abcam

Product datasheet

Anti-SHP2 (phospho Y542) antibody ab17939

2 References 3 Images

Overview

Product name	Anti-SHP2 (phospho Y542) antibody
Description	Rabbit polyclonal to SHP2 (phospho Y542)
Host species	Rabbit
Tested applications	**Suitable for:** IHC-P, ICC/IF, WB
Species reactivity	**Reacts with:** Mouse, Human
Immunogen	Synthetic peptide corresponding to Human SHP2 (phospho Y542). The sequence is conserved in mouse, rat and chicken.
Positive control	WB: NIH3T3 cells treated with PDGF. IHC-P: Human brain tissue. ICC/IF: A-431
General notes	The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Store at -20°C long term.
Storage buffer	pH: 7.30 Preservative: 0.05% Sodium azide Constituents: PBS, 50% Glycerol (glycerin, glycerine), 0.1% BSA
Purity	Immunogen affinity purified
Purification notes	Purified from rabbit serum by sequential epitope-specific chromatography. The antibody has been negatively preadsorbed using a non-phosphopeptide corresponding to the site of phosphorylation to remove antibody that is reactive with non-phosphorylated SHP2. The final product is generated by affinity chromatography using a SHP2 derived peptide that is phosphorylated at tyrosine 542.
Clonality	Polyclonal
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab17939 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
IHC-P		1/20. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
ICC/IF		Use a concentration of 5 µg/ml.
WB		1/1000. Detects a band of approximately 70 kDa (predicted molecular weight: 70 kDa).

Target

Function Acts downstream of various receptor and cytoplasmic protein tyrosine kinases to participate in the signal transduction from the cell surface to the nucleus.

Tissue specificity Widely expressed, with highest levels in heart, brain, and skeletal muscle.

Involvement in disease Defects in PTPN11 are the cause of LEOPARD syndrome type 1 (LEOPARD1) [MIM:151100]. It is an autosomal dominant disorder allelic with Noonan syndrome. The acronym LEOPARD stands for lentigines, electrocardiographic conduction abnormalities, ocular hypertelorism, pulmonic stenosis, abnormalities of genitalia, retardation of growth, and deafness.
Defects in PTPN11 are the cause of Noonan syndrome type 1 (NS1) [MIM:163950]. Noonan syndrome (NS) is a disorder characterized by dysmorphic facial features, short stature, hypertelorism, cardiac anomalies, deafness, motor delay, and a bleeding diathesis. Some patients with Noonan syndrome type 1 develop multiple giant cell lesions of the jaw or other bony or soft tissues, which are classified as pigmented villomoduolar synovitis (PVNS) when occurring in the jaw or joints. Note=Mutations in PTPN11 account for more than 50% of the cases. Rarely, NS is associated with juvenile myelomonocytic leukemia (JMML). NS1 inheritance is autosomal dominant.
Defects in PTPN11 are a cause of juvenile myelomonocytic leukemia (JMML) [MIM:607785]. JMML is a pediatric myelodysplastic syndrome that constitutes approximately 30% of childhood cases of myelodysplastic syndrome (MDS) and 2% of leukemia. It is characterized by leukocytosis with tissue infiltration and in vitro hypersensitivity of myeloid progenitors to granulocyte-macrophage colony stimulating factor.
Defects in PTPN11 are a cause of metachondromatosis (MC) [MIM:156250]. It is a skeletal disorder with radiologic fetarures of both multiple exostoses and Ollier disease, characterized by the presence of multiple enchondromas and osteochondroma-like lesions.

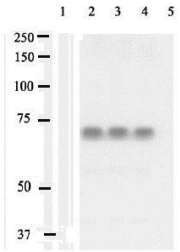
Sequence similarities Belongs to the protein-tyrosine phosphatase family. Non-receptor class 2 subfamily.
Contains 2 SH2 domains.
Contains 1 tyrosine-protein phosphatase domain.

Domain The SH2 domains repress phosphatase activity. Binding of these domains to phosphotyrosine-containing proteins relieves this auto-inhibition, possibly by inducing a conformational change in the enzyme.

Post-translational Phosphorylated on Tyr-546 and Tyr-584 upon receptor protein tyrosine kinase activation; which

modifications	creates a binding site for GRB2 and other SH2-containing proteins.
Cellular localization	Cytoplasm.

