abcam

Product datasheet

Anti-SHIP-1 antibody [EP378Y] - BSA and Azide free ab190551

Recombinant RabMAb

12 Images

Overview

Product name	Anti-SHIP-1 antibody [EP378Y] - BSA and Azide free
Description	Rabbit monoclonal [EP378Y] to SHIP-1 - BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** Flow Cyt (Intra), WB, IHC-P, IP, ICC/IF
Species reactivity	**Reacts with:** Mouse, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	Human spleen tissue, Daudi cell lysate. Flow cyto (intra): Raji cells
General notes	ab190551 is the carrier-free version of **ab45142**. 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. 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**. Rat: We have preliminary internal testing data to indicate this antibody may not react with this

species. Please contact us for more information.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	pH: 7.2 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EP378Y
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab190551 in the following tested applications.

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

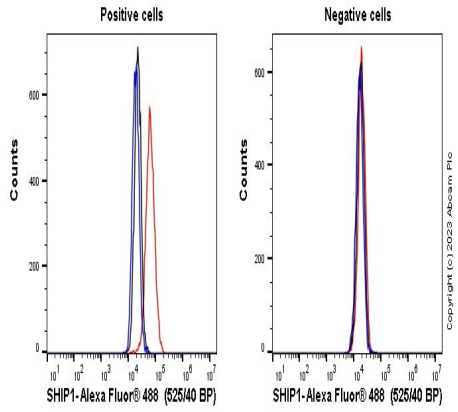
Application	Abreviews	Notes
Flow Cyt (Intra)		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 140 kDa (predicted molecular weight: 133 kDa).
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See **IHC antigen retrieval protocols**.
IP		Use at an assay dependent concentration.
ICC/IF		Use at an assay dependent concentration.

Target

Function Phosphatidylinositol (PtdIns) phosphatase that specifically hydrolyzes the 5-phosphate of phosphatidylinositol-3,4,5-trisphosphate (PtdIns(3,4,5)P3) to produce PtdIns(3,4)P2, thereby negatively regulating the PI3K (phosphoinositide 3-kinase) pathways. Acts as a negative regulator of B-cell antigen receptor signaling. Mediates signaling from the FC-gamma-RIIB receptor (FCGR2B), playing a central role in terminating signal transduction from activating immune/hematopoietic cell receptor systems. Acts as a negative regulator of myeloid cell proliferation/survival and chemotaxis, mast cell degranulation, immune cells homeostasis, integrin alpha-IIb/beta-3 signaling in platelets and JNK signaling in B-cells. Regulates proliferation of osteoclast precursors, macrophage programming, phagocytosis and activation and is required for endotoxin tolerance. Involved in the control of cell-cell junctions, CD32a signaling in neutrophils

	and modulation of EGF-induced phospholipase C activity. Key regulator of neutrophil migration, by governing the formation of the leading edge and polarization required for chemotaxis. Modulates FCGR3/CD16-mediated cytotoxicity in NK cells. Mediates the activin/TGF-beta-induced apoptosis through its Smad-dependent expression. May also hydrolyze PtdIns(1,3,4,5)P4, and could thus affect the levels of the higher inositol polyphosphates like InsP6.
Tissue specificity	Specifically expressed in immune and hematopoietic cells. Expressed in bone marrow and blood cells. Levels vary considerably within this compartment. Present in at least 74% of immature CD34+ cells, whereas within the more mature population of CD33+ cells, it is present in only 10% of cells. Present in the majority of T-cells, while it is present in a minority of B-cells (at protein level).
Sequence similarities	Belongs to the inositol-1,4,5-trisphosphate 5-phosphatase family. Contains 1 SH2 domain.
Domain	The SH2 domain interacts with tyrosine phosphorylated forms of proteins such as SHC1 or PTPN11/SHP-2. It competes with that of GRB2 for binding to phosphorylated SHC1 to inhibit the Ras pathway. It is also required for tyrosine phosphorylation. The NPXY sequence motif found in many tyrosine-phosphorylated proteins is required for the specific binding of the PID domain.
Post-translational modifications	Tyrosine phosphorylated by the members of the SRC family after exposure to a diverse array of extracellular stimuli such as cytokines, growth factors, antibodies, chemokines, integrin ligands and hypertonic and oxidative stress. Phosphorylated upon IgG receptor FCGR2B-binding.
Cellular localization	Cytoplasm. Membrane. Translocates to the plasma membrane when activated, translocation is probably due to different mechanisms depending on the stimulus and cell type. Partly translocated via its SH2 domain which mediates interaction with tyrosine phosphorylated receptors such as the FC-gamma-RIIB receptor (FCGR2B) or CD16/FCGR3. Tyrosine phosphorylation may also participate to membrane localization.

