Technical Data Sheet

Alexa Fluor® 647 Mouse anti-PLC-γ2 (pY759)

Product Information

Material Number:	558498	
Size:	50 Tests	
Vol. per Test:	20 µl	
Clone:	K86-689.37	
Immunogen:	Phosphorylated Human PLC-y2 Peptide	
Isotype:	Mouse IgG1, κ	
Reactivity:	QC Testing: Human	
-	Predicted Reactivity: Mouse, Rat	
Storage Buffer:	Aqueous buffered solution containing BSA and $\leq 0.09\%$ sodium azide.	

Description

The Phospholipase C (PLC) isozymes hydrolyze phosphatidyl inositol biphosphate to inositol triphosphate and diacylglycerol. The former causes release of calcium from the endoplasmic reticulum, while the latter is an activator of Protein Kinase C. Within the PLC family, PLC-Y is the only member that contains SH2 and SH3 domains. These domains enable it to interact with protein tyrosine kinases and become enzymatically activated via phosphorylation. It exists as two isoforms: PLC-y1, which is ubiquitously expressed, and PLC-y2, which is found primarily in the hematopoietic system. PLC-y2-null mice exhibit impaired B lymphocyte maturation and defects in Fc receptor functions. Phosphorylation of PLC-y2 at tyrosines 753 and 759 (Y759) is required for activation of PLC-y2 enzyme activity. PLC-y2 phosphorylation at Y759 can be induced by stimulation of the B cell receptor in Ramos cells, the collagen receptor in platelets, and the T cell receptor in Jurkat cells, and it occurs downstream of Btk and BLNK in the signaling cascade of activated B cells.

The K86-689.37.73 antibody recognizes PLC-y2 phosphorylated at Y759 in the SH2-SH3 linker region.



Analysis of PLC-y2 (pY759) in human Burkitt's lymphoma. Serum-starved Ramos cells were either stimulated (open histogram) by cross-linking of surface IgM with purified Goat anti-Human IgM (SouthernBiotech) at 37°C for 3 minutes or unstimulated (solid histogram). The cells were fixed with pre-warmed BD Phosflow™ Fix Buffer I (Cat. No. 557870) for 10 minutes at 37°C, then permeabilized with BD Phosflow™ Perm Buffer III (Cat. No. 558050) on ice for at least 30 minutes, and then stained with Alexa Fluor® 647 Mouse anti-PLC-γ2 (pY759) (Cat. No. 558498). Flow cytometry was performed on a BD FACSCalibur™ flow cytometry system.

Preparation and Storage

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

The antibody was conjugated to Alexa Fluor® 647 under optimum conditions, and unreacted Alexa Fluor® 647 was removed.

Application Notes

Application						
Intracellular staining (flow cytometry)		Routinely Tested				
Suggested Com	panion Products					
Catalog Number	Name			Size	Clone	_
558050	Perm Buffer III			125 mL	(none)	-
557870	Fix Buffer I			250 mL	(none)	
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United States Canac 877.232.8995 866.93	la Europe Japan 79.9408 32.2.400.98.95 0120.8555.	Asia Pacific 0 65.6861.0633	Latin America/Caribbean 55.11.5185.9995			5 L
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Product Notices

- 1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1×10^{6} cells in a 100-µl experimental sample (a test).
- The Alexa Fluor®, Pacific Blue[™], and Cascade Blue® dye antibody conjugates in this product are sold under license from Molecular Probes, Inc. for research use only, excluding use in combination with microarrays, or as analyte specific reagents. The Alexa Fluor® dyes (except for Alexa Fluor® 430), Pacific Blue[™] dye, and Cascade Blue® dye are covered by pending and issued patents.
- 3. Alexa Fluor® 647 fluorochrome emission is collected at the same instrument settings as for allophycocyanin (APC).
- 4. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
- 5. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
- 6. 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.
- 7. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
- 8. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

Holodick NE, Tumang JR, Rothstein TL.. Continual signaling is responsible for constitutive ERK phosphorylation in B-1a cells.. *Mol Immunol.* 2009; 46(15):3029-3036. (Clone-specific: Flow cytometry)

Irish JM, Myklebust JH, Alizadeh AA, Houot R, Sharman JP, Czerwinski DK, Nolan GP, Levy R.. B-cell signaling networks reveal a negative prognostic human lymphoma cell subset that emerges during tumor progression.. *Proc Natl Acad Sci U S A*. 2010; 107(29):12747-12754. (Clone-specific: Flow cytometry) Kim YJ, Sekiya F, Poulin B, Bae YS, Rhee SG. Mechanism of B-cell receptor-induced phosphorylation and activation of phospholipase C-γ2. *Mol Cell Biol*. 2004; 24(22):9986-9999. (Biology)

Ozdener F, Dangelmaier C, Ashby B, Kunapuli SP, Daniel JL. Activation of phospholipase Cy2 by tyrosine phosphorylation.. *Mol Pharmacol.* 2002; 62(3):672-679. (Biology)

Wang D, Feng J, Wen R, et al. Phospholipase Cy2 is essential in the functions of B cell and several Fc receptors. *Immunity*. 2000; 13:25-35. (Biology) Wen R, Jou S-T, Chen Y, Hoffmeyer A, Wang D. Phospholipase Cy2 is essential for specific functions of FccR and FcyR. *J Immunol*. 2002; 169:6743-6752. (Biology)

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