



# 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.

<b>Cell Type</b>	Mammalian, adherent	<b>Molecules Electroporated</b>	DNA: 11 to 12 kB expression vector, Rep 4.
<b>Species Used</b>	Hamster, CHO -K1, ovary, (requires proline)		

## Before the Pulse

<b>Cell growth medium</b>	F12 + 10% serum (GIBCO/BRL, Sigma)	<b>Growth phase at harvest</b>	Log phase
<b>Wash solution</b>	Phosphate Buffered Saline	<b>Pre-pulse incubation</b>	10 min., on ice.

## The Pulse

<b>Electroporation Temperature</b>	Room temperature (pulse), then ice	<b>Instruments Used</b>	Gene Pulser® apparatus & Capacitance Extender
<b>Electroporation Medium*</b>	Phosphate Buffered Saline	<b>Cuvette Gap</b>	0.4 cm
<b>Cell Density</b>	2 x 10 <sup>(5)</sup> cells / pulse	<b>Voltage</b>	0.20 to 0.40 kV
<b>Volume of Cells</b>	0.8 ml	<b>Field Strength</b>	0.5 to 1.0 kV/cm
<b>DNA Concentration</b>	0.5 µg / pulse	<b>Capacitor</b>	500 & 960 µF
<b>DNA Resuspension Buffer</b>	Not given	<b>Resistor</b>	(Pulse Controller) Ω none
<b>Volume of DNA</b>	Not given	<b>Time Constant</b>	18 to 22 msec

## After the Pulse

<b>Outgrowth Medium</b>	F12 + serum + hygromycin or neomycin	<b>Relevant Publications and/or Comments</b>	<b>Note:</b> exponential values designated in parentheses.
<b>Outgrowth Temperature</b>	37 °C		
<b>Length of Incubation</b>	14 to 21 days		
<b>Selection Method or Assay Used</b>	Hygromycin or neomycin selection		
<b>Electroporation Efficiency</b>	Not done as yet - just beginning		
<b>Per Cent Survival</b>	>10 %		

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