

BD Mouse B-Lymphocyte Kits and Templates

Analysis of B-Lymphocyte Subsets and Activation on the BD Accuri™ C6 Flow Cytometer

Features

Preconfigured kits, protocols, and software templates

Simplify characterization of mouse B-cell subsets and activation markers on the BD Accuri C6

Support studies involving mouse CD45R/B220, CD23, sIgM, CD19, CD25, and CD69

Enable quick and easy setup and analysis using the BD Accuri C6

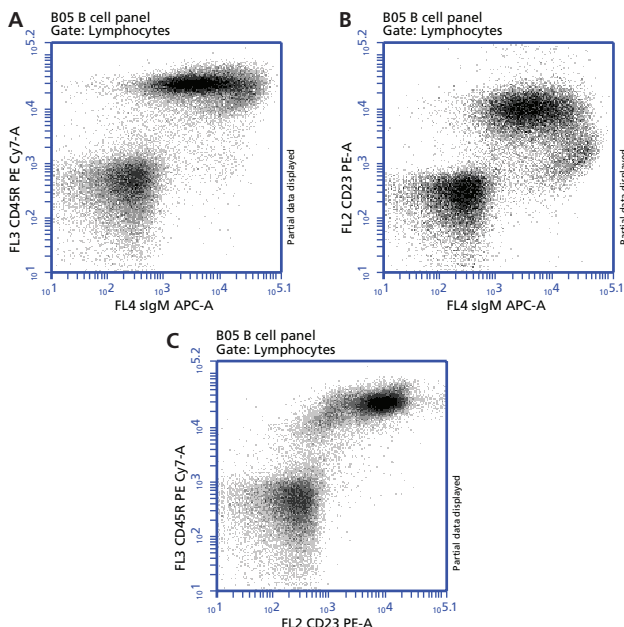


Figure 1. BD Pharmingen Mouse B-Lymphocyte Subset Antibody Cocktail (Cat. No. 558332) analysis on the BD Accuri C6

BALB/c splenocytes were stained with either the Mouse B Lymphocyte Subset Antibody Cocktail (three panels shown) or the Mouse B-Lymphocyte Subset Isotype Control (included in the kit; data not shown) according to the kit procedure. Samples were acquired on a BD Accuri C6 flow cytometer using the kit template and analyzed using BD Accuri C6 software. Scatter plots (not shown) were used to gate on lymphocytes for data analysis. **Results:** Major B-lymphocyte subpopulations can be identified by their levels of expression of CD23 (B, C), CD45R/B220 (A, C), and sIgM (A, B).

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BD mouse B-lymphocyte kits, protocols, and software templates for the BD Accuri™ C6 flow cytometer simplify the identification of mouse B lymphocytes, their subsets, and their activation markers. BD offers two mouse B-lymphocyte kits (for studies involving mouse CD45R/B220, CD23, sIgM, CD19, CD25, and CD69) that include fluorescent antibodies and isotype controls needed for acquisition and analysis. Both panels are compatible with other markers to further characterize B-cell subpopulations. BD Accuri™ C6 software templates matched to each kit include predefined workspaces, markers, regions, gates, and parameter names for quick and easy setup and analysis.

The two kits are listed below. Figures 1 and 2 show data on the BD Accuri C6 using the preconfigured kits and software templates.

The BD Pharmingen™ Mouse B-Lymphocyte Subset Antibody Cocktail (Cat. No. 558332) is a convenient cocktail that enables rapid characterization of major subpopulations of developing and peripheral mouse B lymphocytes based on their expression of CD45R/B220, CD23, and sIgM.

The BD Pharmingen™ Mouse B-Lymphocyte Activation Antibody Cocktail (Cat. No. 558063) enables rapid characterization of activated mouse B cells using the markers CD25 and CD69.

B cells pass through various developmental stages both before and after exposure to antigens. As they mature and differentiate, they give rise to multiple functionally and phenotypically distinct subsets. Major B-cell subsets can be defined by differential expression of cell surface markers including CD45R/B220, CD23 (Fc RII), and surface IgM (sIgM). Using panels of directly conjugated fluorescent antibodies to these markers, multicolor flow cytometry allows researchers to interrogate multiple markers simultaneously on individual cells. This can provide information about the cell lineage and state of differentiation of cell subsets in a particular sample. Relative populations of different B-cell subsets and early and late activation markers such as CD69 and CD25 can provide a useful measurement of immune response to antigens.

In both mouse B-lymphocyte kits, antibodies defining the three key subset or activation markers have been diluted to optimal concentration and pre-mixed to maximize staining performance. The isotype control panels contain equivalent concentrations of fluorochrome- and isotype-matched negative-control immunoglobulins.

Easy to use, simple to maintain, and affordable, the BD Accuri C6 personal flow cytometer is equipped with a blue laser, a red laser, two light scatter detectors, and four fluorescence detectors. A compact design, fixed alignment, and pre-optimized detector settings result in a system that is simple to use. A nonpressurized fluidics system enables kinetic measurements in real time. For walkaway convenience, the optional BD CSampler™ accessory (Cat. No. 653124) offers automated sampling from 24-tube racks or multiwell plates.



