

Basic Information

Product Name	Anti-HSPH1 Antibody (Clone#3D10)	
Gene Name	HSPH1	
Source	Mouse	
Clonality	Monoclonal	
Isotype	IgG1	
Species Reactivity	human	
Tested Application	WB, IHC, ICC/IF, FCM	
Contents	500 ug/ml antibody with PBS, 0.02% NaN ₃ , 1 mg/ml BSA and 50% glycerol.	
Immunogen	E. coli-derived human Hsp105 recombinant protein (Position: Y653-D858).	
Concentration	500 ug/ml	
Purification	protein G purified.	
Observed MW	105 kDa	
Dilution Ratios	Western blot (WB):	1:500-2000
	Immunohistochemistry (IHC):	1:50-400
	Immunocytochemistry/Immunofluorescence (ICC/IF):	1:50-400
	Flow Cytometry (Fixed):	1:50-200
	(Boiling the paraffin sections in 10mM citrate buffer, pH6.0, or PH8.0 EDTA repair liquid for 20 mins is required for the staining of formalin/paraffin sections.) Optimal working dilutions must be determined by end user.	

Storage

12 months from date of receipt, -20°C as supplied. 6 months 2 to 8°C after reconstitution. Avoid repeated freezing and thawing.

Background Information

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 upregulated 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.

Selected Validation Data

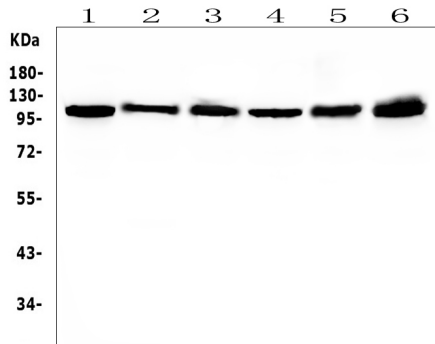


Figure 1. Western blot analysis of HSPH1 using anti-HSPH1 antibody (M04168-1). The sample well of each lane was loaded with 30 ug of sample under reducing conditions.

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 membrane. Then the membrane was incubated with mouse anti-HSPH1 antigen affinity purified monoclonal antibody (M04168-1) at a dilution of 1:1000 and probed with a goat anti-mouse IgG-HRP secondary antibody (Catalog # BA1050). The signal is developed using ECL Plus Western Blotting Substrate (Catalog # AR1197). A specific band was detected for HSPH1 at approximately 105 kDa. The expected band size for HSPH1 is at 97 kDa.

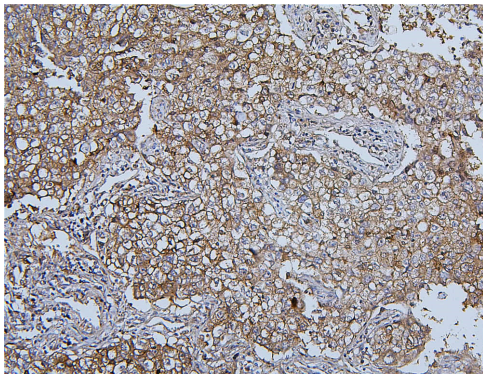


Figure 2. IHC analysis of HSPH1 using anti-HSPH1 antibody (M04168-1).

HSPH1 was detected in a paraffin-embedded section of human lung cancer tissue. Biotinylated goat anti-mouse IgG was used as secondary antibody. The tissue section was incubated with mouse anti-HSPH1 Antibody (M04168-1) at a dilution of 1:200 and developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB (Catalog # AR1027) as the chromogen.

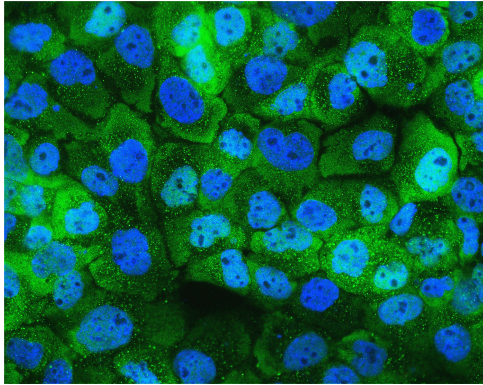


Figure 5. IF analysis of HSPH1 using anti- HSPH1 antibody (M04168-1)

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 tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μ g/mL mouse anti- HSPH1 Antibody (M04168-1) overnight at 4°C. DyLight488 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.

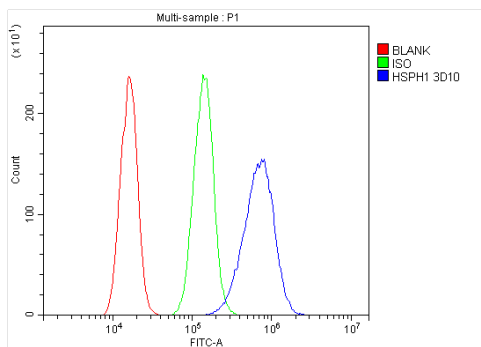


Figure 6. Flow Cytometry analysis of A431 cells using anti-HSPH1 antibody (M04168-1).

Overlay histogram showing A431 cells stained with M04168-1 (Blue line). To facilitate intracellular staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-HSPH1 Antibody (M04168-1) at 1:100 dilution for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126) was used as secondary antibody at 1:100 dilution for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG at 1:100 dilution used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.