

Anti-Alpha A Crystallin Antibody Picoband™ (monoclonal, 8H6)

Catalog # ABO15071

Product Information

Application	WB, FC
Primary Accession	P02489
Host	Mouse
Isotype	Mouse IgG2b
Reactivity	Rat, Human, Mouse
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-Alpha A Crystallin Antibody Picoband™ (monoclonal, 8H6) . Tested in Flow Cytometry, WB applications. This antibody reacts with Human, Mouse, Rat.
Reconstitution	Adding 0.2 ml of distilled water will yield a concentration of 500 µg/ml.

Additional Information

Gene ID	102724652;1409
Other Names	Alpha-crystallin A chain, Heat shock protein beta-4, HspB4, Heat shock protein family B member 4, Alpha-crystallin A(1-172), Alpha-crystallin A(1-168), Alpha-crystallin A(1-162), CRYAA, CRYA1, HSPB4
Calculated MW	19909
Application Details	Western blot, 0.25-0.5 µg/ml, Mouse, Rat Flow Cytometry, 1-3 µg/1x10 ⁶ cells, Human
Source	Eukaryota
Contents	Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na ₂ HPO ₄ .
Clone Names	Clone: 8H6
Immunogen	E. coli-derived human Alpha A Crystallin recombinant protein (Position: M1-S173). Human Alpha A Crystallin shares 94.8% amino acid (aa) sequence identity with both mouse and rat Alpha A Crystallin.
Purification	Immunogen affinity purified.
Storage	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 freezing and thawing.

Protein Information

Name	CRYAA
Synonyms	CRYA1, HSPB4
Function	Contributes to the transparency and refractive index of the lens (PubMed: 18302245). In its oxidized form (absence of intramolecular disulfide bond), acts as a chaperone, preventing aggregation of various proteins under a wide range of stress conditions (PubMed: 18199971 , PubMed: 19595763 , PubMed: 22120592 , PubMed: 31792453). Required for the correct formation of lens intermediate filaments as part of a complex composed of BFSP1, BFSP2 and CRYAA (PubMed: 28935373).
Cellular Location	Cytoplasm. Nucleus. Note=Translocates to the nucleus during heat shock and resides in sub-nuclear structures known as SC35 speckles or nuclear splicing speckles
Tissue Location	Expressed in the eye lens (at protein level).

Background

Alpha-crystallin A chain is a protein that in humans is encoded by the CRYAA gene. Mammalian lens crystallins are divided into alpha, beta, and gamma families. Alpha crystallins are composed of two gene products: alpha-A and alpha-B, for acidic and basic, respectively. Alpha crystallins can be induced by heat shock and are members of the small heat shock protein (HSP20) family. They act as molecular chaperones although they do not renature proteins and release them in the fashion of a true chaperone; instead they hold them in large soluble aggregates. Two additional functions of alpha crystallins are an autokinase activity and participation in the intracellular architecture. The encoded protein has been identified as a moonlighting protein based on its ability to perform mechanistically distinct functions. Alpha-A and alpha-B gene products are differentially expressed; alpha-A is preferentially restricted to the lens and alpha-B is expressed widely in many tissues and organs. Defects in this gene cause autosomal dominant congenital cataract (ADCC).

Images

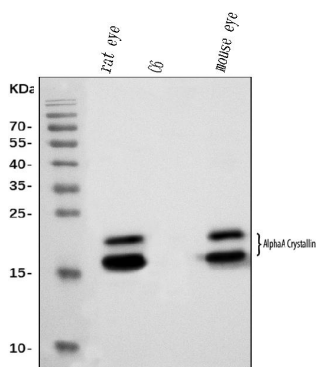


Figure 1. Western blot analysis of Alpha A Crystallin using anti-Alpha A Crystallin antibody (M01900-3).

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: rat eye tissue lysates,
Lane 2: rat C6 whole cell lysates,
Lane 3: mouse eye tissue lysates.

After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA 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-Alpha A Crystallin antigen affinity purified monoclonal antibody (Catalog # M01900-3) at 0.5 µg/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 Alpha A Crystallin at approximately 20-23 kDa. The expected band size for Alpha A Crystallin is at 20-23 kDa.

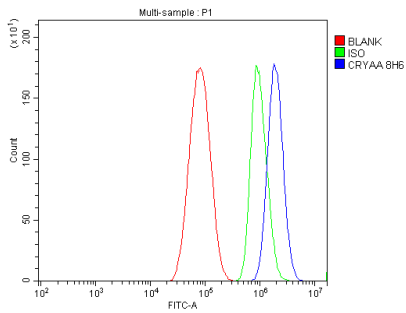


Figure 2. Flow Cytometry analysis of HepG2 cells using anti-Alpha A Crystallin antibody (M01900-3). Overlay histogram showing HepG2 cells stained with M01900-3 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-Alpha A Crystallin Antibody (M01900-3, 1 μ g/ 1×10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 μ g/ 1×10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 μ g/ 1×10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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