Technical Data Sheet

Purified Mouse anti-Syk (pY348)

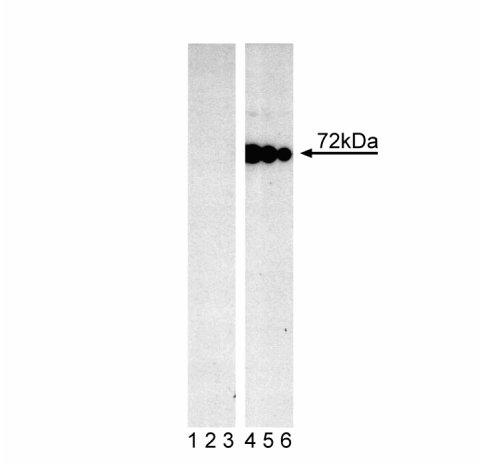
Product Information

Material Number:	**558167**
Size:	50 µg
Concentration:	0.5 mg/ml
Clone:	I120-722
Immunogen:	Phosphorylated Human Syk Peptide
Isotype:	Mouse (BALB/c) IgG1, κ
Reactivity:	QC Testing: Human Predicted due to immunogen sequence identity: Mouse, Rat
Target MW:	72 kDa
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

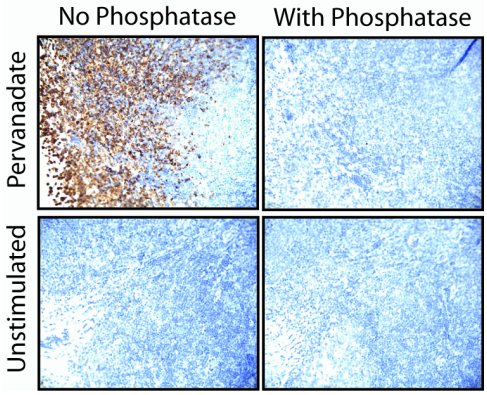
Description

Syk is a non-receptor protein-tyrosine kinase that is closely related to ZAP70 and plays crucial roles in the development and receptor-mediated signaling of most leukocytes and in vascular integrity. Syk is expressed in hematopoietic cells, including B lymphocytes, immature (CD4, CD8 double-negative and double-positive) thymocytes, and myeloid cells, epithelial cell lines, and normal breast tissue. Mature (CD4 or CD8 single-positive) thymocytes and peripheral αβ TCR-bearing T lymphocytes have very low or undetectable levels of Syk. Syk contributes to the signal transduction process by binding to ITAMs (*I*mmunoreceptor *T*yrosine-based *A*ctivation *M*otifs) of immune receptors, including Igα and Igβ (CD79a and b), TCRζ, CD3ε, and FcRγ. Upon receptor activation, Syk binds to phosphorylated ITAMs via its two N-terminal SH2 domains thereby activating Syk and causing tyrosines in the interdomain, between the SH2 and Kinase domains of Syk, to undergo auto-phosphorylation and phosphorylation by Lyn. The tyrosine 348 phosphorylation site (pY348) in human Syk is orthologous to tyrosine 342 in mouse and rat Syk and tyrosine 315 in human ZAP70. This phosphorylated site can act as a binding site for other signaling molecules, such as PLCγ, Vav, and Fgr.

The I120-772 antibody is specific for human Syk (pY348) and does not cross-react with phosphorylated Zap70. The orthologous phosphorylation site in mouse and rat Syk is Y342.



Lysate from control (left panel) and pervanadate-treated (right panel) Ramos cells (Burkitt's lymphoma) were probed with mAb I120-722 at concentrations of 0.25 (lanes 1), 0.125 (lanes 2), and 0.0625 (lanes 3) µg/ml. Syk (pY348) is identified as a strong band of 72 kDa in the pervanadate-treated Ramos cells.



Syk (pY348) staining on tonsil. *Fresh human tonsil, stimulated with a 5 mM Pervanadate solution for 2 hours (top row) or unstimulated (bottom row), was fixed in formalin and processed. Following antigen retrieval with BD Retrievagen A buffer (Cat. no. 550524), the sections were either left untreated (left column) or treated with a phosphatase to eliminate all phosphorylation (right column). The tissue sections were stained with Purified Mouse anti-Syk (pY348) monoclonal antibody with Hematoxylin counterstaining. Original magnification: 20X.*

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Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.
Store undiluted at 4°C.

The purified or conjugated mAb was characterized by flow cytometry (Flow) and western blot (WB) using these model systems:

Method	*Species*	*Cells*	*Treatment*	*Fixation*	*Perm buffer*	*Result*
Flow	Human	PBMC	surface IgM crosslinking	Lyse/Fix or Fix I	Perm I, II, or III	Up-regulated expression on B lymphocytes
		Ramos	surface IgM crosslinking	Lyse/Fix	Perm II	Up-regulated expression
	Mouse	Splenocytes	surface IgM crosslinking	Lyse/Fix	Perm II	Up-regulated expression on B lymphocytes
WB	Human	Ramos	Pervanadate			72-kDa band induced
			Pervanadate + phospho peptide			blocking of 72-kDa band
			Pervanadate + unrelated phospho peptide			no blocking

Application Notes

Application

Western blot	Routinely Tested
Immunohistochemistry-formalin (antigen retrieval required)	Tested During Development
Intracellular staining (flow cytometry)	Tested During Development

Suggested Companion Products

Catalog Number	Name	Size	Clone
554002	HRP Goat Anti-Mouse Ig	1.0 ml	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.

References

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