FlowX™ Mouse Lineage AlexaFluor® 405 Cocktail

Catalog Number: FLC001V

Size: 25 Tests

PRODUCT DESCRIPTION

This product contains a cocktail of AlexaFluor® 405-conjugated antibodies to mouse lineage markers. The cocktail will bind to cells of hematopoietic lineage, including T cells, B cells, monocytes, granulocytes, and erythrocytes, and can identify hematopoietic cells in samples such as mouse bone marrow. Tissue homogenates from spleen, intestine, or lung can also be stained with the lineage cocktail to "gate out" hematopoietic cells from other populations of interest, such as Innate Lymphoid Cells (ILCs).

MATERIALS PROVIDED & STORAGE

Store the unopened kit at 2-8 °C **in the dark.** Use within 6 months of receipt.

	PART #	DESCRIPTION
Mouse Lineage AlexaFluor® 405 Cocktail	898296	500 µL of an AlexaFluor® 405-conjugated antibody cocktail of the following mouse lineage markers: • Anti-mouse CD3 (Clone 17A2) • Anti-mouse B220 (Clone RA3-6B2) • Anti-mouse CD11b (Clone M1/70) • Anti-mouse GR-1 (Clone RB6-8C5) • Anti-mouse Ter-119 (Clone TER119) • Anti-mouse CD5 (Clone 53-7.3)

INTENDED USE

This product is designed for the flow cytometric identification of mouse hematopoietic lineage cells.

STAINING PROTOCOL

1. Wash mouse splenocytes, bone marrow, or other cells (1 x 10^6 cells per sample) with 2 mL of Flow Cytometry Staining Buffer (R&D Systems, Catalog # FC001) or other BSA-containing buffer, by spinning at 300 x g for 5 minutes, using 5 mL flow cytometry tubes.

2. Fc block cells with blocking IgG (1 µg IgG/10^6 cells) for 10 minutes at 2-8 °C.

3. Add 20 µL of Mouse Lineage AlexaFluor® 405 Cocktail, and other staining antibodies as desired.

4. Incubate the mixture for 30-45 minutes at 2-8 °C in the dark.

5. Wash cells two times with Flow Cytometry Staining Buffer.

6. Resuspend the cells in Flow Cytometry Staining Buffer and run on a flow cytometer.

PRECAUTIONS

This antibody cocktail contains sodium azide which may react with lead and copper plumbing to form explosive metallic azides. Flush with large volumes of water during disposal.

TECHNICAL TIPS

• If desired, an isotype control may be used as a negative control. The isotypes of the clones in the cocktail are Rat IgG_{2A} and Rat IgG_{2B}. If desired, stain a separate tube of cells with 10 uL of each AlexaFluor® 405-conjugated Rat IgG_{2A} and Rat IgG_{2B} (R&D Systems Catalog # IC006V and IC013V).

• Depending on starting sample type and desired cell population, it may be beneficial to start with lineage depletion (MagCellect Mouse Hematopoietic Cell Lineage Depletion, R&D Systems Catalog # MAGM209) to get rid of excess hematopoietic cells.

FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES.
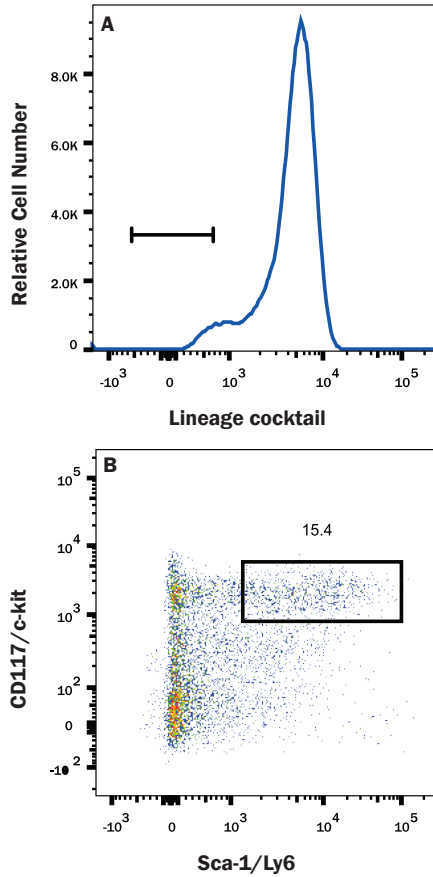
DATA EXAMPLES

Figure 1: Detection of hematopoietic progenitors in mouse bone marrow. Mouse bone marrow cells were stained with Mouse Lineage AlexaFluor® 405 Cocktail, PE-conjugated anti-mouse Sca-1/Ly6 (R&D Systems, Catalog # FAB1226P), and APC-conjugated anti-mouse CD117/c-kit (R&D Systems, Catalog # FAB1356A). Cells in **(B)** were gated on live lymphocytes, then lineage negative cells **(A)**.
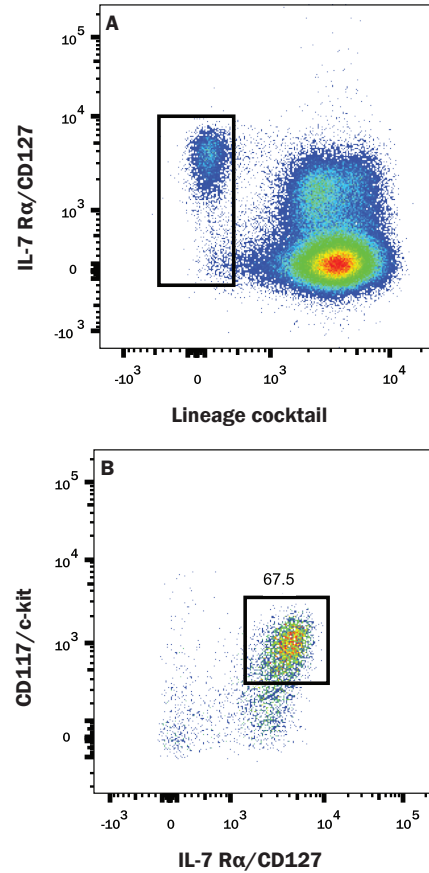

Figure 2: Detection of innate lymphoid cells (ILCs) in mouse Peyer's Patch. Mouse Peyer's Patch cells were stained with Mouse Lineage AlexaFluor® 405 Cocktail, PE-conjugated anti-mouse IL-7 Rα/CD127 (R&D Systems, Catalog # FAB47742P), and APC-conjugated anti-mouse CD117/c-kit (R&D Systems, Catalog # FAB1356A). Cells in **(B)** were gated on live lymphocytes, then lineage negative cells **(A)**.