Flow Cytometry Secondary Reagents

Goat F(ab')$_2$ anti-rat IgG-APC
Catalog Number: F0113
Lot Number: LXO05
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 rat origin, and is of IgG class.

Background Information

This polyclonal antibody preparation has been derived from goats immunized with rat IgG (heavy and light chains). The goat IgG is first purified by affinity chromatography and then adsorbed to eliminate mouse cross reactivity. The IgG fraction is further digested with pepsin to generate F(ab')$_2$ fragments which have a reduced ability to interact with Fc receptors expressed on a variety of cells. The antibody is then conjugated to allophycocyanin (APC) for use in immunofluorescent-type assays.

Reagents Provided

Supplied as 10 μg of antibody in 1 mL saline containing up to 0.5% BSA and 0.1% sodium azide.

Reagent Preparation

Goat anti-rat IgG-APC is produced as the APC derivative of F(ab')$_2$ fragments of goat IgG from animals immunized with rat IgG. The reagent is provided in a ready-to-use liquid format containing phosphate buffered saline with 0.5% BSA and 0.1% NaN_3 as preservatives. allophycocyanin conjugates require 620 - 650 nm laser excitation with a detector optimized to collect peak emissions at 660 - 670 nm. Store reagent at 2° - 8° C. **DO NOT FREEZE**. Dispose of azide-containing liquids with caution and according to local regulations.

Sample Staining

1. Cells of interest (1 - 5 x 10^5 cells) are stained with a rat 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 by centrifugation at 250 x g for 5 minutes.
3. The cell pellet is resuspended in up to 200 μL of PBS and 10 μL of goat anti rat IgG-APC is added to each reaction.
4. 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 μL of PBS for flow cytometric analysis.

N.B. 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.

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