

## Product datasheet

# Anti-Syk antibody [SYK-01] ab3993

★★★★☆ 1 Abreviews 16 References 画像数 5

### 製品の概要

製品名	Anti-Syk antibody [SYK-01]
製品の詳細	Mouse monoclonal [SYK-01] to Syk
由来種	Mouse
特異性	The antibody reacts with Protein tyrosine kinase p72Syk (Syk family tyrosine-specific phosphotransferase).
アプリケーション	<b>適用あり:</b> ICC, WB, Flow Cyt
種交差性	<b>交差種:</b> Mouse, Rat, Human
免疫原	Recombinant fragment, corresponding to amino acids 5-360 of Human Syk.
ポジティブ・コントロール	EB1- cells

### 製品の特性

製品の状態	Liquid
保存方法	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.
バッファー	Preservative: 15mM Sodium Azide Constituents: PBS, pH 7.4
精製度	>95% by SDS-PAGE
ポリ/モノ	モノクローナル
クローン名	SYK-01
アイソタイプ	IgG1

### アプリケーション

Our [Abpromise guarantee](#) covers the use of **ab3993** in the following tested applications.

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

アプリケーション	Abreviews	特記事項
ICC		Use at an assay dependent concentration.

アプリケーション	Abreviews	特記事項
WB	★★★★☆	Use a concentration of 1 µg/ml.
Flow Cyt		Use 1 µg for 10 <sup>6</sup> cells.

## ターゲット情報

### 機能

Non-receptor tyrosine kinase which mediates signal transduction downstream of a variety of transmembrane receptors including classical immunoreceptors like the B-cell receptor (BCR). Regulates several biological processes including innate and adaptive immunity, cell adhesion, osteoclast maturation, platelet activation and vascular development. Assembles into signaling complexes with activated receptors at the plasma membrane via interaction between its SH2 domains and the receptor tyrosine-phosphorylated ITAM domains. The association with the receptor can also be indirect and mediated by adapter proteins containing ITAM or partial hemITAM domains. The phosphorylation of the ITAM domains is generally mediated by SRC subfamily kinases upon engagement of the receptor. More rarely signal transduction via SYK could be ITAM-independent. Direct downstream effectors phosphorylated by SYK include VAV1, PLCG1, PI-3-kinase, LCP2 and BLNK. Initially identified as essential in B-cell receptor (BCR) signaling, it is necessary for the maturation of B-cells most probably at the pro-B to pre-B transition. Activated upon BCR engagement, it phosphorylates and activates BLNK an adapter linking the activated BCR to downstream signaling adapters and effectors. It also phosphorylates and activates PLCG1 and the PKC signaling pathway. It also phosphorylates BTK and regulates its activity in B-cell antigen receptor (BCR)-coupled signaling. In addition to its function downstream of BCR plays also a role in T-cell receptor signaling. Plays also a crucial role in the innate immune response to fungal, bacterial and viral pathogens. It is for instance activated by the membrane lectin CLEC7A. Upon stimulation by fungal proteins, CLEC7A together with SYK activates immune cells inducing the production of ROS. Also activates the inflammasome and NF-kappa-B-mediated transcription of chemokines and cytokines in presence of pathogens. Regulates neutrophil degranulation and phagocytosis through activation of the MAPK signaling cascade. Also mediates the activation of dendritic cells by cell necrosis stimuli. Also involved in mast cells activation. Also functions downstream of receptors mediating cell adhesion. Relays for instance, integrin-mediated neutrophils and macrophages activation and P-selectin receptor/SELPG-mediated recruitment of leukocytes to inflammatory loci. Plays also a role in non-immune processes. It is for instance involved in vascular development where it may regulate blood and lymphatic vascular separation. It is also required for osteoclast development and function. Functions in the activation of platelets by collagen, mediating PLCG2 phosphorylation and activation. May be coupled to the collagen receptor by the ITAM domain-containing FCER1G. Also activated by the membrane lectin CLEC1B that is required for activation of platelets by PDPN/podoplanin. Involved in platelet adhesion being activated by ITGB3 engaged by fibrinogen.

