Gene Pulser® Electroprotocols

* We recommend adapting this protocol to use the Gene Pulser electroporation buffer (catalog #165-2676, 165-2677), which increases cell viability and transfection efficiency in mammalian cell lines.

Cell Type Mammalian, suspension

Species Used Human, red blood cells

Molecules Electroporated Dextrans, various proteins

Before the Pulse

Cell growth medium Harvested from whole blood

Growth phase at harvest Not given

Pre-pulse incubation Held on ice in Phosphate Buffered Saline

Wash solution Isotonic Phosphate Buffered Saline (PBS)

The Pulse

Electroporation Temperature 0 or 25 °C

Electroporation Medium* 20 mM Phosphate Buffered Saline

Cell Density 10 (6) to 10 (7) cells / ml

Volume of Cells 0.4 ml

DNA Concentration 10 (-5) to 10 (-4) M dextran

DNA Resuspension Buffer Protein in Phosphate Buffered Saline

Volume of DNA Not given

Instruments Used Gene Pulser® apparatus

Cuvette Gap 0.2 cm

Voltage 0 to 2.5 kV

Field Strength 0 to 12.5 kV/cm

Capacitor 25 μF

Resistor (Pulse Controller) not used. (SEE NOTE)

Time Constant 1 to 2 msec

After the Pulse

Outgrowth Medium Phosphate Buffered Saline

Outgrowth Temperature 0 °C

Length of Incubation 0 to 5 hr.

Selection Method or Assay Used Flow cytometry, detection of fluorescent molecules

Electroporation Efficiency up to 90% of cells exhibit uptake

Per Cent Survival 25 to 100%

Relevant Publications and/or Comments

Note: exponential values designated in parentheses.

**It is NOT RECOMMENDED to use high voltage with out the Pulse Controller.

PBS: 1x = 8g NaCl, 0.2g KCl, 0.2g KH_2PO_4, 1.15g Na_2HPO_4

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