

Flow Cytometry Buffer Recipes

On this page you find basic recipes of buffers that are regularly used when performing flow cytometric analyses. Buffers may need optimization depending on the cell type and application. Always use buffers that are recommended by the manufacturer.

Flow Cytometry Staining Buffer (FACS Buffer)

This basic FACS Buffer is a buffered saline solution that can be used for immunofluorescent staining protocols, antibody and cell dilution steps, wash steps required for surface staining and flow cytometric analysis. The buffer contains sodium azide as preservative and animal serum proteins (FBS/BSA) to help minimizing non-specific binding of antibodies.

1x PBS

2-5 % (v/v) FBS (or BSA)

2 mM EDTA

2 mM NaN_3

Note: NaN_3 is added as a preservative. Use the buffer without NaN_3 if you want to do functional assays with bacterial cells.

Permeabilization Buffer

This Buffer can be used in flow cytometry, particularly for permeabilization for intracellular staining procedures.

1x PBS

0.1% (w/v) Saponin

2-5 % (v/v) FBS (or BSA)

2 mM EDTA

2 mM NaN_3

Important: Because saponin-mediated cell permeabilization is a reversible process, it is very important to keep the cells in the presence of saponin during intracellular cytokine staining. Perform permeabilization only on fixed cells.

Fixation Buffer

Fixation Buffer can be used in preparation of cells for intracellular staining procedures. The buffer can also be used to preserve light-scattering characteristics and fluorescence stainings of cells that have been stained by immunofluorescence for subsequent flow cytometric analysis.

1x PBS

4 % Paraformaldehyde (PFA)

Important: Fixation with PFA results in reduced forward scatter (size) and tandem dye fluorescent intensities.

Erythrocyte Lysis Buffer (10x)

This Erythrocyte lysis buffer can be used for quick removal of red blood cells from whole blood, tissues and tumor cells with minimal effects on leukocytes. Prepare a fresh 1:10 working solution in deionized water before use.

1.5 M NH₄Cl

100 mM NaHCO₃

10 mM EDTA

pH7.4

Important: Filter this buffer through a 0.2 μm filter (do not autoclave).

AnnexinV Binding buffer

10 mM HEPES pH7.4

140 mM NaCl

2.5 mM CaCl₂ (Calcium is crucial for binding of AnnexinV to PS)

Note: Incubate your cells with AnnexinV in binding buffer at room temperature (not at 4 °C) in the dark.