10320 Camino Santa Fe, Suite G
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Anti-HGS Antibody Picoband™ (monoclonal, 6C2E1)

Catalog # ABO16241

Specification

Anti-HGS Antibody Picoband™ (monoclonal, 6C2E1) - Product Information

Application	**WB, IHC, FC**
Primary Accession	O14964
Host	**Mouse**
Isotype	**Mouse IgG1**
Reactivity	**Rat, Human, Mouse**
Clonality	**Monoclonal**
Format	**Lyophilized**

Description
Anti-HGS Antibody Picoband™ (monoclonal, 6C2E1) . Tested in Flow Cytometry, IHC, WB applications. This antibody reacts with Human, Mouse, Rat.

Reconstitution
Adding 0.2 ml of distilled water will yield a concentration of 500 µg/ml.

Anti-HGS Antibody Picoband™ (monoclonal, 6C2E1) - Additional Information

Gene ID 9146

Other Names
Hepatocyte growth factor-regulated tyrosine kinase substrate, Hrs, Protein pp110, HGS, HRS

Calculated MW
110 kDa KDa

Application Details
Western blot, 0.25-0.5 µg/ml, Human, Mouse, Rat
 Immunohistochemistry(Paraffin-embedded Section), 2-5 µg/ml, Human
 Flow Cytometry, 1-3 µg/1x10^6 cells, Human

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

Immunogen
E.coli-derived human HGS recombinant protein (Position: R3-D777).

Purification
Immunogen affinity purified.

Storage	**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 freezing and thawing.**

Anti-HGS Antibody Picoband™ (monoclonal, 6C2E1) - Protein Information

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Name HGS

Synonyms HRS

Function
Involved in intracellular signal transduction mediated by cytokines and growth factors. When associated with STAM, it suppresses DNA signaling upon stimulation by IL-2 and GM-CSF. Could be a direct effector of PI3-kinase in vesicular pathway via early endosomes and may regulate trafficking to early and late endosomes by recruiting clathrin. May concentrate ubiquitinated receptors within clathrin- coated regions. Involved in down-regulation of receptor tyrosine kinase via multivesicular body (MVBs) when complexed with STAM (ESCRT-0 complex). The ESCRT-0 complex binds ubiquitin and acts as a sorting machinery that recognizes ubiquitinated receptors and transfers them to further sequential lysosomal sorting/trafficking processes. May contribute to the efficient recruitment of SMADs to the activin receptor complex. Involved in receptor recycling via its association with the CART complex, a multiprotein complex required for efficient transferrin receptor recycling but not for EGFR degradation.

Cellular Location
Cytoplasm {ECO:0000250|UniProtKB:Q9JJ50}. Early endosome membrane; Peripheral membrane protein; Cytoplasmic side Endosome, multivesicular body membrane {ECO:0000250|UniProtKB:Q9JJ50}; Peripheral membrane protein {ECO:0000250|UniProtKB:Q9JJ50} Note=Colocalizes with UBQLN1 in ubiquitin-rich cytoplasmic aggregates that are not endocytic compartments.

Tissue Location
Ubiquitous expression in adult and fetal tissues with higher expression in testis and peripheral blood leukocytes

Anti-HGS Antibody Picoband™ (monoclonal, 6C2E1) - 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-HGS Antibody Picoband™ (monoclonal, 6C2E1) - Images

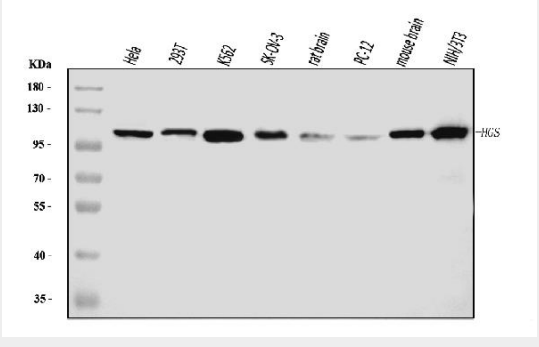

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Figure 1. Western blot analysis of HGS using anti-HGS antibody (M00179-1).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.
Lane 1: human Hela whole cell lysates,
Lane 2: human 293T whole cell lysates,
Lane 3: human K562 whole cell lysates,
Lane 4: rat brain tissue lysates,
Lane 5: rat PC-12 whole cell lysates,
Lane 6: mouse brain tissue lysates,
Lane 7: mouse NIH/3T3 whole cell lysates.
After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA 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-HGS antigen affinity purified monoclonal antibody (Catalog # M00179-1) 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 HGS at approximately 110 kDa. The expected band size for HGS is at 110 kDa.

