

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

Catalog # ABO14879

Specification

Anti-Hsp105/HSPH1 Antibody Picoband[™] (monoclonal, 3D10) - Product Information

Application Primary Accession Host Isotype Reactivity Clonality Format Description WB, IHC, IF, ICC, FC <u>092598</u> Mouse Mouse IgG1 Human Monoclonal Lyophilized

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 μ g/ml.

Anti-Hsp105/HSPH1 Antibody Picoband[™] (monoclonal, 3D10) - 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 105 kDa KDa

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
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Subcellular Localization Cytoplasm.

Tissue Specificity Highly expressed in testis. Present at lower levels in most brain regions, except cerebellum. Overexpressed in cancer cells.

Contents

Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na₂HPO₄, 0.05mg NaN₃.

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.

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

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

Provided below are standard protocols that you may find useful for product applications.

- <u>Western Blot</u>
- Blocking Peptides
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- <u>Cell Culture</u>

Anti-Hsp105/HSPH1 Antibody Picoband™ (monoclonal, 3D10) - 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 μ g/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 Strepavidin-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 Strepavidin-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 Strepavidin-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/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 μ g/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 μ g/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.







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/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 μ g/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 μ g/1x10⁶) 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.

Anti-Hsp105/HSPH1 Antibody Picoband™ (monoclonal, 3D10) - 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 relocalizes from a cytoplasmic to perinuclear position. Besides, HSP110 may thus constitute a major determinant for both prognosis and treatment response in colorectal cancer.