Data Sheet (Cat.No.TMAH-00577)

Anti-HSPA8 Antibody (7O992)

Product Details

Ig Type:	IgG2b
Reactivity:	Human, Rat, Mouse
Conjugation:	Unconjugated
Clone:	7O992
Purification:	Protein A purified

Applications

Verified Activity:

1. Western Blot
-Positive WB detected in: Hela whole cell lysate, NIH/3T3 whole cell lysate, K562 whole cell lysate, HepG2 whole cell lysate, Mouse spleen tissue, Rat spleen tissue, Mouse heart tissue, Rat heart tissue
-All lanes HSPA8 antibody at 1:2000
-Secondary: Goat polyclonal to mouse IgG at 1/50000 dilution
-Predicted band size: 70~75 KDa
-Observed band size: 70~75 KDa
-Exposure time: 10s
2. Western Blot
-Positive WB detected in: Hela whole cell lysate at 20μg, 10μg, 5μg, 2.5μg, 1.25μg, 0.625μg
-All lanes: HSPA8 antibody at 1:2000
-Secondary: Goat polyclonal to mouse IgG at 1/50000 dilution
-Predicted band size: 70~75 KDa
-Observed band size: 70~75 KDa
-Exposure time: 10s
3. Western Blot
-Positive WB detected in: 20μg Hela whole cell lysate
HSPA8 antibody at 1:2000, 1:4000, 1:8000, 1:16000, 1:32000, 1:64000, 1:128000, 1:256000
-Secondary: Goat polyclonal to mouse IgG at 1/50000 dilution
-Predicted band size: 70~75 KDa
-Observed band size: 70~75 KDa
-Exposure time: 10s
4. IHC image of TMAH-00577 diluted at 1:256 and staining in paraffin-embedded human prostate cancer performed on a Leica BondTM system. After dewaxing and hydration, antigen retrieval was mediated by high pressure in a citrate buffer (pH 6.0). Section was blocked with 10% normal goat serum 30min at RT. Then primary antibody (1% BSA) was incubated at 4°C overnight, and detected by a Goat anti-mouse IgG polymer labeled by HRP and visualized using 0.05% DAB.
5. IHC image of TMAH-00577 diluted at 1:256 and staining in paraffin-embedded human prostate cancer performed on a Leica BondTM system. After dewaxing and hydration, antigen retrieval was mediated by high pressure in a citrate buffer (pH 6.0). Section was blocked with 10% normal goat serum 30min at RT. Then primary antibody (1% BSA) was incubated at 4°C overnight, and detected by a Goat anti-mouse IgG polymer labeled by HRP and visualized using 0.05% DAB.
6. Immunoprecipitating HSPA8 in Hela whole cell lysate

-Lane 1: Mouse control IgG2b instead of TMAH-00577 in Hela whole cell lysate
-Lane 2: TMAH-00577 (1.5μl) + Hela whole cell lysate (500μg)
-Lane 3: Hela whole cell lysate (20μg)
For western blotting, the blot was detected with TMAH-00577 at 1:2000, and a HRP-conjugated Protein G antibody was used as the secondary antibody at 1:2000
7. Overlay histogram showing MCF-7 cells stained with TMAH-00577 (red line). The cells were fixed with 70% Ethylalcohol (18h) and then incubated in 10% normal goat serum to block non-specific protein-protein interactions followed by the primary antibody at 1:200 for 1 h at 4°C. The secondary antibody used was FITC goat anti-mouse IgG(H+L) at 1/100 dilution for 30min at 4°C. Isotype control antibody (green line) was mouse IgG2b used under the same conditions. Acquisition of >10,000 events was performed.

Application: ELISA,FCM,IHC,IP,WB

Properties

Purity: >95%

Stability & Storage: Store at -20°C or -80°C for 12 months. Avoid repeated freeze-thaw cycles.

Shipping: Shipping with blue ice.

Antigen Details

Immunogen: Recombinant Protein: Human Heat shock cognate 71 kDa Protein (2-646AA)

Antigen Species: Human

Gene ID: 3312

Uniprot ID: P11142

Biology Area: Immunology

Research Background

Molecular chaperone implicated in a wide variety of cellular processes, including protection of the proteome from stress, folding and transport of newly synthesized polypeptides, activation of proteolysis of misfolded proteins and the formation and dissociation of protein complexes. Plays a pivotal role in the protein quality control system, ensuring the correct folding of proteins, the re-folding of misfolded proteins and controlling the targeting of proteins for subsequent degradation. This is achieved through cycles of ATP binding, ATP hydrolysis and ADP release, mediated by co-chaperones. The co-chaperones have been shown to not only regulate different steps of the ATPase cycle of HSP70, but they also have an individual specificity such that one co-chaperone may promote folding of a substrate while another may promote degradation. The affinity of HSP70 for polypeptides is regulated by its nucleotide bound state. In the ATP-bound form, it has a low affinity for substrate proteins. However, upon hydrolysis of the ATP to ADP, it undergoes a conformational change that increases its affinity for substrate proteins. HSP70 goes through repeated cycles of ATP hydrolysis and nucleotide exchange, which permits cycles of substrate binding and release. The HSP70-associated co-chaperones are of three types: J-domain co-chaperones HSP40s (stimulate ATPase hydrolysis by HSP70), the nucleotide exchange factors (NEF) such as BAG1/2/3 (facilitate conversion of HSP70 from the ADP-bound to the ATP-bound state thereby promoting substrate release), and the TPR domain chaperones such as HOPX and STUB1. Plays a critical role in mitochondrial import, delivers preproteins to the mitochondrial import receptor TOMM70. Acts as a repressor of transcriptional activation. Inhibits the transcriptional coactivator activity of CITED1 on Smad-mediated transcription. Component of the PRP19-CDC5L complex that forms an integral part of the spliceosome and is required for activating pre-mRNA splicing. May have a scaffolding role in the spliceosome assembly as it contacts all other components of the core complex. Binds bacterial lipopolysaccharide (LPS) and mediates LPS-induced inflammatory response, including TNF secretion by monocytes. Participates in the ER-associated degradation (ERAD) quality control pathway in conjunction with J domain-containing co-chaperones and the E3 ligase STUB1. Interacts with VGF-derived peptide TLQP-21.

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