



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	DNA: linearized, (pMSG-derivative)
Species Used	Rat, D202CC, hepatoma; Human, TCCSUP (epithelial-like) bladder carcinoma	Electroporated	

Before the Pulse

Cell growth medium	MEM, 10% Fetal Calf Serum, (+aminopterin, MPA, hypoxanthin, xanthine, thymidine) (GIBCO/BRL, Sigma)	Growth phase at harvest	Log phase
Wash solution	Phosphate Buffered Sucrose	Pre-pulse incubation	10 min. on ice, in Phosphate Buffered Sucrose

The Pulse

Electroporation Temperature	Room temperature	Instruments Used	Gene Pulser® apparatus
Electroporation Medium*	Phosphate Buffered Sucrose (272 mM sucrose, 7 mM potassium phosphate, pH 7.4, 1 mM MgCl ₂)	Cuvette Gap	0.4 cm
Cell Density	1 to 50 x 10 ⁽⁵⁾ cells / ml	Voltage	0.20 to 0.35 kV
Volume of Cells	0.4 ml	Field Strength	0.5 to 0.875 kV/cm
DNA Concentration	Not given	Capacitor	25 μF
DNA Resuspension Buffer	Not given	Resistor	(Pulse Controller) Ω none
Volume of DNA	5 to 20 μg / ml	Time Constant	7 to 12 msec

After the Pulse

Outgrowth Medium	As above	Relevant Publications and/or Comments	
Outgrowth Temperature	Not given	Note:	exponential values designated in parentheses.
Length of Incubation	Stable transfectants		
Selection Method or Assay Used	MPA		
Electroporation Efficiency	1 to 10 x 10 ⁽⁶⁾ cells		
Per Cent Survival	Not given		

Name of Submitter
Institution Address

Telephone Number
Fax Number
Date Submitted 11/13/90
Survey Number 153
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