### 組織特異性

Widely expressed in hematopoietic cells (at protein level). Within the B-cells compartment it is for instance expressed for pro-B-cells to plasma cells.

### 配列類似性

Belongs to the protein kinase superfamily. Tyr protein kinase family. SYK/ZAP-70 subfamily. Contains 1 protein kinase domain. Contains 2 SH2 domains.

### ドメイン

The SH2 domains mediate the interaction of SYK with the phosphorylated ITAM domains of transmembrane proteins. Some proteins like CLEC1B have a partial ITAM domain (also called hemITAM) containing a single YxxL motif. The interaction with SYK requires CLEC1B

homodimerization.

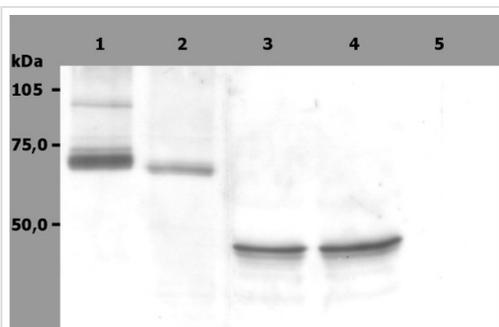
## 翻訳後修飾

Ubiquitinated by CBLB after BCR activation; which promotes proteasomal degradation. Autophosphorylated. Phosphorylated on tyrosine residues by LYN following receptors engagement. Phosphorylation on Tyr-323 creates a binding site for CBL, an adapter protein that serves as a negative regulator of BCR-stimulated calcium ion signaling. Phosphorylation at Tyr-348 creates a binding site for VAV1. Phosphorylation on Tyr-348 and Tyr-352 enhances the phosphorylation and activation of phospholipase C-gamma and the early phase of calcium ion mobilization via a phosphoinositide 3-kinase-independent pathway (By similarity). Phosphorylation on Ser-297 is very common, it peaks 5 minutes after BCR stimulation, and creates a binding site for YWHAG. Phosphorylation at Tyr-630 creates a binding site for BLNK. Dephosphorylated by PTPN6.

## 細胞内局在

Cell membrane. Cytoplasm, cytosol.

## 画像



Western blot - Anti-Syk antibody [SYK-01] (ab3993)

**Lanes 1-2** : Anti-Syk antibody [SYK-01]

(ab3993) at 2 µg/ml

**Lanes 3-4** : anti-Human Cytokeratin 18

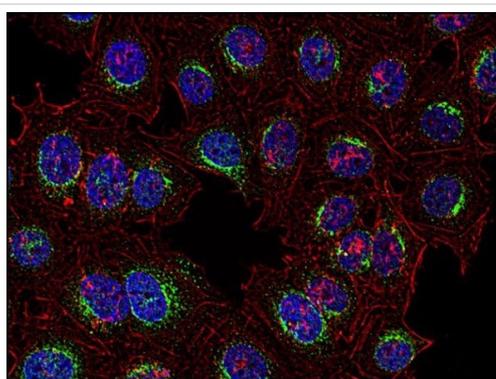
**Lane 1** : Ramos (human Burkitt's lymphoma cell line) cell lysate

**Lane 2** : RBL (rat basophilic leukemia cell line) cell lysate

**Lanes 3-4** : HeLa (human epithelial cell line from cervix adenocarcinoma) cell lysate

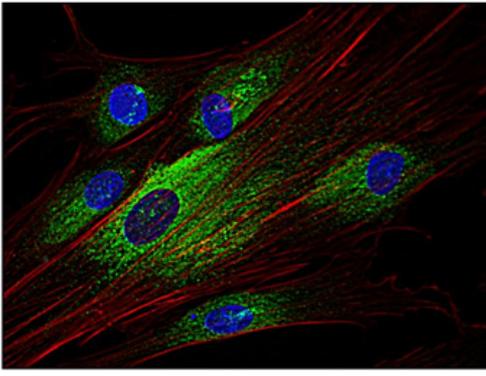
**Lane 5** : negative control

Performed under non-reducing conditions.



