Nucleofection™ Handling – Optimized Protocols

**Step 1**
Harvest cells of interest.

**Step 2**
Mix & combine.
- Nucleofector™ Solution with Supplement
- Cells
- DNA or siRNA

Transfer to a Lonza Certified Cuvette.

**Step 3**
Select Nucleofector™ Program. Insert cuvette. Press start button » X «.

**Step 4**
Rinse cuvette with culture medium.

**Step 5**
Transfer to culture dish. Expression can be detected as soon as 3 – 8 hours post Nucleofection™.

www.lonza.com/research
Tips to Get More Out of Your Nucleofection™

- Prepare multiwell plates with fresh medium and pre-equilibrate at 37 °C prior to experiment

- Use cells at low passage number and at the recommended confluence or density (logarithmic growth)

- Limit time of exposure to trypsin, carefully monitor cell detachment

- Count cells and use appropriate cell number according to optimized protocol; using fewer cells can result in increased mortality

- Use high quality DNA, purified with an endotoxin removing kit; please check the purity of each plasmid preparation by measurement of the A260 : A280 ratio

- Centrifuge at room temperature at the centrifugation speed (80 – 100 xg) and for the time specified in the protocol

- After centrifugation and addition of Nucleofector™ Solution, swirl solution /cell pellet for single cell suspension and avoid manipulating or pipetting pellet

- Following Nucleofection™, add ~ 500 µl of pre-warmed medium on top of cells in cuvette with disposable pipette
  - Gently bring pipette tip to the bottom of cuvette and collect cells
  - Gently seed the cell suspension into prepared multiwell-plate
  - Do NOT mix cells by repeated aspiration

For more information please check our Optimized Protocols at www.lonza.com/cell-database or contact our Scientific Support Team.