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Anti-HAUSP/USP7 Antibody Picoband™ (monoclonal, 5E2)

Catalog # ABO14847

Product Information

Application	WB, IHC, IF, ICC, FC
Primary Accession	Q93009
Host	Mouse
Isotype	Mouse IgG2b
Reactivity	Rat, Human, Mouse
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-HAUSP/USP7 Antibody Picoband™ (monoclonal, 5E2) . 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 500 □g/ml.

Additional Information

Gene ID	7874
Other Names	Ubiquitin carboxyl-terminal hydrolase 7, 3.4.19.12, Deubiquitinating enzyme 7, Herpesvirus-associated ubiquitin-specific protease, Ubiquitin thioesterase 7, Ubiquitin-specific-processing protease 7, USP7 {ECO:0000303|PubMed:12093161, ECO:0000312|HGNC:HGNC:12630}
Calculated MW	128302
Application Details	Western blot, 0.1-0.5 □g/ml Immunohistochemistry (Paraffin-embedded Section), 0.5-1 □g/ml Immunocytochemistry/Immunofluorescence, 2 □g/ml Flow Cytometry, 1-3 □g/1x10^6 cells
Subcellular Localization	Nucleus. PML body. Cytoplasm. Chromosome.
Tissue Specificity	Widely expressed. Overexpressed in prostate cancer.
Source	Eukaryota
Contents	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.05mg NaN3.
Clone Names	Clone: Clone: 5E2
Immunogen	E.coli-derived human HAUSP/USP7 recombinant protein (Position: L258-D483).

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	USP7 {ECO:0000303|PubMed:12093161, ECO:0000312|HGNC:HGNC:12630}
Function	Hydrolase that deubiquitinates target proteins such as ARMC5, FOXO4, DEPTOR, KAT5, p53/TP53, MDM2, ERCC6, DNMT1, UHRF1, PTEN, KMT2E/MLL5 and DAXX (PubMed:11923872, PubMed:15053880, PubMed:16964248, PubMed:18716620, PubMed:25283148, PubMed:25865756, PubMed:26678539, PubMed:28655758, PubMed:33544460, PubMed:35216969). Together with DAXX, prevents MDM2 self-ubiquitination and enhances the E3 ligase activity of MDM2 towards p53/TP53, thereby promoting p53/TP53 ubiquitination and proteasomal degradation (PubMed:15053880, PubMed:16845383, PubMed:18566590, PubMed:20153724). Deubiquitinates p53/TP53, preventing degradation of p53/TP53, and enhances p53/TP53-dependent transcription regulation, cell growth repression and apoptosis (PubMed:25283148). Deubiquitinates p53/TP53 and MDM2 and strongly stabilizes p53/TP53 even in the presence of excess MDM2, and also induces p53/TP53-dependent cell growth repression and apoptosis (PubMed:11923872, PubMed:26786098). Deubiquitination of FOXO4 in presence of hydrogen peroxide is not dependent on p53/TP53 and inhibits FOXO4-induced transcriptional activity (PubMed:16964248). In association with DAXX, is involved in the deubiquitination and translocation of PTEN from the nucleus to the cytoplasm, both processes that are counteracted by PML (PubMed:18716620). Deubiquitinates KMT2E/MLL5 preventing KMT2E/MLL5 proteasomal-mediated degradation (PubMed:26678539). Involved in cell proliferation during early embryonic development. Involved in transcription-coupled nucleotide excision repair (TC-NER) in response to UV damage: recruited to DNA damage sites following interaction with KIAA1530/UVSSA and promotes deubiquitination of ERCC6, preventing UV- induced degradation of ERCC6 (PubMed:22466611, PubMed:22466612). Involved in maintenance of DNA methylation via its interaction with UHRF1 and DNMT1: acts by mediating deubiquitination of UHRF1 and DNMT1, preventing their degradation and promoting DNA methylation by DNMT1 (PubMed:21745816, PubMed:22411829). Deubiquitinates alkylation repair enzyme ALKBH3. OTUD4 recruits USP7 and USP9X to stabilize ALKBH3, thereby promoting the repair of alkylated DNA lesions (PubMed:25944111). Acts as a chromatin regulator via its association with the Polycomb group (PcG) multiprotein PRC1-like complex; may act by deubiquitinating components of the PRC1-like complex (PubMed:20601937). Able to mediate deubiquitination of histone H2B; it is however unsure whether this activity takes place in vivo (PubMed:20601937). Exhibits a preference towards 'Lys-48'-linked ubiquitin chains (PubMed:22689415). Increases regulatory T-cells (Treg) suppressive capacity by deubiquitinating and stabilizing the transcription factor FOXP3 which is crucial for Treg cell function (PubMed:23973222). Plays a role in the maintenance of the circadian clock periodicity via deubiquitination and stabilization of the CRY1 and CRY2 proteins (PubMed:27123980). Deubiquitinates REST, thereby stabilizing REST and promoting the maintenance of neural progenitor cells (PubMed:21258371). Deubiquitinates SIRT7, inhibiting SIRT7 histone deacetylase activity and regulating gluconeogenesis (PubMed:28655758). Involved in the regulation of WASH-dependent actin polymerization at the surface of endosomes and the regulation of endosomal protein recycling (PubMed:26365382). It maintains optimal WASH complex activity and precise

