Product Datasheet

Hsc70 Mouse Monoclonal Antibody (orb623830)

Catalog Number	orb623830
Category	Antibodies
Description	Anti-Hsc70 Antibody (monoclonal, B5D8). Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
Target	Chromosome-associated kinesin KIF4A/4B
Clonality	Monoclonal
Species/Host	Mouse
Isotype	Mouse IgG2b
Conjugation	Unconjugated
Reactivity	Human, Mouse, Rat
Form/Appearance	Lyophilized
Concentration	500 µg/ml
Buffer/Preservatives	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.05mg NaN3.
Reconstitution	Add 0.2ml of distilled water will yield a concentration of 500µg/ml.
Purification	Immunogen affinity purified.
Immunogen	E.coli-derived human Hsc70 recombinant protein (Position: Q520-A614). Human Hsc70 shares 98.9% amino acid (aa) sequence identity with both mouse and rat Hsc70.
UniProt ID	P11142

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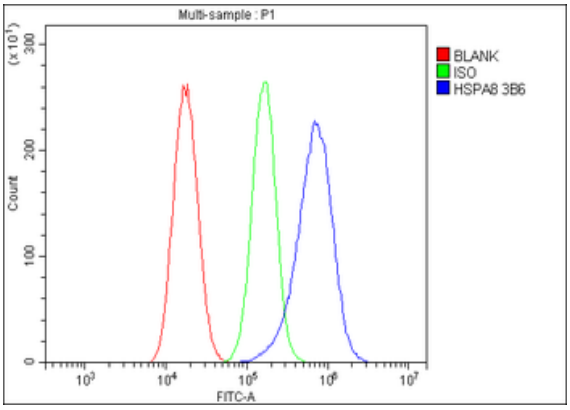
MW	71 kDa
Tested applications	FC, ICC, IF, IHC, WB
Dilution range	Western blot, 0.1-0.5µg/ml, Human, Mouse, Rat Immunohistochemistry (Paraffin-embedded Section), 0.5-1µg/ml, Human, Rat Immunocytochemistry/Immunofluorescence, 2µg/ml, Human Flow Cytometry (Fixed), 1-3µg/1x10^6, Human, Mouse
Specificity	No cross reactivity with other proteins.
Cross Reactivity	No cross-reactivity with other proteins.
Antibody Type	Primary Antibody
Clone Number	B5D8
Storage	Maintain refrigerated at 2-8°C for up to 2 weeks. For long term storage store at -20°C in small aliquots to prevent freeze-thaw cycles.
Note	For research use only
Expiration Date	12 months from date of receipt.



Flow Cytometry analysis of A549 cells using anti-Hsc70 antibody. Overlay histogram showing A549 cells (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-Hsc70 Antibody (1 µg/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (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 without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.

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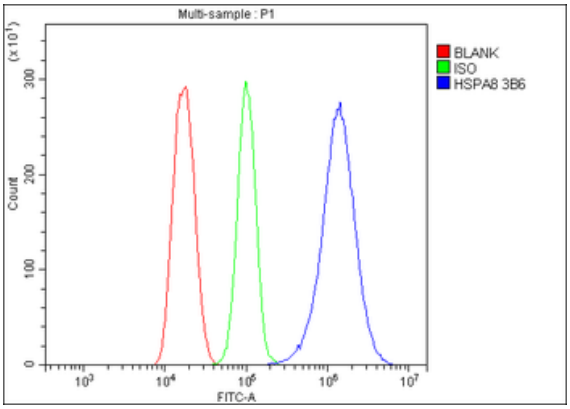
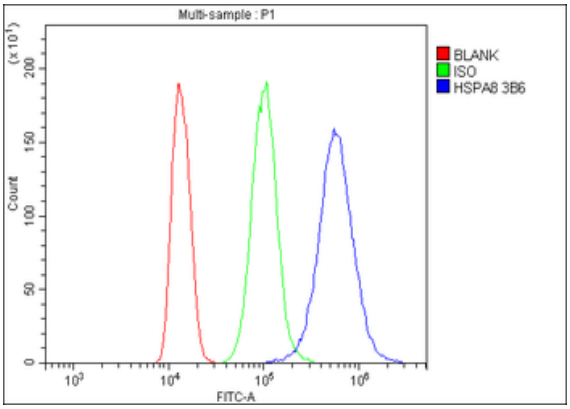


Flow Cytometry analysis of HEPA1-6 cells using anti-Hsc70 antibody. Overlay histogram showing HEPA1-6 cells (Blue line). To facilitate intracellular staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-Hsc70 Antibody (1 µg/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (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 without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.



