Technical Data Sheet

PE Rat Anti-Mouse Early B Lineage

Product Information	
Material Number:	558039
Size:	0.1 mg
Concentration:	0.2 mg/ml
Clone:	AA4.1
Immunogen:	Pre-B lymphoma 70Z/3, derived from (C57BL/6 x DBA/2)F1 mouse
Isotype:	Rat (SD) IgG2b, ĸ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing $\leq 0.09\%$ sodium azide.

Description

The AA4.1 antibody reacts with a 130-140-kDa type I transmembrane protein expressed on immature B lymphocytes in the adult bone marrow; and on hematopoietic progenitors and stem cells in adult bone marrow, fetal liver, and embryonic yolk sac. Although staining of splenic immature/transitional B cells has been reported, we find that the antigen density is much lower in the spleen than in the bone marrow. Staining of spleen requires amplification through the use of a second step. The FITC conjugate of mAb AA4.1, while ideal for bone marrow staining, is not effective in the spleen (please see Usage comments below). It has been observed that the staining pattern of mAb 493 (Cat. Nos. 550433 and 550434 for the purified and biotinylated formats, respectively) is similar to that of mAb AA4.1, that both antibodies precipitate molecules of the same molecular weight, and that staining by mAb AA4.1 is not blocked by mAb 493, suggesting that the antibodies recognize separate epitopes of the same Early B Lineage antigen.



The expression of Early B Lineage antigen on developing and peripheral B lymphocytes. BALB/c bone marrow cells (top panels) and splenocytes (bottom panels) were stained with FITC anti-mouse CD45R/B220 mAb RA3-6B2 (Cat. No. 553087/53008) and either PE rat IgG2b, κ isotype control mAb A95-1 (Cat. No. 553989, left panels) or PE mAb AA4.1 (right panels). Viable cells were selected by exclusion of propidium iodide, and flow cytometry was performed on a BD FACSCalibur™ flow cytometry system.

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography. The antibody was conjugated with R-PE under optimum conditions, and unconjugated antibody and free PE were removed. Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

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Application Notes

Application

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ſ	Flow cytometry	Routinely Tested			

Recommended Assay Procedure:

For detection of the Early B Lineage antigen in the spleen, we recommend amplification of the staining signal through the use of biotinylated mAb 493 (Cat. No. 550434), followed by a "bright" second-step reagent, such as Streptavidin-PE (Cat. No. 554061).

Suggested Companion Products

Catalog Number	Name	Size	Clone	
554680	PE Mouse IgG1, κ Isotype Control	0.1 mg	MOPC-21	
553087	FITC Rat Anti-Mouse CD45R/B220	0.1 mg	RA3-6B2	
553989	PE Rat IgG2b, κ Isotype Control	0.1 mg	A95-1	

Product Notices

- 1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- 2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
- 3. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/pharmingen/colors.
- 4. 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.

References

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