10320 Camino Santa Fe, Suite G
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Anti-hnRNP D/AUF1/HNRNPD Antibody Picoband™ (monoclonal, 2B12)

Catalog # ABO14968

Specification

Anti-hnRNP D/AUF1/HNRNPD Antibody Picoband™ (monoclonal, 2B12) - Product Information

Application	**WB, IHC, IF, ICC, FC**
Primary Accession	Q14103
Host	**Mouse**
Isotype	**Mouse IgG2a**
Reactivity	**Rat, Human, Mouse**
Clonality	**Monoclonal**
Format	**Lyophilized**

Description
Anti-hnRNP D/AUF1/HNRNPD Antibody Picoband™ (monoclonal, 2B12) . Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.

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

Anti-hnRNP D/AUF1/HNRNPD Antibody Picoband™ (monoclonal, 2B12) - Additional Information

Gene ID 3184

Other Names
Heterogeneous nuclear ribonucleoprotein D0, hnRNP D0, AU-rich element RNA-binding protein 1, HNRNPD, AUF1, HNRPD

Calculated MW
43-45 kDa KDa

Application Details
Western blot, 0.1-0.5 µg/ml, Human, Mouse, Rat
 Immunohistochemistry (Paraffin-embedded Section), 0.5-1 µg/ml, Human, Mouse, Rat
Immunocytochemistry/Immunofluorescence, 4 µg/ml, Human
Immunofluorescence, 4 µg/ml, Human
Flow Cytometry, 1-3 µg/1x10^6 cells, Human

Contents
Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.01mg NaN3.

Immunogen
E.coli-derived human hnRNP D/AUF1/HNRNPD recombinant protein (Position: E88-N246).

Purification
Immunogen affinity purified.

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-hnRNP D/AUF1/HNRNPD Antibody Picoband™ (monoclonal, 2B12) - Protein Information

Name HNRNPD

Synonyms AUF1, HNRPD

Function
Binds with high affinity to RNA molecules that contain AU- rich elements (AREs) found within the 3'-UTR of many proto-oncogenes and cytokine mRNAs. Also binds to double- and single-stranded DNA sequences in a specific manner and functions a transcription factor. Each of the RNA-binding domains specifically can bind solely to a single-stranded non-monotonous 5'-UUAG-3' sequence and also weaker to the single-stranded 5'-TTAGGG-3' telomeric DNA repeat. Binds RNA oligonucleotides with 5'-UUAGGG-3' repeats more tightly than the telomeric single-stranded DNA 5'-TTAGGG-3' repeats. Binding of RRM1 to DNA inhibits the formation of DNA quadruplex structure which may play a role in telomere elongation. May be involved in translationally coupled mRNA turnover. Implicated with other RNA-binding proteins in the cytoplasmic deadenylation/translational and decay interplay of the FOS mRNA mediated by the major coding-region determinant of instability (mCRD) domain. May play a role in the regulation of the rhythmic expression of circadian clock core genes. Directly binds to the 3'UTR of CRY1 mRNA and induces CRY1 rhythmic translation. May also be involved in the regulation of PER2 translation.

Cellular Location
Nucleus. Cytoplasm. Note=Localized in cytoplasmic mRNP granules containing untranslated mRNAs. Component of ribonucleosomes. Cytoplasmic localization oscillates diurnally

Anti-hnRNP D/AUF1/HNRNPD Antibody Picoband™ (monoclonal, 2B12) - 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-hnRNP D/AUF1/HNRNPD Antibody Picoband™ (monoclonal, 2B12) - Images

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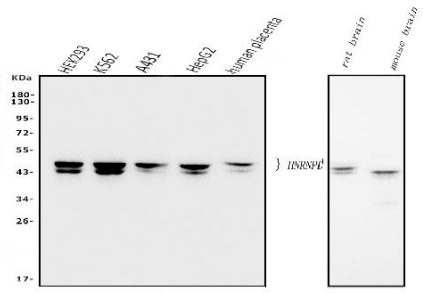

Figure 1. Western blot analysis of hnRNP D/AUF1/HNRNPD using anti-hnRNP D/AUF1/HNRNPD antibody (M09982).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.
Lane 1: human HEK293 whole cell lysates,
Lane 2: human K562 whole cell lysates,
Lane 3: human A431 whole cell lysates,
Lane 4: human HEPG2 whole cell lysates,
Lane 5: human placenta tissue lysates,
Lane 6: rat brain tissue lysates,
Lane 7: mouse brain 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-hnRNP D/AUF1/HNRNPD antigen affinity purified monoclonal antibody (Catalog # M09982) 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 hnRNP D/AUF1/HNRNPD at approximately 43-45KD. The expected band size for hnRNP D/AUF1/HNRNPD is at 38KD.

