# abcam

#### Product datasheet

## Anti-SHP1 antibody [EPR5519] ab124942





RabMAb

### 9 References 7 Images

#### Overview

Product name Anti-SHP1 antibody [EPR5519]

**Description** Rabbit monoclonal [EPR5519] to SHP1

Host species Rabbit

Tested applications Suitable for: Flow Cyt (Intra), WB, IHC-P, ICC/IF

Species reactivity Reacts with: Human

Does not react with: Mouse, Rat

**Immunogen** Synthetic peptide within Human SHP1 aa 1-100. The exact sequence is proprietary.

Database link: P29350

Positive control IHC-P: Human tonsil tissue. WB: THP1, Jurkat, Raji and lymph node lysates. ICC: Raji cells. Flow

Cyt (intra): Jurkat 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<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**<sup>®</sup> **patents**.

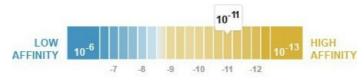
#### **Properties**

Form Liquid

**Storage instructions** Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C.

pH: 7.2

**Dissociation constant (K<sub>D</sub>)**  $K_D = 4.95 \times 10^{-11} M$ 



Learn more about K<sub>D</sub>

Storage buffer

1

Preservative: 0.01% Sodium azide

Constituents: 9% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA, 50% Tissue culture

supernatant

Purity Tissue culture supernatant

Clonality Monoclonal
Clone number EPR5519

**Isotype** IgG

#### **Applications**

The Abpromise guarantee Our Abpromise guarantee covers the use of ab124942 in the following tested applications.

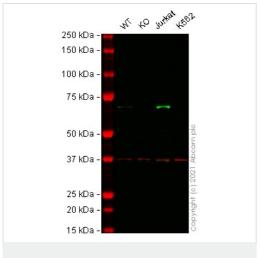
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		1/10 - 1/100.  ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB		1/1000 - 1/10000. Predicted molecular weight: 68 kDa.
IHC-P		1/1000 - 1/2000. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
ICC/IF		1/250 - 1/500.

rarget		
Function	Plays a key role in hematopoiesis. This PTPase activity may directly link growth factor receptors and other signaling proteins through protein-tyrosine phosphorylation. The SH2 regions may interact with other cellular components to modulate its own phosphatase activity against interacting substrates. Together with MTUS1, induces UBE2V2 expression upon angiotensin II stimulation.	
Tissue specificity	lsoform 1 is expressed in hematopoietic cells. Isoform 2 is expressed in non-hematopoietic cells	
Sequence similarities	Belongs to the protein-tyrosine phosphatase family. Non-receptor class 2 subfamily. Contains 2 SH2 domains.  Contains 1 tyrosine-protein phosphatase domain.	
Post-translational modifications	Phosphorylated on serine and tyrosine residues.	
Cellular localization	Cytoplasm. Nucleus. In neurons, translocates into the nucleus after treatment with angiotensin II.	

### **Images**

Target



Western blot - Anti-SHP1 antibody [EPR5519] (ab124942)

**All lanes :** Anti-SHP1 antibody [EPR5519] (ab124942) at 1/1000 dilution

Lane 1: Wild-type THP-1 cell lysate

Lane 2: PTPN6 knockout THP-1 cell lysate

Lane 3 : Jurkat cell lysate
Lane 4 : K562 cell lysate

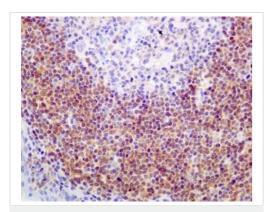
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**Predicted band size:** 68 kDa **Observed band size:** 70 kDa

**Lanes 1 - 4:** Merged signal (red and green). Green - ab124942 observed at 70 kDa. Red - loading control <u>ab8245</u> (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

