

# Anti-HSPB8/Hsp22 Antibody Picoband<sup>™</sup> (monoclonal, 7D8)

Catalog # ABO14941

#### Specification

## Anti-HSPB8/Hsp22 Antibody Picoband<sup>™</sup> (monoclonal, 7D8) - Product Information

Application Primary Accession Host Isotype Reactivity Clonality Format Description WB, IF, ICC, FC <u>O9UJY1</u> Mouse Mouse IgG2b Rat, Human Monoclonal Lyophilized

Anti-HSPB8/Hsp22 Antibody Picoband<sup>™</sup> (monoclonal, 7D8) . Tested in Flow Cytometry, IF, ICC, WB applications. This antibody reacts with Human, Rat.

Reconstitution

Add 0.2ml of distilled water will yield a concentration of 500ug/ml.

### Anti-HSPB8/Hsp22 Antibody Picoband<sup>™</sup> (monoclonal, 7D8) - Additional Information

Gene ID 26353

**Other Names** Heat shock protein beta-8, HspB8, Alpha-crystallin C chain, E2-induced gene 1 protein, Heat shock protein family B member 8, Protein kinase H11, Small stress protein-like protein HSP22, HSPB8, CRYAC, E2IG1, HSP22

Calculated MW 22 kDa KDa

**Application Details** Western blot, 0.1-0.5 μg/ml, Human, Rat<br> Immunocytochemistry/Immunofluorescence, 5 μg/ml, Human<br> Flow Cytometry, 1-3 μg/1x10<sup>6</sup> cells, Human<br>

Subcellular Localization Nucleus. Cytoplasm.

**Tissue Specificity** Predominantly expressed in skeletal muscle and heart.

Contents

Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na<sub>2</sub>HPO<sub>4</sub>, 0.05mg NaN<sub>3</sub>.

Immunogen

E.coli-derived human HSPB8/Hsp22 recombinant protein (Position: M1-T196). Human HSPB8/Hsp22 shares 94.4% and 95.4% amino acid (aa) sequence identity with mouse and rat HSPB8/Hsp22, respectively.



**Purification** Immunogen affinity purified.

**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-HSPB8/Hsp22 Antibody Picoband<sup>™</sup> (monoclonal, 7D8) - Protein Information

Name HSPB8

Synonyms CRYAC, E2IG1, HSP22

Function

Involved in the chaperone-assisted selective autophagy (CASA), a crucial process for protein quality control, particularly in mechanical strained cells and tissues such as muscle. Displays temperature-dependent chaperone activity.

**Cellular Location** 

Cytoplasm. Nucleus Note=Translocates to nuclear foci during heat shock

**Tissue Location** 

Predominantly expressed in skeletal muscle and heart.

## Anti-HSPB8/Hsp22 Antibody Picoband™ (monoclonal, 7D8) - Protocols

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

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

Anti-HSPB8/Hsp22 Antibody Picoband™ (monoclonal, 7D8) - Images





Figure 1. Western blot analysis of HSPB8/Hsp22 using anti-HSPB8/Hsp22 antibody (M02492-2). 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 Hela whole cell lysates;

Lane 2: human T-47D whole cell lysates;

Lane 3: rat RH35 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-HSPB8/Hsp22 antigen affinity purified monoclonal antibody (Catalog # M02492-2) at 0.5  $\mu$ 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 HSPB8/Hsp22 at approximately 22KD. The expected band size for HSPB8/Hsp22 is at 22KD.



Figure 2. Flow Cytometry analysis of CACO-2 cells using anti-HSPB8/Hsp22 antibody (M02492-2). Overlay histogram showing CACO-2 cells stained with M02492-2 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-HSPB8/Hsp22 Antibody (M02492-2, 1  $\mu$ g/1x10<sup>6</sup> cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10  $\mu$ g/1x10<sup>6</sup> cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1  $\mu$ g/1x10<sup>6</sup>) used under the same conditions. Unlabelled sample (Red line) was also used as a control.





Figure 3. Flow Cytometry analysis of U20S cells using anti-HSPB8/Hsp22 antibody (M02492-2). Overlay histogram showing U20S cells stained with M02492-2 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-HSPB8/Hsp22 Antibody (M02492-2, 1  $\mu$ g/1x10<sup>6</sup> cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10  $\mu$ g/1x10<sup>6</sup> cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1  $\mu$ g/1x10<sup>6</sup>) used under the same conditions. Unlabelled sample (Red line) was also used as a control.



Figure 4. IF analysis of HSPB8/Hsp22 using anti-HSPB8/Hsp22 antibody (M02492-2).

HSPB8/Hsp22 was detected in immunocytochemical section of A431 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 5  $\mu$ g/mL mouse anti-HSPB8/Hsp22 Antibody (M02492-2) 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-HSPB8/Hsp22 Antibody Picoband<sup>™</sup> (monoclonal, 7D8) - Background

Heat shock protein beta-8 is a protein that in humans is encoded by the HSPB8 gene. The protein encoded by this gene belongs to the superfamily of small heat-shock proteins containing a conservative alpha-crystallin domain at the C-terminal part of the molecule. The expression of this gene in induced by estrogen in estrogen receptor-positive breast cancer cells, and this protein also functions as a chaperone in association with Bag3, a stimulator of macroautophagy. Thus, this gene appears to be involved in regulation of cell proliferation, apoptosis, and carcinogenesis, and mutations in this gene have been associated with different neuromuscular diseases, including Charcot-Marie-Tooth disease.