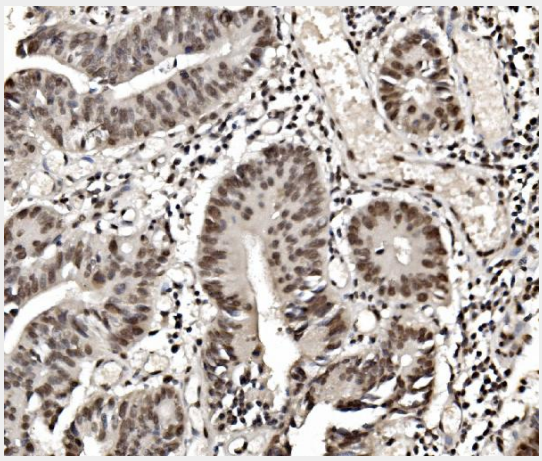

Figure 2. IHC analysis of hnRNP D/AUF1/HNRNPD using anti-hnRNP D/AUF1/HNRNPD antibody (M09982).
hnRNP D/AUF1/HNRNPD was detected in paraffin-embedded section of human rectal cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval

solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-hnRNP D/AUF1/HNRNPD Antibody (M09982) 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.

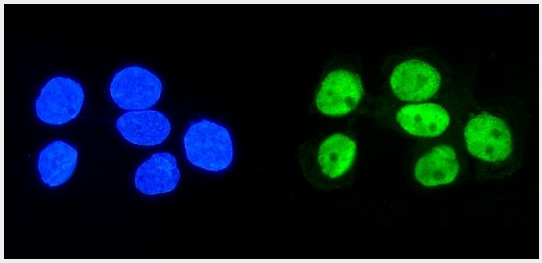

Figure 3. IF analysis of hnRNP D/AUF1/HNRNPD using anti-hnRNP D/AUF1/HNRNPD antibody (M09982).
hnRNP D/AUF1/HNRNPD was detected in immunocytochemical section of A431 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 4 µg/mL mouse anti-hnRNP D/AUF1/HNRNPD Antibody (M09982) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Mouse IgG (BA1126) was used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

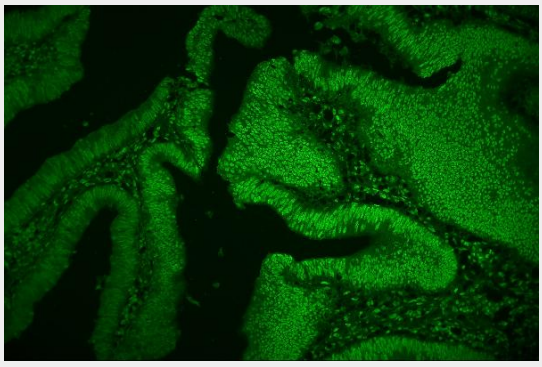

Figure 4. IF analysis of hnRNP D/AUF1/HNRNPD using anti-hnRNP D/AUF1/HNRNPD antibody (M09982).
hnRNP D/AUF1/HNRNPD was detected in paraffin-embedded section of human rectal cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 4 µg/mL mouse anti-hnRNP D/AUF1/HNRNPD Antibody (M09982) overnight at 4°C. Biotin conjugated goat anti-mouse IgG (BA1001) was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using DyLight®488 Conjugated Avidin (BA1128). The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

