

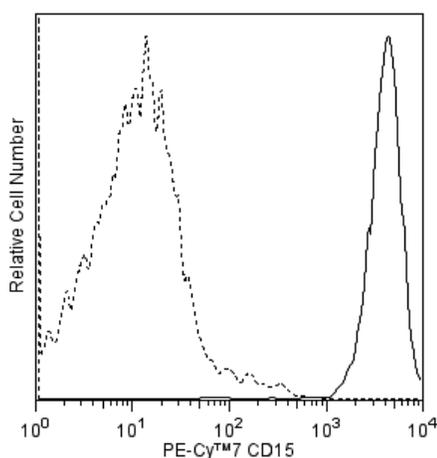
Technical Data Sheet

PE-Cy™7 Mouse Anti-Human CD15**Product Information**

Material Number:	560827
Alternate Name:	Lewis X; Le-X; X-Hapten; SSEA-1; 3-FAL
Size:	50 Tests
Vol. per Test:	5 µl
Clone:	HI98 (also known as HIM1)
Immunogen:	Human Mononuclear Cells
Isotype:	Mouse IgM, κ
Reactivity:	QC Testing: Human
Workshop:	IV M141
Storage Buffer:	Aqueous buffered solution containing BSA, protein stabilizer, and ≤0.09% sodium azide.

Description

The HI98 monoclonal antibody specifically reacts with 3-fucosyl-N-acetylglucosamine (3-FAL), a 220 kDa carbohydrate structure, also called X-hapten, SSEA-1, CD15 or Lewis X. This structure is found on a variety of cell surface glycolipids and glycoproteins. 3-FAL is expressed on >95% of granulocytes, including neutrophils and eosinophils, and to a varying degree on monocytes, but not on lymphocytes or basophils. CD15 plays a role in mediating phagocytosis, bactericidal activity and chemotaxis. This antibody is also suitable for staining formalin-fixed, paraffin-embedded tissue sections without pretreatment. Since the Abs are recognizing a carbohydrate epitope (3-fucosyl-N-acetylglucosamine) they also should work across species and not only for human.



Flow cytometric analysis of CD15 expression on human peripheral blood granulocytes. Whole blood was stained with PE-Cy™7 Mouse Anti-Human CD15 (Cat. No. 560827; solid line histogram) or with a PE-Cy™7 Mouse IgM, κ Isotype Control (Cat. No. 560856; dashed line histogram). The erythrocytes were lysed with Lysing Buffer (Cat. No. 555899). The fluorescence histograms were derived from gated events with the forward and side light-scattering characteristics of viable granulocytes. Flow cytometry was performed on a BD™ LSR II.

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 PE-Cy7 under optimum conditions, and unconjugated antibody and free PE-Cy7 were removed.

Application Notes**Application**

Flow cytometry	Routinely Tested
Immunohistochemistry-paraffin	Tested During Development

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Suggested Companion Products

<u>Catalog Number</u>	<u>Name</u>	<u>Size</u>	<u>Clone</u>
560856	PE-Cy TM 7 Mouse IgM, κ Isotype Control	0.1 mg	G155-228
555899	Lysing Buffer	100 mL	(none)
554656	Stain Buffer (FBS)	500 mL	(none)
554657	Stain Buffer (BSA)	500 mL	(none)
349202	BD FACST TM Lysing Solution	100 mL	(none)

Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1×10^6 cells in a 100- μ l experimental sample (a test).
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. 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.
5. Cy is a trademark of GE Healthcare.
6. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
7. 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.
8. PE-Cy7 is a tandem fluorochrome composed of R-phycoerythrin (PE), which is excited by 488-nm light and serves as an energy donor, coupled to the cyanine dye Cy7, which acts as an energy acceptor and fluoresces maximally at 780 nm. PE-Cy7 tandem fluorochrome emission is collected in a detector for fluorescence wavelengths of 750 nm and higher. Although every effort is made to minimize the lot-to-lot variation in the efficiency of the fluorochrome energy transfer, differences in the residual emission from PE may be observed. Therefore, we recommend that individual compensation controls be performed for every PE-Cy7 conjugate. PE-Cy7 is optimized for use with a single argon ion laser emitting 488-nm light, and there is no significant overlap between PE-Cy7 and FITC emission spectra. When using dual-laser cytometers, which may directly excite both PE and Cy7, we recommend the use of cross-beam compensation during data acquisition or software compensation during data analysis.
9. Warning: Some APC-Cy7 and PE-Cy7 conjugates show changes in their emission spectrum with prolonged exposure to formaldehyde. If you are unable to analyze fixed samples within four hours, we recommend that you use BDTM Stabilizing Fixative (Cat. No. 338036).
10. Please refer to www.bdbiosciences.com/pharming/protocols for technical protocols.

References

Barclay NA, Brown MH, Birkeland ML, et al, ed. *The Leukocyte Antigen FactsBook*. San Diego, CA: Academic Press; 1997(Biology)
Knapp W. W. Knapp .. et al., ed. *Leucocyte typing IV : white cell differentiation antigens*. Oxford New York: Oxford University Press; 1989:1-1182. (Biology)
Lund-Johansen F, Olweus J, Horejsi V, et al. Activation of human phagocytes through carbohydrate antigens (CD15, sialyl-CD15, CDw17, and CDw65). *J Immunol*. 1992; 148(10):3221-9. (Biology)