abcam

Product datasheet

Anti-Hsp27 antibody [EPR5477] - BSA and Azide free ab229442

敲除验证 重组 RabMAb

5 图像

概述

产品名称	Anti-Hsp27抗体[EPR5477] - BSA and Azide free
描述	兔单克隆抗体[EPR5477] to Hsp27 - BSA and Azide free
宿主	Rabbit
经测试应用	**适用于:** WB, ICC/IF, Flow Cyt (Intra)
种属反应性	**与反应:** Human, African green monkey **预测可用于:** Rat
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
阳性对照	WB: HeLa, HAP1, COS-1, BxPC-3, and HT-1376 cell lysates. ICC/IF: HeLa cells. Flow Cyt (intra): HAP1 cells.
常规说明	ab229442 is the carrier-free version of **ab109376**. Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications. Use our **conjugation kits** for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold. This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C. Do Not Freeze.
存储溶液	pH: 7.20 Constituent: PBS

无载体	**是**
纯度	Protein A purified
克隆	单克隆
克隆编号	EPR5477
同种型	IgG

应用

The Abpromise guarantee **Abpromise™承诺保证使用ab229442于以下的经测试应用**

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

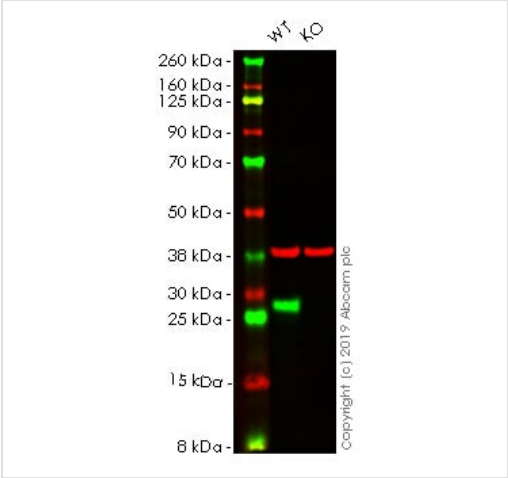
应用	Ab评论	说明
WB		Use at an assay dependent concentration. Predicted molecular weight: 23 kDa.
ICC/IF		Use at an assay dependent concentration.
Flow Cyt (Intra)		Use at an assay dependent concentration. **ab199376** -Rabbit monoclonal IgG (Low endotoxin, Azide free), is suitable for use as an isotype control with this antibody.

靶标

功能	Involved in stress resistance and actin organization.
组织特异性	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.
疾病相关	Defects 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.
序列相似性	Belongs to the small heat shock protein (HSP20) family.
翻译后修饰	Phosphorylated in MCF-7 cells on exposure to protein kinase C activators and heat shock.
细胞定位	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.

图片



Western blot - Anti-Hsp27 antibody [EPR5477] - BSA and Azide free (ab229442)

All lanes : Anti-Hsp27 antibody [EPR5477] (**ab109376**) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : HSPB1 knockout HeLa cell lysate

Lysates/proteins at 20 µg per lane.

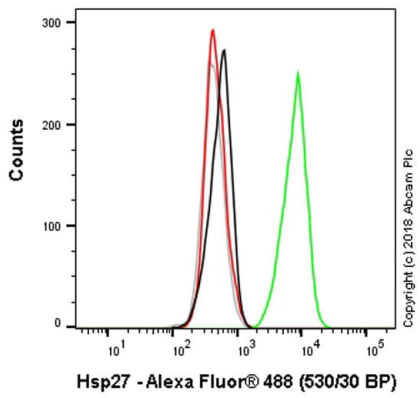
Performed under reducing conditions.

Predicted band size: 23 kDa
Observed band size: 23 kDa

This data was developed using the same antibody clone in a different buffer formulation (**ab109376**).

Lanes 1- 2: Merged signal (red and green). Green - **ab109376** observed at 23 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

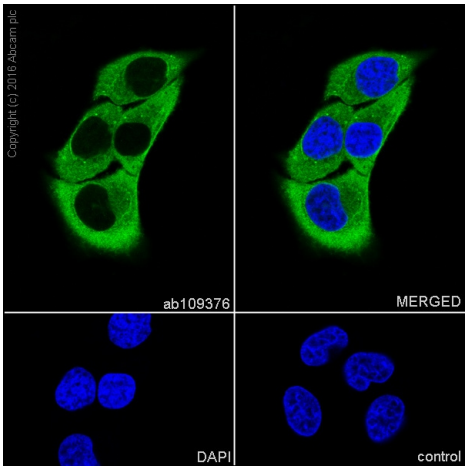
ab109376 was shown to react with Hsp27 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line **ab261738** (knockout cell lysate **ab256945**) was used. Wild-type HeLa and HSPB1 knockout HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. **ab109376** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye®800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye®680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Flow Cytometry (Intracellular) - Anti-Hsp27 antibody [EPR5477] - BSA and Azide free (ab229442)