	F-actin levels via deubiquitination of TRIM27 and WASHC1 (PubMed:26365382). Mediates the deubiquitination of phosphorylated DEPTOR, promoting its stability and leading to decreased mTORC1 signaling (PubMed:35216969). Deubiquitinates H2AB1, H2BC4 and H2BC22 (By similarity).
Cellular Location	Nucleus. Cytoplasm Nucleus, PML body. Chromosome. Note=Present in a minority of ND10 nuclear bodies (PubMed:9034339). Association with ICP0/VMW110 at early times of infection leads to an increased proportion of USP7-containing ND10 (PubMed:9034339). Colocalizes with ATXN1 in the nucleus (PubMed:12093161). Colocalized with DAXX in speckled structures (PubMed:16845383). Colocalized with PML and PTEN in promyelocytic leukemia protein (PML) nuclear bodies (PubMed:18716620). Interaction with FAM170A promotes its nuclear translocation (By similarity) {ECO:0000250|UniProtKB:Q6A4J8, ECO:0000269|PubMed:12093161, ECO:0000269|PubMed:16845383, ECO:0000269|PubMed:18716620, ECO:0000269|PubMed:9034339}
Tissue Location	Expressed in neural progenitor cells (at protein level) (PubMed:21258371). Widely expressed. Overexpressed in prostate cancer.

Background

Ubiquitin-specific-processing protease 7 (USP7), also known as ubiquitin carboxyl-terminal hydrolase 7 or herpesvirus-associated ubiquitin-specific protease (HAUSP), is an enzyme that in humans is encoded by the USP7 gene. The protein encoded by this gene belongs to the peptidase C19 family, which includes ubiquitinyl hydrolases. This protein deubiquitinates target proteins such as p53 (a tumor suppressor protein) and WASH (essential for endosomal protein recycling), and regulates their activities by counteracting the opposing ubiquitin ligase activity of proteins such as HDM2 and TRIM27, involved in the respective process. Mutations in this gene have been implicated in a neurodevelopmental disorder.

Images

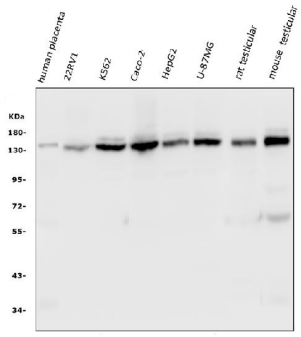


Figure 1. Western blot analysis of HAUSP/USP7 using anti-HAUSP/USP7 antibody (M01239-1).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours.
The sample well of each lane was loaded with 50ug of sample under reducing conditions.
Lane 1: human placenta tissue lysates;
Lane 2: human 22RV1 whole cell lysates;
Lane 3: human K562 whole cell lysates;
Lane 4: human Caco-2 whole cell lysates;
Lane 5: human HepG2 whole cell lysates;
Lane 6: human U-87MG whole cell lysates;
Lane 7: rat testicular tissue lysates;
Lane 8: mouse testicular 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-HAUSP/USP7 antigen affinity purified monoclonal antibody (Catalog # M01239-1) 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 HAUSP/USP7 at approximately 140KD. The expected band size for HAUSP/USP7 is at 128KD.