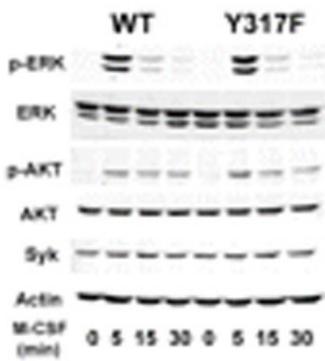
Immunocytochemistry/ Immunofluorescence - Anti-Syk antibody [SYK-01] (ab3993)

Immunofluorescence staining of Syk in human HeLa (human epithelial cell line from cervix adenocarcinoma) cell line using ab3993 (green) at 5 µg/ml. Actin cytoskeleton was decorated by phalloidin (red) and cell nuclei stained with DAPI (blue).



Immunocytochemistry - Anti-Syk antibody [SYK-01] (ab3993)

Immunofluorescence staining of Syk in human primary fibroblasts using ab3993 (green) at 5  $\mu\text{g/ml}$ . Actin cytoskeleton was decorated by phalloidin (red) and cell nuclei stained with DAPI (blue).

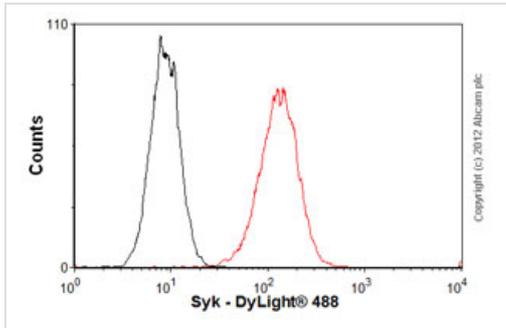


Western blot - Anti-Syk antibody [SYK-01] (ab3993)

Image from Zou W et al, J Biol Chem. 2009 Jul 10;284(28):18833-9. Epub 2009 May 6, Fig 1. DOI 10.1074/jbc.M109.012385

Transduced Syk<sup>-/-</sup> BMMs, cultured in 50 ng/ml M-CSF for 3 days, were serum- and cytokine-starved overnight. The cells were then exposed to either 100 ng/ml M-CSF with time. Signaling molecules were identified by immunoblotting. Actin serves as a loading control.

Cultured cells were washed twice with ice-cold PBS and lysed in RIPA buffer containing 20 mm Tris, pH 7.5, 150 mm NaCl, 1 mm EDTA, 1 mm EGTA, 1% Triton X-100, 2.5 mm sodium pyrophosphate, 1 mm  $\beta$ -glycerophosphate, 1 mm Na<sub>3</sub>VO<sub>4</sub>, 1 mm NaF, and 1 $\times$  protease inhibitor mixture. After incubation on ice for 10 minutes, cell lysates were clarified by centrifugation at 15,000 rpm for 10 minutes. 40 $\mu\text{g}$  of total lysates were subjected to 8% SDS-PAGE and transferred onto polyvinylidene difluoride membranes. Filters were blocked in 0.1% casein in PBS for 1 hour and incubated with primary antibodies at 4  $^{\circ}\text{C}$  overnight followed by probing with fluorescence-labeled secondary antibodies. Proteins were detected with the Odyssey infr



Flow Cytometry-Anti-Syk antibody [SYK-01]  
(ab3993)

Overlay histogram showing Ramos cells stained with ab3993 (red line). The cells were fixed with 4% paraformaldehyde (10 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab3993, 1µg/1x10<sup>6</sup> cells) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-mouse IgG (H+L) (ab96879) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse IgG1 [ICIGG1] (ab91353, 2µg/1x10<sup>6</sup> cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in Ramos cells fixed with 80% methanol (5 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.

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