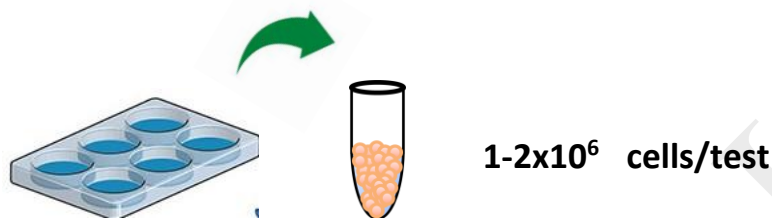


Cell Cycle Analysis by Propidium Iodide Staining



Harvest cells in the appropriate manner and wash in PBS⁻ (1500 rpm for 5min) at 4°C, discard the supernatant (Cells confluences should be about 60-70 %)

Add 1ml cold 70% ethanol drop wise to cell pellet, Vortex tube gently

Incubate cells at 4° C for 2 hours
[Cells can be stored at -20° C for up to several months]

Wash cells twice with 5-10 ml PBS⁻ (1500 rpm, for 5 min, 4°C), remove supernatant

Add 100 µl ribonuclease (100 µg/ml DNase free) to pelleted cells

Incubate cells for 20 minutes at 37oC

Add 200-400 µl of PI (final concentration 50 µg/ml) and incubate at RT for 5 min.
[PI is potentially mutagenic so wear glove]

Analyze by flow cytometry