Flow Cytometry analysis of K562 cells using anti-Hsc70 antibody. Overlay histogram showing K562 cells (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-Hsc70 Antibody (1 µg/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (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 without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.

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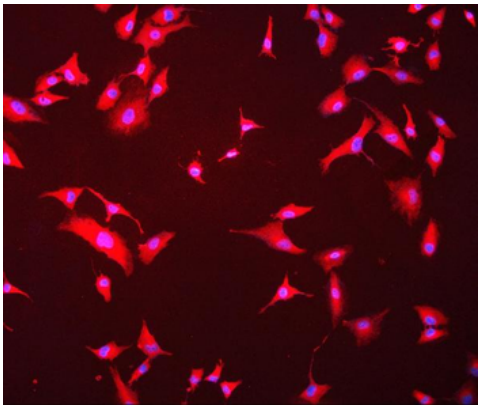
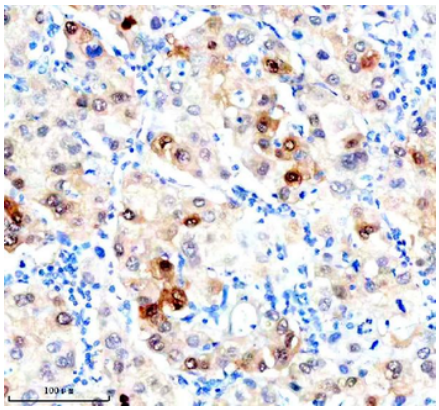
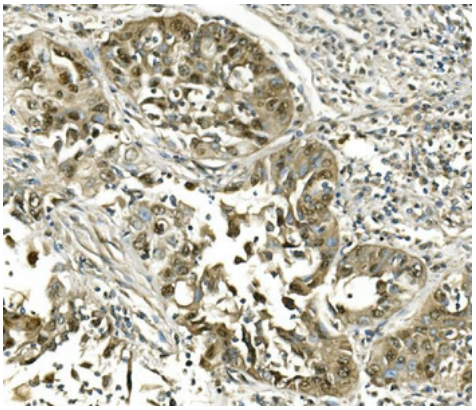


IF analysis of Hsc70 using anti-Hsc70 antibody. Hsc70 was detected in immunocytochemical section of A549 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 2 µg/mL mouse anti-Hsc70 Antibody overnight at 4°C. DyLight®594 Conjugated Goat Anti-Mouse IgG 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.



IHC analysis of Hsc70 using anti-Hsc70 antibody. Hsc70 was detected in a paraffin-embedded section of human liver 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 1:200 mouse anti-Hsc70 Antibody overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit with DAB as the chromogen.



IHC analysis of Hsc70 using anti-Hsc70 antibody. Hsc70 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 1 µg/ml mouse anti-Hsc70 Antibody 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) with DAB as the chromogen.

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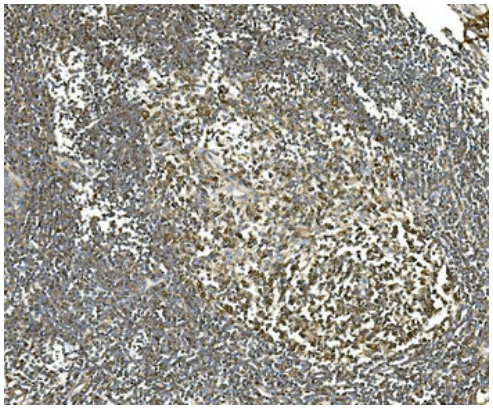
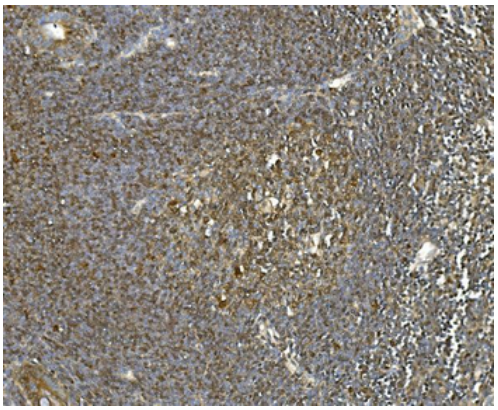
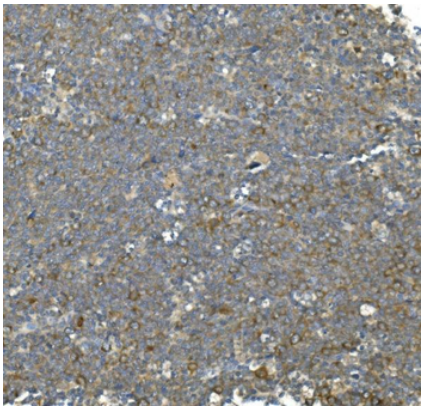


