Technical Data Sheet

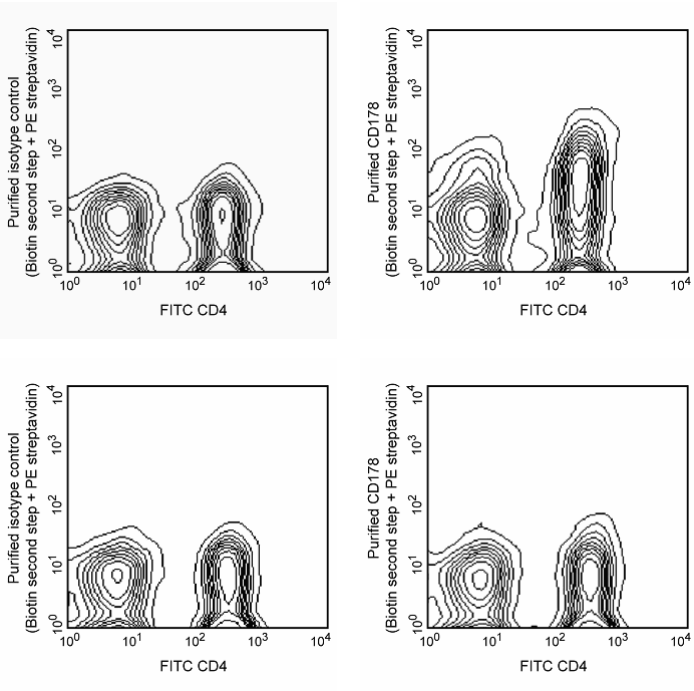
Purified Hamster Anti-Mouse and Rat CD178

Product Information

Material Number:	**555022**
Alternate Name:	Fas Ligand, CD95 Ligand
Size:	0.5 mg
Concentration:	0.5 mg/ml
Clone:	MFL4
Immunogen:	Mouse FasL transfected cells
Isotype:	Armenian Hamster IgG3, κ
Reactivity:	QC Testing: Rat Tested in Development: Mouse
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The MFL4 antibody, generated against mouse CD178 (Fas Ligand, CD95 Ligand), cross-reacts with the rat Fas Ligand antigen. MFL4 mAb detects rat Fas Ligand expressed by transfected cells, but not the parental cell line. In addition, the epitope detected by MFL4 can be induced on activated rat splenic T lymphocytes. The MFL4 mAb has been reported to inhibit killing of mouse lymphoma line A20.2J by rat *FasL*-transfected cells. The tissue distribution of mouse Fas Ligand has been well characterized. It is expressed at the mRNA and protein levels in a variety of tissues and cell types, including spleen, bone marrow, testis, uterus, eye, and activated lymphocytes. The expression of Fas Ligand in the rat has been detected in liver Kupffer cells, T cells in the central nervous system, spleen, and corpus luteum. MFL4 antibody reacts with the mouse Fas Ligand protein in a pattern similar to that of MFL3 antibody (Cat. No. 555291). Fas Ligand is a type-II transmembrane protein, a member of the TNF/NGF superfamily and a ligand for Fas (CD95). Fas Ligand and its counter-receptor CD95 are believed to participate in T-cell development, the regulation of immune responses, and cell-mediated cytotoxic mechanisms. It has been reported that human Fas Ligand is released from the surface of activated lymphocytes by metalloproteinase.



Expression of Fas Ligand on activated T lymphocytes. *T lymphocytes from LOU spleen (rat T Cell Enrichment Column, R&D Systems, Minneapolis, MN) were cultured for 7 hours in the presence of plate-bound R73 and JJ316 antibodies (anti-αβ TCR, Cat. No. 554910, and anti-CD28, Cat. No. 554992, respectively, top panels) or on uncoated plates (bottom panels). They were simultaneously stained with FITC-conjugated OX-35 (anti-rat CD4, Cat. No. 554837, all panels) and purified MFL4 (right panels) or purified A19-4 (isotype control, Cat. No. 553977, left panels) antibodies, followed by biotinylated goat anti-Armenian hamster IgG (Jackson Immunoresearch, West Grove, PA), then Streptavidin-PE (Cat. No. 554061, all panels). Flow cytometry was performed on a BD FACScan™ flow cytometry system.*

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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
Blocking	Reported

Recommended Assay Procedure:

We have found that enriched splenic T cells are induced to express Fas Ligand by 6 - 8-hour culture with plate-bound anti-αβ TCR mAb R73 (Cat. No. 554910) and anti-rat CD28 mAb JJ316 (Cat. No. 554992). Because Fas Ligand is expressed at low density on activated cells, we recommend the use of a biotinylated second-step antibody, such as polyclonal goat anti-hamster IgG, with a "bright" third-step reagent, such as Streptavidin-PE (Cat. No. 554061). Other reported applications include blocking of the cytotoxic activities of *mFasL*-transfected L5178Y T lymphoma, *rFasL*-transfected COS cells,
and *rFasL*-transfected L5178Y cells.

Suggested Companion Products

Catalog Number	Name	Size	Clone
554910	Purified NA/LE Mouse Anti-Rat αβ T-Cell Receptor	0.5 mg	R73
554992	Purified NA/LE Mouse Anti-Rat CD28	0.5 mg	JJ316
554837	FITC Mouse Anti-Rat CD4	0.5 mg	OX-35
554061	PE Streptavidin	0.5 mg	(none)

Product Notices

1. 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. 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.
4. 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 affect the results of functional studies, we recommend the NA/LE (No Azide/Low Endotoxin) antibody format, if available, for in vitro and in vivo use.
5. 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/pharmingen/hamster_chart_11x17.pdf.

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