abcam

Product datasheet

Alexa Fluor® 594 Anti-Hsp27 antibody [EPR5477] ab215328



Recombinant

RabMAb

3 Images

Overview

Product name Alexa Fluor® 594 Anti-Hsp27 antibody [EPR5477]

Alexa Fluor® 594 Rabbit monoclonal [EPR5477] to Hsp27 **Description**

Host species Rabbit

Alexa Fluor® 594, Ex: 590nm, Em: 617nm Conjugation

Tested applications Suitable for: ICC/IF, Flow Cyt (Intra)

Species reactivity Reacts with: Human

Predicted to work with: Mouse, Rat

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control ICC/IF: HeLa cells Flow Cyt (intra): HAP1-WT cells

General notes This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity
- Long-term security of supply
- Animal-free production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

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Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Avoid freeze / thaw cycle. Store In the Dark.

Storage buffer pH: 7.40

Preservative: 0.02% Sodium azide

Constituents: 1% BSA, 30% Glycerol (glycerin, glycerine), PBS

Purity Protein A purified

ClonalityMonoclonalClone numberEPR5477

Isotype IgG

Applications

The Abpromise guarantee Our Abpromise

Our <u>Abpromise guarantee</u> covers the use of ab215328 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
ICC/IF		1/100. This product gave a positive signal in HeLa cells fixed with 4% formaldehyde (10 min) and 100% methanol (5 min)
Flow Cyt (Intra)		Use a concentration of 1 µg/ml.

Target

Function Involved in stress resistance and actin organization.

Tissue specificity Detected in all tissues tested: skeletal muscle, heart, aorta, large intestine, small intestine,

stomach, esophagus, bladder, adrenal gland, thyroid, pancreas, testis, adipose tissue, kidney, liver, spleen, cerebral cortex, blood serum and cerebrospinal fluid. Highest levels are found in the

heart and in tissues composed of striated and smooth muscle.

Involvement in diseaseDefects in HSPB1 are the cause of Charcot-Marie-Tooth disease type 2F (CMT2F)

[MIM:606595]. CMT2F is a form of Charcot-Marie-Tooth disease, the most common inherited disorder of the peripheral nervous system. Charcot-Marie-Tooth disease is classified in two main groups on the basis of electrophysiologic properties and histopathology: primary peripheral demyelinating neuropathy or CMT1, and primary peripheral axonal neuropathy or CMT2. Neuropathies of the CMT2 group are characterized by signs of axonal regeneration in the absence of obvious myelin alterations, normal or slightly reduced nerve conduction velocities, and progressive distal muscle weakness and atrophy. Nerve conduction velocities are normal or slightly reduced. CMT2F onset is between 15 and 25 years with muscle weakness and atrophy usually beginning in feet and legs (peroneal distribution). Upper limb involvement occurs later.

CMT2F inheritance is autosomal dominant.

Defects in HSPB1 are a cause of distal hereditary motor neuronopathy type 2B (HMN2B) [MIM:608634]. Distal hereditary motor neuronopathies constitute a heterogeneous group of neuromuscular disorders caused by selective impairment of motor neurons in the anterior horn of the spinal cord, without sensory deficit in the posterior horn. The overall clinical picture consists of a classical distal muscular atrophy syndrome in the legs without clinical sensory loss. The disease starts with weakness and wasting of distal muscles of the anterior tibial and peroneal compartments of the legs. Later on, weakness and atrophy may expand to the proximal muscles of the lower limbs and/or to the distal upper limbs.

Sequence similarities

Post-translational modifications

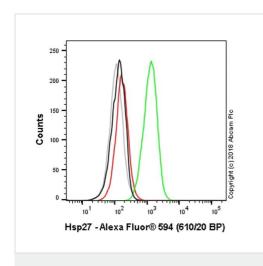
Cellular localization

Belongs to the small heat shock protein (HSP20) family.

Phosphorylated in MCF-7 cells on exposure to protein kinase C activators and heat shock.

Cytoplasm. Nucleus. Cytoplasm > cytoskeleton > spindle. Cytoplasmic in interphase cells. Colocalizes with mitotic spindles in mitotic cells. Translocates to the nucleus during heat shock and resides in sub-nuclear structures known as SC35 speckles or nuclear splicing speckles.

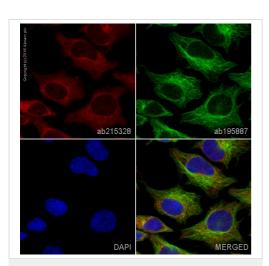
Images



Flow Cytometry (Intracellular) - Alexa Fluor® 594 Anti-Hsp27 antibody [EPR5477] (ab215328) Overlay histogram showing HAP1 wildtype (green line) and HAP1-HSPB1 knockout cells (red line) stained with ab215328. The cells were fixed with 4% formaldehyde (10 min) and then permeabilized with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS / 10% normal goat serum to block non-specific protein-protein interactions followed by the antibody (ab215328, 1µg/ml dilution) for 30 min at 22°C.

A rabbit monoclonal IgG isotype control antibody (<u>ab208568</u>) was used at the same concentration and conditions as the primary antibody (HAP1 wildtype - black line, HAP1-HSPB1 knockout - grey line). Unlabelled sample was also used as a control (this line is not shown for the purpose of simplicity).

Acquisition of >5,000 events were collected using a 50 mW Yellow/Green laser (561nm) and 610/20 bandpass filter.

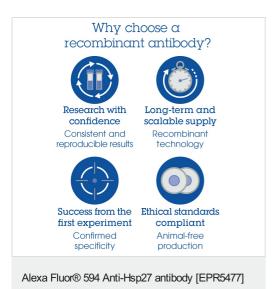


Immunocytochemistry/ Immunofluorescence - Alexa Fluor® 594 Anti-Hsp27 antibody [EPR5477] (ab215328)

ab215328 staining Hsp27 in HeLa cells. The cells were fixed with 100% methanol (5 min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with ab215328 at 1/100 dilution (pseudocolored in red) and ab195887, Mouse monoclonal to alpha Tubulin (Alexa Fluor® 488), at 1/250 dilution (shown in green). Nuclear DNA was labelled with DAPI (shown in blue).

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

This product also gave a positive signal under the same testing conditions in HeLa cells fixed with 4% formaldehyde (10 min).



(ab215328)

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