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Anti-SAMHD1 Antibody Picoband™ (monoclonal, 3B9)

Catalog # ABO15035

Product Information

Application	WB, IHC, IF, ICC, FC
Primary Accession	Q9Y3Z3
Host	Mouse
Isotype	Mouse IgG1
Reactivity	Human
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-SAMHD1 Antibody Picoband™ (monoclonal, 3B9) . Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human.
Reconstitution	Add 0.2ml of distilled water will yield a concentration of 500ug/ml.

Additional Information

Gene ID	25939
Other Names	Deoxynucleoside triphosphate triphosphohydrolase SAMHD1, dNTPase, 3.1.5.-, Dendritic cell-derived IFNG-induced protein, DCIP, Monocyte protein 5 {ECO:0000303|Ref.2}, MOP-5 {ECO:0000303|Ref.2}, SAM domain and HD domain-containing protein 1, hSAMHD1, SAMHD1 (HGNC:15925)
Calculated MW	72201
Application Details	Western blot, 0.25-0.5 □g/ml, Human Immunohistochemistry (Paraffin-embedded Section), 2-5 □g/ml, Human Immunocytochemistry/Immunofluorescence, 5 □g/ml, Human Flow Cytometry, 1-3 □g/1x10^6 cells, Human
Source	Eukaryota
Contents	Each vial contains 4mg Trehalose, 0.9mg NaCl and 0.2mg Na2HPO4.
Clone Names	Clone: 3B9
Immunogen	E.coli-derived human SAMHD1 recombinant protein (Position: E37-M626).
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.

Protein Information

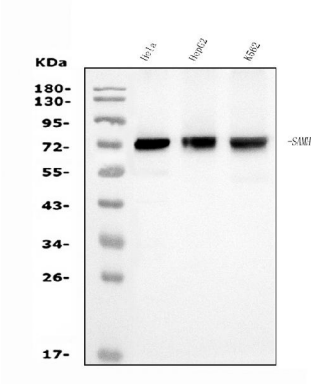
Name	SAMHD1 (HGNC:15925)
Function	Protein that acts both as a host restriction factor involved in defense response to virus and as a regulator of DNA end resection at stalled replication forks (PubMed:19525956, PubMed:21613998, PubMed:21720370, PubMed:22056990, PubMed:23601106, PubMed:23602554, PubMed:24336198, PubMed:26294762, PubMed:26431200, PubMed:28229507, PubMed:28834754, PubMed:29670289). Has deoxynucleoside triphosphate (dNTPase) activity, which is required to restrict infection by viruses, such as HIV-1: dNTPase activity reduces cellular dNTP levels to levels too low for retroviral reverse transcription to occur, blocking early- stage virus replication in dendritic and other myeloid cells (PubMed:19525956, PubMed:21613998, PubMed:21720370, PubMed:22056990, PubMed:23364794, PubMed:23601106, PubMed:23602554, PubMed:24336198, PubMed:25038827, PubMed:26101257, PubMed:26294762, PubMed:26431200, PubMed:28229507). Likewise, suppresses LINE-1 retrotransposon activity (PubMed:24035396, PubMed:24217394, PubMed:29610582). Not able to restrict infection by HIV-2 virus; because restriction activity is counteracted by HIV-2 viral protein Vpx (PubMed:21613998, PubMed:21720370). In addition to virus restriction, dNTPase activity acts as a regulator of DNA precursor pools by regulating dNTP pools (PubMed:23858451). Phosphorylation at Thr-592 acts as a switch to control dNTPase-dependent and -independent functions: it inhibits dNTPase activity and ability to restrict infection by viruses, while it promotes DNA end resection at stalled replication forks (PubMed:23601106, PubMed:23602554, PubMed:29610582, PubMed:29670289). Functions during S phase at stalled DNA replication forks to promote the resection of gapped or reversed forks: acts by stimulating the exonuclease activity of MRE11, activating the ATR-CHK1 pathway and allowing the forks to restart replication (PubMed:29670289). Its ability to promote degradation of nascent DNA at stalled replication forks is required to prevent induction of type I interferons, thereby preventing chronic inflammation (PubMed:27477283, PubMed:29670289). Ability to promote DNA end resection at stalled replication forks is independent of dNTPase activity (PubMed:29670289). Enhances immunoglobulin hypermutation in B-lymphocytes by promoting transversion mutation (By similarity).
Cellular Location	Nucleus. Chromosome Note=Localizes to sites of DNA double-strand breaks in response to DNA damage.
Tissue Location	Expressed in heart, skeletal muscle, spleen, liver, small intestine, placenta, lung and peripheral blood leukocytes (PubMed:11064105). No expression is seen in brain and thymus (PubMed:11064105).

