Technical Data Sheet

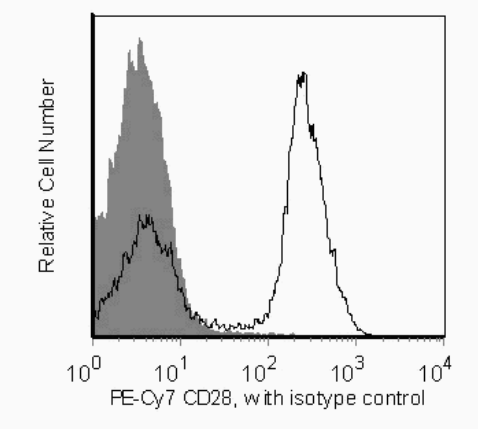
PE-Cy™7 Mouse Anti-Human CD28

Product Information

Material Number:	**560684**
Alternate Name:	CD28 antigen; T44; Tp44; TP44
Size:	50 Tests
Vol. per Test:	5 µl
Clone:	CD28.2
Immunogen:	Human CD28 Transfected Cell Line
Isotype:	Mouse (C3H x BALB/c) IgG1, κ
Reactivity:	QC Testing: Human
Workshop:	V 5T CD28.05
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The CD28.2 monoclonal antibody specifically binds to CD28, a 44 kDa homodimeric transmembrane glycoprotein present on most mature T cells, thymocytes and plasma cells. CD28 is a costimulatory receptor that binds CD80 and CD86 as ligands and plays a very important role in T cell-B cell interactions. It has been suggested that CD28 initiates and regulates a separate and distinct signal transduction pathway from those stimulated by the TCR complex. Additionally, it has been reported that CD28 antibody clones vary in their ability to stimulate T cells to produce IL-2 and increase intracellular Ca2+ concentration. This finding suggests the existence of functionally distinct subregions on the CD28 molecule. CD28.2 has been demonstrated to bind to the same molecule as clone L293, another CD28 mAb, and has been reported to induce Ca2+ influx in Jurkat T cells.



Flow cytometric analysis of CD28 on human lysed whole blood. *Human lysed whole blood was stained with the PE-Cy™7 Mouse Anti-Human CD28 antibody (unshaded) or with a PE-Cy™7 Mouse IgG1, κ isotype control (shaded). Histograms were derived from gated events based on light scattering characteristics for lymphocytes. Flow cytometry was performed on a BD™ LSR II 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 with PE-Cy7 under optimum conditions, and unconjugated antibody and free PE-Cy7 were removed.

Application Notes

Application

Flow cytometry	Routinely Tested

Suggested Companion Products

Catalog Number	Name	Size	Clone
557872	PE-Cy™7 Mouse IgG1 κ Isotype Control	100 Tests	MOPC-21
555899	Lysing Buffer	100 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. Warning: Some APC-Cy7 and PE-Cy7 conjugates show changes in their emission spectrum with prolonged exposure to formaldehyde. If you are unable to analyze fixed samples within four hours, we recommend that you use BD™ Stabilizing Fixative (Cat. No. 338036).
4. PE-Cy7 is a tandem fluorochrome composed of R-phycoerythrin (PE), which is excited by 488-nm light and serves as an energy donor, coupled to the cyanine dye Cy7, which acts as an energy acceptor and fluoresces maximally at 780 nm. PE-Cy7 tandem fluorochrome emission is collected in a detector for fluorescence wavelengths of 750 nm and higher. Although every effort is made to minimize the lot-to-lot variation in the efficiency of the fluorochrome energy transfer, differences in the residual emission from PE may be observed. Therefore, we recommend that individual compensation controls be performed for every PE-Cy7 conjugate. PE-Cy7 is optimized for use with a single argon ion laser emitting 488-nm light, and there is no significant overlap between PE-Cy7 and FITC emission spectra. When using dual-laser cytometers, which may directly excite both PE and Cy7, we recommend the use of cross-beam compensation during data acquisition or software compensation during data analysis.
5. Please observe the following precautions: Absorption of visible light can significantly alter the energy transfer occurring in any tandem fluorochrome conjugate; therefore, we recommend that special precautions be taken (such as wrapping vials, tubes, or racks in aluminum foil) to prevent exposure of conjugated reagents, including cells stained with those reagents, to room illumination.
6. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
7. Cy is a trademark of Amersham Biosciences Limited.
8. 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.
9. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
10. This product is sold under license. Purchase of this product does not include rights to (i) incorporate this product into the purchaser's own products for resale to end-users, or (ii) use this product to conduct for-profit research for or on behalf of another party. For information on obtaining a license to this product for such prohibited uses, contact INSERM, 7 rue Watt, 75013 Paris. Telephone: +33 1 55 03 01 60. Facsimile: +33 1 55 03 01 18. Email: techtransfert@inserm-transfert.fr
11. An isotype control should be used at the same concentration as the antibody of interest.

References

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