Technical Data Sheet

Alexa Fluor® 488 Mouse anti-PLC-γ2 (pY759)

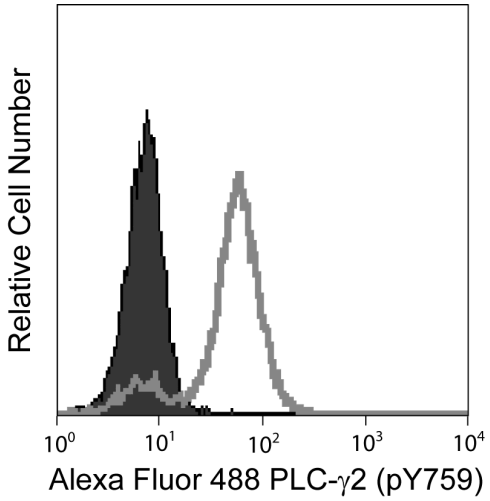
Product Information

Material Number:	**558507**
Size:	50 Tests
Vol. per Test:	20 µl
Clone:	K86-689.37
Immunogen:	Phosphorylated Human PLC-γ2 Peptide
Isotype:	Mouse IgG1, κ
Reactivity:	QC Testing: Human Predicted Reactivity: Mouse, Rat
Storage Buffer:	Aqueous buffered solution containing BSA and ≤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-γ 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-γ1, which is ubiquitously expressed, and PLC-γ2, which is found primarily in the hematopoietic system. PLC-γ2-null mice exhibit impaired B lymphocyte maturation and defects in Fc receptor functions. Phosphorylation of PLC-γ2 at tyrosines 753 and 759 (Y759) is required for activation of PLC-γ2 enzyme activity. PLC-γ2 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-γ2 phosphorylated at Y759 in the SH2-SH3 linker region.



Analysis of PLC-γ2 (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 37ºC, then permeabilized with BD Phosflow™ Perm Buffer III (Cat. No. 558050) on ice for at least 30 minutes, and then stained with Alexa Fluor® 488 Mouse anti-PLC-γ2 (pY759). 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® 488 under optimum conditions, and unreacted Alexa Fluor® 488 was removed.

Application Notes

Application

Intracellular staining (flow cytometry)	Routinely Tested

Suggested Companion Products

Catalog Number	Name	Size	Clone
557870	Fix Buffer I	250 mL	(none)
558050	Perm Buffer III	125 mL	(none)
554656	Stain Buffer (FBS)	500 mL	(none)

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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).
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. 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.
4. Alexa Fluor® 488 fluorochrome emission is collected at the same instrument settings as for fluorescein isothiocyanate (FITC).
5. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
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. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
8. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
9. Species testing during development may have been performed with a different format of the same clone. Selected applications have been tested for cross-reactivity.

References

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 Cγ2 by tyrosine phosphorylation.. *Mol Pharmacol.* 2002; 62(3):672-679. (Biology)

Wang D, Feng J, Wen R, et al. Phospholipase Cγ2 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 Cγ2 is essential for specific functions of FcεR and FcγR. *J Immunol.* 2002; 169:6743-6752. (Biology)

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