

## Anti-Grp75 Monoclonal Antibody

Catalog Number: M02561

### About HSPA9

HSP27s belong to an abundant and ubiquitous family of small heat shock proteins (sHSP). It is an important HSP found in both normal human cells and cancer cells. The basic structure of most sHSPs is a homologous and highly conserved amino acid sequence, with an alpha-crystallin domain at the C-terminus and the WD/EPF domain at the less conserved N-terminus. This N-terminus is essential for the development of high molecular oligomers (1, 2). HSP27-oligomers consist of stable dimers formed by as many as 8-40 HSP27 protein monomers (3). The oligomerization status is connected with the chaperone activity: aggregates of large oligomers have high chaperone activity, whereas dimers have no chaperone activity (4). HSP27 is localized to the cytoplasm of unstressed cells but can redistribute to the nucleus in response to stress, where it may function to stabilize DNA and/or the nuclear membrane. Other functions include chaperone activity (as mentioned above), thermo tolerance in vivo, inhibition of apoptosis, and signal transduction. Specifically, in vitro, it acts as an ATP independent chaperone by inhibiting protein aggregation and by stabilizing partially denatured proteins, which ensures refolding of the HSP70 complex. HSP27 is also involved in the apoptotic signaling pathway because it interferes with the activation of cytochrome c/Apaf-1/dATP complex, thereby inhibiting the activation of procaspase-9. It is also hypothesized that HSP27 may serve some role in cross-bridge formation between actin and myosin (5). And finally, HSP27 is also thought to be involved in the process of cell differentiation. The up-regulation of HSP27 correlates with the rate of phosphorylation and with an increase of large oligomers. It is possible that HSP27 may play a crucial role in termination of growth (6).

### Overview

|                      |  |
|----------------------|--|
| Product Name         | Anti-Grp75 Monoclonal Antibody   |
| Reactive Species     | Human, Mouse, Rat  |
| Description          | Boster Bio Anti-Grp75 Monoclonal Antibody catalog # M02561. Tested in ELISA, IP, IF, WB applications. This antibody reacts with Human, Mouse, Rat. |
| Application          | ELISA, IP, IF, WB  |
| Clonality            | Monoclonal S52A-42   |
| Formulation          | Antibody concentration: 1 mg/ml, stored in PBS pH7.2, 50% glycerol, 0.09% sodium azide   |
| Storage Instructions | Store at -20°C for one year. Avoid repeated freeze-thaw cycles.  |
| Host                 | Mouse  |
| Uniprot ID           | P38646   |

### Technical Details

|                            |   |
|----------------------------|---|
| Immunogen                  | Fusion protein amino acids 551-766 of mouse SALM2.  |
| Predicted Reactive Species | Chimpanzee, Hamster   |
| Cross Reactivity           | Detects ~22kDa. Detects endogenous and exogenous HSP22 in monomeric, dimeric and tetrameric |

|                     |   |
|---------------------|---|
|                     | forms in WB. Does not cross react with alpha crystallin.  |
| Isotype             | IgG1 Kappa  |
| Form                | liquid  |
| Concentration       | 0.5-1mg/ml, actual concentration vary by lot. Use suggested dilution ratio to decide dilution procedure.  |
| Purification        | Protein G Purified  |
| Suggested Dilutions | <p>Dilute the sample so that the expected range of concentrations fall within the detection range of this kit.</p> <p>If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples.</p> <p>Some PubMed article(s) citing the expression level of this target are as follows:</p> <p>Boster Bio's internal QC testing used:</p> <p>WB (1:1000), ICC/IF (1:200) ; optimal dilutions for assays should be determined by the user.</p> |

## Anti-Grp75 Monoclonal Antibody (M02561) Images

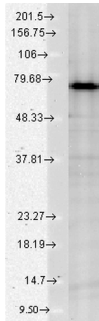
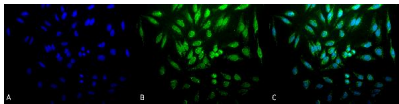


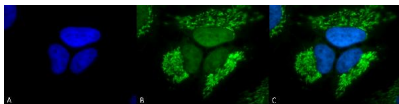
Figure 2. Western blot analysis of HSPA9 using anti-HSPA9 antibody (M02561).

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.

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 rabbit anti-HSPA9 antigen affinity purified polyclonal antibody (Catalog # M02561) at 0.5 ug/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 # SA1021) with Tanon 5200 system. A specific band was detected for HSPA9.



Immunocytochemistry/Immunofluorescence analysis using Mouse Anti-Grp75 Monoclonal Antibody, Clone S52A-42 (M02561) . Tissue: Heat Shocked HeLa Cells. Species: Human. Fixation: 2% Formaldehyde for 20 min at RT. Primary Antibody: Mouse Anti-Grp75 Monoclonal Antibody (M02561) at 1:100 for 12 hours at 4°C. Secondary Antibody: FITC Goat Anti-Mouse (green) at 1:200 for 2 hours at RT. Counterstain: DAPI (blue) nuclear stain at 1:40000 for 2 hours at RT. Localization: Mitochondria. Magnification: 20x. (A) DAPI (blue) nuclear stain. (B) Anti-Grp75 Antibody. (C) Composite. Heat Shocked at 42°C for 1h.



Immunocytochemistry/Immunofluorescence analysis using Mouse Anti-Grp75 Monoclonal Antibody, Clone S52A-42 (M02561) . Tissue: Heat Shocked HeLa Cells. Species: Human. Fixation: 2% Formaldehyde for 20 min at RT. Primary Antibody: Mouse Anti-Grp75 Monoclonal Antibody (M02561) at 1:100 for 12 hours at 4°C. Secondary Antibody: FITC Goat Anti-Mouse (green) at 1:200 for 2 hours at RT. Counterstain: DAPI (blue) nuclear stain at 1:40000 for 2 hours at RT. Localization: Mitochondria. Magnification: 100x. (A) DAPI (blue) nuclear stain. (B) Anti-Grp75 Antibody. (C) Composite. Heat Shocked at 42°C for 1h.

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