Technical Data Sheet

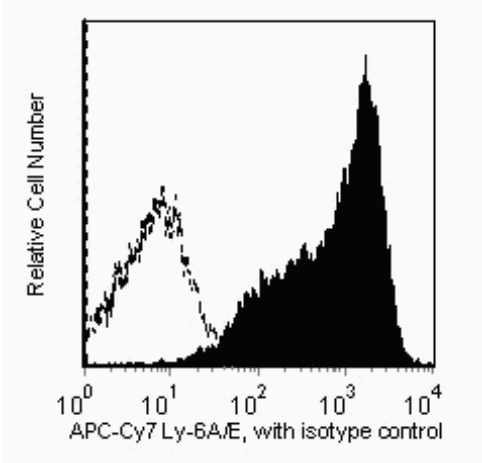
APC-Cy™7 Rat Anti-Mouse Ly-6A/E

Product Information

Material Number:	**560654**
Alternate Name:	Ly-6A/E; Lymphocyte antigen 6A-2/6E-1; Ly-6A.2/Ly-6E.1; Sca-1; TAP
Size:	50 µg
Concentration:	0.2 mg/ml
Clone:	D7
Immunogen:	IL-2-dependent mouse T-cell line CTL-L
Isotype:	Rat (LEW) IgG2a, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing protein stabilizer and ≤0.09% sodium azide.

Description

The D7 antibody reacts with Ly-6A.2 and Ly-6E.1, which are allelic members of the Ly-6 multigene family. Sca-1 (Ly6A/E), a phosphatidylinositol-anchored protein of about 18 kDa, is expressed on the multipotent hematopoietic stem cells (HSC) in the bone marrow of mice with both Ly-6 haplotypes. In mice expressing the Ly-6.2 haplotype (e.g., AKR, C57BL, C57BR, C57L, C58, DBA/2, PL, SJL, SWR, 129), Ly-6A/E is also expressed on distinct subpopulations of bone marrow and peripheral B lymphocytes and thymic and peripheral T lymphocytes. Strains with the Ly-6.1 haplotype (e.g., A, BALB/c, CBA, C3H/He, DBA/1, NZB) have few Ly-6A/E+ resting peripheral lymphocytes, whereas activation of lymphocytes from mice of both Ly-6 haplotypes leads to strong expression of the Sca-1 antigen. Studies with the D7 antibody have demonstrated that Ly-6A/E may be involved in the regulation of B and T lymphocyte responses, and it appears to be required for T-cell receptor-mediated T-cell activation. Purified E13-161.7 mAb (anti-Ly-6A/E, Cat. No. 553333) can block binding of FITC-conjugated D7 antibody (Cat. No. 557405) to mouse splenocytes, but purified mAb D7 is unable to block binding of FITC-conjugated E13-161.7 antibody (Cat. No. 553335). Anti-Ly-6A/E (Sca-1) mAb may be used in combination with the Mouse Lineage Panel (Cat. No. 559971) to identify HSC.



Flow cytometric analysis of Ly-6A/E on stimulated mouse splenocytes. *BALB/c splenocytes were stimulated with 2.5-5.0 µg/mL concanavilin A (Sigma-Aldrich cat. no. C2010) for 48 hours and were subsequently stained either with a APC-Cy™7 Rat IgG2a, κ isotype control (unshaded) or with the APC-Cy™7 Rat Anti-Mouse Ly-6A/E antibody (shaded). Histograms were derived from gated events based on light scattering characteristics for splenocytes. 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

Suggested Companion Products

Catalog Number	Name	Size	Clone
552770	APC-Cy™7 Rat IgG2a, κ Isotype Contol	0.1 mg	R35-95

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Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
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.
7. 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.
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.

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