BD Mouse B-Lymphocyte Kits and Templates

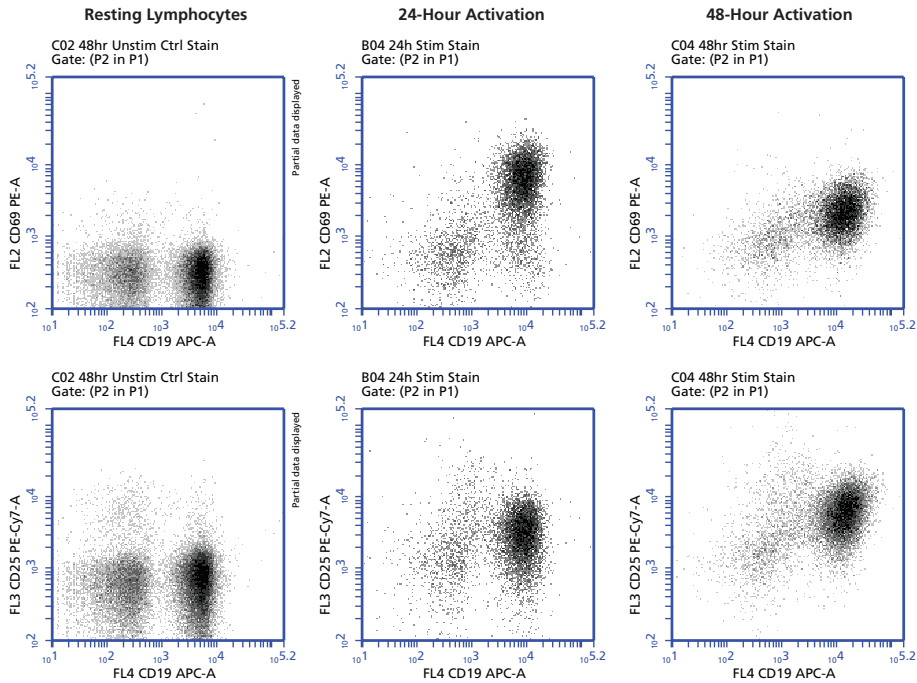


Figure 2. BD Pharmingen Mouse B-Lymphocyte Activation Antibody Cocktail (Cat. No. 558063) analysis on the BD Accuri C6

BALB/c splenocytes were activated for 24 or 48 hours on plate-bound anti-IgM antibody (Jackson ImmunoResearch). The activated splenocytes, along with resting BALB/c splenocytes, were then stained with either the Mouse B-Lymphocyte Activation Antibody Cocktail (six panels shown) or Mouse B-Lymphocyte Activation Isotype Control (included in the kit; data not shown) according to the kit procedure. Samples were acquired on a BD Accuri C6 using the kit template and analyzed using BD Accuri C6 software. Scatter plots (not shown) were used to select either resting lymphocytes (left panels) or activated lymphoblasts (middle and right panels) for data analysis, and gates were used to remove doublets (P2 in P1). **Results:** Density plots show the CD19⁺ B lymphocytes that express the activation antigens CD69 (top plots) and CD25 (bottom plots). As expected, activated B lymphoblasts were more likely than resting lymphocytes to express both activation antigens, with CD69 activation peaking earlier than CD25 activation.

Ordering Information

All kits and their associated software templates are available at bdbiosciences.com/go/templates.

Description	Clone	Quantity	Number of Tests	Cat. No.
BD Pharmingen™ Mouse B-Lymphocyte Subset Antibody Cocktail with Isotype Control, containing:				
Mouse CD45R/B220 PE-Cy7™7	RA3-6B2	20 µL/test	100 tests	558332
Mouse CD23 PE	B3B4			
Mouse sIgM APC	II/41			
Isotype Controls as detailed in technical data sheet		20 µL/test	100 tests	

BD Pharmingen™ Mouse B-Lymphocyte Activation Antibody Cocktail with Isotype Control, containing:				
Mouse CD25 PE-Cy7	PC61	20 µL/test	50 tests	558063
Mouse CD69 PE	H1.2F3			
Mouse CD19 APC	1D3			
Isotype Controls as detailed in technical data sheet		20 µL/test	100 tests	

Related Products

Description	Cat. No.
BD Pharmingen™ Mouse T-Lymphocyte Subset Antibody Cocktail	558431
BD Pharmingen™ Mouse T-Lymphocyte Activation Antibody Cocktail	557908
BD Pharmingen™ Mouse Th17/Treg Phenotyping Kit	560767
BD Pharmingen™ Mouse Th1/Th2/Th17 Phenotyping Kit	560758
BD Accuri™ C6 Flow Cytometer System	653118
BD CSampler™ Automated Sampling System	653124

Class 1 Laser Product.

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23-16693-00



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