IHC analysis of Hsc70 using anti-Hsc70 antibody. Hsc70 was detected in paraffin-embedded section of human rectal cancer (lymph node) tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-Hsc70 Antibody 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) with DAB as the chromogen.



IHC analysis of Hsc70 using anti-Hsc70 antibody. Hsc70 was detected in paraffin-embedded section of human rectal cancer (lymph node) 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 1 µg/ml mouse anti-Hsc70 Antibody 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) with DAB as the chromogen.



IHC analysis of Hsc70 using anti-Hsc70 antibody. Hsc70 was detected in paraffin-embedded section of human tonsil 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 1 µg/ml mouse anti-Hsc70 Antibody 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) with DAB as the chromogen.

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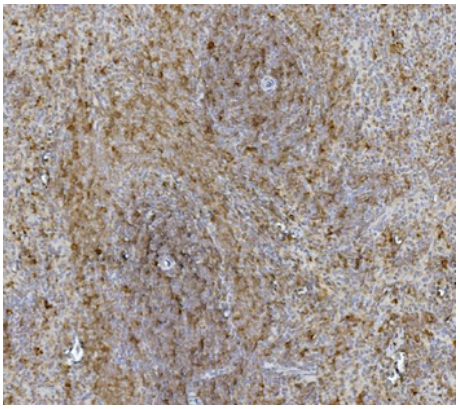
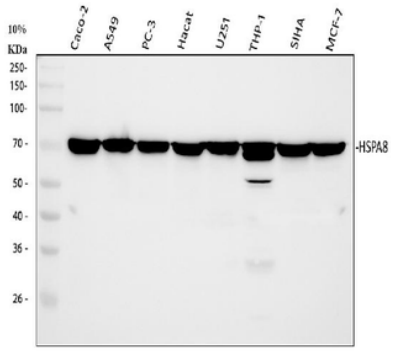


IHC analysis of Hsc70 using anti-Hsc70 antibody. Hsc70 was detected in paraffin-embedded section of rat spleen tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-Hsc70 Antibody 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) with DAB as the chromogen.



Western blot analysis of HSC70/HSPA8 using anti-HSC70/HSPA8 antibody. 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 Caco-2 whole cell lysates, Lane 2: human A549 whole cell lysates, Lane 3: human PC-3 whole cell lysates, Lane 4: human Hacat whole cell lysates, Lane 5: human U251 whole cell lysates, Lane 6: human THP-1 whole cell lysates, Lane 7: human SiHa whole cell lysates, Lane 8: human MCF-7 whole cell lysates. After electrophoresis, proteins were transferred to a membrane. Then the membrane was incubated with mouse anti-HSC70/HSPA8 antigen affinity purified monoclonal antibody at 1:500 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 1:500 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit with Tanon 5200 system. A specific band was detected for HSC70/HSPA8 at approximately 71 kDa. The expected band size for HSC70/HSPA8 is at 71 kDa.

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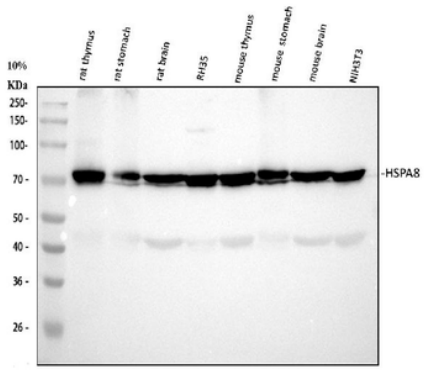

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