Images



Flow Cytometry (Intracellular) - Anti-SHIP-1 antibody [EP378Y] - BSA and Azide free (ab190551)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab45142**).

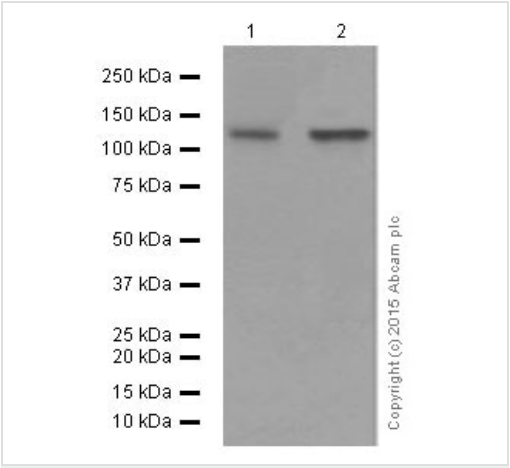
Flow cytometry overlay histogram showing left Raji positive cells and right negative MCF7 stained with **ab45142** (red line). The cells were fixed with 80% methanol (5 min) and then permeabilised with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS containing 10µg/ml human IgG and 10% normal goat serum to block FC receptors and non-specific protein-protein interaction followed by the antibody (**ab45142**) (1x 10^6 in 100µl at 0.008µg/ml (1/275000)) for 30min at 22°C.

The secondary antibody Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min at 22°C

Isotype control antibody (black line) was Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control used at the same concentration and conditions as the primary antibody. Unlabelled

sample (blue line) was also used as a control.

Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter.



Western blot - Anti-SHIP-1 antibody [EP378Y] - BSA and Azide free (ab190551)

All lanes : Anti-SHIP-1 antibody [EP378Y] (**ab45142**) at 1/5000 dilution (purified)

Lane 1 : Daudi cell lysate
Lane 2 : Ramos cell lysate

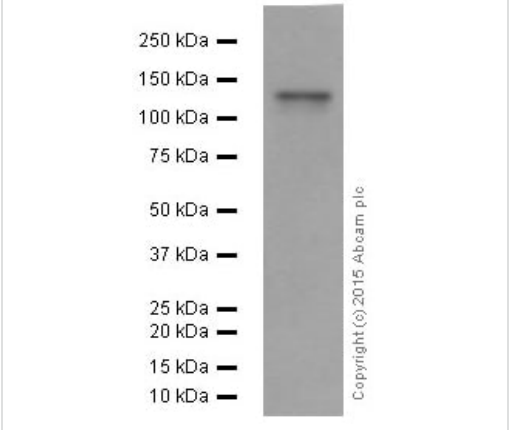
Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Peroxidase-conjugated goat anti-rabbit IgG, (H+L) at 1/1000 dilution

Predicted band size: 133 kDa
Observed band size: 140 kDa

This data was developed using **ab45142**, the same antibody clone in a different buffer formulation.

Blocking and dilution buffer: 5% NFDM/TBST.



Western blot - Anti-SHIP-1 antibody [EP378Y] - BSA and Azide free (ab190551)

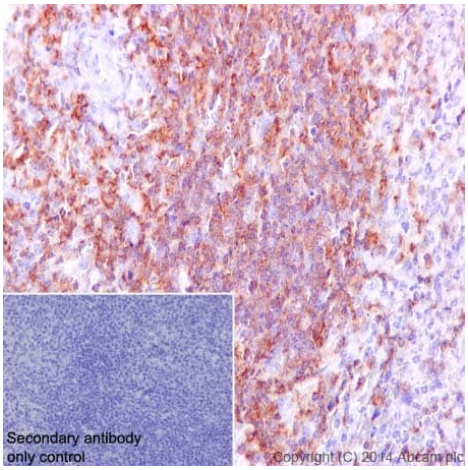
Anti-SHIP-1 antibody [EP378Y] (**ab45142**) at 1/1000 dilution (purified) + KM3 cell lysate at 20 µg

Secondary
Peroxidase-conjugated goat anti-rabbit IgG, (H+L) at 1/1000 dilution

Predicted band size: 133 kDa
Observed band size: 140 kDa

This data was developed using **ab45142**, the same antibody clone in a different buffer formulation.