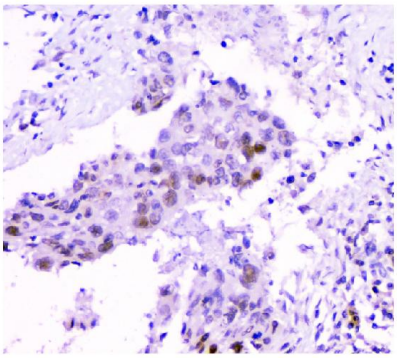


Figure 2. IHC analysis of HAUSP/USP7 using anti-HAUSP/USP7 antibody (M01239-1). HAUSP/USP7 was detected in paraffin-embedded section of human lung cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-HAUSP/USP7 Antibody (M01239-1) 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.

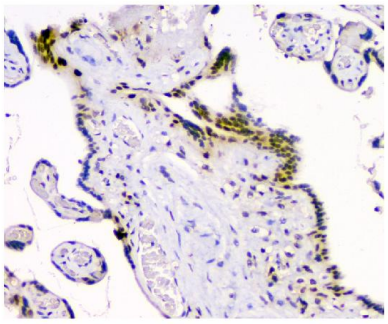


Figure 3. IHC analysis of HAUSP/USP7 using anti-HAUSP/USP7 antibody (M01239-1). HAUSP/USP7 was detected in paraffin-embedded section of human placenta tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-HAUSP/USP7 Antibody (M01239-1) 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.

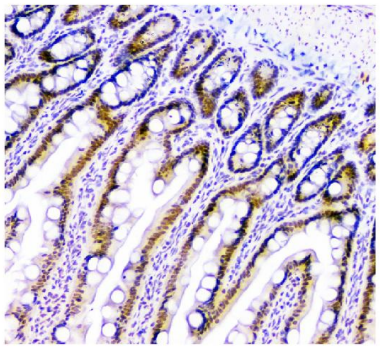


Figure 4. IHC analysis of HAUSP/USP7 using anti-HAUSP/USP7 antibody (M01239-1). HAUSP/USP7 was detected in paraffin-embedded section of rat intestine tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-HAUSP/USP7 Antibody (M01239-1) 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.

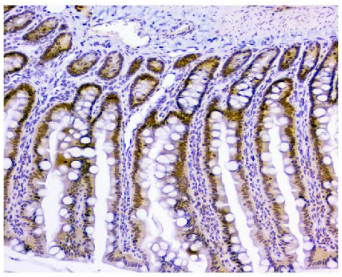


Figure 5. IHC analysis of HAUSP/USP7 using anti-HAUSP/USP7 antibody (M01239-1). HAUSP/USP7 was detected in paraffin-embedded section of rat intestine tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-HAUSP/USP7 Antibody (M01239-1) 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.

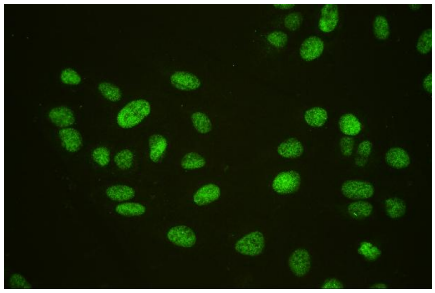


Figure 6. IF analysis of HAUSP/USP7 using anti-HAUSP/USP7 antibody (M01239-1).
HAUSP/USP7 was detected in immunocytochemical section of U20S cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 2 µg/mL mouse anti-HAUSP/USP7 Antibody (M01239-1) 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. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

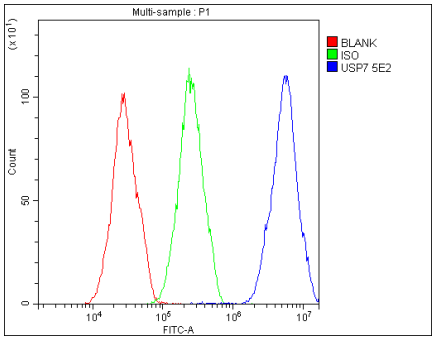


Figure 7. Flow Cytometry analysis of HepG2 cells using anti-HAUSP/USP7 antibody (M01239-1).
Overlay histogram showing HepG2 cells stained with M01239-1 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-HAUSP/USP7 Antibody (M01239-1, 1 µg/1x10^6 cells) for 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.

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