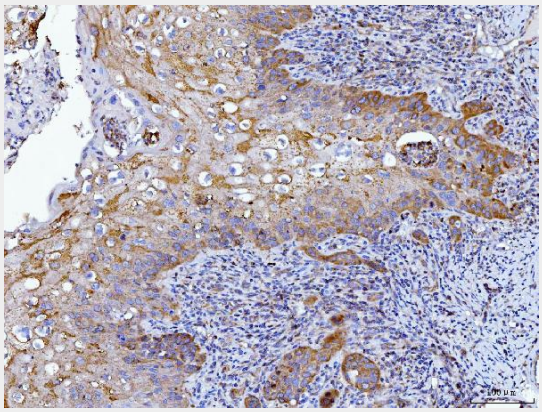

Figure 2. IHC analysis of HGS using anti-HGS antibody (M01174-1).
HGS was detected in a paraffin-embedded section of human laryngeal squamous cell carcinomas tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-HGS Antibody (M01174-1) 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.

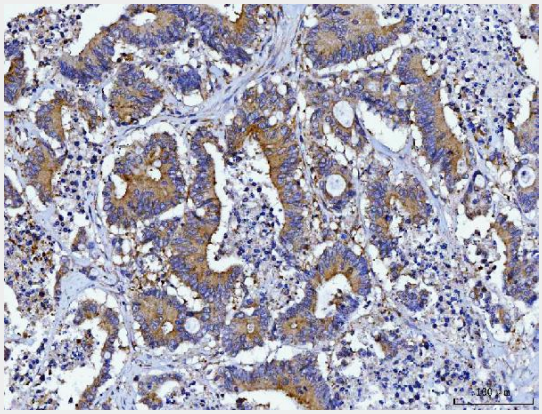

Figure 3. IHC analysis of HGS using anti-HGS antibody (M01174-1).

HGS was detected in a paraffin-embedded section of human colorectal adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-HGS Antibody (M01174-1) 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.

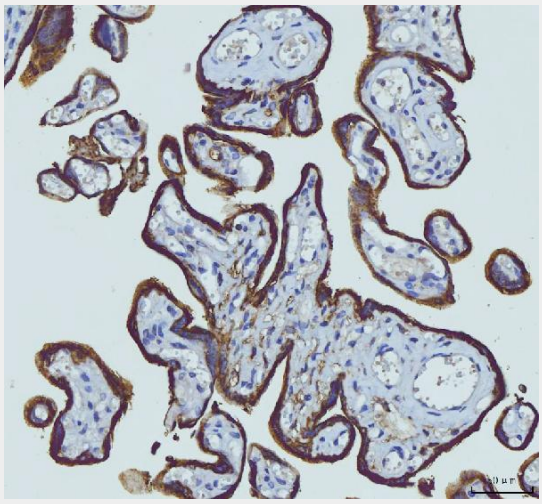

Figure 4. IHC analysis of HGS using anti-HGS antibody (M01174-1).
HGS was detected in a paraffin-embedded section of human placenta tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-HGS Antibody (M01174-1) 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.

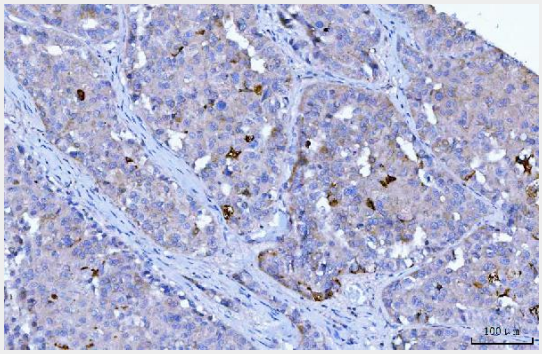

Figure 5. IHC analysis of HGS using anti-HGS antibody (M01174-1).
HGS was detected in a paraffin-embedded section of human hepatocellular carcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-HGS Antibody (M01174-1) 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.

