



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	Molecules Electroporated	DNA: Antibody genes with selectable markers.
Species Used	Hybridomas		

Before the Pulse

Cell growth medium	DMEM + 15% Hourse Serum + L- glutamine (GIBCO/BRL, Sigma)	Growth phase at harvest	Mid- log
Wash solution	Cold Phosphate Buffered Saline	Pre-pulse incubation	10 minutes on ice.

The Pulse

Electroporation Temperature	4°C	Instruments Used	Gene Pulser® apparatus & Capacitance Extender, and Pulse Controller
Electroporation Medium*	Phosphate Buffered Saline	Cuvette Gap	0.4 cm
Cell Density	10 (7) cells / ml	Voltage	0.160 kV
Volume of Cells	0.8 ml	Field Strength	0.4 kV/cm
DNA Concentration	20 µg / ml	Capacitor	960 µF
DNA Resuspension Buffer	Phosphate Buffered Saline	Resistor	(Pulse Controller) Ω none
Volume of DNA	Not given	Time Constant	15 msec

After the Pulse

Outgrowth Medium	DMEM + 15% Fetal Calf Serum + Selector	Relevant Publications and/or Comments
Outgrowth Temperature	37 °C	Note: exponential values designated in parentheses.
Length of Incubation	> 1 week	<i>Immunol.</i> , Jan 15, 1991.
Selection Method or Assay Used	G418 or mycophenolic acid	
Electroporation Efficiency	0.5 to 1 transfectants / µg	
Per Cent Survival	Not given	

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