

ReadiUse™ Preactivated APC-iFluor™ 750 Tandem

Catalog number: 2571
Unit size: 1 mg

| Component | Storage | Amount |
|--|--|-----------------|
| A: ReadiUse™ Preactivated APC-iFluor™ 750 Tandem | Refrigerated (2-8 °C), Minimize light exposure | 1 vial (1 mg) |
| B: Buccutite™ MTA | Freeze (< -15 °C), Minimize light exposure | 1 vial (100 µg) |
| C: Spin Desalting Column | | Not Included |

OVERVIEW

Allophycocyanin (APC) is a phycobiliprotein isolated from *Spirulina* sp., a blue-green alga. Like other phycobiliproteins, APC is fluorescent, with an extremely high absorptivity and a high quantum efficiency. Our APC-iFluor™ 750 Tandem is an excellent replacement for APC-Cy7® and APC-Alexa Fluor® 750 Tandem due to their similar spectral properties. Our APC-iFluor™ 750 Tandem is more photostable than APC-Cy7® Tandem. ReadiUse™ Preactivated APC-iFluor™ 750 Tandem is an activated APC protein, and can be easily conjugated to antibodies with much higher conjugation yield than the conventional APC.

SAMPLE EXPERIMENTAL PROTOCOL

Preparation of pre-activated antibody with Buccutite™ MTA

1. Reconstitute Buccutite™ MTA in DMSO at ~10 mg/mL.
Note Please store unused Buccutite™ MTA at -20 °C and could be used up to two freeze and thaw cycles.
2. Prepare target antibody (Ab) in pH = 8.5 - 9.0 buffer at concentration above 1 mg/mL.
3. Add Buccutite™ MTA to Ab solution at the ratio of 8 - 10 µg Buccutite™ MTA/100 µg Ab.
4. Mix well and react at room temperature for 60 minutes, rotating during the reaction.
5. Purify the reaction mixture with desalting column to remove unreacted Buccutite™ MTA and exchange buffer to PBS or buffer of your choice.
6. Collect the Buccutite™ MTA-activated Ab, and estimate the concentration by 70% yield of the original starting amount.

Conjugation with pre-activated APC-iFluor™ 750 Tandem

1. Reconstitute pre-activated APC-iFluor™ 750 Tandem in 100 µL ddH₂O to 10 mg/mL.
Note Reconstituted pre-activated APC-iFluor™ 750 Tandem could be stored at 4 °C for one month, kept from light.
2. Add APC-iFluor™ 750 Tandem directly to MTA-activated target Ab solution at the ratio of 130 µg APC-iFluor™ 750 Tandem/100 µg MTA-activated Ab.
3. Rotate the mixture for 60 minutes at room temperature.
4. The Ab/APC-iFluor™ 750 Tandem conjugates are now ready to use.

Note The antibody conjugate should be stored at > 0.5 mg/mL in the presence of a carrier protein (e.g., 0.1% bovine serum albumin) and 0.02% - 0.05% sodium azide. The Ab/APC-iFluor™ 750 Tandem conjugates solution could be stored at 4 °C for up to two months, and kept from light.

5. (Optional) Ab/APC-iFluor™ 750 Tandem conjugates could be further purified through size exclusion chromatography to get best performance.

EXAMPLE DATA ANALYSIS AND FIGURES

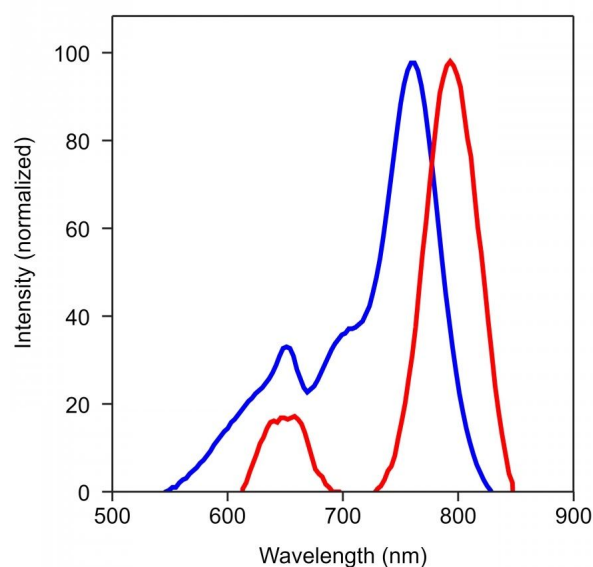


Figure 1.
Spectrum for APC-iFluor 750

DISCLAIMER

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