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General Flow Cytometry Protocol

Protocol may need local adjustments

Direct Surface staining

1. Prepare the cells and adjust cell suspension to a concentration of $1 – 5 \times 10^6$ cells/ml in cell staining buffer (eg PBS with NaN_3 and BSA).
2. Ensure viability to be around 95%, not less than 90%.
3. Aliquot 100 µl of cell suspension into as many test tubes as required.
4. Add the fluorochrome conjugated antibody (see the specific datasheets for recommended dilution).
5. Mix well and incubate for 15-30 min at room temperature or 4°C. This step will require optimization and must be done in the dark.
6. Wash the cells 1-3 times with staining buffer and centrifuge each time at 400 g for 5 minutes and discard the resulting supernatant.
7. Resuspend in remaining buffer and adjust volume with staining buffer.
8. Keep the cells in the dark on ice or at 4°C until acquisition.