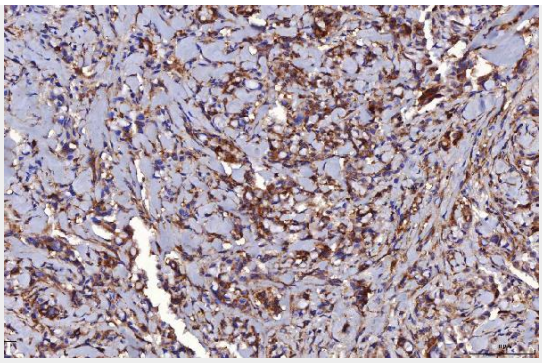

Figure 6. IHC analysis of HGS using anti-HGS antibody (M01174-1).
HGS was detected in a paraffin-embedded section of human breast infiltrating ductal carcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-HGS Antibody (M01174-1) 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.

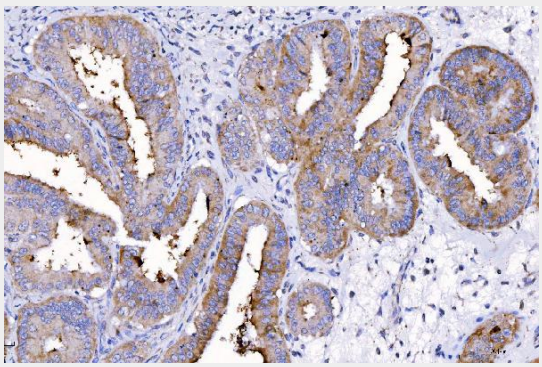

Figure 7. IHC analysis of HGS using anti-HGS antibody (M01174-1).
HGS was detected in a paraffin-embedded section of human endometrial cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-HGS Antibody (M01174-1) 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.

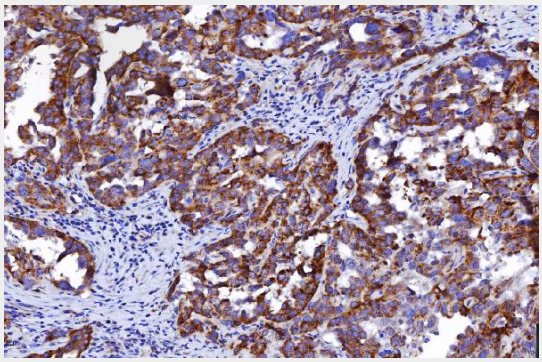

Figure 8. IHC analysis of HGS using anti-HGS antibody (M01174-1).
HGS was detected in a paraffin-embedded section of human ovarian cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue

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section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-HGS Antibody (M01174-1) 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.

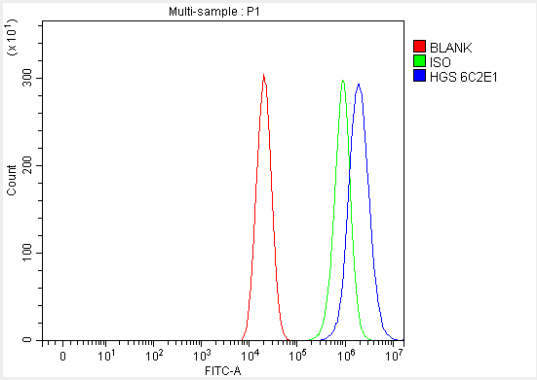

Figure 9. Flow Cytometry analysis of Caco-2 cells using anti-HGS antibody (M01174-1).
Overlay histogram showing Caco-2 cells stained with M01174-1 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-HGS Antibody (M01174-1, 1 µg/1x10^6 cells) for 30 min at 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.

Anti-HGS Antibody Picoband™ (monoclonal, 6C2E1) - Background

Hepatocyte growth factor-regulated tyrosine kinase substrate is an enzyme that in humans is encoded by the HGS gene. It is mapped to 17q25.3. The protein encoded by this gene regulates endosomal sorting and plays a critical role in the recycling and degradation of membrane receptors. The encoded protein sorts monoubiquitinated membrane proteins into the multivesicular body, targeting these proteins for lysosome-dependent degradation.