



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⁽⁹⁾ 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 MgCl₂, 10 mM MgSO₄, 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⁽⁹⁾ 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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