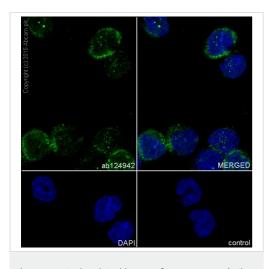
ab124942 was shown to react with SHP1 in wild-type THP-1 cells in Western blot with loss of signal observed in PTPN6 knockout sample. Wild-type THP-1 and PTPN6 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween<sup>®</sup>) before incubation with ab124942 and ab8245 (Mouse anti-GAPDH antibody [6C5]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit lgG H&L (IRDye<sup>®</sup> 800CW) preabsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye<sup>®</sup> 680RD) preabsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-SHP1 antibody
[EPR5519] (ab124942)

ab124942, at a 1/1000 dilution, staining SHP1 in paraffinembedded Human tonsil tissue by immunohistochemistry

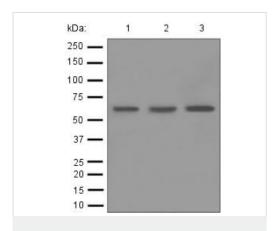
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-SHP1 antibody [EPR5519] (ab124942)

Immunocytochemistry/Immunofluorescence analysis of Raji (human Burkitt's lymphoma) cells labelling SHP1 with purified ab124942 at a dilution of 1/500. Cells were fixed with 4% Paraformaldehyde and permeabilised with 0.1% tritonX-100. An Alexa Fluor® 488-conjugated goat anti-rabbit lgG (ab150077) at dilution of 1/1000 was used as the secondary antibody. Nuclei counterstained with DAPI (blue).

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



Western blot - Anti-SHP1 antibody [EPR5519] (ab124942)

**All lanes :** Anti-SHP1 antibody [EPR5519] (ab124942) at 1/1000 dilution

Lane 1 : Jurkat cell lysate

Lane 2 : Raji cell lysate

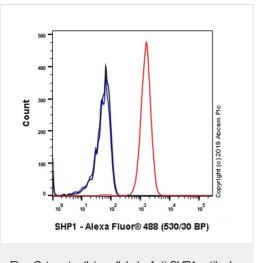
Lane 3: lymph node lysate

Lysates/proteins at 10 µg per lane.

#### **Secondary**

All lanes: standard HRP labelled goat anti-rabbit at 1/2000 dilution

Predicted band size: 68 kDa



Intracellular Flow Cytometry analysis of Jurkat (Human T cell leukemia T lymphocyte) cells labeling SHP1 with purified ab124942 at 1/100 dilution (10µg/mL) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit lgG (Alexa Fluor® 488, **ab150077**) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal lgG (Black). Unlabeled control - Cells without incubation with primary antibody and secondary antibody (Blue).

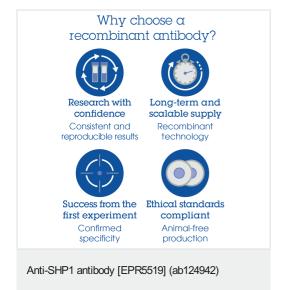
Flow Cytometry (Intracellular) - Anti-SHP1 antibody [EPR5519] (ab124942)

SHP-1 Probe Conc **EPR5519** \_\_\_ 60 nM 12.0 33 nM Surface Density (ng/mm²) 10.0 11 nM \_\_\_ 3.7 nM 8.0 6.0 Fit Summary  $\begin{array}{lll} & \text{kon [1/(sec-M)]} & = & (7.97 \pm 0.02) \, \text{E4} \\ & \text{koff [1/sec]} & = & (3.94 \pm 0.08) \, \text{E-6} \\ & \text{Kd [M]} & = & (4.95 \pm 0.10) \, \text{E-11} \end{array}$ 4.0 2.0 Fit RMSE [ng/mm²] = 1.8 E-1 0.0 -2.0 -4.0 0 30 60 90 135 Time (min)

OI-RD Scanning - Anti-SHP1 antibody [EPR5519] (ab124942)

Equilibrium disassociation constant ( $K_D$ ) Learn more about  $K_D$ 

Click here to learn more about K<sub>D</sub>



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