

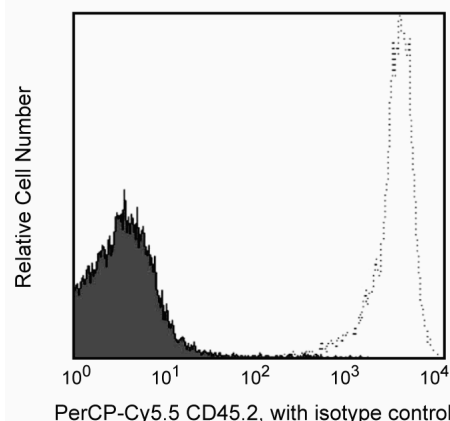
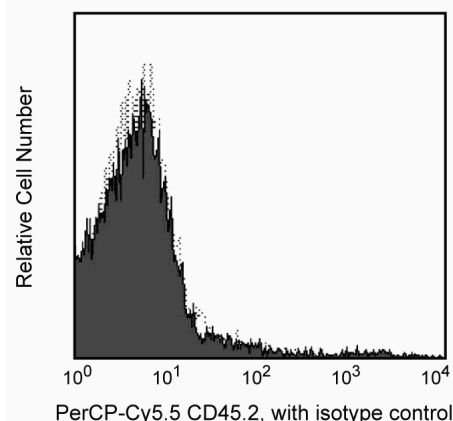
Technical Data Sheet

PerCP-Cy™ 5.5 Mouse Anti-Mouse CD45.2**Product Information**

Material Number:	561096
Alternate Name:	Ly-5.2; T200; LCA; Leukocyte common antigen; Ptprc
Size:	25 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 ≤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.



Differential expression of CD45.2 in SJL and BALB/c spleen. Splenocytes from SJL (left panel) and BALB/c AnN (right panel) mice were stained with either PerCP-Cy™ 5.5-conjugated mouse IgG2a, κ isotype control clone G155-178 (Cat. No. 550927, solid histograms) or PerCP-Cy™ 5.5-conjugated anti-mouse CD45.2 clone 104 (open histograms). Flow cytometry was performed on a BD FACSCalibur™ 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 PerCP-Cy5.5 under optimum conditions, and unconjugated antibody and free PerCP-Cy5.5 were removed. Storage of PerCP-Cy5.5 conjugates in unoptimized diluent is not recommended and may result in loss of signal intensity.

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Application Notes

Application

Flow cytometry

Routinely Tested

Suggested Companion Products

Catalog Number	Name	Size	Clone
550927	PerCP-Cy TM 5.5 Mouse IgG2a, κ Isotype Control	0.1 mg	G155-178

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharming/en/protocols for technical protocols.
3. This product is subject to proprietary rights of Amersham Biosciences Corp. and Carnegie Mellon University and made and sold under license from Amersham Biosciences Corp. This product is licensed for sale only for research. It is not licensed for any other use. If you require a commercial license to use this product and do not have one return this material, unopened to BD Biosciences, 10975 Torreyana Rd, San Diego, CA 92121 and any money paid for the material will be refunded.
4. PerCP-Cy5.5 is optimized for use with a single argon ion laser emitting 488-nm light. Because of the broad absorption spectrum of the tandem fluorochrome, extra care must be taken when using dual-laser cytometers, which may directly excite both PerCP and Cy5.5TM. We recommend the use of cross-beam compensation during data acquisition or software compensation during data analysis.
5. PerCP-Cy5.5-labelled antibodies can be used with FITC- and R-PE-labelled reagents in single-laser flow cytometers with no significant spectral overlap of PerCP-Cy5.5, FITC, and R-PE fluorescence.
6. PerCP is a photosynthetic accessory pigment from Glenodinium species of dinoflagellates, which is excited by the 488-nm light of an Argon ion laser and fluoresces at 675 nm. Therefore, PerCP-labelled antibodies can be used with FITC- and R-PE-labelled reagents in most single-laser flow cytometers with no significant spectral overlap of PerCP fluorescence with that of FITC or R-PE. PerCP has been reported to undergo significant photobleaching, the magnitude of which increases as laser power is increased or beam focus is narrowed. For third-color flow-cytometric analysis using ≥ 25 -mW laser power, we recommend PE-Cy5-, PE-Cy7-, or PerCP-Cy5.5-conjugated reagents.
7. 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.
8. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
9. 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.
10. 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.
11. This PerCP-conjugated product is sold under license to the following patent: US Patent No. 4,876,190.

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⁺]) fluxes among mouse lymph node B- and T-lymphocyte subsets. *Cytometry*. 1996; 23(3):205-217. (Methodology: Flow cytometry)

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)