Overlay histogram showing HAP1 wildtype (green line) and HAP1-HSPB1 knockout cells (red line) stained with **ab109376**. The cells were fixed with 4% formaldehyde (10 min)ï¿½ 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 (**ab109376**, 1ï¿½g/ml) for 30 min at 22ï¿½C. The secondary antibody used was Alexa Fluorï¿½ 488 goat anti-rabbit IgG (H&L) presorbed (**ab150081**) at 1/2000 dilution for 30 min at 22ï¿½C. A rabbit IgG isotype control antibodyï¿½ (**ab172730**) 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 Blue laser (488nm) and 530/30 bandpass filter.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109376**).

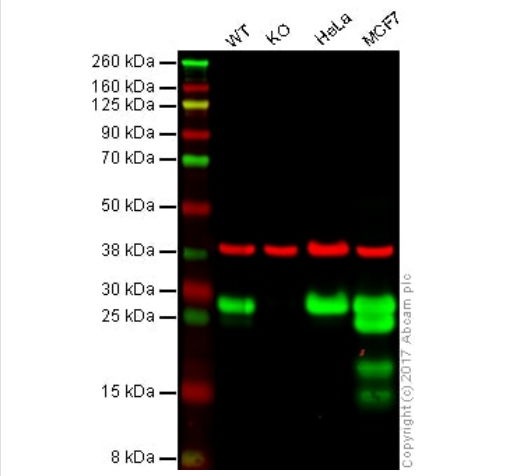


Immunocytochemistry/ Immunofluorescence - Anti-Hsp27 antibody [EPR5477] - BSA and Azide free (ab229442)

Immunocytochemistry/Immunofluorescence analysis of HeLa (human cervix adenocarcinoma) cells labelling Hsp27 with purified **ab109376** at 1/500. Cells were fixed with 100% methanol. **ab150077**, Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000) was used as the secondary antibody. Nuclei were counterstained with DAPI (blue).

Secondary Only Control: PBS was used instead of the primary antibody as the negative control.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109376**).



Western blot - Anti-Hsp27 antibody [EPR5477] - BSA and Azide free (ab229442)

All lanes : Anti-Hsp27 antibody [EPR5477] (**ab109376**) at 1/1000 dilution

Lane 1 : Wild-type HAP1 whole cell lysate
Lane 2 : Hsp27 knockout HAP1 whole cell lysate
Lane 3 : HeLa whole cell lysate
Lane 4 : MCF7 whole cell lysate

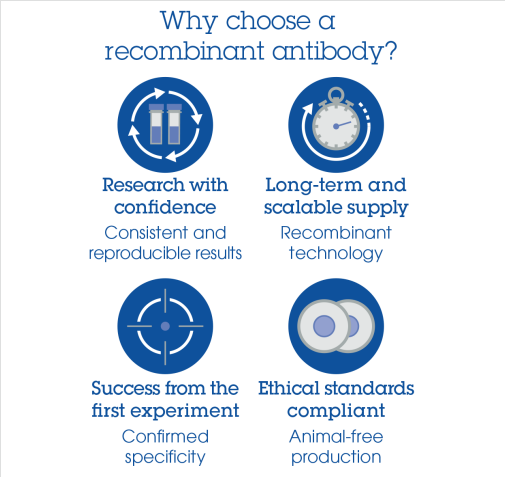
Lysates/proteins at 20 µg per lane.

Predicted band size: 23 kDa

This WB data was generated using the same anti-Hsp27 antibody clone, EPR5477, in a different buffer formulation (cat# **ab109376**).

Lanes 1 - 4: Merged signal (red and green). Green - **ab109376** observed at 27 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab109376 was shown to specifically react with Hsp27 in wild-type cells as signal was lost in Hsp27 knockout HEP1 cells. Wild-type and Hsp27 knockout samples were subjected to SDS-PAGE. Ab109376 and **ab8245** (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed **ab216773** and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed **ab216776** secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



Anti-Hsp27 antibody [EPR5477] - BSA and Azide free (ab229442)

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