

Anti-Hsp105/HSPH1 Antibody Picoband™ (monoclonal, 3D10)

Catalog # ABO14879

Product Information

Application	WB, IHC, IF, ICC, FC
Primary Accession	Q92598
Host	Mouse
Isotype	Mouse IgG1
Reactivity	Human
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-Hsp105/HSPH1 Antibody Picoband™ (monoclonal, 3D10) . 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 500 µg/ml.

Additional Information

Gene ID	10808
Other Names	Heat shock protein 105 kDa, Antigen NY-CO-25, Heat shock 110 kDa protein, Heat shock protein family H member 1, HSPH1, HSP105, HSP110, KIAA0201
Calculated MW	96865
Application Details	Western blot, 0.1-0.5 µg/ml, Human Immunohistochemistry (Paraffin-embedded Section), 0.5-1 µg/ml, Human, By Heat Immunocytochemistry/Immunofluorescence, 2 µg/ml, Human Flow Cytometry, 1-3 µg/1x10 ⁶ cells, Human
Subcellular Localization	Cytoplasm.
Tissue Specificity	Highly expressed in testis. Present at lower levels in most brain regions, except cerebellum. Overexpressed in cancer cells.
Source	Eukaryota
Contents	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na ₂ HPO ₄ , 0.05mg NaN ₃ .
Clone Names	Clone: 3D10
Immunogen	E. coli-derived human Hsp105 recombinant protein (Position: Y653-D858).
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.
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Protein Information

Name	HSPH1
Synonyms	HSP105, HSP110, KIAA0201
Function	Acts as a nucleotide-exchange factor (NEF) for chaperone proteins HSPA1A and HSPA1B, promoting the release of ADP from HSPA1A/B thereby triggering client/substrate protein release (PubMed: 24318877). Prevents the aggregation of denatured proteins in cells under severe stress, on which the ATP levels decrease markedly. Inhibits HSPA8/HSC70 ATPase and chaperone activities (By similarity).
Cellular Location	Cytoplasm.
Tissue Location	Highly expressed in testis. Present at lower levels in most brain regions, except cerebellum. Overexpressed in cancer cells.

Background

HSP105 (HEAT-SHOCK 105/110-KD PROTEIN 1), also called HSPH1 or HSP110, is a protein that in humans is encoded by the HSPH1 gene. Immunohistochemical analysis localizes HSP105 mainly in the cytoplasm. Database analysis indicates that both HSP105 isoforms are highly conserved during evolution. By analysis of radiation hybrids and human/rodent hybrid cell lines, the HSPH1 gene is mapped to chromosome 13. Both HSP105-alpha and HSP105-beta are upregulated in HeLa cells exposed to heat shock. HSP105-alpha, but not HSP105-beta, is also upregulate in response to other cell stresses. Following heat shock, HSP105 relocates from a cytoplasmic to perinuclear position. Besides, HSP110 may thus constitute a major determinant for both prognosis and treatment response in colorectal cancer.

Images

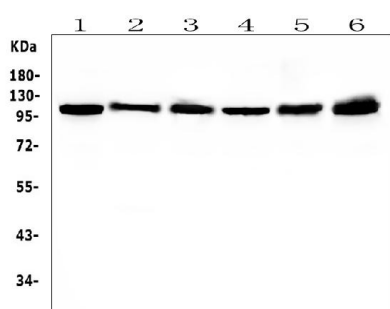


Figure 1. Western blot analysis of HSPH1 using anti-HSPH1 antibody (M04168-1).

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 Caco-2 whole cell lysates

Lane 2: human K562 whole cell lysates

Lane 3: human A549 whole cell lysates

Lane 4: human HepG2 whole cell lysates

Lane 5: human PANC-1 whole cell lysates

Lane 6: human SGC-7901 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-HSPH1 antigen affinity purified monoclonal antibody (Catalog # M04168-1) 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 HSPH1 at approximately 105KD. The expected band size for HSPH1 is at 105KD.

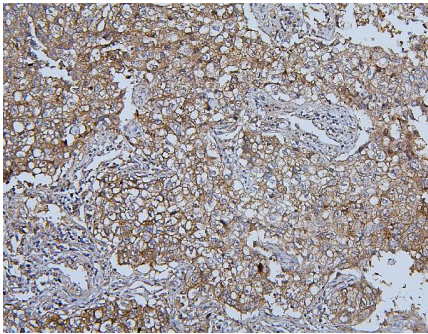


Figure 2. IHC analysis of HSPH1 using anti-HSPH1 antibody (M04168-1). HSPH1 was detected in paraffin-embedded section of human lung cancer tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 μ g/ml mouse anti-HSPH1 Antibody (M04168-1) 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 Streptavidin-Biotin-Complex (SABC)(Catalog # SA1021) with DAB as the chromogen.

