

Technical Data Sheet

PerCP-Cy™ 5.5 Streptavidin

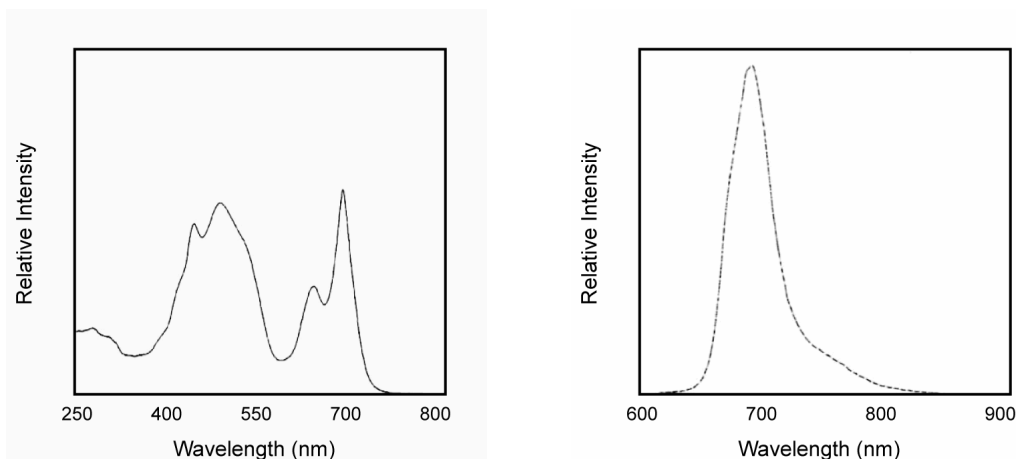
Product Information

Material Number:	551419
Size:	0.1 mg
Concentration:	0.2 mg/ml
Storage Buffer:	Aqueous buffered solution containing protein stabilizer and ≤0.09% sodium azide.

Description

PerCP-Cy5.5 is a tandem fluorochrome composed of peridinin chlorophyll protein (PerCP), which is excited by the 488-nm line of an Argon ion laser and serves as the energy donor, coupled to the cyanine dye Cy™5.5, which acts as the energy acceptor and fluoresces at 695 nm.

SAv-PerCP-Cy5.5 is a useful second-step reagent for the indirect immunofluorescent staining of cells in combination with biotinylated primary antibodies for flow cytometric analysis. PerCP-Cy5.5 tandem fluorochrome emission is collected in the Fluorescence-3 (FL3) channel of BD FACScan™ and BD FACSCalibur™ flow cytometry systems.



PerCP-Cy5.5 spectra. The absorption spectrum of Streptavidin-PerCP-Cy5.5 is presented in the left panel. The corresponding emission spectrum, at the excitation wavelength of 488 nm, appears in the right panel.

Preparation and Storage

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

Streptavidin was conjugated with dye under optimum conditions, and unconjugated Streptavidin and free dye were removed.

Application Notes

Application

Flow cytometry	Routinely Tested
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Recommended Assay Procedure:

PerCP has been reported to undergo significant photobleaching, the magnitude of which increases as laser power is increased or beam focus is narrowed. For tandem conjugates incorporating PerCP (e.g., PerCP-Cy5.5), the excitation and emission properties of PerCP and the kinetics of energy exchange between the fluorochromes of the tandem dye may limit their effectiveness on high-speed and/or sorting flow cytometers. Therefore, for third-color flow-cytometric analysis using ≥ 25-mW laser power, we recommend PE-Cy5 (formerly BD Cy-Chrome™)-conjugated Streptavidin (Cat. No. 554062).

It is recommended that a 712/20-nm band-pass filter be used with stream-in-air instruments such as the BD FACStar™ and BD FACSVerse™ flow cytometry systems.

Suggested Companion Products

Catalog Number	Name	Size	Clone
554657	Stain Buffer (BSA)	500 mL	(none)
554656	Stain Buffer (FBS)	500 mL	(none)
554062	PE-Cy™5 Streptavidin	0.1 mg	(none)

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Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. 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.
3. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
4. 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.
5. 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 Cy5.5™. We recommend the use of cross-beam compensation during data acquisition or software compensation during data analysis.
6. Cy is a trademark of GE Healthcare.
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. Please refer to <http://regdocs.bd.com> to access safety data sheets (SDS).
9. Please refer to www.bdbiosciences.com/us/s/resources 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 ($[Ca^{2+}]_i$) fluxes among mouse lymph node B- and T-lymphocyte subsets. *Cytometry*. 1996; 23(3):205-217. (Biology)

Shapiro HM. *Practical Flow Cytometry, 3rd Edition*. New York: Wiley-Liss, Inc; 1995:280-281. (Biology)