

Technical Data Sheet

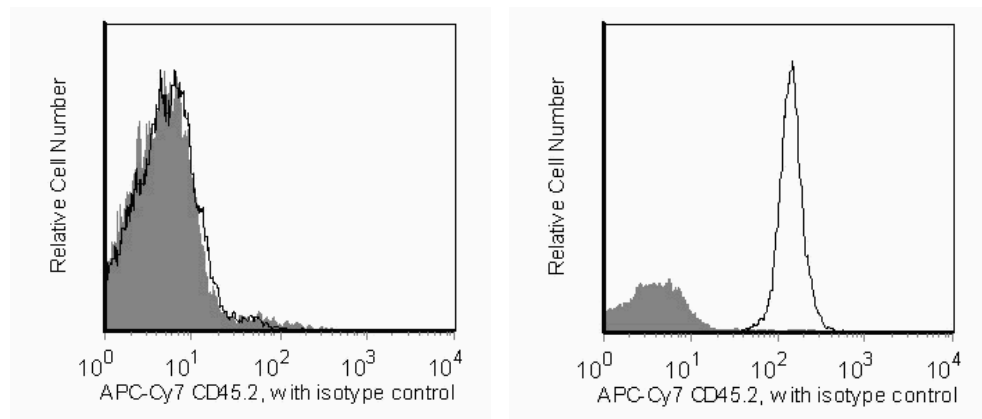
APC-Cy™7 Mouse Anti-Mouse CD45.2

Product Information

Material Number:	560694
Alternate Name:	Ly-5.2; T200; LCA; Leukocyte common antigen; Ptpcr
Size:	50 µg
Concentration:	0.2 mg/ml
Clone:	104
Immunogen:	B10.S mouse thymocytes and splenocytes
Isotype:	Mouse (SJL) IgG2a, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing protein stabilizer and ≤0.09% sodium azide.

Description

The 104 clone has been reported to react with CD45 (Leukocyte Common Antigen) on all leukocytes of most mouse strains (eg, A, AKR, BALB/c, CBA/Ca, CBA/J, C3H/He, C57BL, C57BR, C57L, C58, DBA/1, DBA/2, NZB, SWR, 129). This alloantigen was originally named Ly-5.1, and this was the designation at the time that the antibody was characterized. The designation was later changed from Ly-5.1 to Ly-5.2 to conform with the convention that the .2 alloantigen designations be assigned to the C57BL/6 strain. mAb 104 has been reported not to react with leukocytes of the mouse strains expressing the CD45.1 alloantigen (eg, RIII, SJL/J, STS/A, and DA). CD45 is a member of the Protein Tyrosine Phosphatase (PTP) family: its intracellular (COOH-terminal) region contains two PTP catalytic domains, and the extracellular region is highly variable due to alternative splicing of exons 4, 5, and 6 (designated A, B, and C, respectively), plus differing levels of glycosylation. The CD45 isoforms detected in the mouse are cell type-, maturation-, and activation state-specific. The CD45 isoforms play complex roles in T-cell and B-cell antigen receptor signal transduction. The 104 antibody has been reported to inhibit some responses of B cells, from mice expressing the CD45.2 alloantigen, to certain antigens and LPS. In addition, reduction of serum IgG levels and amelioration of autoimmune renal pathology were reported in mAb 104-treated systemic lupus erythematosus-prone mice.



Flow cytometric analysis of CD45.2 on mouse splenocytes. Splenocytes from SJL/J mice (left panel) or C57BL/6 mice (right panel) were stained either with a APC-Cy™7 Mouse IgG2a, κ isotype control (shaded) or with the APC-Cy™7 Mouse Anti-Mouse CD45.2 antibody (unshaded). Histograms were derived from gated events based on light scattering characteristics for lymphocytes. Flow cytometry was performed on a BD™ LSR II flow cytometry system.

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 with APC-Cy7 under optimum conditions, and unconjugated antibody and free APC-Cy7 were removed.

Application Notes

Application

Flow cytometry	Routinely Tested
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Suggested Companion Products

Catalog Number	Name	Size	Clone
557751	APC-Cy™7 Mouse IgG2a, κ Isotype Control	100 tests	G155-178
553141	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.1 mg	2.4G2

Product Notices

- Since applications vary, each investigator should titrate the reagent to obtain optimal results.

