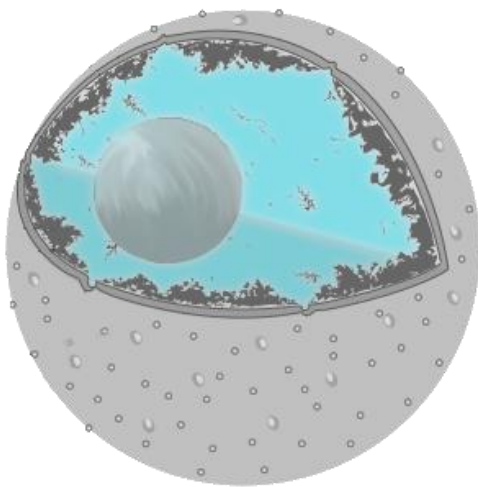


Intracellular Staining of Cells for ChipCytometry Quick Guide

Below are guidelines for the permeabilization of cells or tissues fixed on ZellSafe™ chips for staining of intracellular markers with the True Nuclear™ 10X Perm Buffer (Part: 73162) of the True-Nuclear Transcription Factor Buffer Set (CAT 424401) from BioLegend.



This protocol is for the intracellular staining of markers. For intranuclear staining, see our Intranuclear Staining of Cellular Markers protocol.

A. Preparing Solutions and Chip

1. Prepare a 1x working solution of the Perm Buffer by diluting the 10x Perm Buffer with **ZELLKRAFTWERK wash buffer**.
2. Rinse chip that contains fixed cells/tissues with at least 200 μL of **ZELLKRAFTWERK wash buffer** to remove air bubbles and ensure channel flow through.

B. Permeabilizing the Cells

1. Wash chip with 1000 μL of 1x Perm Buffer.
2. Dilute antibodies as titrated in appropriate volume of 1x Perm buffer and mix.
 - For ZellSafe™ Cell chips, use a total volume of 300 μL .
 - For ZellSafe™ Rare or Tissue chips, use a total volume of 600 μL for staining.
3. Incubate at room temp for 15 min.
4. Wash with 1000 μL of 1x Perm buffer.
5. Wash at least 5 min with ZELLKRAFTWERK wash buffer (~5 mL). Wait 2 min and wash again for 5 min with ZELLKRAFTWERK wash buffer (~5 mL) right before scanning.
6. Scan on ZellScanner ONE. Repeat washing if back diffusion of antibody is observed during the scan.