

Technical Data Sheet

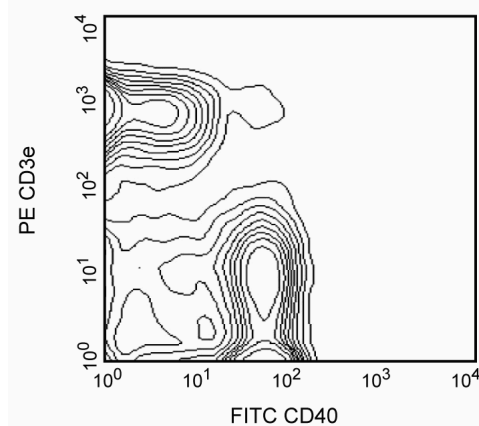
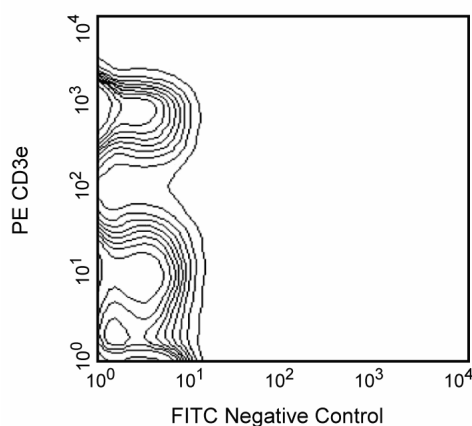
FITC Rat Anti-Mouse CD40

Product Information

Material Number:	561845
Size:	0.1 mg
Concentration:	0.5 mg/ml
Clone:	3/23
Immunogen:	Mouse CD40 Recombinant Protein
Isotype:	Rat (LOU) IgG2a, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing $\leq 0.09\%$ sodium azide.

Description

The 3/23 clone reacts with CD40, a 40-50 kDa glycoprotein expressed on B lymphocytes and other antigen-presenting cells. CD40 has been reported to be transiently expressed on activated CD4⁺ and CD8⁺ T cells and in some mouse strains, the 3/23 mAb has been reported to react with 5-10% of T lymphocytes in adult mouse, but not neonatal, spleen. CD40 plays a key role in B-cell growth and differentiation where interactions of CD40 with its ligand, CD154, are involved in the initiation, effector, and memory stages of cell-mediated immune responses. In addition, CD40 has been reported to be involved with the triggering of NK cells and NK-T cells. Ligation of CD40 with the 3/23 antibody has been reported to induce splenic B cells to express the costimulatory molecule CD86 (B7-2). In addition, although the 3/23 antibody by itself is a weak B-cell mitogen, it has been reported to synergize markedly with mitogenic anti-IgM, anti-IgD mAb or IL-4 to promote B-cell proliferation.



Two-color analysis of the expression of CD40 on mouse spleen cells. BALB/c splenocytes were simultaneously stained with PE-conjugated anti-mouse CD3e mAb 145-2C11 (Cat. No. 553063) and FITC-conjugated mAb 3/23 (right panel). Please note that staining of a T-cell subset by mAb 3/23 has not been consistently observed. Flow cytometry was performed on a BD FACScan™ 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 FITC under optimum conditions, and unreacted FITC was removed.

Application Notes

Application

Flow cytometry	Routinely Tested
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Recommended Assay Procedure:

Flow Cytometry: The staining intensity of FITC-conjugated 3/23 mAb is severely reduced when used on leukocytes that have been fixed (≤ 2.5 hours with $\sim 0.5\%$ formaldehyde). Therefore, **investigators are advised not to fix cells prior to staining for flow cytometry.** Freshly-isolated leukocytes and cell lines may wait for analysis in wash buffer at 4°C, without fixation, for up to 18 hours post-staining without loss of viability. Activated lymphocytes may lose viability rapidly, and data should be collected within 5 hours post-staining. For applications requiring fixation of cells prior to staining, an alternate mAb to mouse CD40, clone HM40-3 (Cat. No. 553723), is recommended.

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Suggested Companion Products

Catalog Number	Name	Size	Clone
553063	PE Hamster Anti-Mouse CD3e	0.1 mg	145-2C11
553723	FITC Hamster Anti-Mouse CD40	0.5 mg	HM40-3
553929	FITC Rat IgG2a, κ Isotype Control	0.25 mg	R35-95
554656	Stain Buffer (FBS)	500 ml	(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. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
5. An isotype control should be used at the same concentration as the antibody of interest.

References

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