

Technical Data Sheet

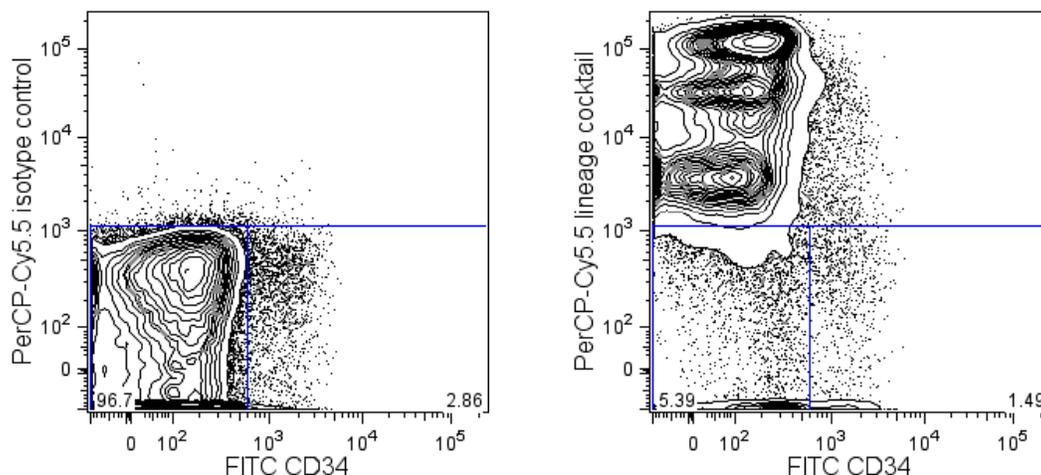
PerCP-Cy™5.5 Mouse Lineage Antibody Cocktail, with Isotype Control**Product Information**

Material Number:	561317
Size:	100 tests
Vol. per Test:	20 µl
Reactivity:	QC Testing: Mouse
Component:	51-9006964
Description:	PerCP-Cy™5.5 Mouse Lineage Antibody Cocktail
Size:	100 tests (1 ea)
Vol. per Test:	20 µl
Storage Buffer:	Aqueous buffered solution containing BSA, protein stabilizer, and ≤0.09% sodium azide.

Component:	51-9006977
Description:	PerCP-Cy™5.5 Mouse Lineage Isotype Control Cocktail
Size:	100 tests (1 ea)
Vol. per Test:	20 µl
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The PerCP-Cy™5.5 Mouse Lineage Antibody Cocktail has been designed to react with cells from the major hematopoietic lineages, such as T lymphocytes, B lymphocytes, monocytes/macrophages, NK cells, erythrocytes, and granulocytes. This pre-diluted Cocktail of five PerCP-Cy™5.5-conjugated antibodies is designed to label lineage marker-positive cells for exclusion to facilitate the flow cytometric identification of lineage marker-negative hematopoietic progenitors in mouse bone marrow. Components include clone 145-2C11, which recognizes Mouse CD3e; M1/70, which recognizes CD11b; RA3-6B2, which recognizes CD45R/B220; TER-119, which recognizes Ly-76, mouse erythroid cells; and RB6-8C5, which recognizes Ly-6G and Ly-6C. PerCP-Cy™5.5 Mouse Lineage Isotype Control Cocktail contains equivalent concentrations of isotype-matched negative-control immunoglobulin. Additional fluorochrome-labeled reagents may be combined with the PerCP-Cy™5.5 Mouse Lineage Antibody Cocktail, and the PerCP-Cy™5.5 Mouse Lineage Isotype Control Cocktail, to further characterize hematopoietic progenitor subpopulations.



Identification of CD34+ and CD34- subpopulations of hematopoietic progenitor cells. BALB/c bone marrow cells were treated with Mouse BD Fc Block™ Purified Rat anti-CD16/CD32 mAb 2.4G2 (Cat. No. 553141/553142) and stained with either FITC Rat IgG2a κ Isotype Control (Cat. No. 554688; data not shown) or FITC Rat anti-Mouse CD34 mAb RAM34 (Cat. No. 553733) and with either PerCP-Cy™ 5.5 Mouse Lineage Isotype Control Cocktail (Left Panel) or PerCP-Cy™ 5.5 Mouse Lineage Antibody Cocktail (Right Panel). Dead cells were excluded from analysis by staining with 7-AAD (7-Amino-Actinomycin D; Cat. No. 559925). Flow cytometry was performed on a BD LSR™II Flow Cytometry 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 PerCP-Cy5.5 under optimum conditions, and unconjugated antibody and free PerCP-Cy5.5 were removed. Storage of PerCP-Cy5.5 conjugates in unoptimized diluent is not recommended and may result in loss of signal intensity.

Application Notes

Application

Flow cytometry	Routinely Tested
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Recommended Assay Procedure:

20 µl of the antibody or isotype control cocktail is sufficient to stain each sample of 10⁶ leukocytes for flow cytometric analysis. We recommend the use of Mouse BD Fc Block™ purified anti-mouse CD16/Cd32 mAb 2.4G2 (Cat. No. 553141/553142) for optimal staining. The PerCP-Cy5.5 Mouse Lineage Antibody Cocktail can be used to deplete cells bearing the hematopoietic lineage markers by flow cytometric sorting. The PerCP-Cy5.5 fluorochrome is excited by laser lines from 595 to 647 nm, and its emission is collected in a detector for fluorescence wavelengths between 640 and 680 nm.

Suggested Companion Products

Catalog Number	Name	Size	Clone
553141	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.1 mg	2.4G2
554688	FITC Rat IgG2a, κ Isotype Control	0.1 mg	R35-95
553733	FITC Rat anti-Mouse CD34	0.5 mg	RAM34
559925	7-AAD	2.0 ml	(none)

Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1 × 10⁶ cells in a 100-µl experimental sample (a test).
2. Please refer to www.bdbiosciences.com/pharming/en/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. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
5. Cy is a trademark of Amersham Biosciences Limited. This conjugated product is sold under license to the following patents: US Patent Nos. 5,486,616; 5,569,587; 5,569,766; 5,627,027.
6. 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.
7. PerCP-Cy5.5-labelled antibodies can be used with FITC- and R-PE-labelled reagents in single-laser flow cytometers with no significant spectral overlap of PerCP-Cy5.5, FITC, and R-PE fluorescence.
8. PerCP-Cy5.5 is optimized for use with a single argon ion laser emitting 488-nm light. Because of the broad absorption spectrum of the tandem fluorochrome, extra care must be taken when using dual-laser cytometers, which may directly excite both PerCP and Cy5.5™. We recommend the use of cross-beam compensation during data acquisition or software compensation during data analysis.
9. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
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References

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