### Technical Data Sheet

# **Purified NA/LE Hamster Anti-Mouse CD28**

#### **Product Information**

Material Number: 553294

Alternate Name: Cd28; CD28 antigen; T-cell-specific surface glycoprotein CD28

 Size:
 0.5 mg

 Concentration:
 1.0 mg/ml

 Clone:
 37.51

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

Isotype:Syrian Hamster IgG2, λ1Reactivity:QC Testing: Mouse

Storage Buffer: No azide/low endotoxin: Aqueous buffered solution containing protein

stabilizer, no preservative, 0.2 $\mu$ m sterile filtered. Endotoxin level is  $\leq$ 0.01 EU/ $\mu$ g ( $\leq$ 0.001 ng/ $\mu$ g) of protein as determined by the LAL assay.

#### 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.

#### **Preparation and Storage**

Store undiluted at 4°C.

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

This preparation contains no preservatives, thus it should be handled under aseptic conditions.

## **Application Notes**

#### Application

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Flow cytometry	Routinely Tested
Immunohistochemistry-frozen	Tested During Development
(Co)-stimulation	Reported
Blocking	Reported
Immunoprecipitation	Reported
Cytotoxicity	Reported

#### **Recommended Assay Procedure:**

Flow Cytometry: For multicolor staining of cell suspensions from peripheral lymphoid tissues (e.g detection of T lymphocytes and/or NK cells), investigators are encouraged to utilize Purified Rat Anti-Mouse CD16/CD32 [Mouse BD Fc Block™] (Cat. No. 553141) to minimize non-specific binding. In addition, since CD28 may be expressed at low density on resting peripheral T lymphocytes, investigators may wish to amplify the staining signal by using a biotinylated second-step antibody followed by a "bright" third-step reagent, such as PE Streptavidin (Cat. No. 554061). If Mouse BD Fc Block™ is used, investigators need to be aware of and account for any potential crossreactivity that may occur between the second-step or third-step reagents chosen with the Mouse BD Fc Block™. Biotin Mouse Anti-Syrian Hamster IgG2 (Cat. No. 554029) would be a suggested second-step antibody for investigators to consider utilizing.

#### **Suggested Companion Products**

Catalog Number	Name	Size	<u>Clone</u>
553141	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block <sup>TM</sup> )	0.1 mg	2.4G2
554061	PE Streptavidin	0.5 mg	(none)
554029	Biotin Mouse Anti-Syrian Hamster IgG2	0.5 mg	G192-3
553961	Purified NA/LE Hamster IgG2, λ1 Isotype Control	0.5 mg	Ha4/8

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#### **Product Notices**

- 1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- 2. 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 http://www.bdbiosciences.com/documents/hamster\_chart\_11x17.pdf.
- 3. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

#### References

Bluestone JA. New perspectives of CD28-B7-mediated T cell costimulation. Immunity. 1995; 2(6):555-559. (Biology)

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: (Co)-stimulation)

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: (Co)-stimulation)

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: (Co) -stimulation)

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: (Co)-stimulation)

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)

Masten BJ, Yates JL, Pollard Koga AM, Lipscomb MF. Characterization of accessory molecules in murine lung dendritic cell function: roles for CD80, CD86, CD54, and CD40L. *Am J Respir Cell Mol Biol.* 1997; 16(3):335-342. (Biology: Blocking)

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: (Co)-stimulation)

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 lysis. *J Immunol.* 1996; 157(10):4347-4353. (Biology: (Co)-stimulation, Cytotoxicity)

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: (Co)-stimulation)

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: (Co)-stimulation)

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