



## Gene Pulser® Electroprotocols

**Cell Type** Bacterial, gram positive  
**Species Used** *Lactobacillus* sp., strain 100-33

**Molecules Electroported** DNA: plasmid pLAR33, 18kB, supercoiled.

### Before the Pulse

<b>Cell growth medium</b>	MRS broth (DIFCO)	<b>Growth phase at harvest</b>	O.D. (600) = 0.6 to 1.0 (log)
<b>Wash solution</b>	2% glycerol in 10 mM NaPO <sub>4</sub> (pH 6.0)	<b>Pre-pulse incubation</b>	none

### The Pulse

<b>Electroporation Temperature</b>	0 °C	<b>Instruments Used</b>	Gene Pulser® apparatus Pulse Controller
<b>Electroporation Medium</b>	2% glycerol in 10 mM NaPO <sub>4</sub> (pH 6.0)	<b>Cuvette Gap</b>	0.2 cm
<b>Cell Density</b>	100x growth when harvested	<b>Voltage</b>	2.5 kV
<b>Volume of Cells</b>	50 µl	<b>Field Strength</b>	12.5 kV/cm
<b>DNA Concentration</b>	1 µg	<b>Capacitor</b>	25 µF
<b>DNA Resuspension Buffer</b>	Not given	<b>Resistor</b>	(Pulse Controller) 200 Ω
<b>Volume of DNA</b>	1 to 5 µl	<b>Time Constant</b>	1 to 3 msec

### After the Pulse

<b>Outgrowth Medium</b>	MRS broth
<b>Outgrowth Temperature</b>	37 °C
<b>Length of Incubation</b>	1 to 3 hrs, depends on selection.
<b>Selection Method or Assay Used</b>	Antibiotic resistance (erythromycin)
<b>Electroporation Efficiency</b>	2 X 10 <sup>(5)</sup> transformants / µg DNA
<b>Per Cent Survival</b>	1%

#### Relevant Publications and/or Comments

**Note:** exponential values designated in parentheses.

We have found that the electroporation of plasmids into our intestinal *Lactobacilli* isolates is highly strain-dependent. Our efficiencies range from 0 to 2 X 10<sup>(5)</sup>.

**Name of Submitter**  
**Institution Address**

**Telephone Number**

**Fax Number**

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**Survey Number** 073

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