Technical Data Sheet

PE-CF594 Rat Anti-Mouse Siglec-F

Product Information

Material Number: 562757

Alternate Name: Siglec5; sialic acid binding Ig-like lectin 5; SIGL5; Siglec-5; CD170

 Size:
 50 µg

 Concentration:
 0.2 mg/ml

 Clone:
 E50-2440

Immunogen: Mouse Siglec-F and human IgG Fc recombinant fusion protein

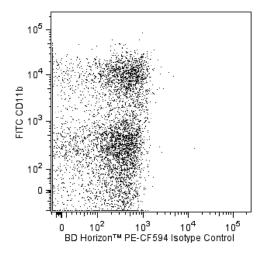
Isotype:Rat (LOU) IgG2a, κ Reactivity:QC Testing: Mouse

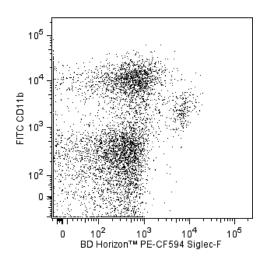
Storage Buffer: Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The E50-2440 antibody reacts with Siglec-F, the sixth siglec protein to be reported in the mouse. Siglecs are the sialic acid-binding immunoglobulin superfamily <u>lec</u>tins defined in the human, each of which has a distinctive expression pattern in the hematopoietic system and at least some of which are known to mediate cell-cell interactions. Orthologous proteins of human Siglec-1 (Sialoadhesin or CD169), Siglec-2 (CD22), and Siglec-4 (myelin-associated glycoprotein) have been characterized in the mouse. Human Siglec-3 (CD33) and Siglecs-5 through -10 are encoded by a cluster of closely related genes, and each has two cytoplasmic ITIM (Immunoreceptor Tyrosine-based Inhibitory Motifs). Similarly, mouse Siglec-F is encoded by a gene in a syntenic cluster in the mouse, and the protein has sialic acid-binding activity and an intracytoplasmic ITIM. Its expression pattern differs from those of the human Siglec-3-related proteins in that it is found on immature cells of the myelomonocytic lineage, with reduced expression on mature neutrophils and monocytes, and not on lymphoid cells. It has been proposed that mAb E50-2440 may be used for identification of immature myelomonocytic cells in the mouse.

This antibody is conjugated to BD HorizonTM 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 Siglec-F expression on BALB/c mouse bone marrow cells. Bone marrow cells were stained simultaneously with FITC Rat Anti-Mouse CD11b antibody (Cat. No. 553310/557396/561688) and with either BD Horizon™ PE-CF594 Rat IgG2a, κ Isotype Control (Cat. No. 562302; Left Panel) or BD Horizon™ PE-CF594 Rat Anti-Mouse Siglec-F (Cat. No. 562757; Right Panel) in the presence of Mouse Fc Block™ (purified anti-mouse CD16/CD32 mAb 2.4G2, Cat. No. 553141/553142). Two-color flow cytometric dot plots show the correlated expression patterns of Siglec-F (or Ig isotype control staining) versus CD11b for gated events with the forward and side light-scatter characteristics of viable bone marrow cells. Flow cytometry was performed using a BD™ 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 antibody was conjugated with BD Horizon™ PE-CF594 under optimum conditions, and unconjugated antibody and free PE-CF594 were removed

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

Application Notes

Application

Flow cytometry	Routinely Tested	
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Suggested Companion Products

Catalog Number	Name Name	Size	Clone	
562302	PE-CF594 Rat IgG2a, κ Isotype Control	0.1 mg	R35-95	
554656	Stain Buffer (FBS)	500 ml	(none)	
553141	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block TM)	0.1 mg	2.4G2	
553142	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block TM)	0.5 mg	2.4G2	
553310	FITC Rat Anti-Mouse CD11b	0.5 mg	M1/70	
557396	FITC Rat Anti-Mouse CD11b	0.1 mg	M1/70	
561688	FITC Rat Anti-Mouse CD11b	25 μg	M1/70	

Product Notices

- 1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- 2. An isotype control should be used at the same concentration as the antibody of interest.
- 3. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
- 4. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
- 5. 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.
- 7. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
- 8. Texas Red is a registered trademark of Molecular Probes, Inc., Eugene, OR.
- 9. CFTM is a trademark of Biotium, Inc.
- 10. When excited by the yellow-green (561-nm) laser, the fluorescence may be brighter than when excited by the blue (488-nm) laser.
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- 12. 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.

References

Angata T, Hingorani R, Varki NM, Varki A. Cloning and characterization of a novel mouse Siglec, mSiglec-F: differential evolution of the mouse and human (CD33) Siglec-3-related gene clusters. *J Biol Chem.* 2001; 276(48):45128-45136. (Immunogen: Flow cytometry, Fluorescence activated cell sorting) Crocker PR, Varki A. Siglecs, sialic acids and innate immunity. *Trends Immunol.* 2001; 22(6):337-342. (Biology)

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