Technical Data Sheet

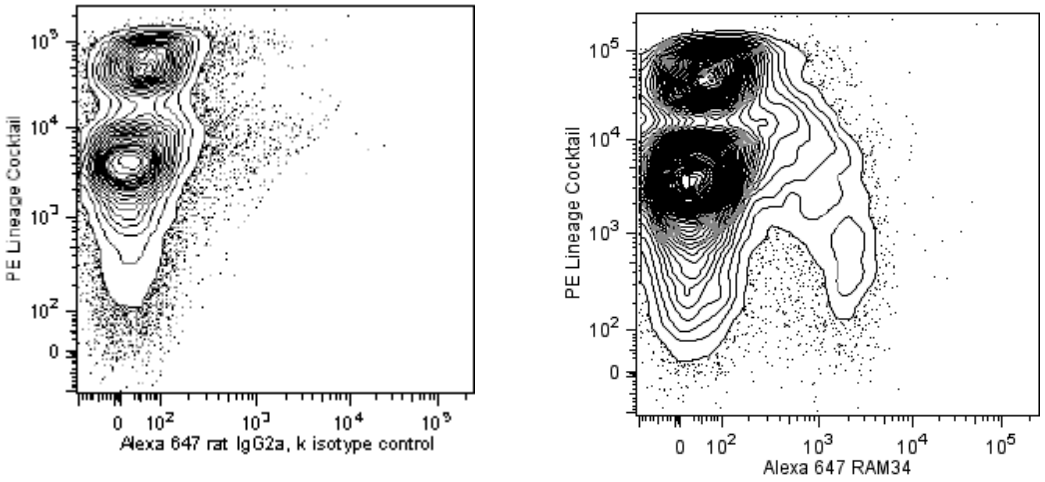
Alexa Fluor® 647 Rat anti-Mouse CD34

Product Information

Material Number:	**560230**
Size:	0.1 mg
Concentration:	0.2 mg/ml
Clone:	RAM34
Immunogen:	Recombinant Mouse CD34
Isotype:	Rat IgG2a, κ
Reactivity:	QC Testing: mouse
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The RAM34 antibody reacts with the CD34 glycoprotein on the surface of three independently derived mouse CD34-transfected cell lines. RAM34 antibody also reacts with the mouse cell lines PA6, 416B, Swiss 3T6, NIH, 3T3, DA1, and M1, all of which are positive for expression of mouse CD34 mRNA. Cell lines shown to be negative for CD34 transcript, including WEHI-3B, EL4, 18.8, and CMT64/61, are also negative for surface expression of CD34 as determined by RAM34 staining. Normal thymocytes and splenocytes are negative for CD34 expression. In the bone marrow, 7-10% of cells are stained with RAM34 mAb, including most of the Ly-6A/E (Sca-1)+ CD90 (Thy-1)low Lineage Marker- hematopoietic stem cell-enriched subpopulation and myeloerythroid progenitors. CD34 is also expressed on a small percentage of fetal liver cells, including NK-cell progenitors. CD34 has been reported to be expressed on the endothelium of capillaries and, in this form, to function as a ligand for L-selectin. Consistent with this observation, RAM34 antibody stains endothelial cells in spleen, thymus, and postcapillary HEVs in the lymph nodes. It is reported that RAM34 antibody can be used to select CD34+ CD117 (c-Kit)+ Ly-6A/E (Sca-1)+ Lineage Marker- bone marrow-derived hematopoietic stem cells, capable of short-term multi-lineage reconstitution of lethally irradiated mice; while the CD34- CD117+ Sca-1+ Lineage Marker- population contains self-renewing hematopoietic stem cells. Similarly, the bone marrow population with high dye-efflux capacity and which is highly enriched for long-term reconstituting hematopoietic stem cells is CD34- CD117 (c-Kit)+ Ly-6A/E (Sca-1)+ Lineage Marker-.



Identification of CD34+ and CD34- subpopulations of hematopoietic progenitors. *BALB/c bone marrow cells were treated with Mouse BD Fc Block™ purified anti-CD16/CD32 mAb 2.4G2 (Cat. No. 553141/553142) and stained with either Alexa Fluor® 647-conjugated anti-mouse CD34 (Clone RAM34, right panel) or a Alexa Fluor® 647-conjugated rat IgG2a, κ isotype control (clone R35-95, Cat. No. 557690, left panel), followed by staining with a PE Lineage Cocktail consisting of CD3e, CD11b, CD45R/B220, Gr-1, Ly6G, Ly6C mAb to identify major lineage committed cells. The contour plots were derived from the gated events based on light scattering characteristics of leukocytes and fluorescence characteristics of the CD34 and Lineage markers . Flow cytometry was performed on a BD FACSCanto™ System.*

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Preparation and Storage

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

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

The antibody was conjugated to Alexa Fluor® 647 under optimum conditions, and unreacted Alexa Fluor® 647 was removed.

Application Notes

Application

Flow cytometry	Routinely Tested

Suggested Companion Products

Catalog Number	Name	Size	Clone
557690	Alexa Fluor® 647 Rat IgG2a, κ Isotype Control	0.1 mg	R35-95
553142	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.5 mg	2.4G2
553064	PE Hamster Anti-Mouse CD3e	0.2 mg	145-2C11
553311	PE Rat Anti-Mouse CD11b	0.2 mg	M1/70
553090	PE Rat Anti-Mouse CD45R/B220	0.2 mg	RA3-6B2
553128	PE Rat Anti-Mouse Ly-6G and Ly-6C	0.1 mg	RB6-8C5
553673	PE Rat Anti-Mouse TER-119/Erythroid Cells	0.2 mg	TER-119

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Alexa Fluor® 647 fluorochrome emission is collected at the same instrument settings as for allophycocyanin (APC).
3. The Alexa Fluor®, Pacific Blue™, and Cascade Blue® dye antibody conjugates in this product are sold under license from Molecular Probes, Inc. for research use only, excluding use in combination with microarrays, or as analyte specific reagents. The Alexa Fluor® dyes (except for Alexa Fluor® 430), Pacific Blue™ dye, and Cascade Blue® dye are covered by pending and issued patents.
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.
5. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
6. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
7. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.

References

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