

Technical Data Sheet

Purified Hamster Anti-Mouse CD28

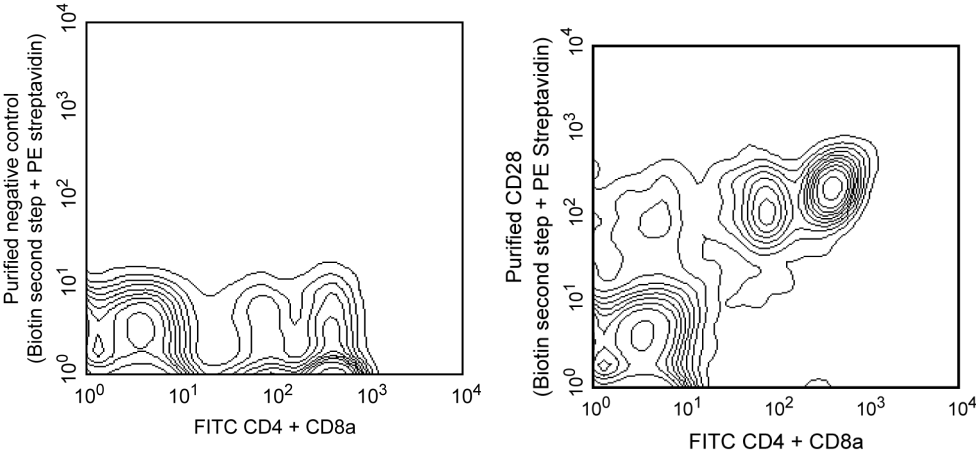
Product Information

Material Number:	553295
Size:	0.5 mg
Concentration:	0.5 mg/ml
Clone:	37.51
Immunogen:	Mouse EL-4 (T-cell lymphoma) Cells
Isotype:	Syrian Hamster IgG2, λ1
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

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 of splenic T lymphocytes. BALB/c splenocytes were preincubated with purified anti-mouse CD16/CD32 mAb 2.4G2 [Mouse BD Fc Block™] (Cat. No. 553141), then stained simultaneously 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 purified anti-mouse CD28 clone 37.51 (right panel). Staining of CD28-positive cells was detected with biotinylated anti-Syrian hamster IgG2 mAb G192-3 (Cat. No. 554029), 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. Store undiluted at 4° C.

Application Notes

Application

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

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Recommended Assay Procedure:

Precautions for 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™] (Cat. No. 553141) be used. Since this antigen is expressed at low density on resting peripheral T lymphocytes, it may be desirable to amplify staining by using a biotinylated second-step antibody followed by a "bright" third-step reagent, such as streptavidin-PE (Cat. No. 554061). If Mouse BD Fc Block™ is used, it is important that the second-step anti-hamster IgG antibody does not cross-react with the Mouse BD Fc Block™. Biotinylated anti-Syrian hamster IgG2 mAb G192-3 (Cat. No. 554029) would be a suggested second-step antibody to consider.

Caution: Sodium azide is a reversible inhibitor of oxidative metabolism; therefore, antibody preparations containing this preservative agent must not be used in cell cultures nor injected into animals. Sodium azide may be removed by washing stained cells or plate-bound antibody or dialyzing soluble antibody in sodium azide-free buffer. Since endotoxin may also effect the results of functional studies, we recommend the NA/LE™ (No Azide/Low Endotoxin) antibody format for in vitro and in vivo use.

Suggested Companion Products

Catalog Number	Name	Size	Clone
554061	Streptavidin PE	0.5 mg	(none)
553962	Purified Hamster IgG2, λ 1 Isotype Control	0.5 mg	Ha4/8
553030	FITC Rat Anti-Mouse CD8a	0.1 mg	53-6.7
553141	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.1 mg	2.4G2
553046	FITC Rat Anti-Mouse CD4	0.1 mg	RM4-5
554029	Biotin Mouse Anti-Syrian Hamster IgG2	0.5 mg	G192-3

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharming/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 http://www.bdbiosciences.com/pharming/hamster_chart_11x17.pdf.
4. 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

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