Images

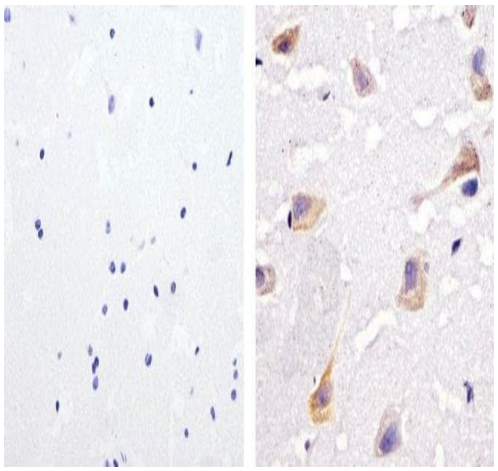


Western blot - Anti-SHP2 (phospho Y542) antibody (ab17939)

Western blot using ab17939 on 10-30µg NIH3T3 cell lysate. Lane 1: untreated cells. Lane 2: cells treated with PDGF. Lane 3: cells treated with PDGF. Antibody blocked with non-phosphorylated immunopeptide. Lane 4: cells treated with PDGF. Antibody blocked with a generic tyrosine-phosphorylated peptide. Lane 5: cells treated with PDGF. Antibody blocked with phosphorylated immunopeptide.

Western blot using ab17939 on 10-30µg NIH3T3 cell lysate.

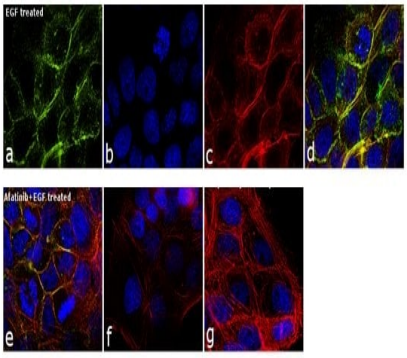
Lane 1: untreated cells.
Lane 2: cells treated with PDGF.
Lane 3: cells treated with PDGF. Antibody blocked with non-phosphorylated immunopeptide.
Lane 4: cells treated with PDGF. Antibody blocked with a generic tyrosine-phosphorylated peptide.
Lane 5: cells treated with PDGF. Antibody blocked with phosphorylated immunopeptide.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-SHP2 (phospho Y542) antibody (ab17939)

Immunohistochemical analysis of paraffin-embedded human brain labeling SHP2 (phospho Y542) with ab17939 at 1/20 dilution (right) compared to a negative control without primary antibody (left).

To expose target proteins, antigen retrieval was performed using 10mM sodium citrate (pH 6.0), microwaved for 8-15 min. Following antigen retrieval, tissues were blocked in 3% H2O2-methanol for 15 min at room temperature, washed with ddH2O and PBS, and then probed with ab17939 diluted in 3% BSA-PBS at a dilution of 1/20 overnight at 4°C in a humidified chamber. Tissues were washed extensively in PBST and detection was performed using an HRP-conjugated secondary antibody followed by colorimetric detection using a DAB kit. Tissues were counterstained with hematoxylin and dehydrated with ethanol and xylene to prep for mounting.



Immunocytochemistry/ Immunofluorescence - Anti-SHP2 (phospho Y542) antibody (ab17939)

Immunofluorescence analysis of 90% confluent log phase A-431 (Human epidermoid carcinoma cell line) cells treated with 0.2 ug/mL of EGF for 10 minutes labeling SHP2 (phospho Y542) with ab17939 at 1/250 dilution.

The cells were fixed with 4% paraformaldehyde for 10 minutes, permeabilized with 0.1% Triton™ X-100 for 10 minutes, and blocked with 1% BSA for 1 hour at room temperature. The cells were labeled with ab17939 at 1/250 dilution in 0.1% BSA and incubated overnight at 4 degree and then labeled with Goat anti-Rabbit IgG (H+L) Superclonal™ Secondary Antibody, Alexa Fluor® 488 conjugate at a dilution of 1/2000 for 45 minutes at room temperature (Panel a: green). Nuclei (Panel b: blue) were stained with SlowFade® Gold Antifade Mountant with DAPI. F-actin (Panel c: red) was stained with Rhodamine Phalloidin at 1/300 dilution. Panel d represents the merged image showing membrane localization. Panel e represents cells treated with antagonist, Afatinib (0.5 uM for 6hrs) followed by EGF (0.2 ug/mL for 10 minutes), showing reduced expression of SHP2 (phospho Y542). Panel f shows untreated cells with no signal. Panel g represents control cells with no primary antibody to assess background. The images were captured at 60X magnification.

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