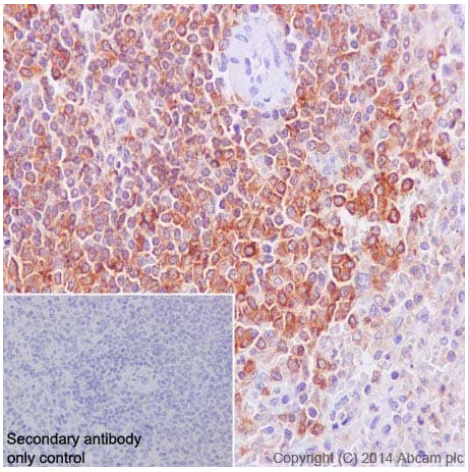
Blocking and dilution buffer: 5% NFDM/TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-SHIP-1 antibody [EP378Y] - BSA and Azide free (ab190551)

This data was developed using **ab45142**, the same antibody clone in a different buffer formulation.

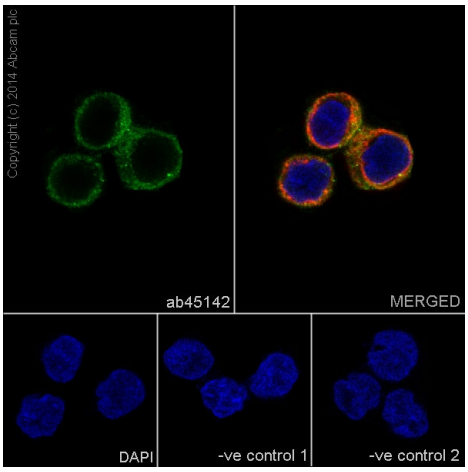
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse spleen tissue labelling SHIP-1 with purified **ab45142** at 1/500. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. **ab97051**, a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-SHIP-1 antibody [EP378Y] - BSA and Azide free (ab190551)

This data was developed using **ab45142**, the same antibody clone in a different buffer formulation.

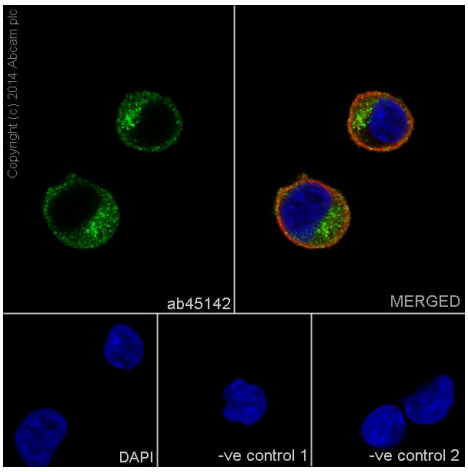
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human spleen tissue labelling SHIP-1 with purified **ab45142** at 1/500. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. **ab97051**, a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.



Immunocytochemistry/ Immunofluorescence - Anti-SHIP-1 antibody [EP378Y] - BSA and Azide free (ab190551)

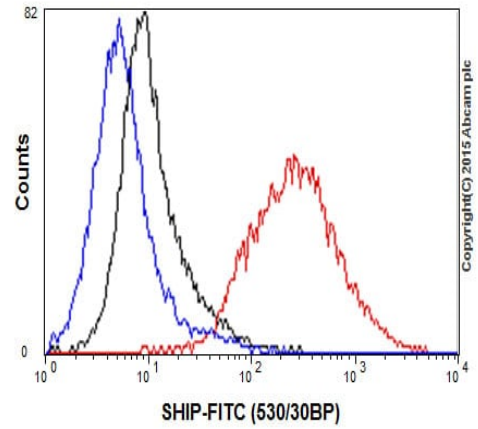
This data was developed using **ab45142**, the same antibody clone in a different buffer formulation.

