# **Technical Data Sheet**

# **Biotin Hamster Anti-Mouse CD28**

### **Product Information**

**Material Number:** 553296 0.5 mg Size: 0.5 mg/mlConcentration: 37.51 Clone:

Mouse EL-4 (T-cell lymphoma) Cells Immunogen:

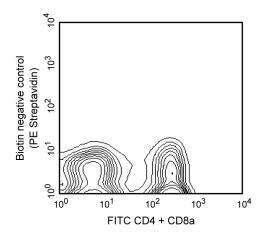
Syrian Hamster IgG2, λ1 Isotype: QC Testing: Mouse Reactivity:

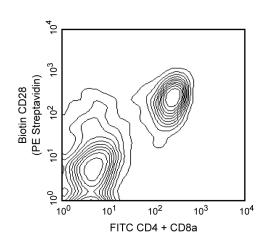
Storage Buffer: Aqueous buffered solution containing protein stabilizer and ≤0.09% sodium

### Description

The 37.51 antibody reacts with CD28, which is expressed on most thymocytes, at low density on nearly all CD4+ and CD8+ peripheral T cells, and at even lower density on NK cells. The expression of CD28, in splenocytes and thymocytes, has been reported to increase after activation. CD28 transcripts are found in mast cells, and cell-surface expression of CD28 is induced upon maturation or activation of mast cells. It has been reported that CD28 is not expressed on some populations of intraepithelial T lymphocytes. CD28 is a costimulatory receptor; its ligands include CD80 (B7-1) and CD86 (B7-2). The 37.51 mAb augments proliferation and cytokine production by activated T and NK cells and can provide a costimulatory signal for CTL induction. There is considerable evidence that CD28 is a costimulatory receptor involved in many, but not all, T cell-dependent immune responses.

This antibody is routinely tested by flow cytometric analysis. Other applications were tested at BD Biosciences Pharmingen during antibody development only or reported in the literature.





Two-color analysis of CD28 expression on splenic T lymphocytes. After pre-incubation with purified anti-mouse CD16/CD32 mAb 2.4G2 [Mouse BD Fc Block™] (Cat. No. 553141), BALB/c splenocytes were simultaneously stained with FITC-conjugated anti-mouse CD4 mAb RM4-5 (Cat. No. 553046) and FITC-conjugated anti-mouse CD8a mAb 53-6.7 (Cat. No. 553030), in addition to staining with biotinylated anti-mouse CD28 clone 37.51 (right panel), followed by streptavidin-PE (Cat No. 554061). Flow cytometry was performed on a BD FACScan™ flow cytometry system.

# **Preparation and Storage**

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography. The antibody was conjugated with biotin under optimum conditions, and unreacted biotin was removed.

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

# **Application Notes**

Application

Flow cytometry Routinely Tested

## **Recommended Assay Procedure:**

# **BD Biosciences**

bdbiosciences.com

**United States** Asia Pacific Latin America/Caribbean Europe 877.232.8995 888.259.0187 32.53.720.550 0120.8555.90 65.6861.0633 55.11.5185.9995

For country-specific contact information, visit bdbiosciences.com/how\_to\_order/

Conditions: The information disclosed herein is not to be construed as a recommendation to use the above product in violation cof any patents. BD Biosciences will not be held responsible for patent infringement or other violations that may occur with the use of our products. Purchase does not include or carry any right to resell or transfer this product either as a stand-alone product or as a component of another product. Any use of this product other than the permitted use without the express written authorization of Becton Dickinson and Company is strictly prohibited.

For Research Use Only. Not for use in diagnostic or therapeutic procedures. Not for resale.

BD, BD Logo and all other trademarks are the property of Becton, Dickinson and Company. ©2008 BD



Flow Cytometry: For flow cytometry of cell suspensions from peripheral lymphoid tissues, it is recommended that multicolor staining be performed to identify T lymphocytes and/or NK cells and that purified anti-mouse CD16/CD32 mAb 2.4G2 [Mouse BD Fc Block<sup>TM</sup>] (Cat. No. 553141) be used. Since this antigen is expressed at low density on resting peripheral T lymphocytes, it is recommended that a "bright" second-step reagent, such as Streptavidin-PE (Cat. No. 554061), be used for flow cytometric staining of these cell populations.