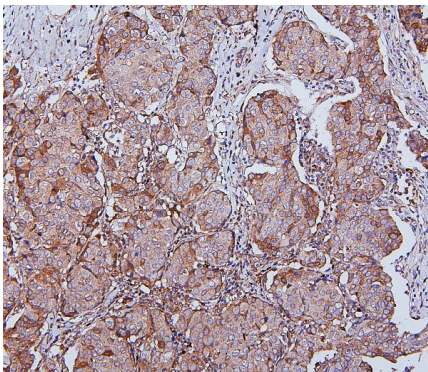


Figure 3. IHC analysis of HSPH1 using anti-HSPH1 antibody (M04168-1). HSPH1 was detected in paraffin-embedded section of human mammary cancer tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 μ g/ml mouse anti-HSPH1 Antibody (M04168-1) 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 Streptavidin-Biotin-Complex (SABC)(Catalog # SA1021) with DAB as the chromogen.

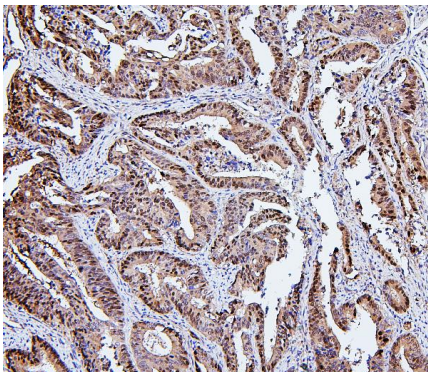


Figure 4. IHC analysis of HSPH1 using anti-HSPH1 antibody (M04168-1). HSPH1 was detected in paraffin-embedded section of human intestinal cancer tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 μ g/ml mouse anti-HSPH1 Antibody (M04168-1) 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 Streptavidin-Biotin-Complex (SABC)(Catalog # SA1021) with DAB as the chromogen.

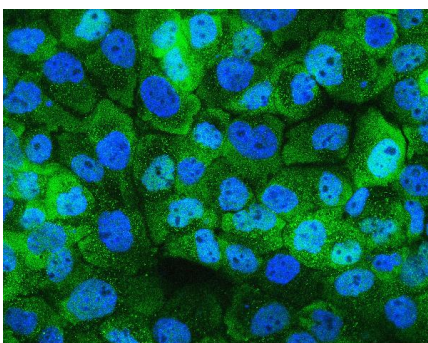


Figure 5. IF analysis of HSPH1 using anti-HSPH1 antibody (M04168-1). HSPH1 was detected in immunocytochemical section of A431 cell. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 2 μ g/mL mouse anti-HSPH1 Antibody (M04168-1) 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. The section was counterstained with DAPI.

Visualize using a fluorescence microscope and filter sets appropriate for the label used.

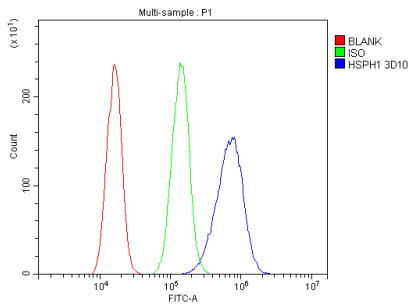


Figure 6. Flow Cytometry analysis of A431 cells using anti-HSPH1 antibody (M04168-1). Overlay histogram showing A431 cells stained with M04168-1 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-HSPH1 Antibody (M04168-1, 1 μ g/ 1×10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 μ g/ 1×10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 μ g/ 1×10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

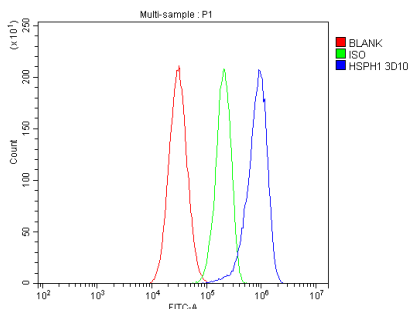


Figure 7. Flow Cytometry analysis of HepG2 cells using anti-HSPH1 antibody (M04168-1). Overlay histogram showing HepG2 cells stained with M04168-1 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-HSPH1 Antibody (M04168-1, 1 μ g/ 1×10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 μ g/ 1×10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 μ g/ 1×10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

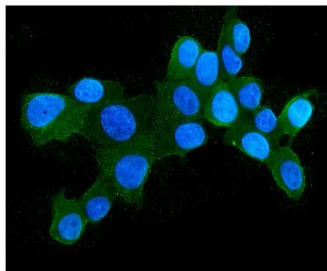


Figure 8. IF analysis of HSPH1 using anti-HSPH1 antibody (M04168-1). HSPH1 was detected in immunocytochemical section of MCF7 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 2 μ g/mL mouse anti-HSPH1 Antibody (M04168-1) 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. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

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