

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. Aliquot cell samples to tubes or wells at a cell density of 1×10^6 cells in 100 μ L of 1x Flow Cytometry Staining Buffer.
3. Add the recommended amount of primary antibody and incubate for 20-40 minutes at 4°C in the dark.
4. 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 7.
5. Resuspend the cells in 100 μ L of diluted fluorochrome-conjugated secondary antibody and incubate for 15-30 minutes at 4°C in the dark.
6. Wash the cells with 1x PBS by centrifugation at 350-500 x g for 5 minutes, discard the supernatant.
7. Resuspend the cells in 200-500 μ L of 1x Flow Cytometry Staining Buffer and analyze on flow cytometer.