Background

SAM domain and HD domain-containing protein 1 is a protein that in humans is encoded by the SAMHD1 gene. This gene may play a role in regulation of the innate immune response. The encoded protein is upregulated in response to viral infection and may be involved in mediation of tumor necrosis factor-alpha proinflammatory responses. Mutations in this gene have been associated with Aicardi-Goutieres syndrome.

Images

Figure 1. Western blot analysis of SAMHD1 using anti-SAMHD1 antibody (M00592-2).
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 30ug of sample under reducing conditions.
Lane 1: human Hela whole cell lysates,
Lane 2: human HepG2 whole cell lysates,
Lane 3: human K562 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-SAMHD1 antigen affinity purified monoclonal antibody (Catalog # M00592-2) 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 SAMHD1 at approximately 72KD. The expected band size for SAMHD1 is at 72KD.

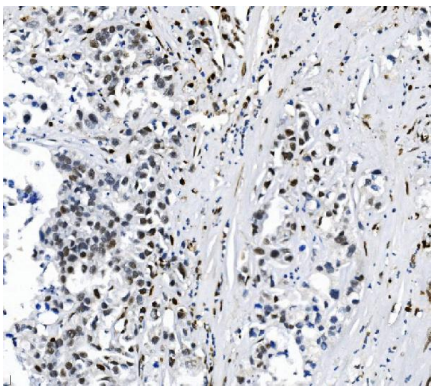


Figure 2. IHC analysis of SAMHD1 using anti-SAMHD1 antibody (M00592-2).
SAMHD1 was detected in paraffin-embedded section of human gastric adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-SAMHD1 Antibody (M00592-2) 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.

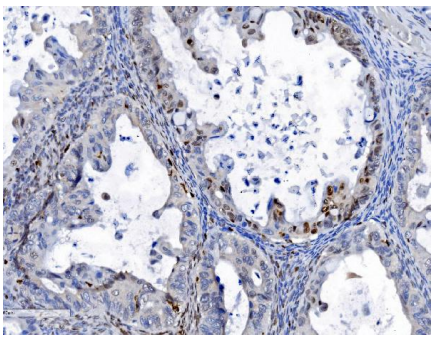


Figure 3. IHC analysis of SAMHD1 using anti-SAMHD1 antibody (M00592-2).
SAMHD1 was detected in paraffin-embedded section of human ovarian cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-SAMHD1 Antibody (M00592-2) 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.

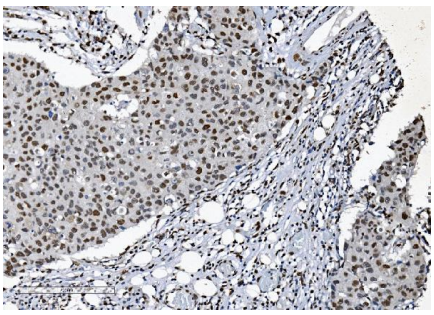


Figure 4. IHC analysis of SAMHD1 using anti-SAMHD1 antibody (M00592-2).
SAMHD1 was detected in paraffin-embedded section of human breast cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-SAMHD1 Antibody (M00592-2) 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.

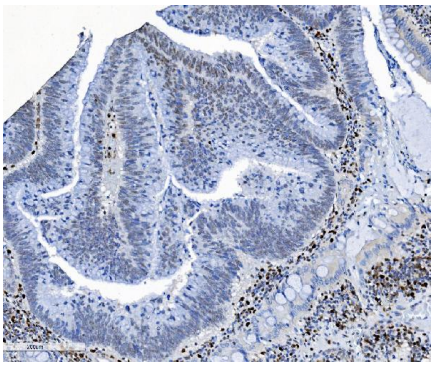


Figure 5. IHC analysis of SAMHD1 using anti-SAMHD1 antibody (M00592-2).
SAMHD1 was detected in paraffin-embedded section of human rectal cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-SAMHD1 Antibody (M00592-2) 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.

