

Technical Data Sheet

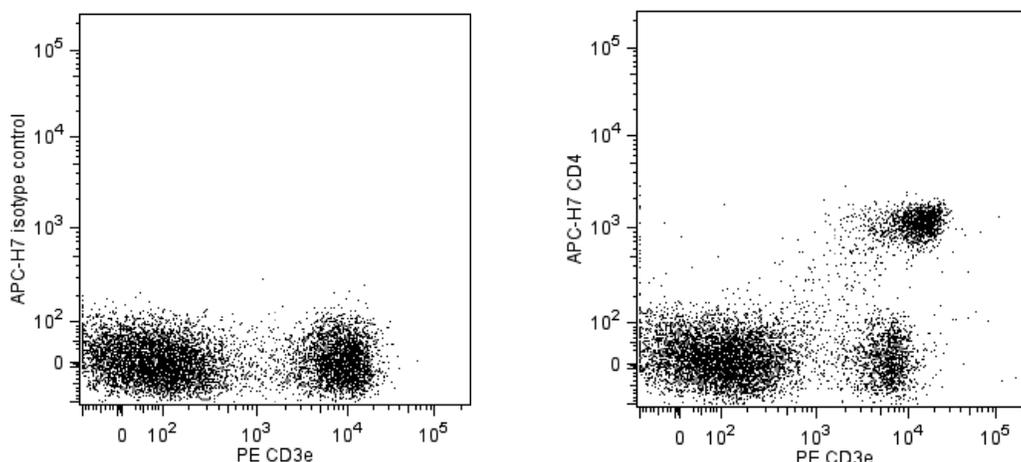
APC-H7 Rat anti-Mouse CD4

Product Information

Material Number:	560246
Alternate Name:	Cd4; CD4 antigen; L3T4; Ly-4; T-cell surface antigen T4/Leu-3
Size:	25 µg
Concentration:	0.2 mg/ml
Clone:	GK1.5
Immunogen:	Mouse CTL clone V4
Isotype:	Rat (LEW) IgG2b, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing protein stabilizer and ≤0.09% sodium azide.

Description

The GK1.5 monoclonal antibody specifically binds to the mouse CD4 (L3T4) differentiation antigen. CD4 is expressed on most thymocytes, a subpopulation of mature T lymphocytes (i.e., MHC class II-restricted T cells, including most T helper cells), and a subset of NK-T cells. In addition, CD4 has also been reported to be detectable on pluripotent hematopoietic stem cells, bone marrow myeloid and B-lymphocyte precursors, intrathymic lymphoid precursors, and a subset of splenic dendritic cells. CD4 has also been reported to be expressed on the plasma membrane of mouse egg cells and is involved in adhesion of the egg to MHC class II-bearing sperm. CD4 is an antigen coreceptor on the T-cell surface which interacts with MHC class II molecules on antigen-presenting cells. It participates in T-cell activation through its association with the T-cell receptor complex and protein tyrosine kinase lck. The GK1.5 antibody reportedly blocks binding of the RM4-5 and H129.19, but not RM4-4 mouse CD4-specific antibodies.



Two-color flow cytometric analysis of CD4 expression on T lymphocytes. BALB/c splenocytes were simultaneously stained with PE Hamster Anti-Mouse CD3e (Cat. No. 553063/553064), and APC-H7 Rat anti-Mouse CD4 (Cat. No. 560246; right panel) or APC-H7 Rat IgG2b, κ Isotype Control (Cat. No. 560200, left panel). Fluorescence dot plot was derived from gated events with the forward and side light-scattering characteristics of viable splenocytes. Flow cytometry was performed on a BD™ LSR II.

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-H7 under optimum conditions, and unconjugated antibody and APC-H7 were removed.

Application Notes

Application

Flow cytometry	Routinely Tested
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Suggested Companion Products

Catalog Number	Name	Size	Clone
560200	APC-H7 Rat IgG2b, κ Isotype Control	0.1 mg	A95-1
553063	PE Hamster Anti-Mouse CD3e	0.1 mg	145-2C11
554656	Stain Buffer (FBS)	500 mL	(none)
554657	Stain Buffer (BSA)	500 mL	(none)
553064	PE Hamster Anti-Mouse CD3e	0.2 mg	145-2C11
560181	APC-H7 Rat anti-Mouse CD4	0.1 mg	GK1.5

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. 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.
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. BD APC-H7 is a tandem conjugate and an analog of APC-Cy7 with the same spectral properties. It has decreased intensity but it is engineered for greater stability and less spillover in the APC channel and consequently offers better performance than APC-Cy7. It has an absorption maximum of approximately 650 nm. When excited by light from a red laser, the APC fluorochrome can transfer energy to the cyanine dye, which then emits at a longer wavelength. The resulting fluorescent emission maximum is approximately 767 nm. BD recommends that a 750-nm longpass filter be used along with a red-sensitive detector such as the Hamamatsu R3896 PMT. As with APC-Cy7 special filters are required when using APC-H7 in conjunction with APC.
Note: Although our APC-H7 products demonstrate higher lot-to-lot consistency than other APC tandem conjugate products, and 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-H7 conjugate.
6. Although BD APC-H7 is engineered to minimize spillover to the APC channel and is more stable and less affected by light, temperature, and formaldehyde-based fixatives, compared to other APC-cyanine tandem dyes, it is still good practice to minimize as much as possible, any light, temperature and fixative exposure when working with all fluorescent conjugates.
7. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
8. Please refer to www.bdbiosciences.com/pharming/protocols for technical protocols.

References

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