

Technical Data Sheet

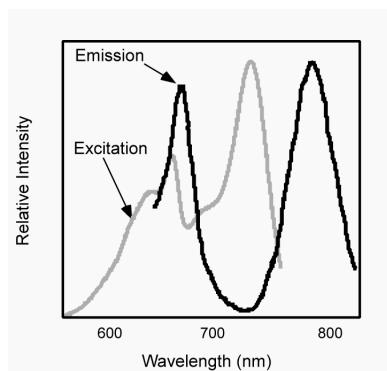
Streptavidin APC-Cy7™7

Product Information

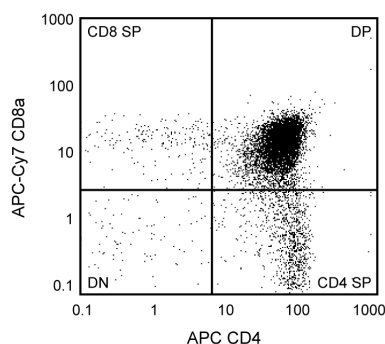
Material Number:	554063
Size:	0.1 mg
Concentration:	0.2 mg/ml
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

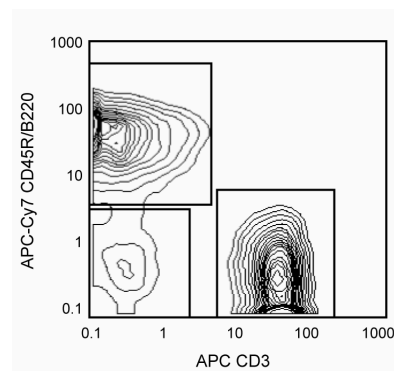
This reagent is routinely tested by flow cytometric analysis. Other applications were tested at BD Biosciences Pharmingen during reagent development only or reported in the literature.



APC-Cy7 spectra. The absorption spectrum of SAV-APC-Cy7 is presented with the corresponding emission spectrum, at the excitation wavelength of 635 nm.



Sample staining on thymocytes (left panel) and splenocytes (right panel). BALB/c mouse thymocytes were stained with APC-conjugated anti-mouse CD4 (clone RM4-5, Cat. No. 553051) and biotinylated anti-mouse CD8a (clone 53-6.7, Cat. No. 553028/553029) (left panel). BALB/c mouse splenocytes were stained with APC-conjugated anti-mouse CD3e (clone 145-2C11, Cat. No. 553066) and biotinylated anti-mouse CD45R/B220 (clone RA3-6B2, Cat. No. 553085/553086) (right panel). The biotin conjugates were revealed with SAV-APC-Cy7. Experiments were performed on a dual-laser FACSVantage™ flow cytometry system equipped with a HeNe laser. APC-Cy7 fluorescence was collected in the FL4 PMT channel, and APC fluorescence was collected in the FL5 PMT channel.



Preparation and Storage

The antibody was conjugated with APC-Cy7 under optimum conditions, and unconjugated antibody and free APC-Cy7 were removed. The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography. Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

Application Notes

Application

Flow cytometry	Routinely Tested
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Recommended Assay Procedure:

Sav-APC-Cy7 is a useful second-step reagent for the indirect immunofluorescent staining of cells in combination with biotinylated primary antibodies for flow cytometric analysis. SAV-APC-Cy7/biotin conjugates can be used with APC-conjugated reagents to provide two independent staining parameters from a HeNe laser. When choosing reagents for a multicolor staining protocol, we recommend that the APC-Cy7 fluorochrome be reserved for detection of high-density antigens to assure adequate discrimination of antigen expression.

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.

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4. 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.
5. 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.
6. APC-Cy7 tandem fluorochrome emission is collected in a detector for fluorescence wavelengths of 750 nm and higher.
7. 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.
8. 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).
9. 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.
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.

References

- Beavis AJ, Pennline KJ. Allo-7: a new fluorescent tandem dye for use in flow cytometry. *Cytometry*. 1996; 24(4):390-395. (Biology)
- Roederer M, Kantor AB, Parks DR, Herzenberg LA. Cy7PE and Cy7APC: bright new probes for immunofluorescence. *Cytometry*. 1996; 24(3):191-197. (Biology)