BD Stemflow Mouse Hematopoietic Stem and Progenitor Cell Isolation Kit

Features

Enables high purity isolation of HPC and HSC subpopulations from mouse bone marrow

Delivers a streamlined solution for consistent experiments with fluorochrome-conjugated antibodies, compensation beads, protocols, and analysis guidelines

Facilitates compensation and scatter setup and conserves cells with BD CompBead microparticles

Provides flexibility to drop in additional fluorochromeconjugated antibodies. Compatible with side population staining and upfront magnetic bead enrichment.

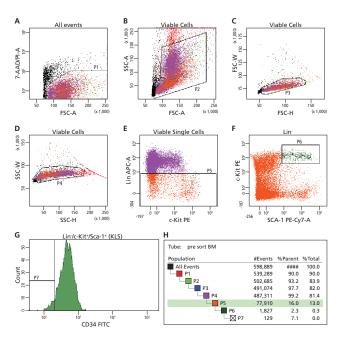


Figure 1. Gating strategy for sorting KSL/CD34-/dim cells.

B6 bone marrow was stained with APC lineage cocktail, c-Kit PE, Sca-1 PE-Cy7, and CD34 FITC. Figure 1A shows the discrimination of dead cells, followed by gating of bone marrow cells based on scatter properties (Figure 1B). Doublet discrimination is shown in Figures 1C and 1D. Lineage positive cells are gated out in Figure 1E, and HSCs and HPCs are gated in Figure 1F (Sca-1*/c-Kit*). Finally, enrichment of HSCs is achieved by gating on the CD34*/dim cells.

The BD Stemflow™ Mouse Hematopoietic Stem and Progenitor Cell Isolation Kit provides a comprehensive research system for the reliable, high purity isolation of hematopoietic progenitor and stem cells (HPCs and HSCs) from bone marrow.

The ready-to-use kit reduces experiment complexity and improves dependability by integrating many of the relevant materials and methods required for cell sorting by flow cytometry. These components include fluorochrome conjugated monoclonal antibodies to markers for HSCs, progenitors, and mature hematopoietic cells, as well as compensation beads, verified protocols, and software and analysis guidelines.

To provide flexibility to explore alternative HSC enrichment options, an open design offers compatibility with side population analysis and magnetic enrichment, as well as the easy addition of supplementary antibodies.

Multicolor flow cytometry for in-depth analysis

By capitalizing on flow cytometry's powerful capabilities for single-cell sorting based on the classical KLS (c-Kit+ Lin- Sca-1+) cell surface markers, the isolation kit delivers optimal purity for isolating multipotent long-term HSC (LT-HSC), short-term HSC (ST-HSC), or multipotent progenitor (MPP) subpopulations from heterogeneous bone marrow cells.

A comprehensive system for simplified, consistent setup

To improve productivity and reduce assay-to-assay variability, the kit contains pre-titrated, pre-conjugated monoclonal antibodies to cell surface markers for self-renewal (c-Kit PE, Sca-1 PE-CyTM7), a marker for progenitors (CD34 FITC), and a cocktail of markers for the hematopoietic lineage (APC Lineage Cocktail). To further streamline the isolation process, Mouse BD Fc BlockTM, 7-AAD vital dye, and isotype controls are also provided.

BDTM CompBead microparticles simplify and standardize experimental setup by facilitating compensation for multicolor analysis.

Optimized protocols and guidelines for processing, sorting, and recovering cells using BD flow cytometry instruments and software provide easy-to-follow guidance and standardization.

Visit bdbiosciences.com/stemcellsource for more information.



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Modular and open to accommodate specific needs

For simple customization and more advanced analysis, the open, modular architecture of the kit allows for the easy addition of supplementary fluorochrome conjugated monoclonal antibodies against critical cell-surface markers. BD CompBead microparticles can also be used as compensation controls for additional antibody drop-ins to this kit. This is particularly useful when testing for markers that might or might not be expressed on cells of interest.

The kit is compatible with side population staining to further enrich for LT-HSCs. In addition, upfront magnetic bead enrichment can be used for preparative applications.

A resource for stem cell research

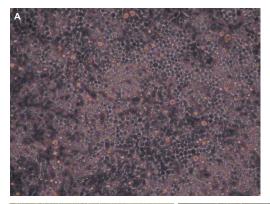
BD Biosciences continues to support innovation in the area of stem cell research with more than 20 years of experience in the field. Inspired by in-depth understanding of the complexities of biological experiments, the BD Stemflow Mouse Hematopoietic Stem and Progenitor Cell Isolation Kit is designed to make it easier for researchers to obtain accurate results, increase research productivity, and accelerate discoveries.

Ordering Information

Description	Cat.No.
BD Stemflow [™] Mouse Hematopoietic Stem and Progenitor Cell Isolation Kit (100 tests: 10 sorts of the bone marrow isolated and pooled from 10 mice)	560492

BD Stemflow Mouse Hematopoietic Stem and Progenitor Cell Isolation Kit contents

Monoclonal Antibodies Mouse c-Kit (CD117) PE Mouse CD34 FITC Mouse Sca-1 PE-Cy7 Mouse APC Lineage Cocktail Isotype Controls and Other Staining Tools Rat $\lg G_{2b'} \kappa$ Isotype Control PE Rat $\lg G_{2a'} \kappa$ Isotype Control FITC Rat $\lg G_{2a'} \kappa$ Isotype Control PE-Cy7 Rat/Hamster Isotype Control Cocktail APC Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block TM) 7-AAD vital dye
Mouse CD34 FITC Mouse Sca-1 PE-Cy7 Mouse APC Lineage Cocktail Isotype Controls and Other Staining Tools Rat $IgG_{2b'}$ κ Isotype Control PE Rat $IgG_{2a'}$ κ Isotype Control FITC Rat $IgG_{2a'}$ κ Isotype Control PE-Cy7 Rat/Hamster Isotype Control Cocktail APC Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block TM)
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Rat/Hamster Isotype Control Cocktail APC Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)
Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)
7-AAD vital dye
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Flow Cytometry Compensation Beads
BD™ CompBead Plus Anti-Rat/Hamster Ig, κ
Protocols and Analysis Guidelines
Protocol for cell sorting and recovery
Protocol for analysis, including creating BD FACSDiva™ templates



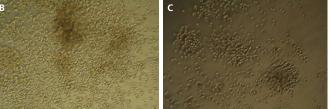


Figure 2. Cobblestone forming and colony forming assays on sorted cells.

Flow cytometry sorted HSCs and HPCs were plated in a cobblestone forming assay (a surrogate LT-HSC assay) for 3 weeks. Images were taken at multiple time points. The characteristic cobblestone pattern was observed (Figure 2A). To show that the cobblestone cells had multilineage potential, they were further plated in a colony forming assay. Colonies were imaged after 12 days (Figures 2B and 2C).



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