**Reagents Provided**

**Streptavidin-Allophycocyanin:**
Supplied as 5 μg of SA-APC in 1 mL saline containing up to 0.5% BSA and 0.09% sodium azide.

**Intended Use**

Used as a secondary reagent in immunofluorescent assays using biotinylated primary labeling reagents. This product has been optimized for use with biotin-conjugated monoclonal antibodies.

**Background Information**

Streptavidin, a protein of 55,000 Daltons, is derived from *Streptomyces avidinii* and can bind 4 moles of biotin per mole of protein. The dissociation constant for biotin is approximately $10^{-15}$ M. The streptavidin-biotin complex is stable over a wide range of pH and temperatures. Streptavidin lacks carbohydrate residues present in the avidin molecule. This tends to reduce non-specific interactions with surface molecules and, therefore, streptavidin is preferred over avidin in many immunologic assays. Streptavidin can be covalently conjugated to fluorescent dyes and then used as a developer where the primary reagent was biotinylated.

**Reagent Preparation**

**Streptavidin-Allophycocyanin** (SA-APC) is provided in a ready-to-use liquid format containing up to 0.5% BSA and 0.09% sodium azide. Allophycocyanin has an absorption spectrum from 620 - 650 nm and has optimal emission at 660 - 670 nm. Store reagent at 2 - 8° C. **DO NOT FREEZE.** Reagent is stable for at least 6 months after purchase.

**Sample Staining**

Ten μL of Streptavidin-Allophycocyanin is added to a maximum of 1 x $10^6$ cells in 100 - 200 μL that have been optimally stained previously with a biotinylated primary reagent. The reaction is then allowed to proceed another 30 - 45 minutes at 2 - 8° C **in the dark.** The cell mixture is then washed twice with 10 mM PBS. The final cell pellet is resuspended in 200 - 300 μL of 10 mM PBS for flow cytometric analysis. SA-APC stained cells should be kept in the dark if storage is required prior to flow cytometric analysis.

**Note:** Contains sodium azide as a preservative - sodium azide may react with lead and copper plumbing to form explosive metal azides. Flush with large volumes of water during disposal.