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Anti-RENT1/hUPF1 Antibody Picoband™ (monoclonal, 11E7)

Catalog # ABO14937

Product Information

Application	WB, IHC, IF, ICC
Primary Accession	Q92900
Host	Mouse
Isotype	Mouse IgG2b
Reactivity	Rat, Human, Mouse
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-RENT1/hUPF1 Antibody Picoband™ (monoclonal, 11E7) . Tested in 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.

Additional Information

Gene ID	5976
Other Names	Regulator of nonsense transcripts 1 {ECO:0000312|HGNC:HGNC:9962}, 3.6.4.12, 3.6.4.13, ATP-dependent helicase RENT1, Up-frameshift suppressor 1 homolog, hUpf1, UPF1 (HGNC:9962)
Calculated MW	124345
Application Details	Western blot, 0.1-0.5 □g/ml, Human, Mouse, Rat Immunohistochemistry (Paraffin-embedded Section), 0.5-1 □g/ml, Human, Mouse, Rat Immunocytochemistry/Immunofluorescence, 2 □g/ml, Human
Subcellular Localization	Nucleus. Cytoplasm. P-body.
Tissue Specificity	Ubiquitous.
Source	Eukaryota
Contents	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na_2HPO_4, 0.05mg NaN_3.
Clone Names	Clone: 11E7
Immunogen	A synthetic peptide corresponding to a sequence in the middle region of human RENT1/hUPF1, identical to the related mouse and rat sequences.
Purification	Immunogen affinity purified.

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	UPF1 (HGNC:9962)
Function	RNA-dependent helicase required for nonsense-mediated decay (NMD) of aberrant mRNAs containing premature stop codons and which modulates the expression level of normal mRNAs (PubMed:11163187, PubMed:16086026, PubMed:18172165, PubMed:21145460, PubMed:21419344, PubMed:24726324). ATP-dependent 5'-3' helicase active on RNA:DNA substrates with a 5'-RNA extension (PubMed:18066079, PubMed:21419344, PubMed:23275559, PubMed:26138914). ATP-dependent 5'-3' helicase active on RNA:RNA substrates with a 5'-RNA extension (PubMed:26138914). ATP-dependent 5'-3' helicase active on DNA:DNA substrates with a 5'-DNA extension (PubMed:10999600, PubMed:26138914, PubMed:30218034). Is recruited to mRNAs upon translation termination and undergoes a cycle of phosphorylation and dephosphorylation; its phosphorylation appears to be a key step in NMD (PubMed:11544179, PubMed:25220460). Recruited by release factors to stalled ribosomes together with the SMG1C protein kinase complex to form the transient SURF (SMG1-UPF1-eRF1-eRF3) complex (PubMed:19417104). In EJC-dependent NMD, the SURF complex associates with the exon junction complex (EJC) (located 50-55 or more nucleotides downstream from the termination codon) through UPF2 and allows the formation of an UPF1-UPF2-UPF3 surveillance complex which is believed to activate NMD (PubMed:21419344). Phosphorylated UPF1 is recognized by EST1B/SMG5, SMG6 and SMG7 which are thought to provide a link to the mRNA degradation machinery involving exonucleolytic and endonucleolytic pathways, and to serve as adapters to protein phosphatase 2A (PP2A), thereby triggering UPF1 dephosphorylation and allowing the recycling of NMD factors (PubMed:12554878). UPF1 can also activate NMD without UPF2 or UPF3, and in the absence of the NMD-enhancing downstream EJC indicative for alternative NMD pathways (PubMed:18447585). Plays a role in replication-dependent histone mRNA degradation at the end of phase S; the function is independent of UPF2 (PubMed:16086026, PubMed:18172165). For the recognition of premature termination codons (PTC) and initiation of NMD a competitive interaction between UPF1 and PABPC1 with the ribosome-bound release factors is proposed (PubMed:18447585, PubMed:25220460). The ATPase activity of UPF1 is required for disassembly of mRNPs undergoing NMD (PubMed:21145460). Together with UPF2 and dependent on TDRD6, mediates the degradation of mRNA harboring long 3'UTR by inducing the NMD machinery (By similarity). Has ssDNA-stimulated ATPase activity (poly(dT) and poly(dA) are most stimulatory) (PubMed:10999600). Binds RNA; addition of ATP decreases RNA-binding (PubMed:10999600, PubMed:18066079, PubMed:21419344).
Cellular Location	Cytoplasm. Cytoplasm, P-body. Nucleus. Cytoplasm, perinuclear region {ECO:0000250|UniProtKB:Q9EPU0} Note=Hyperphosphorylated form is targeted to the P-body, while unphosphorylated protein is distributed throughout the cytoplasm Localized in the chromatoid bodies of round spermatids (By similarity) {ECO:0000250|UniProtKB:Q9EPU0}
Tissue Location	Ubiquitous.

Background

Regulator of nonsense transcripts 1 is a protein that in humans is encoded by the UPF1 gene. This gene encodes a protein that is part of a post-splicing multiprotein complex involved in both mRNA nuclear export and mRNA surveillance. mRNA surveillance detects exported mRNAs with truncated open reading frames and initiates nonsense-mediated mRNA decay (NMD). When translation ends upstream from the last exon-exon junction, this triggers NMD to degrade mRNAs containing premature stop codons. And this protein is located only in the cytoplasm. When translation ends, it interacts with the protein that is a functional homolog of yeast Upf2p to trigger mRNA decapping. Use of multiple polyadenylation sites has been noted for this gene. Alternative splicing results in multiple transcript variants.