Immunocytochemistry/Immunofluorescence analysis of Raji cells labelling SHIP-1 with purified **ab45142** at 1/150. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. **ab150077**, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/500) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. **ab7291**, a mouse anti-tubulin (1/1000) and **ab150120**, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/500) were also used. Control 1: primary antibody (1/150) and secondary antibody, **ab150120**, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/500). Control 2: **ab7291** (1/1000) and secondary antibody, **ab150077**, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/500).



Immunocytochemistry/ Immunofluorescence - Anti-SHIP-1 antibody [EP378Y] - BSA and Azide free (ab190551)

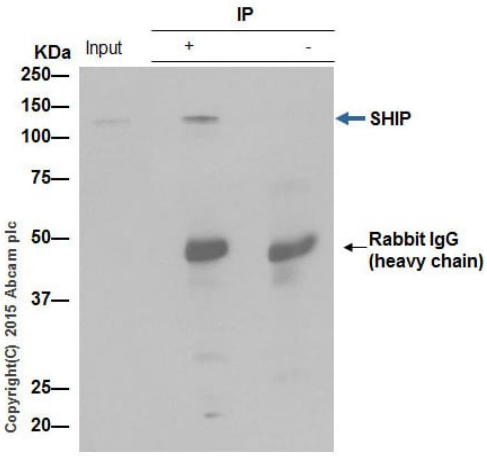
This data was developed using **ab45142**, the same antibody clone in a different buffer formulation.Immunocytochemistry/Immunofluorescence analysis of Jurkat cells labelling SHIP-1 with purified **ab45142** at 1/150. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. **ab150077**, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/500) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. **ab7291**, a mouse anti-tubulin (1/1000) and **ab150120**, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/500) were also used. Control 1: primary antibody (1/150) and secondary antibody, **ab150120**, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/500). Control 2: **ab7291** (1/1000) and secondary antibody, **ab150077**, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/500).



Flow Cytometry (Intracellular) - Anti-SHIP-1 antibody [EP378Y] - BSA and Azide free (ab190551)

This data was developed using **ab45142**, the same antibody clone in a different buffer formulation.

Intracellular Flow Cytometry analysis of Raji cells labelling SHIP-1 with purified **ab45142** at 1/50 (red). Cells were fixed with 2% paraformaldehyde. A FITC-conjugated goat anti-rabbit IgG (1/150) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.



Immunoprecipitation - Anti-SHIP-1 antibody [EP378Y] - BSA and Azide free (ab190551)

This data was developed using **ab45142**, the same antibody clone in a different buffer formulation.

ab45142 (purified) at 1/20 immunoprecipitating SHIP-1 in Daudi whole cell lysate.

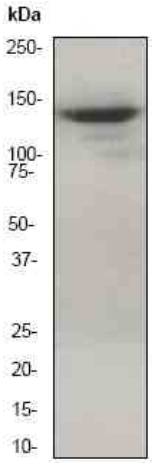
Lane 1 (input): Daudi whole cell lysate (10µg)

Lane 2 (+): **ab45142** + Daudi whole cell lysate (10µg).

Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of **ab45142** in Daudi whole cell lysate.

For western blotting, a HRP-conjugated anti-rabbit IgG, specific to the non-reduced form of IgG was used as the secondary antibody (1/1500).

Blocking and dilution buffer: 5% NFDM/TBST.



Western blot - Anti-SHIP-1 antibody [EP378Y] - BSA and Azide free (ab190551)

Anti-SHIP-1 antibody [EP378Y] (**ab45142**) at 1/50000 dilution (unpurified) + Daudi cell lysate at 10 µg

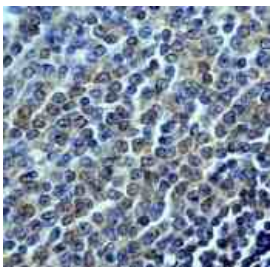
Secondary

HRP-conjugated goat anti-rabbit IgG at 1/2000 dilution

Predicted band size: 133 kDa

Observed band size: 140 kDa

This data was developed using **ab45142**, the same antibody clone in a different buffer formulation.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-SHIP-1 antibody [EP378Y] - BSA and Azide free (ab190551)

This data was developed using **ab45142**, the same antibody clone in a different buffer formulation.Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human spleen tissue labelling SHIP-1 with unpurified **ab45142** at a dilution of 1/100. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

Anti-SHIP-1 antibody [EP378Y] - BSA and Azide free (ab190551)

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