

Description of Methods for the Use of Monosan Antibodies in Indirect Flow Cytometry

Surface Staining:

1. Prepare cell suspension in PBS .
2. Add 10µl of antibody at various dilutions eg 1:10 upwards to 90µl of cell suspension. Incubate for 30 minutes at 4°C (an isotype control is recommended).
3. Wash twice in PBS.
4. Add Secondary FITC Ab (or equivalent) in PBS. Incubate for 30 minutes at 4°C.
5. Wash twice with PBS.
6. Resuspend in 300µl of PBS for analysis.

Intracellular Staining:

1. Prepare cell suspension in PBS (containing 0.5% (w/v) saponin).
2. Add 10µl of antibody at various dilutions eg 1:10 upwards to 90µl of cell suspension. Incubate for 30 minutes at 4°C (an isotype control is recommended).
3. Wash twice in PBS.
4. Add secondary FITC Ab (or equivalent) in PBS. Incubate for 30 minutes at 4°C.
5. Wash twice with PBS.
6. Resuspend in 300µl of PBS for analysis.