### **Suggested Companion Products**

Catalog Number	Name	Size	Clone
553141	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block <sup>TM</sup> )	0.1 mg	2.4G2
553046	FITC Rat Anti-Mouse CD4	0.1 mg	RM4-5
553030	FITC Rat Anti-Mouse CD8a	0.1 mg	53-6.7
554061	PE Streptavidin	0.5 mg	(none)
553963	Biotin Hamster IgG2, λ1 Isotype Control	0.25 mg	Ha4/8

### **Product Notices**

- 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. Although hamster immunoglobulin isotypes have not been well defined, BD Biosciences Pharmingen has grouped Armenian and Syrian hamster IgG monoclonal antibodies according to their reactivity with a panel of mouse anti-hamster IgG mAbs. A table of the hamster IgG groups, Reactivity of Mouse Anti-Hamster Ig mAbs, may be viewed at <a href="http://www.bdbiosciences.com/pharmingen/hamster">http://www.bdbiosciences.com/pharmingen/hamster</a> chart 11x17.pdf.
- 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

Cibotti R, Punt JA, Dash KS, Sharrow SO, Singer A. Surface molecules that drive T cell development in vitro in the absence of thymic epithelium and in the absence of lineage-specific signals. *Immunity*. 1997; 6(3):245-255. (Biology)

Gelfanov V, Lai YG, Gelfanova V, Dong JY, Su JP, Liao NS. Differential requirement of CD28 costimulation for activation of murine CD8+ intestinal intraepithelial lymphocyte subsets and lymph node cells. *J Immunol.* 1995; 155(1):76-82. (Biology)

Gross JA, Callas E, Allison JP. Identification and distribution of the costimulatory receptor CD28 in the mouse. *J Immunol.* 1992; 149(2):380-388. (Immunogen: (Co)-stimulation, Immunoprecipitation)

Harding FA, Allison JP. CD28-B7 interactions allow the induction of CD8+ cytotoxic T lymphocytes in the absence of exogenous help. *J Exp Med.* 1993; 177(6):1791-1796. (Biology)

Harding FA, McArthur JG, Gross JA, Raulet DH, Allison JP. CD28-mediated signalling co-stimulates murine T cells and prevents induction of anergy in T-cell clones. *Nature*. 1992; 356(6370):607-609. (Biology)

June CH, Bluestone JA, Nadler LM, Thompson CB. The B7 and CD28 receptor families. Immunol Today. 1994; 15(7):321-331. (Biology)

Krummel MF, Allison JP. CD28 and CTLA-4 have opposing effects on the response of T cells to stimulation. *J Exp Med.* 1995; 182(2):459-465. (Biology) Lepesant H, Pierres M, Naquet P. Deficient antigen presentation by thymic epithelial cells reveals differential induction of T cell clone effector functions by CD28-mediated costimulation. *Cell Immunol.* 1995; 161(2):279-287. (Biology)

Marietta EV, Weis JJ, Weis JH. CD28 expression by mouse mast cells is modulated by lipopolysaccharide and outer surface protein A lipoprotein from Borrelia burgdorferi. *J Immunol.* 1997; 159(6):2840-2848. (Biology)

Nandi D, Gross JA, Allison JP. CD28-mediated costimulation is necessary for optimal proliferation of murine NK cells. *J Immunol.* 1994; 152(7):3361-3369. (Biology)

Nishio M, Spielman J, Lee RK, Nelson DL, Podack ER. CD80 (B7.1) and CD54 (intracellular adhesion molecule-1) induce target cell susceptibility to promiscuous cytotoxic T cell Ivsis. *J Immunol.* 1996; 157(10):4347-4353. (Biology)

Ong CJ, Lim AS, Teh HS. CD28-induced cytokine production and proliferation by thymocytes are differentially regulated by the p59fyn tyrosine kinase. *J Immunol.* 1997; 159(5):2169-2176. (Biology)

Rakasz E, Hagen M, Sandor M, Lynch RG. Gamma delta T cells of the murine vagina: T cell response in vivo in the absence of the expression of CD2 and CD28 molecules. *Int Immunol.* 1997; 9(1):161-167. (Biology)

Shahinian A, Pfeffer K, Lee KP, et al. Differential T cell costimulatory requirements in CD28-deficient mice. *Science*. 1993; 261(5121):609-612. (Biology) Wells AD, Gudmundsdottir H, Turka LA. Following the fate of individual T cells throughout activation and clonal expansion. Signals from T cell receptor and CD28 differentially regulate the induction and duration of a proliferative response. *J Clin Invest*. 1997; 100(12):3173-3183. (Biology)

553296 Rev. 15 Page 2 of 2