



## Gene Pulser® Electroprotocols

**Cell Type** Bacterial, gram negative  
**Species Used** *E. coli*, DH10B

**Molecules Electroported** DNA: pUC 18

### Before the Pulse

**Cell growth medium** 2x YT broth

**Growth phase at harvest** O.D. (600) = 0.8

**Pre-pulse incubation** 0° C

**Wash solution** Distilled water / 10% glycerol

### The Pulse

**Electroporation Temperature** 0° C

**Instruments Used** Gene Pulser® apparatus and Pulse Controller

**Electroporation Medium** 10% glycerol

**Cell Density** 1 x 10<sup>(9)</sup> cells / ml

**Cuvette Gap** 0.1 cm

**Volume of Cells** 50 µl

**Voltage** 2.5 kV

**DNA Concentration** TE buffer

**Field Strength** 25 kV/cm

**DNA Resuspension Buffer** TE buffer

**Capacitor** 25 µF

**Volume of DNA** 1 µl

**Resistor** 200 Ω (Pulse Controller)

**Time Constant** 4 to 5 msec

### After the Pulse

**Outgrowth Medium** SOC

#### Relevant Publications and/or Comments

**Note:** exponential values designated in parentheses.  
**SOC:** 2% Bacto tryptone, 0.5% Bacto yeast extract, 10mM NaCl, 2.5mM KCl, 10 mM MgCl<sub>2</sub>, 10 mM MgSO<sub>4</sub>, 20 mM glucose.

**Outgrowth Temperature** 37 °C

**Length of Incubation** 1 hour

**Selection Method or Assay Used** Ampicillin resistance

**2xYT:** 1.6% Bacto tryptone, 1.0% Bacto yeast extract, 0.5% NaCl.

**Electroporation Efficiency** 5 x 10<sup>(9)</sup> transformants / µg DNA

**Per Cent Survival** not given

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

**Telephone Number**

**Fax Number**

**Date Submitted** 4/22/91

**Survey Number** 027

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