Images

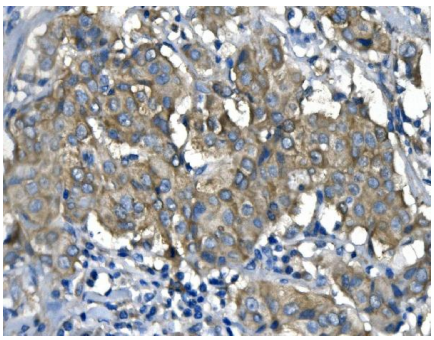


Figure 2. IHC analysis of RENT1/hUPF1 using anti-RENT1/hUPF1 antibody (M00900).
RENT1/hUPF1 was detected in paraffin-embedded section of human mammary cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-RENT1/hUPF1 Antibody (M00900) 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.

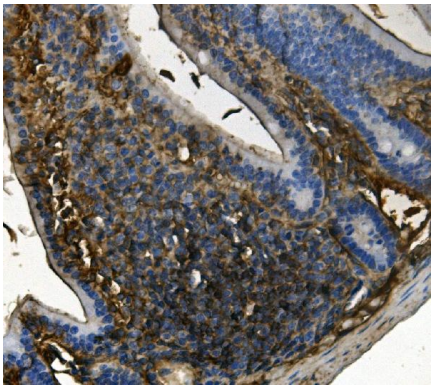


Figure 3. IHC analysis of RENT1/hUPF1 using anti-RENT1/hUPF1 antibody (M00900).
RENT1/hUPF1 was detected in paraffin-embedded section of mouse intestine tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-RENT1/hUPF1 Antibody (M00900) 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.

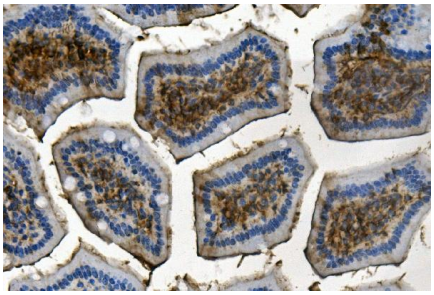
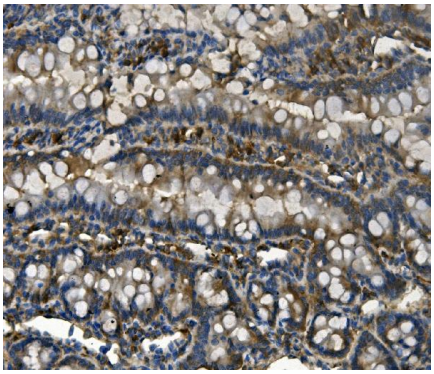


Figure 4. IHC analysis of RENT1/hUPF1 using anti-RENT1/hUPF1 antibody (M00900).
RENT1/hUPF1 was detected in paraffin-embedded section of mouse intestine tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-RENT1/hUPF1 Antibody (M00900) 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 RENT1/hUPF1 using anti-RENT1/hUPF1 antibody (M00900).



RENT1/hUPF1 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-RENT1/hUPF1 Antibody (M00900) 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.

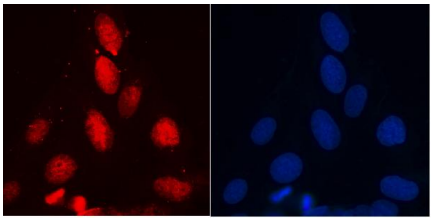


Figure 6. IF analysis of RENT1/hUPF1 using anti-RENT1/hUPF1 antibody (M00900).
RENT1/hUPF1 was detected in immunocytochemical section of U20S cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 2 µg/mL mouse anti-RENT1/hUPF1 Antibody (M00900) overnight at 4°C. DyLight®594 Conjugated Goat Anti-Mouse IgG (BA1141) 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.

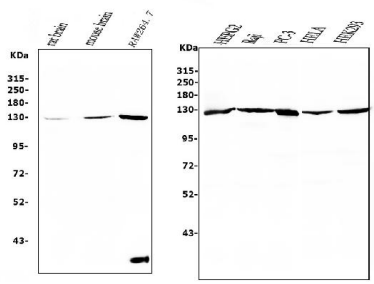


Figure 1. Western blot analysis of RENT1/hUPF1 using anti-RENT1/hUPF1 antibody (M00900).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.
Lane 1: rat brain tissue lysates;
Lane 2: mouse brain tissue lysates;
Lane 3: human RAW264.7 whole cell lysates;
Lane 4: human HepG2 whole cell lysates;
Lane 5: human Raji whole cell lysates;
Lane 6: human PC-3 whole cell lysates;
Lane 7: human Hela whole cell lysates;
Lane 8: human HEK293 whole cell 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-RENT1/hUPF1 antigen affinity purified monoclonal antibody (Catalog # M00900) 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 RENT1/hUPF1 at approximately 130KD. The expected band size for RENT1/hUPF1 is at 130KD.

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