

APC [Allophycocyanin]

Cat #: B-CHM301

Size: 1mg/5mg

Storage: Store at 4°C protected from light.

Product Introduction

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 high quantum efficiency. It is a protein which can be easily linked to antibodies and other proteins by conventional protein cross-linking techniques without altering its spectral characteristics. Allophycocyanin is the least stable among the major phycobiliproteins, susceptible to dissociation at low concentrations including concentrations at which some assays are performed.

Product Properties

Form: APC is supplied with 60% ammonium sulfate, 100 mM sodium phosphate buffer pH 7.0 with 5 mM EDTA, and 2 mM sodium azide.

Spectral properties: Ex / Em = 652 nm / 662 ± 2 nm

Purity: A650/A280 >5, A650/A620 >1.5

Methods

1. Get 10 mg APC in ammonium sulfate solution, centrifuge (10,000 rpm, 10 min, 4°C), remove supernatant, and dissolve, re-suspend the precipitate in 1 ml PBS.
2. Continuously dilute by PBS.
3. To calculate the quality and concentration of APC, must measure the absorbance of stock solution dilution range 0.4-0.8 at 650 nm.

Concentration & purity

1. Determined protein concentration using extinction coefficient:

$$\text{Concentration (mg/ml)} = \frac{\text{Absorbance at } \lambda_{\text{max}} \text{ (OD 650 nm)}}{\text{Extinction coefficient (700000 M}^{-1} \text{ cm}^{-1}) \times \text{Pathlength (cm)}} \times \text{Molecular weight (105000 g/mol)} \times \text{Dilution factor}$$

2. A650/A620 is a general indicator of contamination with R-phycoerythrin (R-PE) as there is only residual absorbance by R-PE at 620 nm. Both the absorbance of R-Phycocyanin (R-PC) or Allophycocyanin (APC) is at 650 nm.

3. A650/A280 indicates purity of the preparation with respect to most forms of contaminating protein. Absorbance at 280 nm in these preparations is primarily due to aromatic amino acids, and thus is roughly proportional to the overall concentration of protein in solution, including Allophycocyanin (APC).

4. Example:

Dilution factor = 64 times

A650 = 0.617; A620 = 0.390; A280 = 0.117

APC concentration = $(0.617 / (700000 * 1)) * 105000 * 64 = 11.84 \text{ mg/ml}$