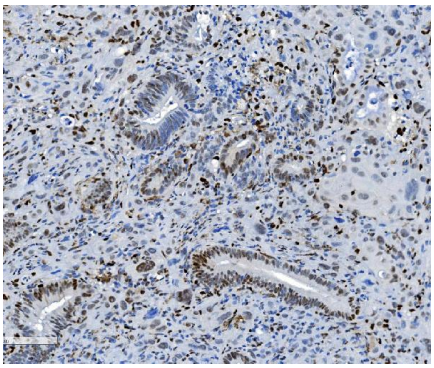


Figure 6. IHC analysis of SAMHD1 using anti-SAMHD1 antibody (M00592-2).
SAMHD1 was detected in paraffin-embedded section of human gallbladder adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-SAMHD1 Antibody (M00592-2) 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.

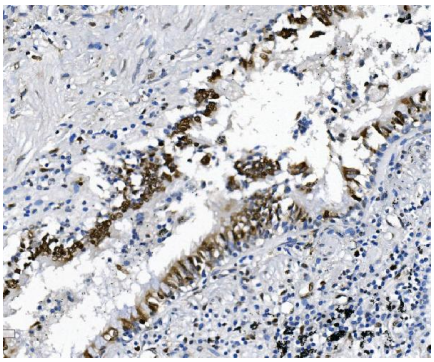


Figure 7. IHC analysis of SAMHD1 using anti-SAMHD1 antibody (M00592-2).
SAMHD1 was detected in paraffin-embedded section of human lung cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-SAMHD1 Antibody (M00592-2) 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.

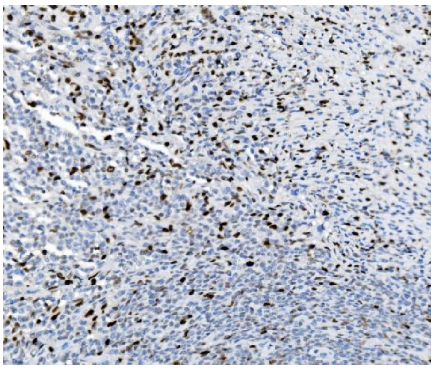


Figure 8. IHC analysis of SAMHD1 using anti-SAMHD1 antibody (M00592-2).
SAMHD1 was detected in paraffin-embedded section of human lymphoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-SAMHD1 Antibody (M00592-2) 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.

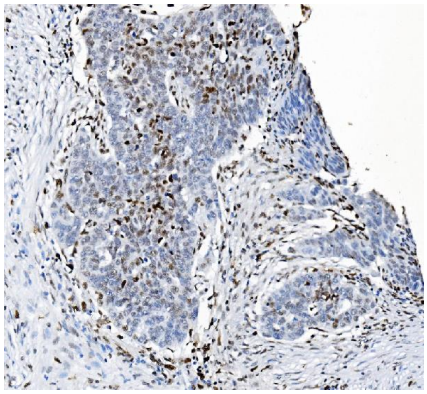


Figure 9. IHC analysis of SAMHD1 using anti-SAMHD1 antibody (M00592-2).
SAMHD1 was detected in paraffin-embedded section of human ovarian serous adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-SAMHD1 Antibody (M00592-2) 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.

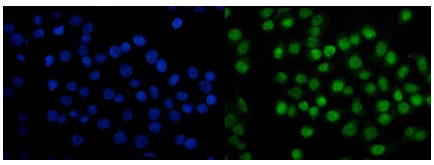


Figure 10. IF analysis of SAMHD1 using anti-SAMHD1 antibody (M00592-2).
SAMHD1 was detected in immunocytochemical section of CACO-2 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 µg/mL mouse anti-SAMHD1 Antibody (M00592-2) 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.

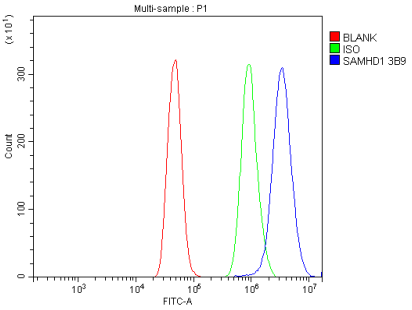


Figure 11. Flow Cytometry analysis of A431 cells using anti-SAMHD1 antibody (M00592-2).
Overlay histogram showing A431 cells stained with M00592-2 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-SAMHD1 Antibody (M00592-2, 1 µg/1x10^6 cells) for 30 min 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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