

# Flow Cytometry Secondary Reagents

Donkey Anti-Sheep IgG (H+L)-PerCP

Catalog Number: F0128 Lot Number: ABSZ02

100 Tests

#### Intended Use

This reagent is designed for use as a secondary developing reagent in immunofluorescent assays, such as flow cytometry, where the primary antibody does not have a fluorescent reporter molecule, is of sheep origin, and is of the IgG class.

# **Background Information**

This polyclonal antibody preparation has been derived from donkey immunized with sheep IgG. Sheep IgG specific IgG is purified by sheep IgG (H+L) affinity chromatography and absorbed against human IgG, and human, mouse, bovine, and rabbit serum to eliminate cross-reactivity. The IgG fraction is then conjugated to PerCP for use in immunofluorescent-type assays.

# Reagents Provided

Supplied as 25  $\mu g$  of antibody in 1 mL saline containing up to 0.5% BSA and 0.1% sodium azide.

### Storage

Reagents are stable for **twelve months** from the date of receipt when stored in the dark at 2° - 8° C.

#### Reagent Preparation

Donkey anti-sheep IgG (H+L)-PerCP is produced as the PerCP derivative of donkey IgG from animals immunized with sheep IgG. The reagent is provided in a ready-to-use liquid format containing phosphate buffered saline with 0.5% BSA and 0.1% NaN<sub>3</sub> as a preservative. Store reagent at 2° - 8° C. DO NOT FREEZE. Dispose of liquids containing azide with caution and according to local regulations.

## Sample Staining

- 1. Cells of interest (up to 1 x 10<sup>6</sup> cells) are stained with a sheep IgG primary antibody according to the antibody manufacturer's instructions.
- 2. After the recommended incubation period, the cells are washed 3 times with a PBS buffer followed each time by centrifugation at 250 x g for 5 minutes.
- 3. The cell pellet is resuspended in up to 200  $\mu$ L of PBS and 10  $\mu$ L of donkey anti-sheep IgG (H+L)-PerCP is added to each reaction.
- The cells are incubated for 30 minutes at 2° 8° C in the dark. The cells are washed 3 times as indicated in step # 2.
- 5. The cell pellet is resuspended in 400  $\mu$ L of PBS for flow cytometric analysis.

**Warning:** 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.