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2. An isotype control should be used at the same concentration as the antibody of interest.
3. Warning: Some APC-Cy7 and PE-Cy7 conjugates show changes in their emission spectrum with prolonged exposure to formaldehyde. If you are unable to analyze fixed samples within four hours, we recommend that you use BD™ Stabilizing Fixative (Cat. No. 338036).
4. Please observe the following precautions: Absorption of visible light can significantly alter the energy transfer occurring in any tandem fluorochrome conjugate; therefore, we recommend that special precautions be taken (such as wrapping vials, tubes, or racks in aluminum foil) to prevent exposure of conjugated reagents, including cells stained with those reagents, to room illumination.
5. This conjugated product is sold under license to the following patent: US Patent No. 5,714,386.
6. Cy is a trademark of Amersham Biosciences Limited. This conjugated product is sold under license to the following patents: US Patent Nos. 5,486,616; 5,569,587; 5,569,766; 5,627,027.
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8. APC-Cy7 tandem fluorochrome emission is collected in a detector for fluorescence wavelengths of 750 nm and higher.
9. APC-Cy7 is a tandem fluorochrome composed of Allophycocyanin (APC), which is excited by laser lines between 595 and 647 nm and serves as an energy donor, coupled to the cyanine dye Cy7™, which acts as an energy acceptor and fluoresces at 780 nm. BD Biosciences Pharmingen has maximized the fluorochrome energy transfer in APC-Cy7, thus maximizing its fluorescence emission intensity, minimizing residual emission from APC, and minimizing required electronic compensation in multilaser-laser flow cytometry systems. Note: Although every effort is made to minimize the lot-to-lot variation in residual emission from APC, it is strongly recommended that every lot be tested for differences in the amount of compensation required and that individual compensation controls are run for each APC-Cy7 conjugate.
10. 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.
11. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
12. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

- Greimers R, Trebak M, Moutschen M, Jacobs N, Boniver J. Improved four-color flow cytometry method using fluo-3 and triple immunofluorescence for analysis of intracellular calcium ion ([Ca²⁺]_i) fluxes among mouse lymph node B- and T-lymphocyte subsets. *Cytometry*. 1996; 23(3):205-217. (Biology)
- Johnson P, Greenbaum L, Bottomly K, Trowbridge IS. Identification of the alternatively spliced exons of murine CD45 (T200) required for reactivity with B220 and other T200-restricted antibodies. *J Exp Med*. 1989; 169(3):1179-1184. (Biology)
- Morse HC 3rd, Shen FW, Hammerling U. Genetic nomenclature for loci controlling mouse lymphocyte antigens. *Immunogenetics*. 1987; 25(2):71-78. (Biology)
- Ogimoto M, Mizuno K, Tate G, et al. Regulation of lipopolysaccharide- and IL-4-induced immunoglobulin heavy chain gene activation: differential roles for CD45 and Lyb-2. *Int Immunol*. 1992; 4(6):651-659. (Biology)
- Shapiro HM. *Practical Flow Cytometry, 3rd Edition*. New York: Wiley-Liss, Inc; 1995:280-281. (Methodology: Flow cytometry)
- Shen FW. Monoclonal antibodies to mouse lymphocyte differentiation alloantigens. In: Hammerling GJ, Hammerling U, Kearney JF, ed. *Monoclonal Antibodies and T-cell Hybridomas; Perspectives and Technical Advances*. 1981:25-31. (Immunogen)
- Shen FW, Tung JS, Boyse EA. Further definition of the Ly-5 system. *Immunogenetics*. 1986; 24(3):146-149. (Biology)
- Suzuki K, Oida T, Hamada H, et al. Gut cryptopatches: direct evidence of extrathymic anatomical sites for intestinal T lymphopoiesis. *Immunity*. 2000; 13(5):691-702. (Biology)
- Yakura H, Ashida T, Kawabata I, Katagiri M. Alleviation of autoimmunity in BXSB mice by monoclonal alloantibody to Ly-5 (CD45). *Eur J Immunol*. 1989; 19(8):1505-1508. (Biology)
- Yakura H, Kawabata I, Ashida T, Katagiri M. Differential regulation by Ly-5 and Lyb-2 of IgG production induced by lipopolysaccharide and B cell stimulatory factor-1 (IL-4). *J Immunol*. 1988; 141(3):875-880. (Biology)
- Yakura H, Kawabata I, Shen FW, Katagiri M. Selective inhibition of lipopolysaccharide-induced polyclonal IgG response by monoclonal Ly-5 antibody. *J Immunol*. 1986; 136(8):2729-2733. (Biology)
- Yakura H, Shen FW, Bourcet E, Boyse EA. On the function of Ly-5 in the regulation of antigen-driven B cell differentiation. Comparison and contrast with Lyb-2. *J Exp Med*. 1983; 157(4):1077-1088. (Biology)