Technical Data Sheet

PE-CF594 Rat Anti-Mouse CD24

Product Information

Material Number: 562477

Alternate Name: CD24a; HSA; Heat Stable Antigen; Ly-52; Nectadrin; R13-Ag

 Size:
 0.1 mg

 Concentration:
 0.2 mg/ml

 Clone:
 M1/69

Immunogen: C57BL/10 Mouse Splenic T Lymphocytes

Isotype:Rat (DA) IgG2b, κ Reactivity:QC Testing: Mouse

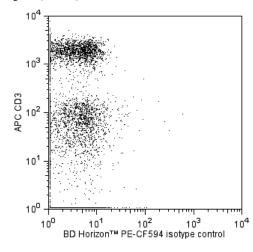
Storage Buffer: Aqueous buffered solution containing BSA, protein stabilizer, and ≤0.09%

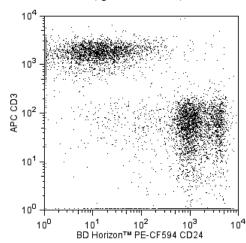
sodium azide.

Description

The M1/69 monoclonal antibody specifically binds to CD24 (Heat-Stable Antigen, HSA or HsAg), a variably glycosylated, glycosyl-phosphatidylinositol-anchored membrane protein expressed on erythrocytes, granulocytes, monocytes, lymphocytes, and neurons. Hematopoietic stem cells of the embryonic yolk sac and fetal liver express CD24. Levels of expression of CD24 vary during differentiation of the T and B cell lineages. In the bone marrow, hematopoietic progenitors acquire CD24 expression upon commitment to the B-lymphocyte lineage. Immature B cells in the bone marrow express low CD24 levels whereas peripheral B lymphocytes express intermediate to high levels of CD24. The level of CD24 expression has been reported to rise upon activation of splenic B cells with LPS, but not with CD154 (CD40 Ligand). The majority of thymocytes express high levels of CD24, while most mature thymic and peripheral T lymphocytes do not express CD24. In contrast, TCR-bearing thymocytes which emigrate to the spleen are CD24+. Dendritic cells of the thymus, spleen, liver, and epidermal Langerhans cells have also been reported to express CD24. CD24 is not expressed by NK cells, as determined by staining with J11d mAb (Cat. No. 553146). CD24 is involved in the costimulation of CD4+ T cells by B cells, it is a "co-inducer" of in vitro thymocyte maturation, and it is a ligand of CD62P (P-selectin). While the monoclonal antibodies 30-F1, M1/69, and J11d all react with CD24, they show subtle differences in the level of staining of different lymphocyte populations. When possible, investigators should continue to use the same monoclonal anti-CD24 antibody as used in previous studies.

This antibody is conjugated to BD Horizon[™] PE-CF594, which has been developed exclusively by BD Biosciences as a better alternative to PE-Texas Red®. PE-CF594 excites and emits at similar wavelengths to PE-Texas Red® yet exhibits improved brightness and spectral characteristics. Due to PE having maximal absorption peaks at 496 nm and 564 nm, PE-CF594 can be excited by the blue (488-nm), green (532-nm) and yellow-green (561-nm) lasers and can be detected with the same filter set as PE-Texas Red® (eg 610/20-nm filter).





Multicolor flow cytometric analysis of CD24 expression on mouse splenocytes. Splenocytes from C57BL/6 mice were stained with APC Hamster Anti-Mouse CD3e antibody (Cat. No. 553066) and with either a BD HorizonTM PE-CF594 Rat [gG2b, κ isotype control (Cat. No. 562308, Left Panel) or with the BD HorizonTM PE-CF594 Rat Anti-Mouse CD24 antibody (Cat. No. 562477, Right Panel). Two-color flow cytometric dot plots showing the correlated expression patterns of CD3 versus CD24 (or lg isotype control staining) were derived from gated events with the forward and side light-scatter characteristics of viable splenocytes. Flow cytometry was performed using a BDTM LSR II Flow Cytometer System.

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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 BD Horizon™ PE-CF594 under optimum conditions, and unconjugated antibody and free PE-CF594 were removed.

Application Notes

Application

Flow cytometry	Routinely Tested	
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Suggested Companion Products

<u>C</u>	Catalog Number	Name	Size	Clone
5	62308	PE-CF594 Rat IgG2b, κ Isotype Control	0.1 mg	A95-1
5	54656	Stain Buffer (FBS)	500 ml	(none)
5	53066	APC Hamster Anti-Mouse CD3e	0.1 mg	145-2C11

Product Notices

- 1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- 2. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
- 3. An isotype control should be used at the same concentration as the antibody of interest.
- 4. Please observe the following precautions: Absorption of visible light can significantly alter the energy transfer occurring in any tandem fluorochrome conjugate; therefore, we recommend that special precautions be taken (such as wrapping vials, tubes, or racks in aluminum foil) to prevent exposure of conjugated reagents, including cells stained with those reagents, to room illumination.
- 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.
- 6. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
- Texas Red is a registered trademark of Molecular Probes, Inc., Eugene, OR.
- CFTM is a trademark of Biotium, Inc.
- 9. When excited by the yellow-green (561-nm) laser, the fluorescence may be brighter than when excited by the blue (488-nm) laser.
- 10. This product is provided under an Agreement between BIOTIUM and BD Biosciences. The manufacture, use, sale, offer for sale, or import of this product is subject to one or more patents or pending applications owned or licensed by Biotium, Inc. This product, and only in the amount purchased by buyer, may be used solely for buyer's own internal research, in a manner consistent with the accompanying product literature. No other right to use, sell or otherwise transfer (a) this product, or (b) its components is hereby granted expressly, by implication or by estoppel. This product is for research use only. Diagnostic uses require a separate license from Biotium, Inc. For information on purchasing a license to this product including for purposes other than research, contact Biotium, Inc., 3159 Corporate Place, Hayward, CA 94545, Tel: (510) 265-1027. Fax: (510) 265-1352. Email: btinfo@biotium.com.
- Because of the broad absorption spectrum of the tandem fluorochrome, extra care must be taken when using multi-laser cytometers, which
 may directly excite both PE and CFTM594.
- 12. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

Alterman LA, Crispe IN, Kinnon C. Characterization of the murine heat-stable antigen: an hematolymphoid differentiation antigen defined by the J11d, M1/69 and B2A2 antibodies. *Eur J Immunol.* 1990; 20(7):1597-1602. (Clone-specific)

Crowley M, Inaba K, Witmer-Pack M, Steinman RM. The cell surface of mouse dendritic cells: FACS analyses of dendritic cells from different tissues including thymus. *Cell Immunol.* 1989; 118(1):108-125. (Clone-specific)

Reichlin A, lizuka K, Yokoyama WM. Isolation of murine natural killer cells. In: Coligan J, Kruisbeek AM, Margulies D, Shevach EM, Strober W, ed. *Current Protocols in Immunology*, New York: John Wiley and Sons; 1999;3.22.1-3.22.6. (Biology)

Springer T, Galfre G, Secher DS, Milstein C. Monoclonal xenogeneic antibodies to murine cell surface antigens: identification of novel leukocyte differentiation antigens. *Eur J Immunol.* 1978; 8(8):539-551. (Immunogen: Immunohistochemistry, Western blot)

Stall AM, Wells SM. FACS analysis of murine B-cell populations. In: Herzenberg LA, Weir DM, Blackwell C, ed. Weir's Handbook of Experimental Immunology. Blackwell Science Publishers; 1997:63.1-63.17. (Biology)

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