abcam

ab40660

Anti-SRC Family (phospho Y418) antibody [EP503Y]

Recombinant | RabMAb | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 50% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EP503Y
Purification technique	Affinity purification Protein A
Specificity	This antibody recognises Lyn phosphorylation on Tyrosine 396.BLAST analysis show 100% homology with Hck (pY410) and Lck (pY393), and 92% homology with Src (pY418), Fyn (pY419) and Yes (pY425) Therefore, the antibody may cross-react with these proteins.
Concentration	0.139 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

ELISA

Tested

Species	Recombinant fragment - Human

Dilution info	-
Notes	-

Expected

Species	Rat, Human
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Mouse
Dilution info	-
Notes	-

WB

Tested

Species	Rat
Dilution info	1/1000 - 1/10000
Notes	Induction of cells with Pervanadate: Prepare 500mM stock Sodium Orthovanadate in PBS. To make 100mM (100x) Pervanadate Na3VO4 right before experiment, add 400ul 500mM Sodium Orthovanadate to 1560ul H2O and then 40ul H2O2, vortex and incubate at room RT for 5-10min. Cells are serum-starved overnight, and then treated with freshly made Pervanadate (100x, final concentration 1mM) for 20min at 37°C.

Species	Human
Dilution info	1/1000 - 1/10000
Notes	Induction of cells with Pervanadate: Prepare 500mM stock Sodium Orthovanadate in PBS. To make 100mM (100x) Pervanadate Na3VO4 right before experiment, add 400ul 500mM Sodium Orthovanadate to 1560ul H2O and then 40ul H2O2, vortex and incubate at room RT for 5-10min. Cells are serum-starved overnight, and then treated with freshly made Pervanadate (100x, final concentration 1mM) for 20min at 37°C.

Expected

Species	Mouse
Dilution info	1/1000 - 1/10000
Notes	Induction of cells with Pervanadate: Prepare 500mM stock Sodium Orthovanadate in PBS. To make 100mM (100x) Pervanadate Na3VO4 right before experiment, add 400ul 500mM Sodium Orthovanadate to 1560ul H2O and then 40ul H2O2, vortex and incubate at room RT for 5-10min. Cells are serum-starved overnight, and then treated with freshly made Pervanadate (100x, final concentration 1mM) for 20min at 37°C.

Not recommended

Species	Recombinant fragment - Human
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Rat, Human
Dilution info	-
Notes	-

Expected

Species	Mouse
Dilution info	Use at an assay dependent concentration.
Notes	-

Not recommended

Species	Recombinant fragment - Human
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/50
Notes	ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Expected

Species	Mouse
Dilution info	1/50
Notes	ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Species	Rat
Dilution info	1/50
Notes	ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Not recommended

Species	Recombinant fragment - Human
Dilution info	-
Notes	-

Target data

See full target information SRC phospho Y418

Function

Non-receptor protein tyrosine kinase which is activated following engagement of many different classes of cellular receptors including immune response receptors, integrins and other adhesion receptors, receptor protein tyrosine kinases, G protein-coupled receptors as well as cytokine receptors (PubMed:34234773). Participates in signaling pathways that control a diverse spectrum of biological activities including gene transcription, immune response, cell adhesion, cell cycle progression, apoptosis, migration, and transformation. Due to functional redundancy between members of the SRC kinase family, identification of the specific role of each SRC kinase is very difficult. SRC appears to be one of the primary kinases activated following engagement of receptors and plays a role in the activation of other protein tyrosine kinase (PTK) families. Receptor clustering or dimerization leads to recruitment of SRC to the receptor complexes where it phosphorylates the tyrosine residues within the receptor cytoplasmic domains. Plays an important role in the regulation of cytoskeletal organization through

