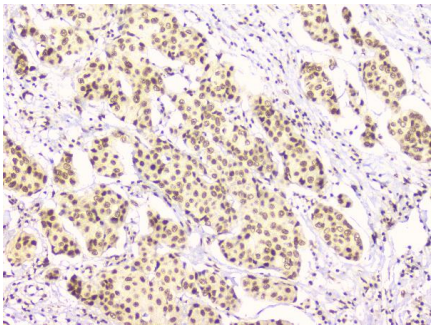
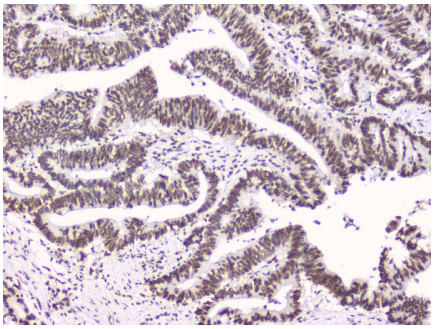


Figure 4. IHC analysis of RbAp48 using anti-RbAp48 antibody (M02702-1).
RbAp48 was detected in paraffin-embedded section of human mammary cancer tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-RbAp48 Antibody (M02702-1) 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 RbAp48 using anti-RbAp48 antibody (M02702-1).
RbAp48 was detected in paraffin-embedded section of human intestinal cancer tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was



blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-RbAp48 Antibody (M02702-1) 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.

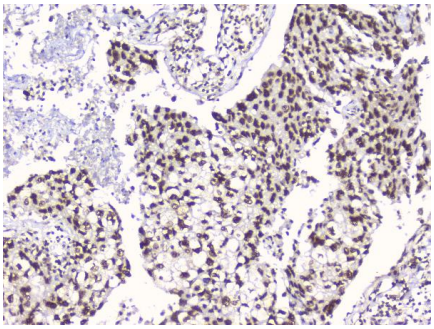


Figure 6. IHC analysis of RbAp48 using anti-RbAp48 antibody (M02702-1).
RbAp48 was detected in paraffin-embedded section of human lung cancer tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-RbAp48 Antibody (M02702-1) 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.

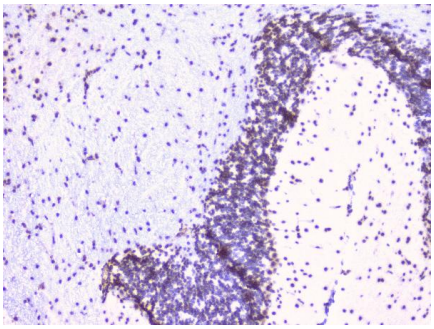


Figure 7. IHC analysis of RbAp48 using anti-RbAp48 antibody (M02702-1).
RbAp48 was detected in paraffin-embedded section of mouse brain tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-RbAp48 Antibody (M02702-1) 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.

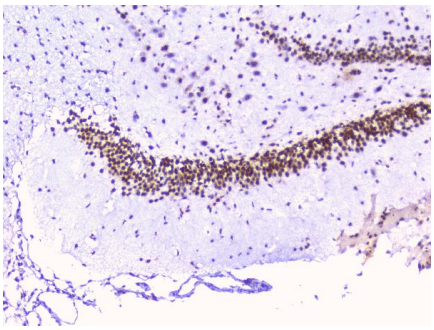
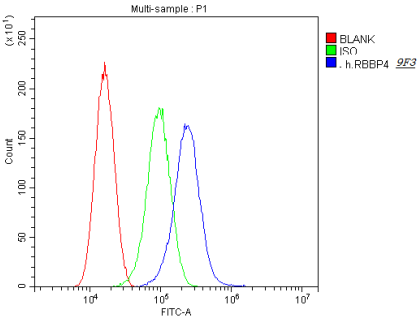


Figure 8. IHC analysis of RbAp48 using anti-RbAp48 antibody (M02702-1).
RbAp48 was detected in paraffin-embedded section of rat brain tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-RbAp48 Antibody (M02702-1) 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 9. Flow Cytometry analysis of SiHa cells using anti-RbAp48 antibody (M02702-1).
Overlay histogram showing SiHa cells stained with M02702-1 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-RbAp48 Antibody (M02702-1,1 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 rabbit IgG (1 µg/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

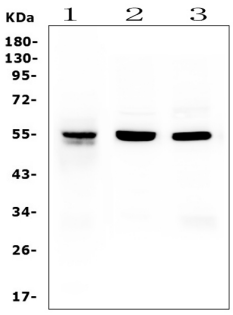


Figure 10. Western blot analysis of RbAp48 using anti-RbAp48 antibody (M02702-1).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.
Lane 1: human Jurkat whole cell lysates
Lane 2: rat thymus tissue lysates
Lane 3: mouse spleen 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-RbAp48 antigen affinity purified monoclonal antibody (Catalog # M02702-1) 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 RbAp48 at approximately 55KD. The expected band size for RbAp48 is at 48KD.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.