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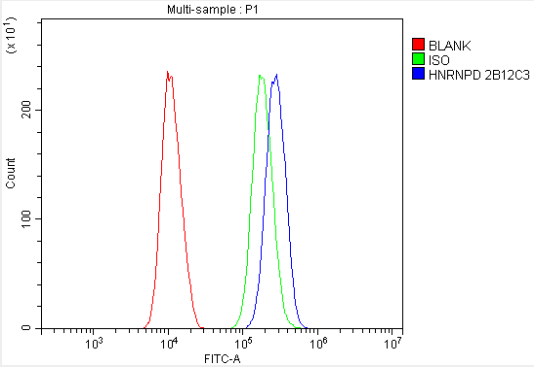

Figure 5. Flow Cytometry analysis of SiHa cells using anti-hnRNP D/AUF1/HNRNPD antibody (M09982).
Overlay histogram showing SiHa cells stained with M09982 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-hnRNP D/AUF1/HNRNPD Antibody (M09982, 1 µg/1x10^6 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.

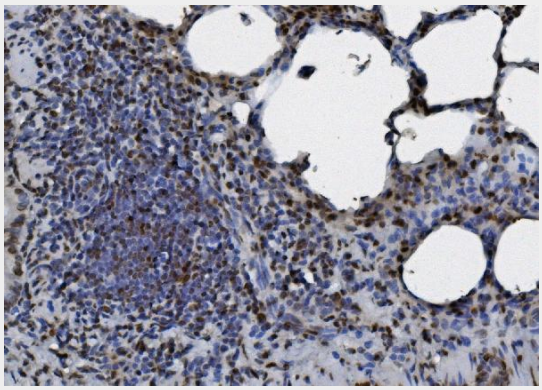

Figure 6. IHC analysis of hnRNP D/AUF1/HNRNPD using anti-hnRNP D/AUF1/HNRNPD antibody (M09982).
hnRNP D/AUF1/HNRNPD was detected in paraffin-embedded section of rat lung tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-hnRNP D/AUF1/HNRNPD Antibody (M09982) 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.

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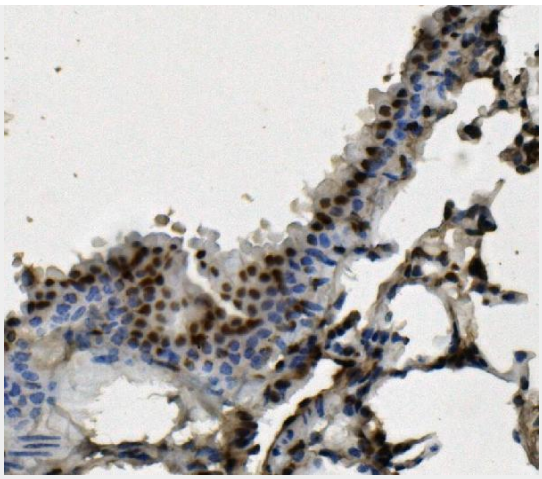

Figure 7. IHC analysis of hnRNP D/AUF1/HNRNPD using anti-hnRNP D/AUF1/HNRNPD antibody (M09982).
hnRNP D/AUF1/HNRNPD was detected in paraffin-embedded section of mouse lung tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-hnRNP D/AUF1/HNRNPD Antibody (M09982) 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.

Anti-hnRNP D/AUF1/HNRNPD Antibody Picoband™ (monoclonal, 2B12) - Background

Heterogeneous nuclear ribonucleoprotein D0 (HNRNPD) also known as AU-rich element RNA-binding protein 1 (AUF1) is a protein that in humans is encoded by the HNRNPD gene. It is mapped to 4q21.22. This gene belongs to the subfamily of ubiquitously expressed heterogeneous nuclear ribonucleoproteins (hnRNPs). The hnRNPs are nucleic acid binding proteins and they complex with heterogeneous nuclear RNA (hnRNA). These proteins are associated with pre-mRNAs in the nucleus and appear to influence pre-mRNA processing and other aspects of mRNA metabolism and transport. While all of the hnRNPs are present in the nucleus, some seem to shuttle between the nucleus and the cytoplasm. The hnRNP proteins have distinct nucleic acid binding properties. The protein encoded by this gene has two repeats of quasi-RRM domains that bind to RNAs. It localizes to both the nucleus and the cytoplasm. This protein is implicated in the regulation of mRNA stability. Alternative splicing of this gene results in four transcript variants.