phosphorylation of specific substrates such as AFAP1. Phosphorylation of AFAP1 allows the SRC SH2 domain to bind AFAP1 and to localize to actin filaments. Cytoskeletal reorganization is also controlled through the phosphorylation of cortactin (CTTN) (Probable). When cells adhere via focal adhesions to the extracellular matrix, signals are transmitted by integrins into the cell resulting in tyrosine phosphorylation of a number of focal adhesion proteins, including PTK2/FAK1 and paxillin (PXN) (PubMed:21411625). In addition to phosphorylating focal adhesion proteins, SRC is also active at the sites of cell-cell contact adherens junctions and phosphorylates substrates such as beta-catenin (CTNNB1), delta-catenin (CTNND1), and plakoglobin (JUP). Another type of cell-cell junction, the gap junction, is also a target for SRC, which phosphorylates connexin-43 (GJA1). SRC is implicated in regulation of pre-mRNA-processing and phosphorylates RNA-binding proteins such as KHDRBS1 (Probable). Phosphorylates PKP3 at 'Tyr-195' in response to reactive oxygen species, which may cause the release of PKP3 from desmosome cell junctions into the cytoplasm (PubMed:25501895). Also plays a role in PDGF-mediated tyrosine phosphorylation of both STAT1 and STAT3, leading to increased DNA binding activity of these transcription factors (By similarity). Involved in the RAS pathway through phosphorylation of RASA1 and RASGRF1 (PubMed:11389730). Plays a role in EGF-mediated calcium-activated chloride channel activation (PubMed:18586953). Required for epidermal growth factor receptor (EGFR) internalization through phosphorylation of clathrin heavy chain (CLTC and CLTCL1) at 'Tyr-1477'. Involved in beta-arrestin (ARRB1 and ARRB2) desensitization through phosphorylation and activation of GRK2, leading to beta-arrestin phosphorylation and internalization. Has a critical role in the stimulation of the CDK20/MAPK3 mitogen-activated protein kinase cascade by epidermal growth factor (Probable). Might be involved not only in mediating the transduction of mitogenic signals at the level of the plasma membrane but also in controlling progression through the cell cycle via interaction with regulatory proteins in the nucleus (PubMed:7853507). Plays an important role in osteoclastic bone resorption in conjunction with PTK2B/PYK2. Both the formation of a SRC-PTK2B/PYK2 complex and SRC kinase activity are necessary for this function. Recruited to activated integrins by PTK2B/PYK2, thereby phosphorylating CBL, which in turn induces the activation and recruitment of phosphatidylinositol 3-kinase to the cell membrane in a signaling pathway that is critical for osteoclast function (PubMed:14585963, PubMed:8755529). Upon activation of the G(q)-dependent KISS1/KISS1R signaling pathway, active SRC is recruited, together with the phosphatase DUSP18, to the KISS1R C-terminus (PubMed:38346942). This leads to DUSP18-mediated SRC dephosphorylation and inactivation, down-regulation of osteoclast differentiation and activity, and consequently suppression of bone resorption (By similarity). Promotes energy production in osteoclasts by activating mitochondrial cytochrome C oxidase (PubMed:12615910). Phosphorylates DDR2 on tyrosine residues, thereby promoting its subsequent autophosphorylation (PubMed:16186108). Phosphorylates RUNX3 and COX2 on tyrosine residues, TNK2 on 'Tyr-284' and CBL on 'Tyr-731' (PubMed:20100835, PubMed:21309750). Enhances RIGI-elicited antiviral signaling (PubMed:19419966). Phosphorylates PDPK1 at 'Tyr-9', 'Tyr-373' and 'Tyr-376' (PubMed:14585963). Phosphorylates BCAR1 at 'Tyr-128' (PubMed:22710723). Phosphorylates CBLC at multiple tyrosine residues, phosphorylation at 'Tyr-341' activates CBLC E3 activity (PubMed:20525694). Phosphorylates synaptic vesicle protein synaptophysin (SYP) (By similarity). Involved in anchorage-independent cell growth (PubMed:19307596). Required for podosome formation (By similarity). Mediates IL6 signaling by activating YAP1-NOTCH pathway to induce inflammation-induced epithelial regeneration (PubMed:25731159). Phosphorylates OTUB1, promoting deubiquitination of RPTOR (PubMed:35927303). Phosphorylates caspase CASP8 at

