abcam

Product datasheet

Anti-Hsp27 antibody [EPR5477] ab109376

KO 評価済 リコンビナント RabMAb

2 Abreviews 7 References 画像数 6

製品の概要

製品名	Anti-Hsp27 antibody [EPR5477]
製品の詳細	Rabbit monoclonal [EPR5477] to Hsp27
由来種	Rabbit
アプリケーション	**適用あり:** Flow Cyt (Intra), ICC/IF, WB
種交差性	**交差種:** Human, African green monkey **交差が予測される動物種:** Rat
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
ポジティブ・コントロール	WB: HeLa, HAP1, MCF7, COS-1, BxPC-3 and HT-1376 cell lysates. ICC/IF: HeLa cells. Flow Cyt (intra): HAP1 cells.
特記事項	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**.

製品の特性

製品の状態	Liquid
保存方法	Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C.
バッファー	pH: 7.20 Preservative: 0.05% Sodium azide Constituents: 0.1% BSA, 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue culture supernatant
精製度	Protein A purified
ポリ/モノ	モノクローナル
クローン名	EPR5477

アイソタイプ	IgG

アプリケーション

The Abpromise guarantee **Abpromise保証は、** 次のテスト済みアプリケーションにおけるab109376の使用に適用されます

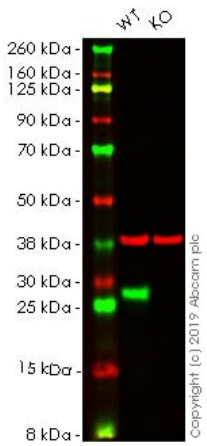
アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
Flow Cyt (Intra)		1/10 - 1/100. **ab172730** -Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
ICC/IF		1/500.
WB	★★★★★ (2)	1/1000 - 1/10000. Predicted molecular weight: 23 kDa.

ターゲット情報

機能	Involved in stress resistance and actin organization.
組織特異性	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.
関連疾患	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.
配列類似性	Belongs to the small heat shock protein (HSP20) family.
翻訳後修飾	Phosphorylated in MCF-7 cells on exposure to protein kinase C activators and heat shock.
細胞内局在	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.

画像



Western blot - Anti-Hsp27 antibody [EPR5477] (ab109376)

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

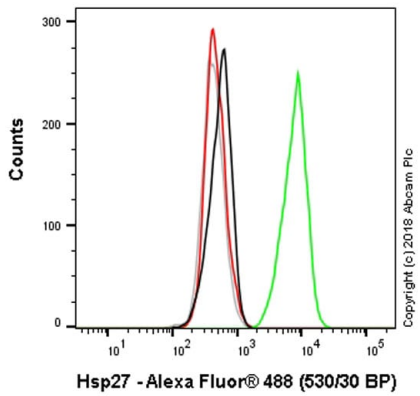
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

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

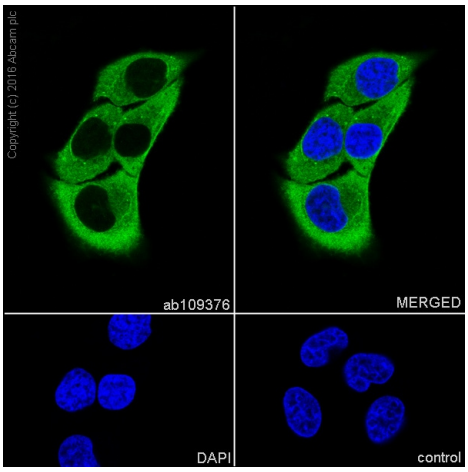
Lanes 1- 2: Merged signal (red and green). Green - ab109376 observed at 23 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

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] (ab109376)

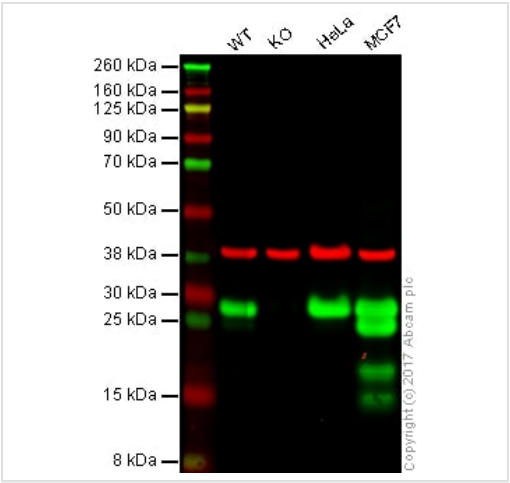
Overlay histogram showing wild-type HAP1 (green line) and HSPB1 knockout HAP1 cells (red line) stained with ab109376. The cells were fixed with 4% formaldehyde (10 min) and then permeabilized with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS / 10% normal goat serum to block non-specific protein-protein interactions followed by the antibody (ab109376, 1µg/ml) for 30 min at 22°C. The secondary antibody used was Alexa Fluorr® 488 goat anti-rabbit IgG (H&L) presorbed (**ab150081**) at 1/2000 dilution for 30 min at 22°C. A rabbit IgG isotype control antibody (**ab172730**) was used at the same concentration and conditions as the primary antibody (HAP1 wildtype - black line, knockout HSPB1 HAP1- 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.



Immunocytochemistry/ Immunofluorescence - Anti-Hsp27 antibody [EPR5477] (ab109376)

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.



Western blot - Anti-Hsp27 antibody [EPR5477] (ab109376)

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

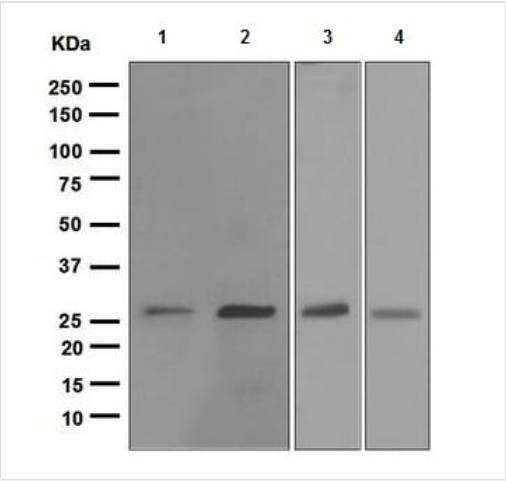
Lane 1 : Wild-type HAP1 whole cell lysate
Lane 2 : Hsp27 knockout HAP1 whole cell lysate
Lane 3 : HeLa whole cell lysate
Lane 4 : MCF7 whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 23 kDa

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 HAP1 cells as signal was lost in Hsp27 knockout HAP1 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.



Western blot - Anti-Hsp27 antibody [EPR5477] (ab109376)

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

Lane 1 : HeLa cell lysate
Lane 2 : COS-1 cell lysate
Lane 3 : BxPC-3 cell lysate
Lane 4 : HT-1376 cell lysate

Lysates/proteins at 10 µg per lane.

Predicted band size: 23 kDa
Observed band size: 27 kDa

Anti-Hsp27 antibody [EPR5477] (ab109376)

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