

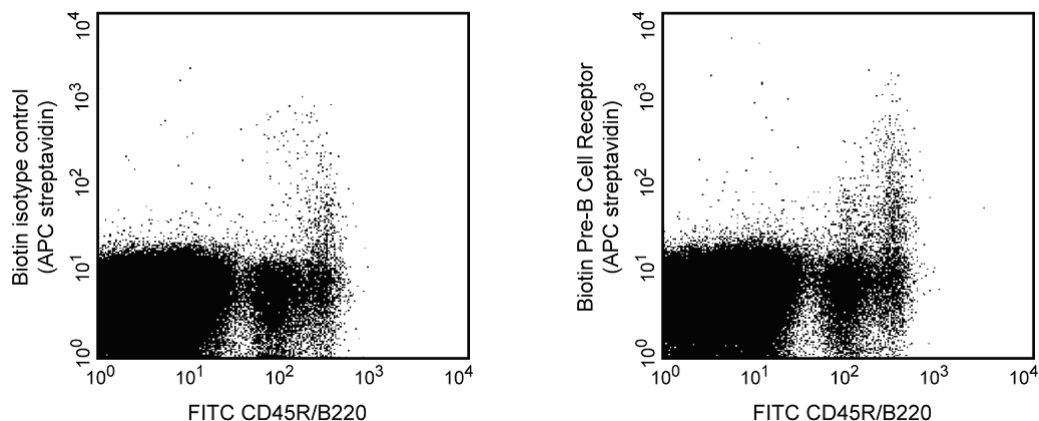
Technical Data Sheet

Biotin Rat Anti-Mouse Pre-B Cell Receptor**Product Information**

Material Number:	551863
Alternate Name:	Pre-BCR
Size:	0.1 mg
Concentration:	0.5 mg/ml
Clone:	SL156
Immunogen:	Purified soluble complex of recombinant μ IgH, λ 5, and VpreB
Isotype:	Rat (LEW) IgG2a, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing $\leq 0.09\%$ sodium azide.

Description

The pre-B cell receptor (pre-BCR) expressed during the early stages of B lymphocyte development is a heterodimer of immunoglobulin heavy chain (IgH) with surrogate light chain, which is an Ig-light-chain-like molecule composed of the non-covalently linked Lambda 5 (λ 5) and VpreB proteins. The pre-BCR is believed to control IgH repertoire selection and proliferation of differentiating B lymphocytes. The SL156 antibody reacts with a conformational epitope of the pre-BCR in transfected X63-Ag8.653 cells but not with surrogate light chain, λ 5, or VpreB in the absence of IgH. It detects pre-BCR on pre-B cell lines, but not on pro-B cell lines or IgM-positive splenocytes. It also detects pre-BCR on the surface of pre-B cells, but not on Ig light chain (IgL)-positive B lymphocytes, from the bone marrow of normal mice. It has been noted that the cell-surface expression of pre-BCR is upregulated after a one-hour incubation of bone-marrow leukocytes in tissue culture medium at 37°C. After immunization, cell-surface pre-BCR is detected on a subset of splenic IgL+ germinal-center B lymphocytes.



Two-color analysis of Pre-B cell receptor (Pre-BCR) expression on bone-marrow pre-B lymphocytes. After immunomagnetic depletion of IgM+ cells using biotinylated anti-mouse IgM[b] mAb AF6-78 (Cat. No. 553519), C57BL/6 bone-marrow leukocytes were incubated for 1 hour in DMEM at 37°C to enhance Pre-BCR expression. Then the leukocytes were stained with either biotinylated rat IgG2a, κ isotype control mAb R35-95 (Cat. No. 553928, left panel) or biotinylated mAb SL156 (right panel) in the presence of Mouse BD Fc Block™ purified anti-mouse CD16/CD32 mAb 2.4G2 (Cat. No. 553141/553142), followed by Streptavidin-APC (Cat. No. 554067) and FITC-conjugated anti-mouse CD45R/B220 mAb (Cat. No. 553087/553088). 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 biotin under optimum conditions, and unreacted biotin was removed.

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

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Application Notes

Application

Flow cytometry	Routinely Tested
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Recommended Assay Procedure:

Because the pre-BCR is expressed at very low levels on bone-marrow-derived early B lineage cells, we recommend the use of Mouse BD Fc Block™ purified anti-mouse CD16/CD32 mAb 2.4G2 (Cat. No. 553141/553142) and amplification of staining by the use of a "bright" second-step reagent, such as Streptavidin-PE (Cat. No. 554061) or Streptavidin-APC (Cat. No. 554067). It is difficult to distinguish the cells expressing pre-BCR among the total bone-marrow population. Therefore, the staining protocol described in the literature. Essential features of this procedure include immunomagnetic depletion of surface IgM+ cells and incubation of the cells at 37°C for 1 hour in tissue culture medium.

Suggested Companion Products

Catalog Number	Name	Size	Clone
553519	Biotin Mouse Anti-Mouse IgM[b]	0.5 mg	AF6-78
553928	Biotin Rat IgG2a κ Isotype Control	0.25 mg	R35-95
554067	APC Streptavidin	0.1 mg	(none)
553141	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.1 mg	2.4G2
553087	FITC Rat Anti-Mouse CD45R/B220	0.1 mg	RA3-6B2
554061	PE Streptavidin	0.5 mg	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to wwwbdbiosciences.com/pharming/en/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.

References

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