Technical Data Sheet

Purified Rat Anti-Mouse CD34

Product Information

553731 **Material Number:** 0.5 mg Size: **Concentration:** 0.5 mg/ml Clone: RAM34

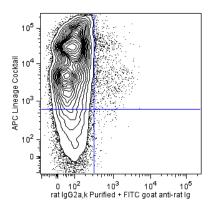
Recombinant Mouse CD34 Immunogen:

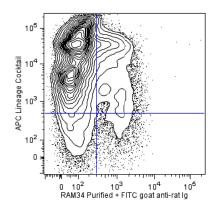
Rat IgG2a, κ Isotype: QC Testing: Mouse Reactivity:

Aqueous buffered solution containing ≤0.09% sodium azide. Storage Buffer:

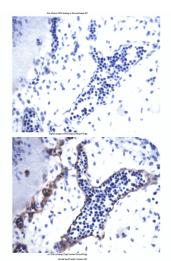
Description

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 mouse bone marrow cells were incubated with either purified monoclonal antibody RAM34 (right panel) or purified rat IgG2a, κ isotype control (clone R35-95, Cat. No. 553927, left panel), followed by second step staining with FITC polyclonal goat-anti-rat Ig (554016). Cells were washed and blocked with mouse Fc Block purified anti-CD16/CD32mAb (553141/553142) and normal rat serum, followed by staining with APC Lineage Cocktail (558074) 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.



Immunohistochemistry application of CD34 antibody. Immunohistochemistry staining of anti-mouse CD34 antibody clone RAM34 on formalin fixed paraffin embedded mouse embryonic (day 11) tissue section with citrate buffer for antigen retrieval. The antibody stains on endothelial cells of vessels and blood stem cells in vessels (lower panel) with isotype control (upper panel). The secondary antibody was used biotin polyclonal goat anti-rat IgG (559286) and followed by Streptavidin -HRP (550946), and DAB detection kit (550880). Original amplification 40X

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Preparation and Storage

Store undiluted at 4°C.

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

Application Notes

Application

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Flow cytometry	Routinely Tested	
Immunohistochemistry-zinc-fixed	Tested During Development	
Immunohistochemistry-frozen	Tested During Development	
Immunohistochemistry-formalin (antigen retrieval required)	Tested During Development	

Suggested Companion Products

Catalog Number	Name	Size	Clone	
553927	Purified Rat IgG2a, κ Isotype Control	0.5 mg	R35-95	
554016	FITC Goat Anti-Rat Ig	0.5 mg	Polyclonal	
553142	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.5 mg	2.4G2	
558074	APC Mouse Lineage Antibody Cocktail, with Isotype Control	100 tests	(none)	

Product Notices

- 1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- 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.
- 3. Sodium azide is a reversible inhibitor of oxidative metabolism; therefore, antibody preparations containing this preservative agent must not be used in cell cultures nor injected into animals. Sodium azide may be removed by washing stained cells or plate-bound antibody or dialyzing soluble antibody in sodium azide-free buffer. Since endotoxin may also affect the results of functional studies, we recommend the NA/LE (No Azide/Low Endotoxin) antibody format, if available, for in vitro and in vivo use.
- 4. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

Akashi K, Traver D, Miyamoto T, Weissman IL. A clonogenic common myeloid progenitor that gives rise to all myeloid lineages. *Nature*. 2000; 404(6774):193-197. (Biology)

Brown J, Greaves MF, Molgaard HV. The gene encoding the stem cell antigen, CD34, is conserved in mouse and expressed in haemopoietic progenitor cell lines, brain, and embryonic fibroblasts. *Int Immunol.* 1991; 3(2):175-184.(Biology)

Goodell MA, Rosenzweig M, Kim H, et al. Dye efflux studies suggest that hematopoietic stem cells expressing low or undetectable levels of CD34 antigen exist in multiple species. *Nat Med.* 1997; 3(12):1337-1345.(Biology)

Lu J, Patrene KD, Herberman RB, Boggs SS. Expression of murine CD34 by fetal liver NK cell progenitors. *Exp Hematol.* 1999; 27(2):272-281.(Biology) Morel F, Szilvassy SJ, Travis M, Chen B, Galy A. Primitive hematopoietic cells in murine bone marrow express the CD34 antigen. *Blood.* 1996; 88(10):3774-3784. (Clone-specific)

Osawa M, Hanada K, Hamada H, Nakauchi H. Long-term lymphohematopoietic reconstitution by a single CD34-low/negative hematopoietic stem cell. *Science*. 1996; 273(5272):242-245.(Immunogen)

Spangrude GJ, Heimfeld S, Weissman IL. Purification and characterization of mouse hematopoietic stem cells. *Science*. 1988; 241(4861):58-62.(Biology) Suda J, Sudo T, Ito M, Ohno N, Yamaguchi Y, Suda T. Two types of murine CD34 mRNA generated by alternative splicing. *Blood*. 1992; 79(9):2288-2295. (Biology)

Suzuki A, Andrew DP, Gonzalo JA, et al. CD34-deficient mice have reduced eosinophil accumulation after allergen exposure and show a novel crossreactive 90-kD protein. *Blood.* 1996; 87(9):3550-3562.(Biology)

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