'Tyr-380' which negatively regulates CASP8 processing and activation, down-regulating CASP8 proapoptotic function (PubMed:16619028). Mediates laminin-induced activation of RAC1 signaling through phosphorylation of syntrophin (By similarity). Isoform 1. Non-receptor protein tyrosine kinase which phosphorylates synaptophysin with high affinity. Isoform 2. Non-receptor protein tyrosine kinase which shows higher basal kinase activity than isoform 1, possibly due to weakened intramolecular interactions which enhance autophosphorylation of Tyr-419 and subsequent activation (By similarity). The SH3 domain shows reduced affinity with the linker sequence between the SH2 and kinase domains which may account for the increased basal activity (By similarity). Displays altered substrate specificity compared to isoform 1, showing weak affinity for synaptophysin and for peptide substrates containing class I or class II SH3 domain-binding motifs (By similarity). Plays a role in L1CAM-mediated neurite elongation, possibly by acting downstream of L1CAM to drive cytoskeletal rearrangements involved in neurite outgrowth (By similarity). Isoform 3. Non-receptor protein tyrosine kinase which shows higher basal kinase activity than isoform 1, possibly due to weakened intramolecular interactions which enhance autophosphorylation of Tyr-419 and subsequent activation (By similarity). The SH3 domain shows reduced affinity with the linker sequence between the SH2 and kinase domains which may account for the increased basal activity (By similarity). Displays altered substrate specificity compared to isoform 1, showing weak affinity for synaptophysin and for peptide substrates containing class I or class II SH3 domain-binding motifs (By similarity). Plays a role in neurite elongation (By similarity).

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

This protein is known to be similar in amino acid sequence to HCK (P08631), LCK (P06239), FYN (P06241), YES1 (P07947), and SRC (P12931). Therefore, cross-reactivity with these homologous proteins may be observed. We would be happy to provide immunogen alignment information upon request.

Patented technology
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?
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 batch production

For more information, read more on recombinant antibodies.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

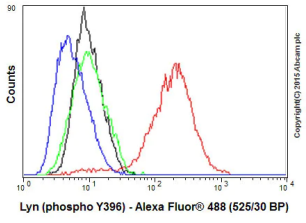
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

7 product images



Flow Cytometry (Intracellular) - Anti-SRC Family (phospho Y418) antibody [EP503Y] (ab40660)

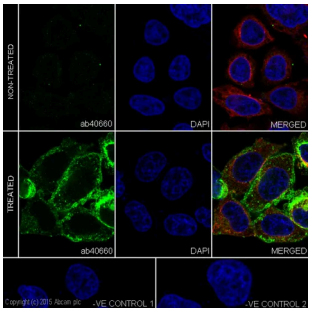
SRC Family (phospho Y418) Flow Cytometry (Intracellular) staining using rabbit Anti-SRC Family (phospho Y418) antibody

Intracellular Flow Cytometry analysis of HeLa cells (treated with 1mM pervanadate for 30 minutes)labelling SRC Family (phospho Y418) with purified ab40660 at 1/80 (red). Cells were fixed with 4% paraformaldehyde. A FITC-conjugated goat anti-rabbit IgG (1/500) was used as the secondary antibody.
Black - Isotype control, rabbit monoclonal IgG with pervanade treated HeLacells.

Blue - Unlabelled control, pervanadate treated HeLa cells without incubation with primary and secondary antibodies.

Red - Pervandate treated HeLa cells with ab40660.

Green - Untreated HeLa cells labelled with ab40660.

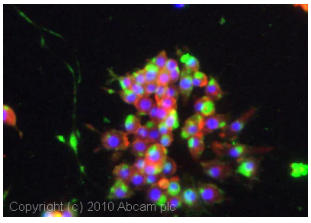


Immunocytochemistry/ Immunofluorescence - Anti-SRC Family (phospho Y418) antibody [EP503Y] (ab40660)

SRC Family (phospho Y418) Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-SRC Family (phospho Y418) antibody

Immunocytochemistry/Immunofluorescence analysis of HeLa cells (untreated and treated with 1mM pervanadate for 15 minutes) labelling SRC Family (phospho Y418) with purified ab40660 at 1/250. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000) 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 (1/1000) were also used. Control 1: primary antibody (1/250) and secondary antibody, ab150120, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/1000).

