Technical Data Sheet

V450 Mouse Lineage Antibody Cocktail, with Isotype Control

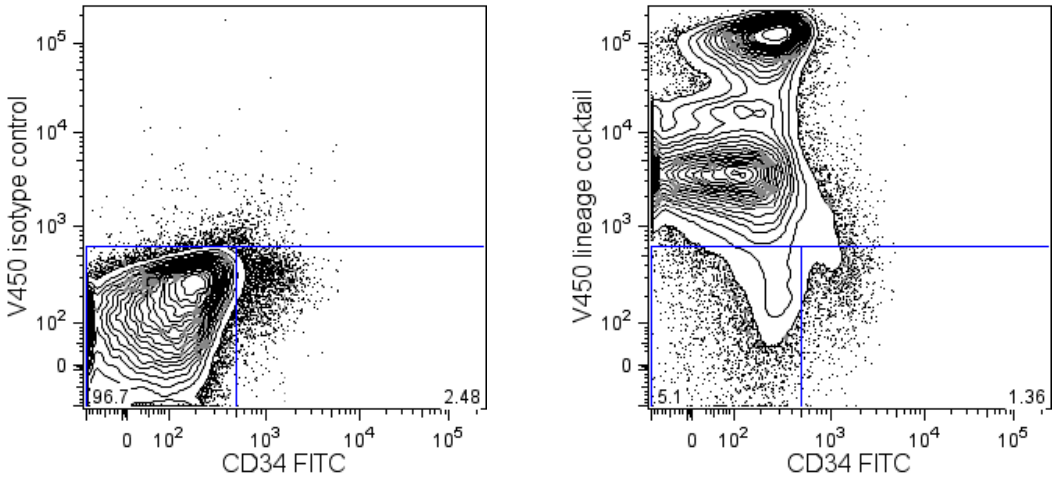
Product Information

Material Number:	**561301**
Size:	100 tests
Vol. per Test:	20 µl
Reactivity:	QC Testing: Mouse
Component:	**51-9006957**
Description:	V450 Mouse Lineage Antibody Cocktail
Size:	100 tests (1 ea)
Vol. per Test:	20 µl
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.
Component:	**51-9006958**
Description:	V450 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 BD Horizon™ V450 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 BD Horizon™ V450-conjugated antibodies is designed to label lineage marker-positive cells for exclusion to facilitate the flow cytometric identification of lineage marker-negative hematopoietic progenitor cells in mouse bone marrow. Components include clone 500A2, 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. The BD Horizon™ V450 Mouse Lineage Isotype Control Cocktail contains equivalent concentrations of isotype-matched negative-control immunoglobulin. Additional fluorochrome-labeled reagents may be combined with the BD Horizon™ V450 Mouse Lineage Antibody Cocktail, and the BD Horizon™ V450 Mouse Lineage Isotype Control Cocktail, to further characterize hematopoietic progenitor subpopulations.

The antibodies are conjugated to BD Horizon™ V450 which has been developed for use in multicolor flow cytometry experiments and is available exclusively from BD Biosciences. It is excited by the Violet laser Ex max of 406 nm and has an Em Max at 450 nm. Conjugates with BD Horizon™ V450 can be used in place of Pacific Blue™ conjugates.



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 BD Horizon™ V450 Mouse Lineage Isotype Control Cocktail (Left Panel) or BD Horizon™ V450 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 using 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 BD Horizon™ V450 under optimum conditions, and unreacted BD Horizon™ V450 was removed.

Application Notes

Application

Flow cytometry	Routinely Tested

Recommended Assay Procedure:

20 µl of the antibody or isotype control cocktail is sufficient to stain each sample of 10^6 leukocytes for flow cytometric analysis. We recommend the use of Mouse BD Fc Block™ Purified Rat anti-Mouse CD16/CD32 mAb 2.4G2 (Cat. No. 553141/553142) for optimal staining. The V450 Mouse Lineage Antibody Cocktail can be used to deplete cells bearing the hematopoietic lineage markers by flow cytometric sorting.

Suggested Companion Products

Catalog Number	Name	Size	Clone
553141	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.1 mg	2.4G2
553733	FITC Rat anti-Mouse CD34	0.5 mg	RAM34
554688	FITC Rat IgG2a, κ Isotype Control	0.1 mg	R35-95
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 × 10^6 cells in a 100-µl experimental sample (a test).
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. BD Horizon™ V450 has a maximum absorption of 406 nm and maximum emission of 450 nm. Before staining with this reagent, please confirm that your flow cytometer is capable of exciting the fluorochrome and discriminating the resulting fluorescence.
6. Pacific Blue™ is a trademark of Molecular Probes, Inc., Eugene, OR.
7. Source of all serum proteins is from USDA inspected abattoirs located in the United States.

References

Goodell MA, Rosenzweig M, Kim H, et al. Dye efflux studies suggest that hematopoietic stem cells expressing low or undetectable levels of CD34 antigen exist in multiple species. *Nat Med.* 1997; 3(12):1337-1345. (Biology)

Morrison SJ, Wandycz AM, Hemmati HD, Wright DE, Weissman IL. Identification of a lineage of multipotent hematopoietic progenitors. *Development.* 1997; 124(10):1929-1939. (Biology)

Okada S, Nakauchi H, Nagayoshi K, Nishikawa S, Miura Y, Suda T. In vivo and in vitro stem cell function of c-kit- and Sca-1-positive murine hematopoietic cells. *Blood.* 1992; 80(12):3044-3050. (Biology)

Osawa M, Tokumoto Y, Nakauchi H. Hematopoietic stem cells. In: Herzenberg LA, Weir DM, Blackwell C, ed. *Weir's Handbook of Experimental Immunology, 5th Edition.* Cambridge: Blackwell Science; 1996:66.1-66.5. (Biology)

Spangrude GJ, Heimfeld S, Weissman IL. Purification and characterization of mouse hematopoietic stem cells. *Science.* 1988; 241(4861):58-62. (Biology)

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