Flow Cytometry Cell Surface Staining Protocol

Reagents required:

Flow Cytometry Staining Buffer (1x) (PF00012)

1x PBS

Flow cytometry antibodies

Experiment procedures:

1. Harvest cells and wash them twice with 1x PBS by centrifugation at 350-500 x g for 5 minutes each time, discard the supernatant.

2. Resuspend the cells in 1x Flow Cytometry Staining Buffer and incubate for 30 minutes at room temperature.

3. Aliquot cell samples to tubes or wells at a cell density of 1 x 10^6 cells in 100 µL of 1x Flow Cytometry Staining Buffer.

4. Add the recommended amount of primary antibody and incubate for 20-40 minutes at 4°C in the dark.

5. Wash the cells with 1x PBS by centrifugation at 350-500 x g for 5 minutes, discard the supernatant.
Note: If using fluorochrome-conjugated primary antibodies, skip to step 8.

6. Resuspend the cells in 100 µL of diluted fluorochrome-conjugated secondary antibody and incubate for 15-30 minutes at 4°C in the dark.

7. Wash the cells with 1x PBS by centrifugation at 350-500 x g for 5 minutes, discard the supernatant.

8. Resuspend the cells in 200-500 µL of 1x Flow Cytometry Staining Buffer and analyze on flow cytometer.