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, adherent	**Molecules Electroporated**	DNA: CMV β-gal , 6 kB, supercoiled
Species Used	Monkey, COS-7, kidney, SV-40 transformed		

Before the Pulse

Cell growth medium	DMEM + 10% Fetal Bovine Serum (GIBCO/BRL, Sigma)	**Growth phase at harvest**	Log phase, 70 to 80% confluent
		Pre-pulse incubation	Room temperature
Wash solution	Trypsinize		

The Pulse

Electroporation Temperature	Room temperature	**Instruments Used**	Gene Pulser® apparatus & Capacitance Extender
Electroporation Medium*	DMEM + 10% Fetal Bovine Serum + 5mM BES, pH 7.2	**Cuvette Gap**	0.4 cm
Cell Density	2 x 10 (7) cells / ml	**Voltage**	0.170 kV
Volume of Cells	10 μg / 250 μl DNA	**Field Strength**	0.425 kV/cm
DNA Concentration	10 mM Tris, 1 mM EDTA, pH 8.0)	**Capacitor**	960 μF
DNA Resuspension Buffer	Not given	**Resistor**	(Pulse Controller) Ω none
Volume of DNA	Not given	**Time Constant**	45 msec

After the Pulse

Outgrowth Medium	Not given
Outgrowth Temperature	Room temperature
Length of Incubation	10 min., then pellet& resuspend in DMEM
Selection Method or Assay Used	Not given
Electroporation Efficiency	25%
Per Cent Survival	about 100 %

Relevant Publications and/or Comments

Note: exponential values designated in parentheses.

See reference: Ustav, M., and Stenlund, A. 1991. *EMBO J.* **10**(2):449-457.

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