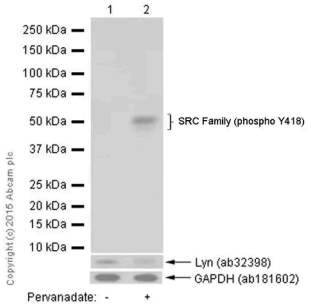
Control 2: ab7291 (1/1000) and secondary antibody, ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000).



Immunocytochemistry/ Immunofluorescence - Anti-SRC Family (phospho Y418) antibody [EP503Y] (ab40660)

SRC Family (phospho Y418) Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-SRC Family (phospho Y418) antibody

ICC/IF image of unpurified ab40660 stained PC12 cells. The cells were 4% formaldehyde fixed (10 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab40660, 5µg/ml) overnight at +4°C. The secondary antibody (green) was Alexa Fluor® 488 goat anti-rabbit IgG (H+L) used at a 1/1000 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.



Western blot - Anti-SRC Family (phospho Y418) antibody [EP503Y] (ab40660)

SRC Family (phospho Y418) Western blot staining using rabbit Anti-SRC Family (phospho Y418) antibody

Blocking and dilution buffer: 5% BSA/TBST.
All lanes:
Western blot - Anti-SRC Family (phospho Y418) antibody [EP503Y] (ab40660) at 1/2000 dilution

Lane 1:
Untreated HeLa whole cell lysate at 10 µg

Lane 2:
Pervanadate treated HeLa whole cell lysate at 10 µg

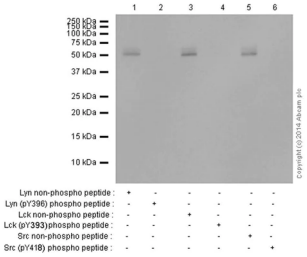
Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 60 kDa

Observed band size: 56-59 kDa

Exposure time: 3s



Western blot - Anti-SRC Family (phospho Y418) antibody [EP503Y] (ab40660)

Blocking and dilution buffer: 5% NFDM/TBST.
All lanes:
Western blot - Anti-SRC Family (phospho Y418) antibody [EP503Y] (ab40660) at 1/1000 dilution

Lane 1:
Pervanadate treated Jurkat cell lysate with Lyn non-phospho peptide

Lane 2:
Pervanadate treated Jurkat cell lysate with Lyn (pY396) phospho peptide

Lane 3:
Pervanadate treated Jurkat cell lysate with Lck non-phospho peptide

Lane 4:
Pervanadate treated Jurkat cell lysate with Lck (pY393) phospho peptide

Lane 5:
Pervanadate treated Jurkat cell lysate with Src non-phospho peptide

Lane 6:
Pervanadate treated Jurkat cell lysate with Src (pY418) phospho peptide

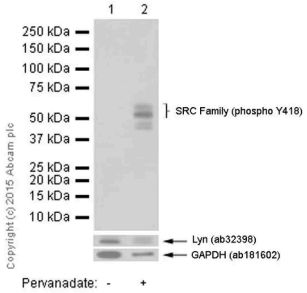
Secondary

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

Predicted band size: 60 kDa

Observed band size: 56-59 kDa

Exposure time: 3min



Western blot - Anti-SRC Family (phospho Y418) antibody [EP503Y] (ab40660)

Blocking and dilution buffer: 5% BSA/TBST.
All lanes:
Western blot - Anti-SRC Family (phospho Y418) antibody [EP503Y] (ab40660) at 1/1000 dilution

Lane 1:
Untreated C6 whole cell lysate at 10 µg

Lane 2:

Pervanadate treated C6 whole cell lysate at 10 µg

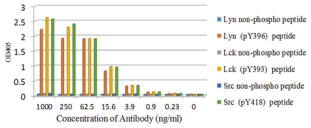
Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 60 kDa

Observed band size: 56-59 kDa

Exposure time: 1s



ELISA - Anti-SRC Family (phospho Y418) antibody [EP503Y] (ab40660)

Direct ELISA antibody dose-response curve using unpurified ab40660 at 0-1000 ng/ml. Antigen concentration of 1000 ng/mL. An alkaline phosphatase-conjugated goat anti-rabbit IgG (H+L) (1/2500) was used as the secondary antibody.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.